Myelodysplastic syndromes

Myelodysplastic syndromes (MDS) are a group of bone marrow disorders that mainly occur in the elderly and are characterized by ineffective hematopoiesis resulting in anemia, neutropenia, and thrombocytopenia. MDS can occur spontaneously (primary or de novo) or can occur as a result from exposure to chemotherapy or chemicals including benzene and others (secondary). MDS is seen in approximately 5 per 100,000 people. Risk factors for MDS include age, previous chemotherapy, and exposure to environmental toxins, tobacco and gender (male). Approximately half of MDS patients are asymptomatic at the time of initial diagnosis and are diagnosed only after routine laboratory tests showing peripheral blood abnormalities. Although MDS can eventually result in neutropenia and/or thrombocytopenia, anemia is the most common characteristic at the time of initial diagnosis.

Prognosis is dependent upon multiple factors, including morphology, number of cytopenias, blast count, and cytogenetics. If untreated, median survival ranges from 0.4 years for high-risk MDS patients to 5.7 years for low-risk patients, with death most often resulting from anemia, infections, or severe bleeding. Approximately one-third of adult MDS patient’s progress to acute myeloid leukemia (AML), with a survival period of 6-12 months. Multiple evidence-based treatment guidelines for MDS have been published. According to these guidelines, the treatment for MDS should be determined by the International Prognostic Scoring System (IPSS) risk category, as well as age and performance status. The IPSS scoring system takes into account the number of cytopenias in the peripheral blood, cytogenetics and the number of blasts in the bone marrow to predict survival and risk of AML transformation. Prognostically, MDS has been classified into low risk and high IPSS risk categories for transformation to AML. Low risk categories include refractory anemia (RA), refractory anemia with ringed sideroblasts (RARS), and some chronic myelomonocytic leukemia (CMML) patients. High risk MDS includes some CMML’s, refractory anemia with excess blasts (RAEB) and refractory anemia with excess blasts in transformation (RAEB-T).
Management of myelodysplastic syndromes based upon risk categories

The management and treatment goals for MDS are varied and dependent upon the risk category for the patient. The goal of treatment in patients with low or Int-1 IPSS risk is to improve hematopoiesis and/or alleviate its potential problems (i.e. anemia, thrombocytopenia, neutropenia). This is because apoptosis remains the main feature of the pathogenesis in the low and Int-1 risk categories. Aggressive therapeutic intervention is usually not required. Treatment of patients in this risk category includes observation, early recognition and treatment of infections, transfusions and/or the use of cytokines (i.e. erythropoietin, darbopoietin, G-CSF), and iron chelation therapy if required. In non-responsive patients or those with del(5q-) abnormalities, more aggressive therapy with azacytidine, decitabine or lenalidomide may be indicated.

The goal of therapy for Int-2 and high risk MDS patients is to improve survival and delay the time to AML transformation. Apoptosis in high risk MDS is usually decreased and epigenetic changes, hypermethylation and chromatin remodeling are considered the main features of the pathogenesis in this category. Therapeutic interventions for this category of MDS may include hematopoietic stem cell transplant, high intensity therapy, azacytidine, decitabine or participation in clinical trials.

DNA methylation

DNA methylation is a common epigenetic change that plays an important role in gene expression in mammalian cells. Methylated genes have been found in many cancers, including MDS, leukemias, prostate, lung, lymphomas, and others. Certain genes may normally be silenced through methylation of cytosine residues in their promoter regions (CpG islands). However, in some hematopoietic malignancies including MDS, DNA hypermethylation can inactivate genes that are essential for the control of normal cell growth, differentiation, or apoptosis. A group of enzymes called DNA methyltransferases (DNMTs) catalyze the methylation of cytosine residues in newly synthesized DNA, thus replicating the methylation signal.

Multiple genes appear to be hypermethylated in MDS, including p15^{INK4B}, which encodes a cell-cycle inhibitor. Studies suggest that p15^{INK4B} methylation is correlated with blastic bone marrow involvement, may allow leukemic cells to proliferate and that it increases during MDS progression to AML. Recently, investigators have studied the effect of inhibition of these enzymes in MDS, and found that this resulted in DNA hypomethylation and re-expression of tumor suppressor genes. Cytosine analogues such as decitabine have been shown to inhibit DNMT.

Decitabine and myelodysplastic syndromes therapy

MDS patients are a challenging treatment group, due to their advanced age, comorbidities, and inability to tolerate intensive therapy. The only potentially curative treatment at this time for MDS is hematopoietic stem cell transplantation (HSCT). Unfortunately, HSCT is available for only a small number of patients (i.e., younger age, histocompatible donor, no significant comorbidities). Although some high-risk MDS patients may be eligible for intensive anti-leukemic chemotherapy, most MDS patients are managed with supportive care, including red blood cell (RBC)/platelet transfusions, growth factors such as recombinant erythropoietin and granulocyte colony-stimulating factors, and antibiotics.

A number of therapeutic options are being evaluated or have been recently approved for the treatment of MDS. These include tipifarnib, lenalidomide (FDA approved in December, 2005 for MDS 5q deletion patients), and imatinib (FDA approved in...
October 2006 for myelodysplastic/myeloproliferative diseases [MDS/MPD]). In addition, the hypomethylating agents decitabine and azacytidine have been recently approved by the FDA for the treatment of MDS.

Decitabine (5-aza-2’-deoxycytidine) is a cytosine analogue that distributes extensively throughout human tissues. Although the exact route of elimination and metabolic fate of decitabine is unknown in humans, one elimination pathway for decitabine appears to be deamination by cytidine deaminase found in the liver as well as in granulocytes, intestinal epithelium and whole blood. Decitabine treatment has been shown to reverse hypermethylation of p15\textsuperscript{INK4B} allowing for re-establishment of normal p15\textsuperscript{INK4B} protein expression.

Decitabine is believed to have a dual mechanism of action depending on dose. At both lower and higher doses, decitabine incorporates into DNA; however, at higher doses, decitabine inhibits cell proliferation through nonreversible covalent linking with DNA methyltransferase and blocking of DNA synthesis. At lower doses, decitabine induces hypomethylation, thereby promoting cell differentiation, re-expression of tumor suppressor genes, stimulation of immune mechanisms, and suppression of tumor growth.

Phase 2 trials of decitabine in MDS patients yielded overall responses of 26-45% and complete responses of 21-28%. These results led to a North American, multicenter phase 3 study of decitabine compared with supportive care in 170 MDS patients, which formed the basis for the FDA approval of decitabine. In this study, patients were stratified by IPSS risk group and MDS type and randomly assigned to receive supportive care alone or decitabine (15 mg/m\textsuperscript{2} as a 3-hour infusion every 8 hours for 3 days, repeated every 6 weeks) plus supportive care. Primary endpoints were overall response rate (ORR) and time to AML transformation or death. Responses were assessed using the International Working Group (IWG) criteria. Response criteria had to be met for at least 8 weeks.

The results of the phase III study indicated that decitabine is clinically effective in patients with MDS. Patient baseline characteristics were well balanced between the two study arms. The overall response rate (ORR) of patients in the decitabine arm was 17% compared with 0% in the supportive care only arm (p<0.001). In decitabine-treated patients with pathologically confirmed MDS at baseline who received at least two cycles of treatment, the ORR was 21% (12/56). Responses were observed across all IPSS risk groups and were durable, with a median duration of response of 10.3 months. Median time to first response was 3.3 months. Hematologic improvement (HI) was observed in an additional 13% of patients in the decitabine group versus 7% in the supportive care arm. The overall improvement rate for patients receiving decitabine was 30% versus 7% for patients receiving supportive care. For patients in the decitabine arm, the median time to AML progression or death that was 4.3 months greater than that of patients in the supportive care only arm (p=0.16). Patients receiving decitabine experienced a longer time to AML or death than patients receiving supportive care only.

All responders in the phase 3 study, defined as patients achieving a CR or PR, became RBC and platelet transfusion independent in the absence of growth factors during the time of the response. The percentage of patients in the decitabine arm who became RBC transfusion independent increased with increased number of treatment cycles, while the percentage of patients in the supportive care arm who required RBC transfusions did not change. Eight of the responding patients had cytoge-
netic abnormalities at baseline. All eight of these responders with cytogenetic abnormalities at baseline were evaluable for cytogenetic response, and all achieved a cytogenetic response (seven major responses and one minor response). The median number of cycles delivered was three, with 43 of 89 patients receiving two or more cycles. Of the 15 patients who responded after decitabine treatment, the median number of courses was six. In contrast, the median number of cycles in the phase 2 studies was four, which may in part explain the slightly higher response rates in the phase 2 studies. Greater benefit may have occurred if the patients continued receiving decitabine therapy for a longer period of time.

Decitabine therapy was well tolerated with manageable adverse effects. Febrile neutropenia occurred in 28% of patients who received decitabine. As a result of these studies, decitabine was FDA-approved in May 2006 for the treatment of patients with MDS, including previously treated or untreated, de novo or secondary MDS of all FAB subtypes and intermediate-1, intermediate-2, and high-risk IPSS groups. FDA-approved decitabine dosing for MDS is 15 mg/m² via a 3-hour continuous infusion three times a day for 3 days for the first treatment cycle, repeated every 6 weeks. It is recommended that patients be treated for a minimum of four cycles with dose adjustments and delays as required; however, it is noted that a complete or partial response may take longer than four cycles. Decitabine treatment is associated with myelosuppression, so regular complete blood counts are recommended. Myelosuppression and worsening neutropenia may occur more frequently in the first or second treatment cycles, and may not necessarily indicate progression of underlying MDS.

Decitabine has also been studied in relapsed MDS patients who had previously responded to the drug. Rüter and colleagues reported on 22 patients who received decitabine retreatment at the time of disease relapse. These patients had initially received a median of six courses of decitabine (range 2-6 courses), which resulted in a CR in 55% (12 of 22 patients), a PR in 27% (6 of 22), and a hematologic improvement in 18% (4 of 22). Decitabine retreatment was initiated at a median of 11 months after the last course of initial treatment. In the retreatment stage, patients received a median of three courses, resulting in 45% (10 of 22 patients) of patients responding (7 HI, 2 PR, and 1 CR). The median duration of second response was 4 months. Because the quality and duration of the second response were inferior to those of the first response, the authors suggest that decitabine responders may benefit from a longer period of initial treatment.

As previously discussed, decitabine is believed to have a dual mechanism of action depending on dose, with higher doses associated with cytotoxicity and lower doses associated with demethylation. Because of this dose-dependent mechanism of action, lower-dose schedules of decitabine may be safer and more effective than higher dose ones. Indeed, early studies of decitabine using high doses of the drug showed activity in various types of hematologic malignancies but with significant prolonged myelosuppression. Issa et al has shown that decitabine appears significantly more active at lower doses compared with higher doses.

Kantarjian and colleagues performed a randomized study of three low-dose schedules in patients with MDS and chronic myelomonocytic leukemia (CMML). The decitabine dose per course was reduced from the total FDA approved dose of 135 mg/m² to 100 mg/m². In addition, the doses of decitabine were delivered every 4 weeks (rather than every 6 to 8 weeks), as long as there was persistent disease and no significant myelosuppression-associated complications, and thera-
py was continued for three or more courses before response was evaluated. The three dosing schedules were as follows: ARM A: 20 mg/m$^2$ intravenously (IV) over 1 hour daily for 5 days, ARM B: 10 mg/m$^2$ IV over 1 hour daily for 10 days, and ARM C: 10 mg/m$^2$ subcutaneously (SQ) given twice daily for 5 days.

The results of this study showed that lower dose schedules of decitabine have significant anti-MDS activity. In total, 32 patients had a complete response (34%), 1 patient had a partial response (1%), 23 patients (24%) had marrow complete response, and 13 patients had hematological improvement (14%). When analyzed by schedule, the complete response rates were 39% for Arm A, 24% for Arm B, and 21% for Arm C. Dosing in Arm A (20 mg/m$^2$ IV over 1 hour daily for 5 days) was found to be superior to the other two regimens and is a therapeutic option in addition to the FDA approved dose.

Azacytidine and myelodysplastic syndromes therapy

Azacytidine is the only other approved hypomethylating agent for the treatment of MDS. Although similar in structure to decitabine, azacytidine is incorporated primarily into RNA and to a much lesser extent into DNA. This difference could account for the approximately 10-fold higher potency of decitabine compared with azacytidine.$^{34}$

In a randomized phase 3 trial of azacytidine in patients with MDS, azacytidine produced results similar to decitabine, with an ORR (CR + PR) in the azacytidine arm of 16.2% compared to 0% in the supportive care arm ($p<0.0001$).$^{35,36}$ The CR rate in patients treated with azacytidine was 6.1% compared with 0% for patients treated with supportive care. Median time to AML or death was increased with azacytidine treatment (21 months compared with 13 months for supportive care). As expected for this class of agent, the most common treatment-related toxicity was myelosuppression.

Because of the lack of any head-to-head trials, it is difficult to compare the efficacy of decitabine and azacytidine. Differences in study design between the two completed phase 3 trials add to this difficulty. Patients in the phase 3 azacytidine study were able to stay on treatment longer, resulting in a median of nine treatment cycles,$^{35}$ compared to the phase 3 decitabine trial who received a median of three treatment cycles.$^{26}$ The median duration of MDS was 7.3 months in the decitabine study compared with 2.8 months in the azacytidine trial, suggesting that the decitabine patients had more aggressive disease. In addition, response criteria in the azacytidine trial were less rigorous, requiring a CR or PR for at least 4 weeks and not requiring disappearance of dysplastic changes, compared with the decitabine study in which response was determined using strict IWG criteria.

Hypomethylating agents could improve overall survival of MDS patients. Azacytidine has been compared to conventional care in higher risk MDS patients for overall survival advantage, and the results were reported by Fenaux et al.$^{37}$ Three hundred fifty eight patients were randomized to either azacytidine or conventional care (i.e. supportive care, low dose Ara-C or standard chemotherapy). After a median follow up for overall survival of 21.1 months in this study, AZA was found to be statistically superior to conventional care in this patient population ($p=0.0001$), with overall survival in the azacytidine arm of 24.4 months versus 15 months with conventional care.

In contrast, preliminary results of a 2002 study comparing decitabine vs. supportive care in terms of overall survival were announced by the EORTC in July of 2008.$^{38}$ In this study, decitabine was administered to elderly patients (≥60 yrs) not eligible for intensive chemotherapy. Preliminary results do not demonstrate a statistically significant survival advantage of
decitabine over supportive care, and final results of this study are pending. The FDA-approved dosage of 15 mg/m² over 3 hrs every 8 hrs x 3 days (total dosage of 135 mg/m² per cycle) was used in this study. We have learned, however, that lower dosages of decitabine (i.e., 100 mg/m² per cycle) may have more efficacy.12,33 The numbers of treatment cycles in this study were also limited. Further study in this area is needed. Ideally, a “head to head” study of decitabine vs. azacytidine on overall survival should be done. A study of this type is in process at our center.

Decitabine in other malignancies

**Acute myeloid leukemia**

Preliminary results from a recent phase 2 study in AML patients not eligible for induction chemotherapy suggest that decitabine is an active first-line treatment.39 Decitabine (135 mg/m² IV over 72 hours repeated every 6 weeks) was given for up to four courses. In decitabine sensitive patients, all-transretinoic acid (ATRA) was also administered (45 mg/m² per day for 28 days during the second course). In the 29 fully evaluable patients, a CR was observed in four patients (14%) and a PR was observed in five patients (17%).

Preliminary studies of decitabine in poor prognosis AML patients suggested its possible effectiveness, and have led to further investigation of its efficacy in AML.40,41 Blum et al. investigated the used of decitabine alone or in combination with valproic acid in 25 acute myeloid leukemia patients, 12 untreated and 13 with relapsed AML.42 Of 21 accessible patients, 11 responded (4 CR, 4 incomplete CR, and 3 PR). In a phase 1/2 study, 54 patients with MDS or AML received 15 mg/m² decitabine IV for 10 days along with increasing doses of valproic acid. Objective responses occurred in 22%, including 10 CR (19%). Results from this study suggest that the combination of decitabine and valproic acid has significant activity in patients with AML and MDS and this response was associated with changes in histone acetylation and DNA hypomethylation.43

De Lima et al. have studied the effectiveness of high dose decitabine, busulfan, cyclophosphamide and allogeneic transplantation for the treatment of leukemia patients (AML, ALL, CML, CMML).44 At 3.3 years (median) post transplant, 26% of the patients were alive and disease free. This group included 40% of the treated AML patients, and 10% (1/9) CML patients. This study suggests that decitabine may have a role in the conditioning regimen prior to stem cell transplant in leukemia patients. Cashen et al reported a case of AML treated with decitabine who achieved a CR and relapsed post therapy. This patient was retreated with decitabine, resulting in another CR. This report suggests the effectiveness of retreatment of relapsed AML patients with another course of decitabine.45

These results, taken in combination with the results for those AML patients included in the previously discussed MDS patient studies, suggest the effectiveness of decitabine alone or in combination with valproic acid as a therapeutic modality in AML patients.

**Chronic myeloid leukemia**

As already mentioned, decitabine may have a role in the therapy of chronic myeloid leukemia patients. Kantarjian et al. treated a total of 130 CML patients with 100 mg/m² over 6 hours every 12 hours for 5 days every 4-8 weeks. Objective responses occurred in 18 of 64 (28%) of the CML patients in the blastic phase, and in 28 of 51 (55%) of the patients in accelerated phase of CML.46 According to the investigators, decitabine appears to have significant activity in CML, and further studies are needed.

In a recent Phase II study by Oki et al., decitabine in combination with imatinib mesy-
late was used to treat 28 patients with accelerated or blastic phase of CML. These patients were treated with 15 mg/m² IV for 5 days a week for 2 weeks, and 600 mg oral imatinib. Nine patients (32%) had complete hematological responses, 1 (4%) had a partial response and 2 (7%) had hematologic improvement. Other studies have been done on the effectiveness of decitabine either alone or in combination with other agents in CML. These results suggest that decitabine has a place in the therapy of advanced CML patients.

Decitabine is also being evaluated in some solid tumors including renal, lung, colorectal, melanoma and cervical cancer.50,51,52

Summary

Hypermethylation has been linked to loss of control of normal cell growth, differentiation and/or apoptosis, leading to abnormal cell proliferation. Decitabine (5-aza-2′-deoxycytidine) is a hypomethylating agent that interferes with the hypermethylation seen in many hematological malignancies, including MDS, leukemias and many other cancers. Its use has been shown to be well tolerated with low toxicity. Decitabine has recently been approved for the treatment of MDS patients in a dosing schedule of 15 mg/m² in a 3 hour infusion every 8 hours for three days in 6 week cycles. Recent studies suggest that the efficacy of decitabine may be further optimized by allowing for multiple treatment cycles and by using lower-dose schedules such as a schedule of 20 mg/m² IV over 1 hour daily for 5 days. It is currently under investigation in other malignancies such as AML, CML and solid tumors such as renal cell, lung colorectal and others.

Further studies are needed in terms of effectiveness on overall survival and its use in combination therapy in MDS and other malignancies.

References
