

## Updates on DNA Hypomethylation in the Treatment of MDS

---

A Review of Selected Abstracts From the 47th Annual  
Meeting of the American Society of Hematology  
December 10–13, 2005  
Atlanta, Georgia

With commentary by:

**Guillermo Garcia-Manero, MD**

The University of Texas M. D. Anderson Cancer Center



# EDITORIAL ADVISORY BOARD

## Editor-in-Chief

**Bruce D. Cheson, MD**  
Georgetown University  
Medical School  
Lombardi Cancer Center

## Section Editors

### Oncology

**James L. Abbruzzese, MD**  
The University of Texas  
M. D. Anderson Cancer Center

**Mark J. Ratain, MD**  
The University of Chicago

### Hematologic Malignancies

**Clara D. Bloomfield, MD**  
Ohio State University  
Comprehensive Cancer Center

### Hematology

**Craig M. Kessler, MD**  
Georgetown University  
Medical School  
Lombardi Cancer Center

**David B. Agus, MD**  
Cedars-Sinai Medical Center  
University of California,  
Los Angeles

**Kenneth C. Anderson, MD**  
Dana-Farber Cancer Institute

**Frederick R. Appelbaum, MD**  
Fred Hutchinson Cancer  
Research Center  
University of Washington

**Bart Barlogie, MD, PhD**  
University of Arkansas  
for Medical Sciences

**James R. Berenson, MD**  
Institute for Myeloma  
& Bone Cancer Research  
West Hollywood, CA

**Howard A. Burris III, MD**  
The Sarah Cannon Cancer Center

**James Bussel, MD**  
Weill Medical College  
of Cornell University

**John Byrd, MD**  
Ohio State University  
Comprehensive Cancer Center

**Mitchell S. Cairo, MD**  
Columbia University

**George P. Canellos, MD**  
Dana-Farber Cancer Institute  
Harvard Medical School

**Michael A. Carducci, MD**  
The Sidney Kimmel  
Comprehensive Cancer Center  
at Johns Hopkins

**Richard E. Champlin, MD**  
The University of Texas  
M. D. Anderson Cancer Center

**Edward Chu, MD**  
Yale Cancer Center,  
Yale University

**Bertrand Coiffier, MD**  
Hospices Civils de Lyon  
Université Claude Bernard, Lyon

**Jeffrey Crawford, MD**  
Duke University Medical Center

**David C. Dale, MD**  
University of Washington

**George D. Demetri, MD**  
Dana-Farber Cancer Institute  
Harvard Medical School

**Brian Durie, MD**  
Cedars-Sinai Comprehensive  
Cancer Center  
International Myeloma Foundation

**Elihu H. Estey, MD**  
The University of Texas  
M. D. Anderson Cancer Center

**David S. Ettinger, MD**  
The Sidney Kimmel  
Comprehensive Cancer Center  
at Johns Hopkins

**James Feusner, MD**  
Children's Hospital Oakland

**Stephen J. Forman, MD**  
City of Hope National  
Medical Center

**Richard M. Goldberg, MD**  
University of North Carolina  
at Chapel Hill

**Michael S. Gordon, MD**  
Arizona Cancer Center,  
University of Arizona

**F. Anthony Greco, MD**  
The Sarah Cannon Cancer Center

**Mark Green, MD**  
Medical University of South Carolina  
Hollings Cancer Center

**Stephanie A. Gregory, MD**  
Rush Medical College  
Rush-Presbyterian-  
St. Luke's Medical Center  
Rush University Medical Center

**Stuart A. Grossman**  
The Sidney Kimmel  
Comprehensive Cancer Center  
at Johns Hopkins

**John D. Hainsworth, MD**  
The Sarah Cannon Cancer Center

**Roy S. Herbst, MD, PhD**  
The University of Texas  
M. D. Anderson Cancer Center

**Hagop M. Kantarjian, MD**  
The University of Texas  
M. D. Anderson Cancer Center

**Lawrence D. Kaplan, MD**  
University of California,  
San Francisco

**Neil E. Kay, MD**  
Mayo Clinic

**Hedy Lee Kindler, MD**  
University of Chicago

**John M. Kirkwood, MD**  
University of Pittsburgh  
Cancer Institute

**John G. Kuhn, PharmD,  
FCCP, BCOP**  
University of Texas  
College of Pharmacy

**Corey J. Langer, MD**  
Fox Chase Cancer Center  
Temple University  
Medical School

**Richard A. Larson, MD**  
University of Chicago

**John P. Leonard, MD**  
Weill Medical College  
of Cornell University  
New York Presbyterian Hospital

**Christopher J. Logothetis, MD**  
The University of Texas  
M. D. Anderson Cancer Center

**John S. Macdonald, MD**  
St. Vincent's Comprehensive  
Cancer Center

**Maurie Markman, MD**  
The University of Texas M. D. Anderson  
Cancer Center

**Robert J. Mayer, MD**  
Dana-Farber Cancer Institute

**Ruth O'Regan, MD**  
Winship Cancer Institute  
Emory University

**Thomas L. Ortel, MD, PhD**  
Duke University Medical Center

**Anders Österborg, MD, PhD**  
Karolinska Hospital

**Marshall R. Posner, MD**  
Dana-Farber Cancer Institute  
Harvard Medical School

**Eric K. Rowinsky, MD**  
University of Texas  
Health Science Center

**Leonard Saltz, MD**  
Memorial Sloan-Kettering  
Cancer Center

**Charles A. Schiffer, MD**  
Karmanos Cancer Institute  
Wayne State University  
School of Medicine

**Richard L. Schilsky, MD**  
University of Chicago

**George W. Sledge Jr., MD**  
Indiana University Cancer Center

**Mark A. Socinski, MD**  
Lineberger Comprehensive  
Cancer Center  
University of North Carolina

**Margaret Tempero, MD**  
University of California,  
San Francisco Comprehensive  
Cancer Center

**Joel E. Tepper, MD**  
University of North Carolina  
School of Medicine

**Alan P. Venook, MD**  
University of California,  
San Francisco Comprehensive  
Cancer Center

**Everett E. Vokes, MD**  
University of Chicago

**Peter H. Wiernik, MD**  
New York Medical College  
Our Lady of Mercy Cancer Center

**John R. Wingard, MD**  
University of Florida  
College of Medicine

Included in Index Medicus/PubMed/Medline

## Disclaimer

Funding for this Abstract Review has been provided through an educational grant from Pharmion Corporation. Sponsorship of this monograph does not imply the sponsor's agreement with the views expressed herein. Every effort has been made to ensure that drug usage and other information are presented accurately; however, the ultimate responsibility rests with the prescribing physician. Millennium Medical Publishing, Inc., the sponsors, and the participants shall not be held responsible for errors or for any consequences arising from the use of information contained herein. Readers are strongly urged to consult any relevant primary literature. No claims or endorsements are made for any drug or compound at present under clinical investigation.

## Table of Contents

Introduction	4
Summaries of Abstracts	6
Commentary by Guillermo Garcia-Manero, MD	14

# Updates on DNA Hypomethylation in the Treatment of MDS

A Review of Selected Abstracts From the  
47th Annual Meeting of the American Society of Hematology  
Atlanta, Georgia  
December 10–13, 2005

## Introduction

Myelodysplastic syndrome (MDS) is a group of clonal hematopoietic stem cell disorders caused by ineffective hematopoiesis. MDS is characterized by cytopenias in the peripheral blood in all three lineages: granulocyte, erythrocyte, and megakaryocyte. It is a clonal disease, arising from a hematopoietic progenitor. A number of chromosomal abnormalities are associated with MDS development, including deletions of chromosomes 7q, 5q, 20q, 6q, 11q, and 13q. Deletions of or within chromosomes 7q and 5q are the most common chromosomal aberrations. Abnormalities within chromosome 7 or multiple abnormalities are associated with a poor prognosis in MDS.<sup>1</sup>

Approximately 15,000 to 20,000 new cases of MDS are diagnosed each year. This disease is most common in older persons, with an incidence of 1 in 500 in those over 60 years of age.<sup>2,3</sup> The incidence of MDS has been steadily rising since 1980.<sup>4</sup> The reasons for this are unknown but could be related to changing classification systems, better diagnosis, an aging population, or a true rise in disease occurrence. MDS progresses to acute myeloid leukemia (AML) in 35–40% of cases.<sup>4</sup> Most patients die from bleeding or infection associated with bone marrow failure.

Until recently, supportive care was the primary therapy for MDS. This approach consists primarily of blood transfusions and antibiotics and its goal is to reduce morbidity from ineffective hematopoiesis and improve quality of life. This remains the treatment of choice in a large percentage of the mostly elderly MDS population. Supportive care has been enhanced by treatment with various growth factors aimed at stimulating blood cell production. Erythropoietin is one of the most commonly used growth factors in MDS therapy. It has efficacy in patients with low serum erythropoietin levels and minimal red blood cell (RBC) transfusion need, but other MDS patients have had response rates of 10% or less.<sup>5,6</sup> Granulocyte colony-stimulating factor (G-CSF) and granulocyte-macrophage colony-stimulating factor have been used to improve neutrophil counts. Although

erythropoietin and G-CSF may not be effective in the treatment of MDS individually, there are data suggesting that combining these therapies may have some benefit for patients with low-risk MDS.<sup>7</sup> Thrombopoietic growth factors are currently being investigated to counteract thrombocytopenia, one of the leading causes of mortality in MDS.<sup>8</sup> The pleiotropic growth factor interleukin-11 is also being studied for this purpose.<sup>5</sup>

More recently, several different approaches have been taken to move beyond supportive care with the goal of decreasing progression to leukemia and increasing overall survival. Allogeneic stem cell transplantation may be the most effective of these approaches, with cure rates between 20% and 40%; however, transplant-related mortality, graft-versus-host disease, and the fact that many patients are not eligible for transplantation (due to age and/or lack of donor availability) mean that the actual transplantation-associated cure rates are much lower. A 1998 study demonstrated 5-year disease-free survival rates of 60%, 36%, and 28% after bone marrow transplant (BMT) in low- and intermediate-1-, intermediate-2-, and high-risk groups, respectively.<sup>9</sup> However, treatment-related mortality rates as high as 43% have been seen in similar populations.<sup>10</sup> These data led to recommendation of BMT for patients with intermediate-1-, intermediate-2-, and high-risk disease only. Even within these risk groups, however, the advanced age of most MDS patients makes BMT a possibility for only a small fraction of individuals. Immunosuppressive drugs may be an alternative for patients who experience T cell-mediated suppression of hematopoiesis.<sup>11</sup> Antithymocyte globulin and cyclosporin A have been used with success in low-risk refractory anemia (RA) patients.<sup>12,13</sup> Other high- and low-dose chemotherapy regimens have been used with varying success in MDS. In general, toxicity and relapse rates have been high and duration of response short.<sup>7</sup> Several more targeted and much less toxic new agents with unique mechanisms of action are now being investigated for the treatment of MDS; foremost among these are agents which target the bone marrow microenvironment and those which thwart DNA methylation.

Thalidomide (Thalomid, Celgene) and its analog lenalidomide (Revlimid, Celgene) have been shown to inhibit both angiogenesis and tumor necrosis factor alpha. In a phase II trial, thalidomide was associated with an almost 20% rate of either (1) RBC transfusion independence or (2) a greater than 50% decrease in required transfusions.<sup>14</sup> Thalidomide was also linked to a number of serious side effects, including dose-limiting neuropathy. The thalidomide derivative lenalidomide may provide many of the same benefits of thalidomide possibly with less neurotoxicity. Lenalidomide has demonstrated activity in the subset of patients with 5q- syndrome.<sup>15</sup> In fact, the Food and Drug Administration (FDA) recently approved lenalidomide for low-risk 5q- MDS, making it the second drug to be indicated specifically for MDS.

The first drug approved by the FDA for the treatment of MDS was azacitidine (Vidaza, Pharmion), which is indicated for the treatment of all MDS patients of all French-American-British (FAB) classification subtypes. This pyrimidine nucleoside analog of cytidine functions as a DNA methyltransferase inhibitor. Azacitidine covalently binds methyltransferase, targeting it for degradation. In MDS, the promoter region of the cyclin-dependent kinase inhibitor *p15INK4B* is methylated in 46–100% of cases.<sup>16</sup> This gene is upregulated by transforming growth factor  $\beta$  and maintains hematopoietic cells in quiescence. Methylation of the *p15INK4B* promoter silences the gene and thus enables hyperproliferation of hematopoietic cells. The frequency of *p15INK4B* methylation is associated with transformation to AML and increased blast counts.<sup>17</sup> By inhibiting this methylation, azacitidine may hinder leukemic transformation of myelodysplastic cells.

Decitabine (Dacogen, MGI Pharma) is a DNA methyltransferase inhibitor currently under review by the FDA for the treatment of MDS. An analog of deoxycytidine, decitabine is a prodrug requiring metabolic activation by deoxycytidine kinase.<sup>18</sup> The antileukemic activity of this agent is related to its reversal of epigenetic silencing of genes that suppress leukemogenesis. This silencing occurs through aberrant DNA methylation.

The following are recent studies of the DNA methyltransferase inhibitors that were presented at the 47th Annual Meeting of the American Society of Hematology, held December 10–13, 2005, in Atlanta, Georgia. They address many of the most topical issues concerning DNA methyltransferase inhibitor use in MDS, including interpreting data in light of changing disease classification, potential adverse effects, alternative dosing schedules, efficacy in high-risk patients, and the molecular mechanisms underlying treatment response.

## References

1. Dansey R. Myelodysplasia. *Curr Opin Oncol.* 2000;12:13-21.
2. Hofmann WK, Ottmann OG, Ganser A, Hoelzer D. Myelodysplastic syndromes: clinical features. *Semin Hematol.* 1996;33:177-185.
3. Hofmann WK, Koeffler HP. Important features of myelodysplastic syndrome. *Int J Hematol.* 2002;76(suppl 2):222-227.
4. Aul C, Germing U, Gattermann N, Minning H. Increasing incidence of myelodysplastic syndromes: real or fictitious? *Leuk Res.* 1998;22:93-100.
5. Seipelt G, Ottmann OG, Hoelzer D. Cytokine therapy for myelodysplastic syndrome. *Curr Opin Hematol.* 2000;7:156-160.
6. Gordon MS. Advances in supportive care of myelodysplastic syndromes. *Semin Hematol.* 1999;36(4 suppl 6):21-24.
7. Jadersten M, Montgomery SM, Dybedal I, Porwit-MacDonald A, Hellstrom-Lindberg E. Long-term outcome of treatment of anemia in MDS with erythropoietin and G-CSF. *Blood.* 2005;106:803-811.
8. Ogata K, Tamura H. Thrombopoietin and myelodysplastic syndromes. *Int J Hematol.* 2000;72:173-177.
9. Appelbaum FR, Anderson J. Allogeneic bone marrow transplantation for myelodysplastic syndrome: outcomes analysis according to IPSS score. *Leukemia.* 1998;12(suppl 1):S25-29.
10. Anderson JE, Appelbaum FR, Fisher LD, et al. Allogeneic bone marrow transplantation for 93 patients with myelodysplastic syndrome. *Blood.* 1993;82:677-681.
11. Kondo Y, Mollidrem JJ. Immune-induced cytopenia: bone marrow failure syndrome. *Curr Hematol Rep.* 2004;3:178-183.
12. Sauntharajah Y, Nakamura R, Nam JM, et al. HLA-DR15 (DR2) is overrepresented in myelodysplastic syndrome and aplastic anemia and predicts a response to immunosuppression in myelodysplastic syndrome. *Blood.* 2002;100:1570-1574.
13. Shimamoto T, Tohyama K, Okamoto T, et al. Cyclosporin A therapy for patients with myelodysplastic syndrome: multicenter pilot studies in Japan. *Leuk Res.* 2003;27:783-788.
14. Bowen D, Culligan D, Jowitt S, et al. Guidelines for the diagnosis and therapy of adult myelodysplastic syndromes. *Br J Haematol.* 2003;120:187-200.
15. List A, Kurtin S, Roe DJ, et al. Efficacy of lenalidomide in myelodysplastic syndromes. *N Engl J Med.* 2005;352:549-557.
16. Aggerholm A, Guldberg P, Hokland M, Hokland P. Extensive intra- and inter-individual heterogeneity of p15INK4B methylation in acute myeloid leukemia. *Cancer Res.* 1999;59:436-441.
17. Quesnel B, Guillerme G, Verecque R, et al. Methylation of the p15(INK4b) gene in myelodysplastic syndromes is frequent and acquired during disease progression. *Blood.* 1998;91:2985-2990.
18. Momparler RL. Pharmacology of 5-aza-2'-deoxycytidine (decitabine). *Semin Hematol.* 2005;42(3 suppl 2):S9-S16.

# Updates on DNA Hypomethylation in the Treatment of MDS

## Abstract Summaries

### 2517 Azacitidine Treatment Response Assessed Using Three Alternative Dosing Schedules in Patients With Myelodysplastic Syndromes<sup>1</sup>

R. Lyons, T. Cosgriff, S. Modi, L. Lintz, C. L. Beach, and J. T. Backstrom

The current standard dosing schedule for azacitidine is 75 mg/m<sup>2</sup> subcutaneously (SC) daily for 7 days, repeated every 28 days for up to 18 cycles.<sup>2</sup> This regimen would be more convenient for patients and clinicians if weekend injections were eliminated. The current study is a randomized, open-label, multicenter, phase II trial in which three different dosing schedules for azacitidine are being evaluated. Sixty-one patients have been randomized thus far to the three treatment arms: (i) AZA 5-2-2 (n=25), azacitidine 75 mg/m<sup>2</sup> daily for 5 days, no treatment for 2 days, azacitidine 75 mg/m<sup>2</sup> for 2 days; (ii) AZA 5-2-5 (n=19), azacitidine 50 mg/m<sup>2</sup> daily for 5 days, no treatment for 2 days, azacitidine 50 mg/m<sup>2</sup>/day for 5 days; and (iii) AZA 5 (n=17), azacitidine 75 mg/m<sup>2</sup>/day for 5 days. The primary response criteria for the study are hematologic improvement, using International Working Group (IWG) criteria,<sup>3</sup> and transfusion independence, defined as a transfusion-free period of at least 56 days.

The primary eligibility criteria include diagnosis of one of the five FAB subtypes of MDS (RA, RA with ringed

sideroblasts [RARS], RA with excess blasts [RAEB], RAEB in transformation [RAEB-t], and chronic myelomonocytic leukemia); Eastern Cooperative Oncology Group (ECOG) performance status of 0–3; serum bilirubin and creatinine levels no more than 1.5 times the upper limit of normal (ULN); and serum aspartate aminotransferase and alanine aminotransferase no more than twice the ULN. The main exclusion criteria are secondary MDS or history of other cancer and folate or vitamin B<sub>12</sub> deficiency. Patients are stratified by FAB risk criteria. Low-risk patients are considered to be RA or RARS. In order to be evaluated, patients must complete at least two cycles of treatment. Thus far, 35 patients are evaluable, 19 on the AZA 5-2-2 arm, 9 on the AZA 5-2-5 arm, and 7 on the AZA 5 arm.

Although the study is still accruing patients and the current number of evaluable patients is small, particularly on the AZA 5-2-5 and AZA 5 arms, all three dosing schedules have shown efficacy. Hematologic improvement from baseline has been seen in 63% of patients on the 5-2-2 arm (95% confidence interval [CI], 38–84%), 44% of patients on the 5-2-5 arm (95% CI, 14–79%), and 43% of patients on the AZA 5 arm (95% CI, 10–82%) (Table 1). On the AZA 5-2-2 arm, patients have shown major hematologic improvement in erythroid, platelet, and neutrophil measures, with response rates of 47%, 26%, and 11%, respectively. On the AZA 5-2-5 arm, 33% of patients have had a major erythroid improvement and platelet and neutrophil improvements have been seen in 11% of patients each. On the AZA 5 arm, 29% and 14% of patients have had major erythroid and neutrophil improvements, respectively, but no patients have yet had major neutrophil improvements.

All three treatment arms have also had a positive impact on transfusion-independence. A majority of patients on all arms who were transfusion-dependent at baseline no longer require RBC transfusions. Independence from RBC transfusion has been seen in 58% of previously dependent patients on the AZA 5-2-2 arm (95% CI, 28–85%) and 67% of patients on both the 5-2-5 and AZA 5 arms (95% CI, 9–99%). Transfusion indepen-

**Table 1.** Hematologic Improvement in Evaluable Patients

Type of Hematologic Improvement	AZA 5-2-2 N=19		AZA 5-2-5 N=9		AZA 5 N=7	
	n (%)	95% CI	n (%)	95% CI	n (%)	95% CI
Erythroid Major	9 (47)	24, 71	3 (33)	8, 70	2 (29)	4, 71
Platelet Major	5 (26)	9, 51	1 (11)	0, 48	1 (14)	0, 58
Neutrophil Major	2 (11)	1, 33	1 (11)	0, 48	0 (0)	0, 41
Major + Minor*	12 (63)	38, 84	4 (44)	14, 79	3 (43)	10, 82

\* Patients with hematologic improvement (major or minor) were counted only once in the overall response.

CI = confidence interval.

dence has been comparable in FAB low- and high-risk patients and patients who were transfusion-dependent or -independent at baseline.

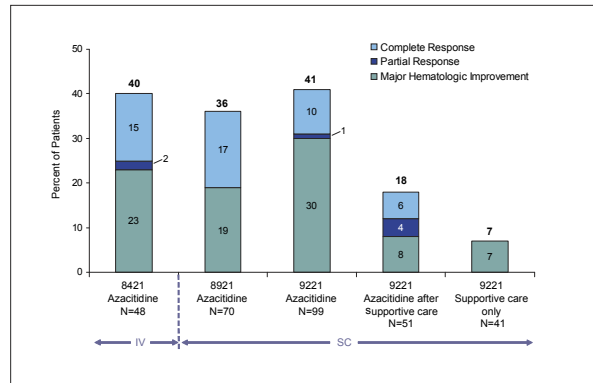
The three alternative dosing schedules proposed in this study have thus far been shown to be efficacious and well tolerated. They confer hematologic improvements and allow for transfusion independence in both FAB low- and high-risk patients. The most common adverse events are injection site reactions and hematologic and gastrointestinal toxicities. In this interim analysis, all three regimens have been shown to be comparable in safety and efficacy to the standard 7-day azacitidine schedule.<sup>2</sup>

## 2526 Response Rates Using International Working Group Criteria in Patients With Myelodysplastic Syndromes Treated With Azacitidine<sup>4</sup>

L. Silverman, D. McKenzie, B. Peterson, C. DeCastro, J. Ellerton, K. Knapp, C. L. Beach, R. Larson; the Cancer and Leukemia Group B

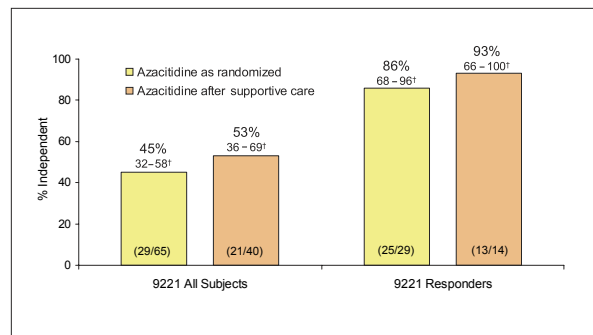
The Cancer and Leukemia Group B (CALGB) studies of azacitidine for the treatment of MDS (CALGB 8421, 8921, and 9221) were published before the release of new response criteria by the IWG. The current study reanalyzed the results of these trials using the revised response criteria. The primary differences between the new IWG response criteria and previously established measures of response were: doubling of the response duration requirement (from at least 4 to at least 8 weeks), and eliminating gender-specific hemoglobin targets.

Treatment response and transfusion independence were analyzed in 309 patients, 268 of whom were treated with azacitidine and 41 with only supportive care. Overall response rates (complete response + partial response + hematologic improvement) with azacitidine were observed in 44% of patients in study 8421, 40% in study 8921, and 47% in study 9221. These can be compared with a response rate of 17% for the supportive care–only arm of study 9221 and 36% for the azacitidine-after-supportive-care (crossover) arm. Trends were similar when examining the percentage of patients with complete response, partial response, and hematologic improvement in each study (Figure 1). Complete responses were observed in 15%, 17%, and 10% of patients on the azacitidine arms of studies 8421, 8921, and 9221, respectively. No complete



**Figure 1.** Response rates (complete response + partial response + major hematologic improvement) using International Working Group response criteria by study.

IV= intravenous; SC = subcutaneous.



**Figure 2.** Red blood cell transfusion independence in patients dependent at baseline according to International Working Group criteria. Numbers in parentheses are n/N.

† 95% confidence interval.

responses were observed on the supportive care arm of study 9221.

Azacitidine elicited impressive response durations, and response was achieved relatively quickly in patients in the three CALGB studies. The median duration of response in patients on azacitidine was 366 days (range, 56–4641), 379 days for complete response (range, 92–4412). Response was achieved after a median of three cycles of therapy, with best response occurring an average of two cycles later. For 75% of responders, response was achieved by cycle 4; 90% experienced a response by the sixth cycle.

The vast majority of patients who were transfusion-dependent at baseline and responded to SC azacitidine demonstrated transfusion independence. Among patients who responded to treatment and were RBC transfusion–dependent at baseline, 80%, 86%, and 93% on study 8921, the 9221 azacitidine arm, and the 9221

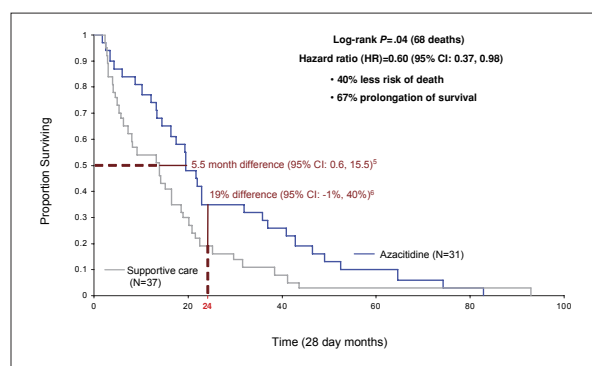
crossover arm, respectively, became transfusion-independent (Figure 2). The rate was lower, 47%, for intravenous (IV) azacitidine patients in study 8421. Median duration of RBC transfusion independence in these patient groups was 132, 244, 252, and 286 days for 8421, 8921, 9221 azacitidine, and 9221 crossover patients, respectively.

Results from this study demonstrated that azacitidine is effective for the treatment of MDS even when more stringent IWG response criteria are applied. In fact, response rates in this study were comparable to those observed using CALGB response criteria.<sup>2</sup> Azacitidine induced response rates of 40–47% and was associated with high rates of transfusion independence among those previously requiring RBC transfusions (47–93%).

## 2524 Azacitidine Prolongs Survival and Time to AML Transformation in High-Risk Myelodysplastic Syndrome Patients $\geq 65$ Years of Age<sup>5</sup>

L. Silverman, D. McKenzie, B. Peterson, E. Demakos, N. Malone, J. Holland, R. Larson, and the CALGB

This analysis identified a high-risk group within the CALGB 9221 study. Older patients with MDS are considered a high-risk group and are often unable to be treated with intensive chemotherapy or transplantation regimens. Thus, elderly patients are left with few treatment options. The phase III CALGB 9221 study demonstrated efficacy of azacitidine for the treatment of MDS but did not include subgroup analyses. This study is a subgroup analysis of patients from CALGB 9221 who were at least 65 years of age and diagnosed as RAEB or RAEB-t at baseline.



**Figure 3.** Overall survival.

The three primary endpoints of this subgroup analysis were: overall survival, time to death or AML transformation, and time to AML transformation. The safety of azacitidine for this high-risk group of patients was also investigated. Of the 191 patients from CALGB 9221, 68 were eligible for this study, 31 from the azacitidine arm and 37 from the supportive care arm. Twenty patients from the supportive care arm crossed over to the azacitidine arm but all of their data were included in the supportive care group for efficacy analysis. The median age of patients in this analysis was 70 years. More than two thirds (69%) of patients were male.

Azacitidine was highly effective in the subset of patients in CALGB 9221 who were 65 years of age or older. Its use was associated with substantial increases in survival and time to progression. Overall survival for patients on the azacitidine arm was increased by a median of 5.5 months ( $P=.04$ ; Figure 3). This was a 67% prolongation of survival. Azacitidine was thus correlated with a 40% decrease in risk of death (hazard ratio [HR] = 0.60; 95% CI, 0.37–0.98). Progression to AML was delayed by a median of more than 2 years (24.3 months;  $P=.04$ ) in patients treated with azacitidine. This represented a 53% decreased risk of transformation to AML (HR = 0.47; 95% CI, 0.23–0.98). The drug was associated with a 9.9 month median increase in time to AML transformation or death ( $P=.008$ ), from a median of 9.2 months in the supportive care group (95% CI, 4.3–14.2) to a median of 19.1 months (95% CI, 12.1–22.9) in the azacitidine group. Thus, time to progression or death was increased by 92%. Overall risk of death or progression to AML was diminished by 48% for patients treated with azacitidine (HR = 0.52; 95% CI, 0.32–0.85).

Azacitidine was generally well tolerated in this subgroup of elderly patients. Toxicities observed in these patients were comparable to those seen in previous studies.<sup>2,6</sup> Adverse events occurred at similar rates in both arms, suggesting that toxicities may be related more to underlying medical conditions than to drug treatment. The most common adverse events were injection site reactions and hematologic and gastrointestinal disorders. Among hematologic toxicities, anemia, thrombocytopenia, and leukopenia were most frequent, occurring in 82%, 69%, and 53% of patients, respectively. Nausea and vomiting were the most common gastrointestinal toxicities, affecting 53% and 41% of patients, respectively.

This study demonstrated that azacitidine is safe and effective in patients aged 65 and over. It was associated with prolonged survival and delayed progression to AML when compared with supportive care therapy. The drug was well tolerated in this elderly patient population, with a toxicity profile similar to that of supportive care. The authors conclude that this study demonstrates the benefit

of azacitidine in the treatment of RAEB and RAEB-t patients 65 years of age and older, expanding the limited treatment options for this high-risk group.

## 2530 Early and Sustained Response to Azacitidine in High-Risk MDS Patients with Monosomy 7 Correlates with Increased Apoptosis and Not *CDKN2B* Demethylation<sup>7</sup>

K. Raj, A. M. John, A. Ho, N. S. B. Thomas, and G. J. Mufti

Tremendous strides have been made in recent years in understanding the role of cytogenetics in MDS disease pathogenesis and progression. A number of studies have focused on identifying genetic and cytogenetic abnormalities characteristic of myelodysplastic cells. Methylation of the *CDKN2B* gene promoter was found in these studies to be characteristic of MDS. This hypermethylation inhibits expression of the cell cycle regulator p15<sup>INK4B</sup>, enabling proliferation of bone marrow blasts. The frequency of *CDKN2B* methylation is associated with increased progression to leukemia.<sup>8</sup> The current study sought to determine whether clinical response to azacitidine correlated with decreased methylation of the *CDKN2B* promoter and increased apoptosis of cells in the bone marrow.

Thirty-one patients were enrolled in the study. They were predominantly male (80%) with a median age of 66 years. All FAB subtypes were included, but 81% of patients were characterized as having RAEB or RAEB-t. The majority of the group was considered intermediate-2- or high-risk by IPSS standards. Cytogenetic abnormalities were present in 80% of patients. Monosomy 7 was the most common abnormality, occurring in 12 patients (39%). Seven of these patients had a derivative chromosome 7 as their only cytogenetic abnormality whereas the remaining 5 had a complex karyotype. Other chromosomal abnormalities represented in the population were: deletions of chromosomes 11q, 5q, 20q, and iso 17p, and trisomy 8. The treatment regimen was standard for azacitidine (75 mg/m<sup>2</sup> daily for 7 days every 28 days). Response was assessed using IWG criteria.

A median of five courses of treatment was received by each patient (range, 1–13). Complete remission was achieved in 7 patients (23%). Complete cytogenetic

remission was observed in 6 patients (19%). Hematologic improvements were seen in 6 patients (19%): platelet response in 6 patients, erythroid response in 2 patients, and neutrophil response in 3 patients. Blast counts were reduced in 5 patients. Azacitidine was associated with increased apoptosis in patients who responded to therapy. Among those 11 responders, apoptosis of bone marrow mononuclear cells occurred at a rate of 6% prior to treatment and increased to 19% following treatment ( $P=.05$ ). Baseline methylation of *CDKN2B* was comparable in CD33+ and CD34+ cells of responders and nonresponders. The gene was more likely to be unmethylated in lymphocytes of responders, however. Azacitidine did not appear to significantly alter the methylation status of the *CDKN2B* gene. Four patients (13%) had demethylated *CDKN2B* following treatment. This demethylation was not correlated with response, although it was associated with decreased blasts.

The efficacy of azacitidine in high-risk MDS patients was demonstrated in this study, with high rates of complete remission and hematologic improvements. Response rates were especially dramatic in isolated monosomy 7 cases, approaching 80%. This group of patients is typically associated with poor prognosis. At the molecular level, *CDKN2B* methylation status could not be correlated with response to azacitidine, but the drug was associated with increased rates of apoptosis in the bone marrow. Study authors recommended future studies using DNA microarrays and other diagnostics to further investigate molecular markers of response to azacitidine, particularly in the high-risk group of patients with chromosome 7 abnormalities.

## 2525 Rates of Infection and Bleeding Are Not Increased in Patients With Myelodysplastic Syndromes Treated With Azacitidine Compared With Supportive Care<sup>9</sup>

L. Silverman, D. McKenzie, B. Peterson, R. Odchimar-Reissig, R. Hinkle, J.T. Backstrom, R. Larson, and the CALGB

Many high-risk MDS patients (RAEB and RAEB-t) experience fatal hemorrhage or infection within 1 year of diagnosis. This study examines whether azacitidine is associated with an increased incidence of bleeding or infection in patients with MDS. Published results from CALGB study 9221 revealed that treatment with azacitidine was correlated with worsening of pre-existing cytopenias in a majority of patients.<sup>2</sup> However, in that publication safety data were reported for the azacitidine arm only. Since then, a more complete database has been compiled with adverse event reporting for all study patients—azacitidine, supportive care, and crossover. The current study uses this comprehensive safety data to analyze the effects of azacitidine on bleeding and infection.

Patients in CALGB 9221 were randomized to either azacitidine 75 mg/m<sup>2</sup>/day × 7 days in a 28-day cycle plus best supportive care or best supportive care only. A total of 191 patients were enrolled. Patients with disease progression were allowed to crossover to the azacitidine arm.

For the purposes of this study, 150 patients were analyzed as azacitidine patients, 99 who had been randomized to drug treatment and 51 who crossed over. The supportive care patients numbered 92, 41 of whom never crossed over to azacitidine. All FAB subtypes of MDS were included in this study. Baseline characteristics were comparable between treatment arms. The vast majority of azacitidine patients (98%) had cytopenias at baseline. Rates of adverse events were calculated as number of patients per patient-year of exposure due to the differences in length of exposure between the azacitidine and supportive care arms.

Overall, infections occurred at a lower rate in the azacitidine group (0.64) compared with the supportive care group (0.95). A similar trend was observed in high-risk patients 65 years or older, where the rate of infection was 0.76 per patient-years of exposure among those receiving supportive care compared with 0.38 for those receiving azacitidine (Table 2). Pneumonia was the most common infection observed, followed by herpes simplex and urinary tract infections. Sepsis was a leading cause of

infection in supportive care patients, both overall and in the high-risk subgroup. Infection was considered to be the cause of death in 2% of patients on both arms.

Bleeding was common in both treatment groups. The rate of bleeding per patient year for azacitidine patients was 0.56, comparable to the rate of 0.60 for supportive care patients. This was likewise true in high-risk patients, where the rates per patient year were 0.32 and 0.44 for the azacitidine and supportive care groups, respectively (see Table 2). The most common sources of bleeding were gastrointestinal and respiratory, thoracic, and mediastinal disorders. These two areas accounted for nearly half of all bleeding. As with infection, bleeding was the cause of death in 2% of patients on both arms.

**Table 2.** Selected Infection and Bleeding Rates in High-Risk Older Patients (RAEB and RAEB-t ≥65 years)

Adverse Event*†	Number of Patients (number of patients per patient-year of exposure)	
	Azacitidine N=51	Supportive Care N=37
<b>Infection—total</b>	<b>31 (0.38)</b>	<b>19 (0.76)</b>
Pneumonia	6 (0.07)	2 (0.08)
Sepsis	2 (0.02)	4 (0.16)
<b>Bleeding—total</b>	<b>26 (0.32)</b>	<b>11 (0.44)</b>
<b>Gastrointestinal disorders—total</b>	<b>11 (0.13)</b>	<b>4 (0.16)</b>
Gastrointestinal hemorrhage	1 (0.01)	0 (0.00)
Gingival bleeding	5 (0.06)	1 (0.04)
Rectal hemorrhage	3 (0.04)	0 (0.00)
<b>Nervous system disorders—total</b>	<b>1 (0.01)</b>	<b>1 (0.04)</b>
Intracranial hemorrhage	1 (0.01)	0 (0.00)
Subdural hematoma	0 (0.0)	1 (0.04)
<b>Respiratory, thoracic, and mediastinal disorders—total</b>	<b>9 (0.11)</b>	<b>5 (0.20)</b>
Epistaxis	8 (0.10)	4 (0.16)
Hemoptysis	1 (0.01)	1 (0.04)

\* Multiple reports of the same adverse event term for a patient are only counted once within each treatment group.

† Includes National Cancer Institute Common Toxicity Criteria grades 1–4.

RAEB = refractory anemia with excess blasts; RAEB-t = RAEB in transformation.

Patients on both arms of this study were characterized by severe hematologic disease at study entry, reflected in the rates of infection and bleeding being highest in the first two treatment cycles. Cytopenias were exacerbated in both groups, with 78% of patients on the azacitidine arm and 70% on the supportive care arm experiencing worsening of this condition. These findings suggest that azacitidine is superior to supportive care in MDS treatment because this drug confers significant therapeutic benefit and does not significantly induce infection or bleeding above levels attributable to MDS disease itself.<sup>2</sup>

## 2515 Response Rates of Phase 2 and Phase 3 Trials of Decitabine in Patients with Myelodysplastic Syndromes<sup>10</sup>

H. Saba, M. Lubbert, P. W. Wijermans

Although not yet approved by the FDA, decitabine is another nucleoside analog that has shown activity in phase II and III trials for the treatment of MDS. Like azacitidine, decitabine induces hypomethylation, enabling transcription of previously silenced genes. The current report summarizes three phase II studies (91–01, 95–11, and 97–19) and one phase III study (D-0007).

All studies were open-label. The phase II studies were single-arm whereas D-0007 was a randomized comparison of decitabine plus supportive care versus supportive care alone. The typical dose of decitabine administered was 15 mg/m<sup>2</sup> administered IV over 4 hours every 8 hours for 3 days, repeated every 6 weeks. The phase II trials involved 4–6 cycles of therapy. In D-0007, patients received a maximum of 10 cycles of therapy. They were removed from therapy after six cycles if partial response was not achieved, after eight cycles in the absence of complete response, or after two consecutive cycles of complete response. All studies evaluated safety, efficacy, and tolerability of decitabine. The primary endpoints of D-0007 were: (i) overall response rate (complete and partial response); (ii) time to AML transformation or death; (iii) hematologic improvements, survival, and quality of life.

A total of 271 patients were treated with decitabine in the four studies. In general patients were older, with a median age range of 68–72. The majority of patients in all studies were male (59–75%). Patients from all IPSS risk categories and FAB subtypes were included. A small proportion of patients in studies 91–01 and 97–19 were diagnosed with AML—7% and 3%, respectively—but no AML patients were included in the other studies.

Responses to decitabine were observed in all four studies. Overall response rates ranged from 17% for the phase III study D-0007 to 45% for trial 91–01 (Table 3). Importantly, overall response in patients randomized to decitabine in D-0007 was significantly greater than in those randomized to supportive care, none of whom exhibited a clinical response ( $P < .001$ ). Complete response was seen in 28%, 21%, 22%, and 9% of patients in studies 91–01, 95–11, 97–19, and D-0007, respectively. Hematologic improvement was also observed in all four studies, ranging from 7% in 91–01 to 15% in 97–19. A median of four treatment cycles were delivered in the phase II studies, with one third of patients receiving at least six cycles. Three median treatment cycles were administered in D-0007, with less than half (48%) of patients receiving four or more cycles. There was a trend toward improved median time to AML transformation or death among decitabine patients in D-0007 (12.1 vs 7.3 months for supportive care), although this improvement was not statistically significant ( $P = .16$ ). When subgroup analyses were performed, however, decitabine patients experienced significantly longer survival and AML-free times. These improvements were more dramatic in patients in higher IPSS risk categories, ranging from an increase of 5 months in median time to AML transformation or death for treatment-naïve patients on decitabine to an increase of 6.5 months for high-risk patients. Indeed, in high-risk patients, median time to AML transformation or death was 9.3 months for decitabine patients compared with 2.8 months for supportive care patients ( $P = .01$ ). In addition, RBC transfusion requirements decreased for decitabine patients but remained unchanged for supportive care patients on the phase III study.

Decitabine was well tolerated in all four studies. Toxicities were comparable to other nucleoside analogs. The most common toxicities in the phase III study were hematologic and included neutropenia, thrombocytopenia, anemia, and febrile neutropenia. The rate of grade 3/4 neutropenia on the decitabine arm was 87%, compared with 50% on the supportive care arm. Grade 3/4 thrombocytopenia was experienced by 85% of decitabine patients, compared with 43% of supportive care patients. Rates of anemia were comparable between arms, 12% for decitabine versus 15% for supportive care. Grade 3/4 neutropenia was observed in 23% of decitabine patients compared with 4% of supportive care patients. Patients on decitabine and supportive care had grade 3/4 pneumonia rates of 15% and 9%, respectively. Death rates and causes of death were comparable between arms.

The safety and efficacy of decitabine for the treatment of MDS was shown in four studies, one of which was a randomized phase III comparison of decitabine versus supportive care. Decitabine induced response rates

**Table 3.** Response Rates for Phase II and III Studies of Decitabine (N=271)<sup>3-5</sup>

	Phase II			Phase III
	91-01	95-11	97-19	D-0007
N	29	66	87	89
ORR (CR + PR), n (%)	13 (45%)	17 (26%)	23 (26%)	15 (17%)
CR, n (%)	8 (28%)	14 (21%)	19 (22%)	8 (9%)
PR, n (%)	5 (17%)	3 (5%)	4 (5%)	7 (8%)
Median duration of response (CR + PR), days	217	250	146	288
HI, n (%)	2 (7%)	8 (12%)	13 (15%)	12 (13%)
Median # cycles	4	4	4	3

CR = complete response; HI = hematologic improvement; ORR = overall response rate (CR + PR); PR = partial response.

of up to 45% and complete response in up to 28% of patients. Decitabine was also associated with increased time to AML and death, particularly among high-risk MDS patients, and may decrease transfusion requirements for MDS patients. Decitabine was well tolerated, with a toxicity profile similar to other drugs in its class. The study authors contend that decitabine is a promising new treatment for MDS and recommend future studies to optimize dosing strength and intervals.

## 2522 Decitabine Low-Dose Schedule in Myelodysplastic Syndrome: Comparison of 3 Different Dose Schedules<sup>11</sup>

H. Kantarjian, S. O'Brien, F. Giles, F. Ravandi-Kashani, S. Faderl, G. Garcia-Manero, J. Davis, J.-P. Issa

While decitabine has demonstrated efficacy for the treatment of MDS and a favorable safety profile, optimal dosing has not been established. The current study was a randomized trial in which three different dosing regimens of decitabine were evaluated for safety and efficacy: (i) 20 mg/m<sup>2</sup> IV over 1 hour daily for 5 days; (ii) 10 mg/m<sup>2</sup>

IV over 1 hour daily for 10 days; or (iii) 10 mg/m<sup>2</sup> SC twice per day for 5 days. Treatment cycles were repeated every 4–6 weeks. Clinical benefit was defined as any of the following: platelet counts increased by at least 50% and above 30 × 10<sup>9</sup>/L, granulocyte counts increased by at least 100% and to above 10<sup>9</sup>/L, hemoglobin increased by at least 2 g/dL or transfusion independence, splenomegaly decreased by 50% or more, or monocytes decreased by 50% or more (if pretreatment counts were >5 × 10<sup>9</sup>/L). Response rates were compared with a historical group of 114 patients with MDS who received intensive chemotherapy from 2000–2004.

The median age of the 96 patients enrolled in the study was 65 years (range 39–90). All patients were considered intermediate- or high-risk by IPSS standards, with comparable distribution among those subgroups. Cytogenetic abnormalities were present in 58% of patients and 41% had marrow blasts greater than 10%. Prior therapies included erythropoietin (52%), G-CSF (19%), thalidomide (11%), azacitidine (4%) and other therapies (8%).

Complete response rates were similar among decitabine and the historical control patients (42% versus 45%, respectively; Table 4), with slightly higher rates of partial response (5% vs 0) and hematologic improvement (2% vs 0) seen with decitabine therapy. Although no marrow complete response and/or clinical benefit was reported for control patients, 34% of patients treated with decitabine achieved these endpoints. The survival rate at 3 and 6 months was also higher among these patients. Grade 3/4 side effects were minimal for all dose groups and included fever, infection, and bleeding. Importantly, the response rates varied among the three decitabine treatment arms, with the 5-day IV decitabine arm showing statistically superior outcomes compared with the other arms after 55 patients had been randomized. Study authors noted that timely and repeated cycles of therapy are needed to facilitate optimal efficacy of decitabine.

**Table 4.** Decitabine vs Intensive Chemotherapy: Responses

	Intensive Chemotherapy, n (%)	Decitabine, n (%)
Total evaluable	114	96
CR	51 (45)	40 (42)
PR + HI	0	7 (7)
Marrow CR/clinical benefit	0	33 (34)
Death within 6 wk	24 (21)	2 (2)
Death within 3 mo	38 (33)	7 (7)

CR = complete response; HI = hematologic improvement; PR = partial response.

## 371 Hypomethylation Therapy of Decitabine in Patients With Myelodysplastic Syndromes Induces Apoptosis and Reduces Proliferation<sup>12</sup>

I. Jilani, H. Kantarjian, M. Gorre, J.-P. Issa, J. Bennett, A. Raza, F. Ravandi, H. Saba, M. Albitar

The depletion of methylcytosine by decitabine results in hypomethylation of target genes in MDS. However, whether this hypomethylation reduces apoptosis, thereby allowing the growth of normal cells, or increases tumor cell apoptosis, is not known.

Jilani and colleagues conducted a study to examine the relationship between apoptosis and proliferation among patients with MDS treated with either decitabine or best supportive care in a randomized clinical trial. Bone marrow and peripheral blood samples were collected at baseline and intermittently during the course of treatment. Apoptosis was measured by annexin V and mitochondrial potential using multiparameter flow cytometry. Proliferation was measured similarly using bromodeoxyuridine incorporation.

There was no significant difference at baseline in apoptosis, proliferation, or percentage of CD34(+) cells between the decitabine and supportive care groups either in the bone marrow or peripheral blood. After 3 months of therapy, patients receiving decitabine had a significant increase in apoptosis of CD34(+) compared to the baseline measurement, whereas no significant increase was observed among patients receiving supportive care ( $P=.01$ ). The increase in apoptosis in the decitabine arm was intensified with additional courses of therapy. Decitabine was also associated with a greater reduction in proliferation of CD34(+) cells compared with supportive care ( $P=.01$ ). Interestingly, patients treated with decitabine who had a higher rate of proliferation at diagnosis experienced a statistically significantly higher complete response rate than patients with a low rate of proliferation ( $P=.03$ ).

The authors concluded that decitabine leads to hypomethylation when used to treat MDS, but the effects of this agent also include high levels of apoptosis, neoplastic cell death, and a related reduction in the proliferation of leukemic cells.

## References

1. Lyons R, Cosgriff T, Modi S, Lintz L, Beach CL, Backstrom JT. Azacitidine (Vidaza) treatment response assessed using three alternative dosing schedules in patients with myelodysplastic syndromes (MDS). *Blood*. 2005;106. Abstract 2517.
2. Silverman LR, Demakos EP, Peterson BL, et al. Randomized controlled trial of azacitidine in patients with the myelodysplastic syndrome: a study of the Cancer and Leukemia Group B. *J Clin Oncol*. 2002;20:2429-2440.
3. Cheson BD, Bennett JM, Kantarjian H, et al; World Health Organization (WHO) international working group. Report of an international working group to standardize response criteria for myelodysplastic syndromes. *Blood*. 2000;96:3671-3674.
4. Silverman L, McKenzie D, Peterson B, DeCastro C, Ellerton J, Knapp K, Beach CL, Larson R, the Cancer and Leukemia Group B (CALGB). Response rates using International Working Group (IWG) criteria in patients with myelodysplastic Syndromes (MDS) Treated with Azacitidine. *Blood*. 2005;106:Abstract 2526.
5. Silverman L, McKenzie D, Peterson B, Demakos E, Malone N, Holland J, Larson R, the Cancer and Leukemia Group B (CALGB). Azacitidine prolongs survival and time to AML transformation in high-risk myelodysplastic syndrome (MDS) patients  $\geq 65$  years of age. *Blood*. 2005;106. Abstract 2524.
6. Silverman LR, Holland JF, Weinberg RS, et al. Effects of treatment with 5-azacytidine on the in vivo and in vitro hematopoiesis in patients with myelodysplastic syndromes. *Leukemia*. 1993;7(suppl 1):21-29.
7. Raj K, John AM, Ho A, Thomas NSB, Mufti GJ. Early and sustained response to azacitidine in high-risk MDS patients with monosomy 7 correlates with increased apoptosis and not *CDKN2B* demethylation. *Blood*. 2005;106. Abstract 2530.
8. Quesnel B, Guillermin G, Verecque R, et al. Methylation of the p15(INK4b) gene in myelodysplastic syndromes is frequent and acquired during disease progression. *Blood*. 1998;91:2985-2990.
9. Silverman L, McKenzie D, Peterson B, Odchimar-Reissig R, Hinkle R, Backstrom JT, Larson R, the Cancer and Leukemia Group B (CALGB). Rates of infection and bleeding are not increased in patients with myelodysplastic syndromes (MDS) treated with azacitidine compared with supportive care. *Blood*. 2005;106. Abstract 2525.
10. Saba H, Lubbert M, Wijermans PW. Response rates of phase 2 and 3 trials of decitabine (DAC) in patients with myelodysplastic syndromes (MDS). *Blood*. 2005;106. Abstract 2515.
11. Issa JP, Ravandi-Kashani F, O'Brien S, Giles F, et al. Decitabine low-dose schedule (100 mg/m<sup>2</sup>/course) in myelodysplastic syndrome (MDS): comparison of 3 different dose schedules. *Blood*. 2005;106. Abstract 2522.
12. Jilani I, Kantarjian H, Gorre M, et al. Hypomethylation therapy of decitabine in patients with myelodysplastic syndromes (MDS) induces apoptosis and reduces proliferation. *Blood*. 2005;106. Abstract 371.

## Commentary

Guillermo Garcia-Manero, MD

Associate Professor of Medicine  
Department of Leukemia  
University of Texas M. D. Anderson Cancer Center

### Introduction

The last 3 years have seen remarkable improvements in therapy for MDS, including the approval of azacitidine for all subtypes of MDS,<sup>1</sup> followed by the approval of lenalidomide for patients with low-risk disease and alterations of chromosome 5q31.<sup>2</sup> Other promising therapies include decitabine, tipifarnib (Zarnestra, Johnson & Johnson), and, potentially, arsenic trioxide (Trisenox, Cephalon) in various combination regimens.

At the 2005 ASH annual meeting, several studies of DNA methyltransferase inhibitors were presented that clearly demonstrate that this class of agents has significant activity in MDS and the capacity to delay the time to transformation to AML. These agents appear to decrease transfusion requirements and to prolong survival.

### Azacitidine

#### *Initial Results of CALGB 9221*

In order to consider the studies of azacitidine and decitabine presented at the 2005 ASH annual meeting, it is logical to review the results of the CALGB 9221 study, a landmark phase III randomized trial of azacitidine versus supportive care in patients with MDS.<sup>3</sup>

This study randomized 191 patients to receive SC azacitidine 75 mg/m<sup>2</sup> daily for 7 days (n=99) or supportive care (n=92). Patients initially randomized to supportive care could cross over to azacitidine, granted that they had remained under observation for 4 months. All MDS categories were included in this study. Median time to transformation to AML was 21 months for azacitidine versus 13 months for supportive care ( $P=.007$ ), and the median survival was 20 months versus 14 months, respectively ( $P=.1$ ). The latter figure was not statistically significant most likely due to the crossover design. To eliminate the confounding effect of the crossover, a landmark analysis after 6 months on study showed a significant improvement in overall survival associated with azacitidine (18 vs 11 months;  $P=.03$ ). In a companion study, azacitidine was shown to significantly improve the quality of life of responding patients.<sup>4</sup> Based on these data, the FDA approved azacitidine for MDS, and subsequently published their analysis of the CALGB 9221 study.<sup>1</sup>

#### *Results Presented at ASH 2005*

In what is probably the most important information regarding the activity of azacitidine in MDS since publication of CALGB 9221,<sup>3</sup> Silverman and colleagues<sup>5</sup> reported that older patients with high-risk MDS experienced a statistically significant benefit in overall survival and time to AML transformation when treated with azacitidine.

Most investigators have since adopted the IWG criteria to assess response in MDS.<sup>6</sup> Following this change, the results of the different CALGB studies with azacitidine were re-evaluated using the IWG criteria by Silverman and coworkers.<sup>7</sup> Azacitidine was still effective for the treatment of MDS even when using the more stringent IWG criteria, with the response rates being comparable between the current study and those using the CALGB response criteria.<sup>6,7</sup> The investigators also used this analysis to evaluate the time to response. Responses were gradual, with a median of two courses of therapy to best response after initial response. These data confirm the need to continue therapy for at least 4–6 courses, even in patients who do not respond initially, and the importance of continuing therapy in order to achieve maximal benefit.

Because the most common toxicity of azacitidine is myelosuppression,<sup>3</sup> there is concern that this agent may exacerbate pre-existing cytopenias, particularly because many patients with MDS die from complications of bleeding or infection rather than from AML transformation. Silverman et al compared the risk of infection and bleeding in patients treated with azacitidine or supportive care in the 9221 study.<sup>8</sup> No difference in worsening of cytopenias was observed between the two arms. A trend toward lower infection rates was observed in patients treated with azacitidine compared to patients receiving supportive care. Two percent of patients died from complications of infection in both arms. The rate of bleeding was similar in both arms. These data indicate that the risk of infection or bleeding is not significantly higher in patients treated with azacitidine.

To study the possibility of using alternative dosing schedules that could avoid the need to administer azacitidine for 7 consecutive days, a clinical trial is currently underway to analyze the safety and efficacy of three different schedules of azacitidine.<sup>9</sup> Preliminary findings were presented at the ASH meeting. Although the sample size is relatively small for this interim analysis, 63% of patients on the AZA 5-2-2 schedule, 44% in the AZA 5-2-5, and 43% in the 5-day schedule had achieved a hematologic improvement at the time of presentation at ASH. Transfusion independence was achieved in 58% of patients who were transfusion-dependent at baseline. This figure was 67% in patients with low-risk disease. No differences were observed in terms of safety or clinical activity between the three different schedules.

## Decitabine

Initial data from a phase I trial had indicated that response to decitabine is dependent on schedule, with a low-dose schedule being optimal in terms of response and toxicity profile.<sup>10</sup> The data from several phase II and III trials of the hypomethylating agent decitabine were summarized and presented at the 2005 ASH meeting.<sup>11</sup> Saba and colleagues summarized the response rates observed in four studies of decitabine in patients with MDS. Responses to decitabine were observed in all studies, with overall response rates of 17–45%. Hematologic improvement was also observed in all studies, with more dramatic improvements seen among higher-risk patients. RBC transfusion requirements decreased for patients receiving decitabine but not for those receiving supportive care in the phase III trial. An increased time to transformation to AML and increased survival were observed among patients receiving decitabine with study D-0007, and these results serve as the basis for the application for FDA approval.

The main limitation of the studies summarized by Saba et al is that the schedules requires hospital admission. Data from the M. D. Anderson Cancer Center had indicated that a low-dose schedule of decitabine infused over 1 hour daily for 10 days was associated with an excellent activity and toxicity profile.<sup>10</sup> Based on these data, Kantarjian and colleagues compared three low-dose schedules of decitabine in the treatment of MDS.<sup>12</sup> All of three schedules tested were associated with a lower 3-month mortality rate compared with historical control patients who received intensive chemotherapy. Complete response rates were similar, but the rate of hematologic improvement was higher for patients who received decitabine compared to the historical control group. Importantly, responses to decitabine were dependent on the schedule used. Patients who received decitabine 20 mg/m<sup>2</sup> IV over 1-hour daily for 5 days had a significantly higher complete response rate (41%) than the other two schedules (28% and 24%, respectively). A multicenter study is ongoing to confirm these important results.

## Conclusion

After the FDA approval of azacitidine for the treatment of MDS, it was important to continue to obtain more data in order to refine the treatment approaches for the wide range of MDS patients. These new studies provide essential insights, such as the possibility of using alternative dosing schedules of azacitidine, and demonstrate that a change in response criteria does not diminish the efficacy seen with this agent in initial studies. The benefit seen with azacitidine seen among older patients with high-risk disease is particularly promising.

Decitabine is also a promising agent for the treatment of MDS. The studies presented at the 2005 ASH annual meeting take new steps in our understanding of this agent, as investigators continue to determine the optimal dosing schedule for this important new agent. In particular, confirmation of the results of the low-dose, 5-day schedule may lead to a major treatment improvement.

The complete and partial remission rates still need to be improved for patients with MDS. New DNA methylation inhibitor dose schedules should continue to be investigated, along with safe and active combinations, perhaps with other epigenetic agents such as histone deacetylase inhibitors.<sup>13</sup> Efforts to identify such combination are ongoing.<sup>14,15</sup>

Finally, all of the investigators involved in these studies, including Dr. Silverman and collaborators at the CALGB, and Drs. Saba, Lubbert, Wijermans, and Kantarjian should be congratulated for their efforts in bringing to clinical practice this new class of agents, and for their continuous efforts to evaluate the activity of these important therapeutic options.

## References

1. Kaminskas E, Farrell A, Abraham S, et al. Approval summary: azacitidine for treatment of myelodysplastic syndrome subtypes. *Clin Cancer Res.* 2005;11:3604-3608.
2. List A, Kurtin S, Roe DJ, et al. Efficacy of lenalidomide in myelodysplastic syndromes. *N Engl J Med.* 2005;352:549-557.
3. Silverman LR, Demakos EP, Peterson BL, et al. Randomized controlled trial of azacitidine in patients with the myelodysplastic syndrome: a study of the cancer and leukemia group B. *J Clin Oncol.* 2002;20:2429-2440.
4. Kornblith AB, Herndon JE, 2nd, Silverman LR, et al. Impact of azacitidine on the quality of life of patients with myelodysplastic syndrome treated in a randomized phase III trial: a Cancer and Leukemia Group B study. *J Clin Oncol.* 2002;20:2441-2452.
5. Silverman LR, McKenzie DR, Peterson BL, et al. Azacitidine prolongs survival and time to AML transformation in high-risk myelodysplastic syndrome patients older than 65 years of age. *Blood.* 2005;106:Abstract 2524.
6. Cheson BD, Bennett JM, Kantarjian H, et al. Report of an international working group to standardize response criteria for myelodysplastic syndromes. *Blood.* 2000;96:3671-3674.
7. Silverman L, McKenzie DR, Peterson BL, et al. Response rates using international working group (IWG) criteria in patients with myelodysplastic syndrome treated with azacitidine. *Blood.* 2005;106:Abstract 2526.
8. Silverman L, McKenzie DR, Peterson BL, et al. Rates of infection and bleeding are not increased in patients with myelodysplastic syndrome treated with azacitidine compared with supportive care. *Blood.* 2005;Abstract 2525.
9. Lyons R, Cosgriff T, Modi S, Lintz L, Beach CL, Backstrom J. Azacitidine (Vidaza) treatment response assessed using three alternative dosing schedules in patients with myelodysplastic syndromes. *Blood.* 2005;106:Abstract 2517.
10. Issa JP, Garcia-Manero G, Giles FJ, et al. Phase I study of low-dose prolonged exposure schedules of the hypomethylating agent 5-aza-2'-deoxycytidine (decitabine) in hematopoietic malignancies. *Blood.* 2004;103:1635-1640.
11. Saba H, Lubbert M, Wijermans PW. Response rates of phase 2 and phase 3 trials of decitabine in patients with myelodysplastic syndromes (MDS). *Blood.* 2005;106:Abstract 2515.
12. Issa JP, Ravandi-Kashani F, O'Brien S, et al. Decitabine low-dose schedule (100 mg/m<sup>2</sup>/course) in myelodysplastic syndrome: comparison of 3 different dose schedules. *Blood.* 2005;106:Abstract 2522.
13. Garcia-Manero G, Issa JP. Histone deacetylase inhibitors: a review of their clinical status as antineoplastic agents. *Cancer Invest.* 2005;23:635-642.
14. Garcia-Manero G, Gore SD. Future directions for the use of hypomethylating agents. *Semin Hematol.* 2005;42:S50-59.
15. Gore SD. Combination therapy with DNA methyltransferase inhibitors in hematologic malignancies. *Nat Clin Pract Oncol.* 2005;2(suppl 1):S30-S35.

