To the editor:

Methylation of SHP1 gene and loss of SHP1 protein expression are frequent in systemic anaplastic large cell lymphoma

Anaplastic large cell lymphoma (ALCL) is a type of non-Hodgkin lymphoma of T/null-cell immunophenotype, characterized by anaplastic cytology and CD30 expression; a subset carries chromosomal abnormalities involving ALK, and these tumors aberrantly express anaplastic lymphoma kinase (ALK) protein. Several cellular signaling pathways are abnormally activated in ALK+ ALCL, owing to the tyrosine kinase activity of ALK. Signal transducer and activator of transcription-3 (STAT3), one of the cellular signaling molecules known to be oncogenic, is activated by the nucleophosmin (NPM)-ALK fusion product. STAT3 is constitutively activated in ALCL, in most ALK+ tumors, and approximately half of ALK- tumors. We also previously showed that Janus kinase-3 (JAK3), one of the normal physiologic activators of STAT3, is activated in ALK+ ALCL cell lines and tumors.

As reported in Blood by Chim et al., gene methylation of SHP1 is frequent in multiple myeloma and is associated with STAT3 activation. Importantly, restoration of Src homology 2 domain-containing protein tyrosine phosphatase (SHP1) expression by 5-azacytidine treatment is associated with down-regulation of the phosphorylated/activated form of STAT3. Loss of SHP-1 expression, which correlates with SHP1 methylation, also has been observed in peripheral T-cell lymphomas and types of B-cell lymphoma.

SHP1 is known as a negative regulator of STATs and JAKs. It is possible that loss of SHP1 may contribute to STAT3 activation in ALCL, similar to the scenario in multiple myeloma. We immunohistochemically surveyed SHP1 expression in ALCL tumors diagnosed according to criteria defined in the WHO classification. Of 44 cases, 36 ALCLs (16 ALK+, 20 ALK-) (82%) were negative for SHP1 (Figure 1A). There were 8 SHP1-positive ALCLs (3 ALK+, 5 ALK-). Using methylation-specific PCR and primer sets described previously, we surveyed 2 ALK+ ALCL cell lines, Karpas 299 and SU-DHL-1, and 14 ALCL tumors (5 ALK+ and 9 ALK-). Both cell lines and 9 ALCL tumors (3 ALK+ and 6 ALK-) (64%) showed SHP1 methylation (Figure 1B). None of the 9 cases with SHP1 methylation were SHP1-positive by immunohistochemistry. By contrast, 2 (40%) of 5 cases without SHP1 methylation were SHP1-positive (P = .027; Fisher exact test). These results indicate that loss of SHP1 expression is frequent in ALCL, regardless of ALK status, and that SHP1 gene methylation likely contributes to loss of SHP1 expression.

We also correlated SHP1 expression and STAT3 activation, assessed by immunohistochemistry for tyrosine phosphorylated STAT3 (pSTAT3). Using a 10% cutoff, 23 of 35 SHP1-negative cases and 5 of 7 SHP1-positive cases were pSTAT3-positive. Among pSTAT3-positive cases, the median percentage of pSTAT3-positive tumor cells was 80% in the SHP1-negative group, compared with 50% in the SHP1-positive group (P = .057; t test). These data suggest that loss of SHP1 expression contributes to increased STAT3 activation in ALCL tumors.

In summary, our data from studies of ALCL parallel those of Chim et al., who studied multiple myeloma. It is likely that lack of SHP1 expression contributes to constitutive activation of the JAK/STAT pathway in other tumor cell types and enhances the oncogenic potential of STAT3. It is of note that 3 cases in our series lacked SHP1 protein expression but had no evidence of SHP1 methylation, suggesting that alternative mechanisms that silence SHP1 expression are also likely to exist.

Joseph D. Khoury, George Z. Rassidakis, L. Jeffrey Medeiros, Hesham M. Amin, and Raymond Lai

Correspondence: Raymond Lai, Department of Laboratory Medicine and Pathology, University of Alberta and Cross Cancer Institute, 481 Walter MacKenzie Health Sciences Center, 8440 112 St, Edmonton, AB, Canada T6G 2B7; e-mail: raymondmail_65@yahoo.com.

References


To the editor:

Elimination of *Aspergillus* infection in allogeneic stem cell transplant recipients with long-term itraconazole prophylaxis: prevention is better than treatment

In their single-center report of antifungal prophylaxis after allogeneic stem cell transplantation (SCT),1 Marr et al refer to our multicenter randomized trial.2 However, the authors failed to note important differences between our study and theirs. In our study,2 we used a dose of oral itraconazole solution (200 mg 2 times daily) that was lower than the dose used by the Seattle group and administered it only after transplantation. Patients unable to tolerate oral medications were switched back to the intravenous form of itraconazole or fluconazole and not removed from the study. Despite a lower dose of itraconazole, mean trough plasma concentrations of itraconazole were greater than the 500-ng/mL level targeted for prophylactic efficacy.3 Proven invasive fungal infections occurred in 9% of the itraconazole patients and 25% of the fluconazole patients during the first 180 days after transplantation. The incidence of *Aspergillus* infection was reduced from 12% in the fluconazole group to 4% in the itraconazole group. Except for more frequent gastrointestinal side effects, itraconazole was well tolerated. The incidences of drug-related hepatotoxicity and renal toxicity were low in our study and much less than those reported by Marr et al. The incidences of hepatotoxicity in both the fluconazole patients and the itraconazole patients reported by the Seattle group were extremely high and much greater than those reported by this group and others in previous studies.3,6 Thus, other factors besides fluconazole or itraconazole were likely responsible for liver dysfunction in most of their patients.

On the basis of our results, we introduced long-term itraconazole as routine antifungal prophylaxis in all adult patients undergoing an allogeneic SCT at the University of California, Los Angeles (UCLA). Intravenous itraconazole (200 mg intravenously every 12 hours for 2 days followed by 200 mg intravenously every 24 hours) is started on day 1 after SCT and continued until time of engraftment. After engraftment, patients receive oral itraconazole solution (200 mg every 12 hours) until day 100 after transplantation. After day 100, oral itraconazole is continued in patients still requiring corticosteroids for prevention or treatment of graft-versus-host disease (GVHD). Both inpatients and outpatients unable to take oral therapy are returned to intravenous itraconazole. From December 2001 to December 2003, 73 allogeneic SCT patients received itraconazole prophylaxis. These patients were at high risk for *Aspergillus* infection (median age, 40 years; range, 18-64 years; advanced disease, 78%; previous SCT, 20%; unrelated donor, 41%; high-dose corticosteroids for prevention or treatment of GVHD, 86%; grades II-IV GVHD, 45%). None of the patients developed *Aspergillus* infection, which is significantly lower than the incidence of *Aspergillus* infection in similar UCLA allogeneic SCT patients receiving fluconazole prophylaxis before December 2001 (0% vs 13%; P = .004). The only invasive fungal infections were 3 cases of candidemia (2 itraconazole sensitive, 1 itraconazole resistant). Overall survival was 55%, but no deaths were related to fungal infection. Except for nausea and vomiting (19% incidence), itraconazole was well tolerated. There were no cases of hepatotoxicity or renal toxicity attributable to itraconazole, and we did not find it necessary to routinely monitor serum drug levels.

In summary, we conclude that *Aspergillus* can be safely eliminated as a significant pathogen in allogeneic SCT recipients when prophylactic itraconazole is administrated at a tolerable dose and started after the day of transplantation. In view of the difficulty of treating established *Aspergillus* infections in highly immunosuppressed patients, we believe that prevention of infection is clearly better than treatment.

Drew J. Winston, Christos Emmanouilides, Kathy Barton, Gary J. Schiller, Ronald Paquette, and Mary C. Territo

Correspondence: Stem Cell Transplant Program, Division of Hematology-Oncology, Department of Medicine, UCLA Medical Center, Los Angeles, CA 90095; e-mail: dwinston@mednet.ucla.edu.

References


Methylation of SHP1 gene and loss of SHP1 protein expression are frequent in systemic anaplastic large cell lymphoma

Joseph D. Khoury, George Z. Rassidakis, L. Jeffrey Medeiros, Hesham M. Amin and Raymond Lai