IDH1 mutations in patients with myelodysplastic syndromes are associated with an unfavorable prognosis

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ABSTRACT

Background
Myelodysplastic syndromes are a heterogeneous group of hematopoietic stem cell disorders with a high propensity to transform into acute myeloid leukemia. Heterozygous missense mutations in *IDH1* at position R132 and in *IDH2* at positions R140 and R172 have recently been reported in acute myeloid leukemia. However, little is known about the incidence and prognostic impact of *IDH1* and *IDH2* mutations in myelodysplastic syndromes.

Design and Methods
We examined 193 patients with myelodysplastic syndromes and 53 patients with acute myeloid leukemia arising from myelodysplastic syndromes for mutations in *IDH1* (R132), *IDH2* (R172 and R140), and *NPM1* by direct sequencing.

Results
We found that mutations in *IDH1* occurred with a frequency of 3.6% in myelodysplastic syndromes (7 mutations in 193 patients) and 7.5% in acute myeloid leukemia following myelodysplastic syndromes (4 mutations in 53 patients). Three mutations in codon R140 of *IDH2* and one mutation in codon R172 were found in patients with acute myeloid leukemia following myelodysplastic syndromes (7.5%). No *IDH2* R140 or R172 mutations were identified in patients with myelodysplastic syndromes. The presence of *IDH1* mutations was associated with a shorter overall survival (HR 3.20; 95% CI 1.47-6.99) and a higher rate of transformation into acute myeloid leukemia (67% *versus* 28%, P=0.04). In multivariate analysis when considering karyotype, transfusion dependence and International Prognostic Scoring System score, *IDH1* mutations remained an independent prognostic marker in myelodysplastic syndromes (HR 3.57; 95% CI 1.59-8.02; P=0.002).

Conclusions
These results suggest that *IDH1* mutations are recurrent molecular aberrations in patients with myelodysplastic syndromes, and may become useful as a poor risk marker in these patients. These findings await validation in prospective trials.

Key words: *IDH1*, *IDH2*, myelodysplastic syndrome, secondary acute myeloid leukemia, prognosis.

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Introduction

Myelodysplastic syndromes (MDS) are a heterogeneous group of hematopoietic stem cell disorders. They are characterized by two cardinal features: ineffective hematopoiesis leading to bone marrow failure and a propensity to transform into acute myeloid leukemia (AML). While significant progress has been made in revealing cytogenetic and molecular changes in AML[1,2] less is known about the molecular changes that can lead to MDS. A number of non-specific recurrent mutations have been described in MDS including *NRAS*,[3] *TP53*,[4] *RUNX1*,[5,6] and *FMS*,[7,8] with the recent additions of *TET2*[9,10] and *ASXL1*.[11,12] However, given the heterogeneity of the disease it is of major importance to characterize MDS patients better at the molecular level, and to evaluate the prognostic relevance of new mutations.

While clinical scoring systems such as the World Health Organization (WHO) adapted Prognostic Scoring System (WPSS) and the International Prognostic Scoring System (IPSS)[13,14] can help to stratify patients according to their risk of death and leukemic progression, good molecular prognostic markers are still lacking.[15]

Isocitrate dehydrogenase 1 gene (*IDH1*) encodes for the protein isocitrate dehydrogenase 1, an enzyme that participates in the citric acid cycle. It catalyzes the carboxylation of isocitrate to alpha-ketoglutarate. Recurrent mutations in *IDH1* have been described in 12% of patients with glioblastomas[16] as well as in 70% of patients with WHO grade II and III astrocytomas and oligodendrogliomas.[17] In these entities *IDH1* mutations affect one single amino acid residue in position 132 leading to a switch from arginine to histidine (R132H).[17] At a lower frequency, mutations in *IDH2* affecting the analogous amino acid (R172) have also been described in oligodendrogliomas and astrocytomas.[17] Interestingly, the same *IDH1* mutation at position 132 was found when sequencing the genome of a patient with AML-M1.[18] Since this first description of *IDH1* mutations in AML several reports have confirmed that *IDH1* mutations occur in patients with cytogenetically normal AML with a frequency of 5.5-11%.[19-21] Additionally, a strong association between *IDH1* mutations with intermediate risk karyotype and concurrent *NPM1* mutations was found.[19-21] A recent study in AML patients showed a low frequency of mutations of codon R172 of *IDH2*.[21] A novel mutation in codon R140 of *IDH2* was identified in two patients with leukemic transformation of myeloproliferative neoplasms as well as in patients with AML.[22-24]

Little is known about the incidence and prognostic impact of *IDH1* and *IDH2* mutations in MDS patients. In this study, we examined the DNA of 193 patients with MDS for the presence of mutations in *IDH1* (R132), *IDH2* (R172 and R140), and *NPM1* by direct sequencing, and evaluated the prognostic impact of these mutations.

Design and Methods

Patients

Samples from 193 patients with MDS and 53 with AML with a prior history of MDS were collected at the time of enrollment in clinical trials. All patients with MDS were enrolled in multicenter treatment trials that investigated the use of all-*trans* retinoic acid,[25] antithymocyte globulin,[26] deferasirox,[27] lenalidomide, or thalidomide for the treatment of MDS while demethylating agents were not employed in this cohort of patients.

The diagnosis of AML arising from MDS was based on history, cytogenetics and morphology. All patients with secondary AML were treated within a trial of 5-aza-2'-deoxycytidine (decitabine) (Clinical Trials Identifier NCT00866073) or multicenter treatment trials AML SHG 0295 and AML SHG 0199 (ClinicalTrials Identifier NCT00209833). Details of the treatment protocols have been reported previously.[28,29] Clinical and hematologic data were recorded after patients had given their informed consent in accordance with the Declaration of Helsinki, and the scientific analysis of the samples was approved by the institutional review board of Hannover Medical School (n. 2467). According to the WHO classification, patients were classified as having refractory anemia (n=38), refractory anemia with ringed sideroblasts (n= 20), MDS with isolated del(5q) (n= 18); refractory cytopenia with multilineage dysplasia (n=30); refractory anemia with excess blasts-1 (n=22), refractory anemia with excess blasts-2 (n=31) and MDS-unclassifiable (n=7). Information on WHO subtype was not available for 27 patients. The IPSS stratification was low in 39 patients, intermediate-1 in 57, intermediate-2 in 38, and high in 13 (information on IPSS score was not available for 46 patients). Follow-up samples were available for 35 patients (median follow-up, 226 days; range, 13-988 days). Seven patients had had samples taken while they had MDS and also after their disease had progressed to AML (Figure 1). Follow-up information was available for 153 of the 193 patients with MDS. The follow-up information was updated by means of clinic visits as well as telephone calls to patients, their doctors, and local registry offices.

Cytogenetic analysis and mutation analysis of IDH1/2 and NPM1

Cytogenetic analysis was performed centrally by G- and R-banding analysis. Mutation analysis was performed as described previously.[30] Mononuclear cells from patients' samples were enriched by Ficoll density gradient centrifugation and were stored at -196°C in liquid nitrogen until use. Genomic DNA was extracted from samples using the All Prep DNA/RNA Kit (Qiagen, Hilden, Germany) according to the manufacturer's recommendations. The genomic region that spans the wild-type R132 of *IDH1* (exon 4) was amplified using polymerase chain reaction (PCR) with the following primers: 5`TGTGTTGAGATGGACGCCTATTTG and 5`TGCCACCAACGACCAAGTCA as previously described.[16] The following primers were used for amplification of the genomic region that spans wild-type R140 and R172 of *IDH2* (exon 4) using PCR: 5`GGGGTTCAAATTCTGGTTGA and 5`CTAGGCGAGGAGCTCCAGT. PCR fragments were directly sequenced, and were analyzed using Sequencing Analysis 5.3.1 software (Applied Biosystems, Darmstadt, Germany) and Vector NTI Advance 10 software (Invitrogen, Karlsruhe, Germany). Point mutations were confirmed in independent experiments. Genomic DNA was analyzed for *NPM1* mutations as previously described.[31]

Statistical analysis

Overall survival end-points, measured from the date of first sample collection, were death (failure) and alive at last follow-up (censored). Event-free survival end-points, measured from the date of first sample collection, were progression to AML or death (failure), and alive without progression of disease to AML at time of last follow-up (censored). Progression to AML was defined according to the 2008 WHO classification The median follow-up times for overall survival and event-free survival were calculated according to the method of Korn.[32] The primary analysis was performed on overall survival. Sensitivity analysis was performed on event-free survival, and the results are dis-

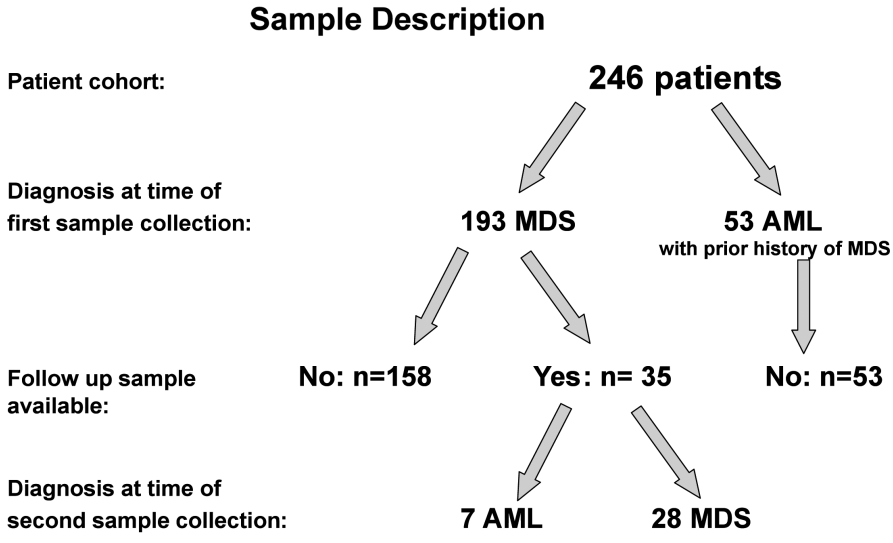

Figure 1. Description of patients' samples. MDS, myelodysplastic syndrome; AML, acute myeloid leukemia

played for exploratory purposes. Pair-wise comparisons were performed using two-sided Kolmogorov-Smirnov tests for continuous variables and by two-sided χ^2 tests for categorical variables, and are provided for exploratory purposes. For multivariate analysis, a Cox proportional hazards model was constructed for overall survival and event-free survival, adjusting for potential confounding covariates.[33] Variables considered for inclusion in the model were karyotype (favorable *versus* intermediate risk *versus* high risk), IPSS (low/int-1 *versus* int-2/high), transfusion dependence (yes *versus* no), ferritin level (above or below 1000 μg/L), age (below *versus* above median), number of therapies (best supportive care *versus* at least one other treatment), and *IDH1* mutation status. Variables with a *P* value of 0.05 or less in the univariate analysis for overall survival or event-free survival were included in the model. The two-sided level of significance was set at *P* less than 0.05. The statistical analyses were performed with the statistical software package SPSS 17.0 (SPSS Science, Chicago, IL, USA).

Results

Mutation status of IDH1/2 in patients with myelodysplastic syndromes

Among 193 patients with MDS, seven patients (3.6%) had a heterozygous mutation in codon 132 of *IDH1*. Six patients showed conversion of CGT to TGT leading to an Arg132Cys substitution and one patient had a CGT to CAT conversion leading to an Arg132His substitution (Table 1). Follow-up samples were available for 35 of these 193 patients (median follow-up, 226 days) and examined in addition to the first sample. One of these patients had an identical mutation in *IDH1* codon 132 at the time of MDS (refractory anemia) and in the follow-up sample at the time of AML transformation 2.7 years later. In the remaining 34 cases with follow-up samples, including six patients for whom we had a follow-up sample at the time of progression to AML, no *IDH1* mutation was found. Mutations in codon 140 or 172 of *IDH2* were not identified in any of the 193 MDS patients. Only one *NPM1* mutation was found in a MDS patient with wild-type *IDH1* and *IDH2*.

Mutation status of IDH1/2 in patients with acute myeloid leukemia following myelodysplastic syndrome

Among the 53 AML patients with a previous history of MDS, four patients (7.5%) had *IDH1* mutations in codon 132. Two patients showed conversion of CGT to CAT, one patient had conversion of CGT to TGT, and one patient had conversion of CGT to AGT leading to an Arg132Ser substitution. Three out of 53 patients with AML following MDS were found to have mutations in codon R140 of *IDH2*. All showed a conversion of CGG to CAG leading to an arginine to glutamine substitution. One patient with AML following MDS had a mutation in codon 172 of *IDH2* causing a conversion of AGG to AGT which leads to an Arg172Ser substitution (Table 2). Thus, 15% of AML patients with a history of MDS had mutated *IDH1* or *IDH2*. Mutated *NPM1* was found in three of 33 studied patients with AML following MDS; of these, one also had mutated *IDH1*.

Patients' characteristics in relation to IDH1/2 mutations

The clinical and hematologic characteristics of patients with and without mutations are compared in Table 3. There were no differences in age, sex, WHO classification, karyotype, bone marrow blasts, hemoglobin, transfusion dependence, ferritin, IPSS score, or number of treatments between the patients with *IDH1* mutations and those with the wild-type gene.

Prognostic impact of IDH1 mutations

The prognostic impact of *IDH1* mutations was evaluated in MDS patients for whom follow-up information was available (n=153). The median follow-up of patients alive was 3 years. In univariate analysis the overall survival of patients with *IDH1* mutation was significantly shorter than that of patients with wild-type *IDH1* (HR 3.20; 95% CI 1.47-6.99; Figure 2A). The 2-year survival rate was 52% and 14% for MDS patients with wild-type and mutated R132 *IDH1*, respectively. In univariate analysis IPSS score (high/int-2 *versus* low/int-1; HR 2.05; 95% CI 1.28-3.74; *P*=0.003), transfusion dependence (dependent *versus* independent; HR 3.62; 95% CI 1.65-7.93; P=0.001) and karyotype (high *versus* intermediate *versus* low risk; HR 1.91;

95%CI 1.44-2.52; *P*<0.001) were also identified as prognostic factors for overall survival. In multivariate analysis including *IDH1* mutation status, IPSS score, transfusion dependence, and karyotype, the presence of an *IDH1* mutation was found to be an independent unfavorable prognostic factor for overall survival (Table 4). Among the 186 MDS patients with wild-type *IDH1*, information about progression to AML was available for 145 patients. Among these, 41 developed AML (28.4%). Of the seven MDS patients with *IDH1* mutations, information about progression to AML was available for six patients; of these, four developed documented AML (67%, *P*=0.04). Among the remaining two patients without documented progression to AML, one patient had a leukocyte count of 70.2×10^9/L with 1% blasts in the differential count at the time of death. Due to critical illness a bone marrow biopsy was not obtained to confirm the diagnosis of AML, and the patient died shortly thereafter. In univariate analysis patients with mutations in codon R132 of *IDH1* had significantly lower event-free survival (HR 2.37; 95%CI 1.10-5.11) (Figure 2B). Furthermore, in univariate analysis for event-free survival, IPSS score (high/int-2 *versus* low/int-1; HR 2.00; 95% CI 1.39-2.86; *P*<0.001), transfusion dependence (dependent *versus* independent; HR 2.29; 95% CI 1.46-3.52; *P*<0.001), and karyotype (high *versus* intermediate versus low risk; HR 1.68; 95% CI 1.33-2.12; *P*<0.001) were identified as prognostic factors for event-free survival. In multivariate analysis including *IDH1* mutation status, IPSS score, transfusion dependence, and karyotype, the presence of *IDH1* mutations was found to be an independent unfavorable prognostic factor for event-free survival (Table 4).

Table 1. Details of *IDH1* mutations in patients with myelodysplastic syndromes.

Patient	Age	Sex	WHO classification	FAB classification	IPSS classification	Karyotype	Nucleotide change	sAML	Survival (years), status#	Therapy
1	68	F	RCMD	RA	Low	46,XX	CGT(R) > TGT(C)	No	1.05 (d)	Deferasirox
2	50	M	RCMD	RA	Int-1	46,XY	CGT (R) > TGT(C)	Likely*	0.34 (d)	ATG
3	70	F	del(5q)	RA	Int-1	46,XX,del(5)(q?14q?34)	CGT (R) > TGT(C)	Yes	0.11 (d)	ATRA
4	58	M	RA	RA	Low	46,XY	CGT (R)> CAT(H)	Yes	4.31 (d)	ATG
5	71	F	RAEB-2	RAEB	High	46,XX	CGT (R) > TGT(C)	Yes	1.18 (d)	Thalidomide
6	75	M	RAEB-1	RAEB	Int-1	46,XY	CGT (R) > TGT(C)	Unknown	2.08 (d)	ATG
7	63	M	RAEB-2	RAEB	High	45,XY,-7	CGT (R) > TGT(C)	Yes	0.32 (d)	ATG

*F: female; M: male; RA: refractory anemia; del5q, MDS with isolated del(5q); RCMD, refractory cytopenia with multilineage dysplasia; RAEB-1, refractory anemia with excess blasts-1; RAEB-2, refractory anemia with excess blasts-2; IPSS, International Prognostic Scoring System; int-1, intermediate-1; sAML, secondary acute myeloid leukemia after a prior diagnosis of myelodysplastic syndrome; (R), Arginine; (C), Cysteine; (H), Histidine; (S), Serine; # survival status; d: dead; ATG: antithymocyte globulin; ATRA: all-*trans *retinoic acid *At time of death patient's white blood cell count was 70.2x10^9/L with 1% peripheral blood blasts, 7% myelocytes, and 8% metamyelocytes. Bone marrow was not biopsied prior to the patient's death. Thus, the development of AML is likely, but not proven by bone marrow biopsy.*

Table 2. Details of *IDH1/2* mutations in acute myeloid leukemia patients with a previous history of myelodysplastic syndromes.

Patient	Age	Sex	Karyotype	Gene	Nucleotide change	Survival (years), status#	Treatment
1	66	M	55,XY,+X,+Y,+12,+13,+14,+19,+20,+21,+22[35]	*IDH1*	CGT(R)>TGT(C)	0.13 (d)	Decitabine
2	52	F	46,XX[36] 47,XX,+4[35]	*IDH1*	CGT(R)>AGT(S)	0.58 (d)	Conventional chemotherapy
3	59	M	46,XY	*IDH1*	CGT(R)>CAT(H)	0.52 (d)	Conventional chemotherapy
4	43	M	46, XY	*IDH1*	CGT(R)>CAT(H)	7.99 (a)	Conventional chemotherapy
5	59	M	46,XY	*IDH2 R140*	CGG(R)>CAG(Q)	1.70(d)	Conventional chemotherapy
6	59	M	46,XY	*IDH2 R140*	CGG(R)>CAG(Q)	0.07(d)	Conventional chemotherapy
7	75	M	45,XY,-7	*IDH2 R140*	CGG(R)>CAG(Q)	0.67(d)	Decitabine
8	48	F	46, XX	*IDH2 R172*	AGG(R)>AGT(S)	0.58 (d)	Conventional chemotherapy

F,female; M, male; (R), Arginine; (C), Cysteine; (H), Histidine; (S), Serine; (Q), Glutamine; # survival status; d, dead; a, alive.

Discussion

In the present study we found that *IDH1* mutations were a recurring molecular aberration in MDS patients, occurring with a frequency of 3.6%. Moreover, mutated *IDH1* was associated with a high rate of leukemic transformation, and poor event-free and overall survival rates. The rate of *IDH1* R132 mutations in MDS patients was lower than the rate in patients with AML arising from MDS or the reported rate in patients with *de novo* AML.[18,20,34] We did not identify any *IDH2* R172 or R140 mutations in MDS patients. However, we demonstrated that mutations of *IDH2* occur in AML patients with a prior history of MDS. We, therefore, showed that *IDH1* mutations are rare but recurrent molecular aberrations in MDS patients, and establish *IDH1/2* mutations as one of the most frequent mutations in AML arising from MDS (15%). In our cohort of MDS patients, *NPM1* mutations were not identified in patients with *IDH1* mutations, and the rate of *NPM1* mutations was very low. Given the low frequency of *NPM1* mutations in our cohort of patients we could not evaluate an association of *NPM1* and *IDH* mutations previously found in AML patients.

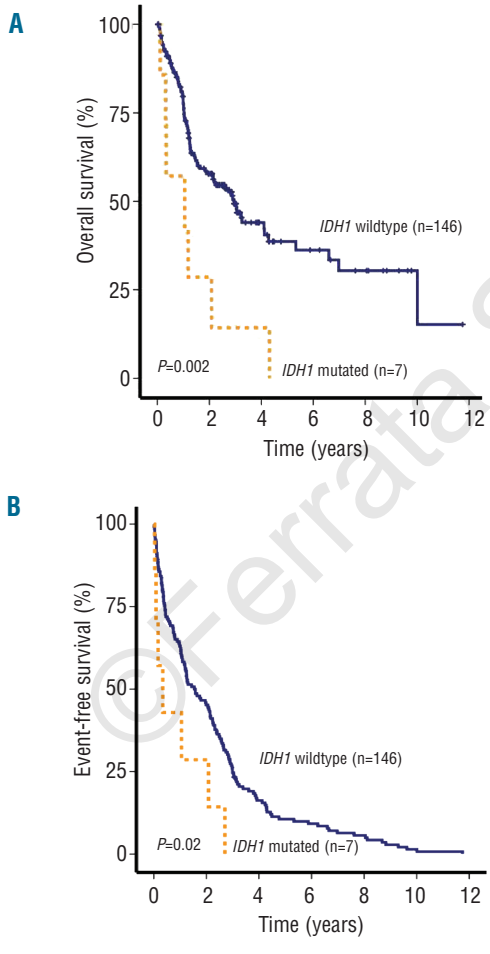

Figure 2. Kaplan-Meier curves for overall and event-free survival. (A) Overall survival in MDS patients with mutated (n= 7) and unmutated (n=146) *IDH1* (log-rank test, *P*=0.002). (B) Event-free survival in MDS patients (n=153) with mutated (n= 7) and unmutated (n=146) *IDH1* (log-rank test, *P*=0.02).

In this study, mutated *IDH1* was an independent unfavorable prognostic marker for both event-free survival and overall survival of patients with MDS. Different

Table 3. Comparison of clinical and molecular characteristics of MDS patients with mutated or wild-type *IDH1*.

Characteristic	All n=193	*IDH1* mutated n=7 (3.6%)	*IDH1* wildtype n=186 (96.4%)	*P*
Age, years				
Median		68	66	0.8
Range	36-92	50-75	36-92	
Sex				0.8
Male - n. (%)	119	4 (57)	115 (62)	
Female - n. (%)	74	3 (42)	71 (38)	
WHO-Subtype				0.7
RA - n. (%)	38	1 (14)	37 (20)	
RARS - n. (%)	20	0 (0)	20 (11)	
del5q- - n. (%)	18	1 (14)	17 (9)	
RCMD - n. (%)	30	2 (29)	28 (15)	
RAEB-1 - n. (%)	22	1 (14)	21 (11)	
RAEB-2 - n. (%)	31	2 (29)	29 (16)	
MDS-U (%)	7	0 (0)	7 (4)	
Missing data - n. (%)	27	0 (0)	27 (14)	
Karyotype risk				0.56
Low - n. (%)	109	6 (86)	103 (55)	
Intermediate - n (%)	20	0 (0)	20 (11)	
High - n (%)	23	1 (14)	22 (12)	
Missing data - n. (%)	41	0 (0)	41 (22)	
Bone marrow blasts				0.78
<5% - n. (%)	108	4 (57)	105 (56)	
>5% and <10% - n. (%)	24	1 (14)	21 (11)	
10-20% - n. (%)	30	2 (29)	29 (16)	
Missing data - n. (%)	31	0 (0)	31 (17)	
Hemoglobin				0.91
<8 g/L (%)	35	2 (29)	33 (18)	
8-10 g/L – n. (%)	78	3 (42)	75 (40)	
>10 g/L - n. (%)	44	2 (29)	42 (23)	
Missing data - n. (%)	36	0 (0)	36 (19)	
Transfusion dependence				0.18
Yes - n (%)	124	7 (100)	117 (63)	
No - n. (%)	30	0 (0)	30 (16)	
Missing data - n. (%)	39	0 (0)	39 (21)	
Ferritin				0.24
Median – µg/L (%)		764	881	
<1000 µg/L- n. (%)	72	4 (57)	68 (36)	
>1000 µg/L- n. (%)	60	1 (14)	59 (32)	
Missing data – n. (%)	61	2 (29)	59 (32)	
Transformation into AML				0.02
Yes (%)	45	4 (57)	41 (22)	
No (%)	129	2 (29)	127 (68)	
Missing data - n. (%)	19	1 (14)	18 (10)	
IPSS - n.				0.16
Low risk (%)	39	2 (29)	37 (20)	
Int-1 (%)	57	3 (42)	54 (29)	
Int-2 (%)	38	0 (0)	38 (20)	
High (%)	13	2 (29)	11 (6)	
Missing data - n. (%)	46	0 (0)	46 (25)	
Median number of treatments other than supportive care				0.95
Median	1.00	1.00	1.00	
Range	0-4	0-2	0-4	
Missing – n. (%)	42	0	42 (23)	

RA: refractory anemia; RARS: refractory anemia with ringed sideroblasts; del5q-, MDS with isolated del(5q); RCMD: refractory cytopenia with multilineage dysplasia; RAEB-1: refractory anemia with excess blasts- 1; RAEB-2: refractory anemia with excess blasts-2; MDS-U: MDS-unclassifiable; IPSS: International Prognostic Scoring System; AML: acute myeloid leukemia.

Table 4. Cox regression analysis for overall survival and event-free survival in MDS patients with mutated (n=7) or unmutated (n=146) *IDH1*.

	Overall survival			Event-free survival		
	HR	95% CI	*P*	HR	95% CI	*P*
***IDH1* mutation status**						
mutated *vs.* unmutated	3.57	1.59-8.02	0.002	2.66	1.21-5.83	0.02
IPSS-based karyotype						
high *vs.* intermediate *vs.* favorable risk	1.89	1.34-2.66	<0.002	1.52	1.16-1.99	0.002
Transfusion dependence						
dependent *vs.* independent	2.79	1.24-6.27	0.01	2.00	1.24-3.21	0.004
IPSS score						
high/int-2 *vs.* low/int-1	1.11	0.6-2.01	0.74	1.44	0.93-2.22	0.11

Hazard ratios greater than 1 indicate an increased risk of an event for the first category listed. HR: hazard ratio; CI: confidence interval.

treatment regimens did not influence prognosis in our cohort (*data not shown*), suggesting that treatment differences between patients were unlikely to have had a confounding effect on our analysis. The present study illustrates a potential new unfavorable prognostic marker in MDS patients, especially in patients with normal cytogenetics, which may become useful for treatment stratification in the future. Additional studies in larger cohorts of patients are warranted.

The intriguing finding that *IDH1* and *IDH2* mutations occur in the leukemic transformation of myeloproliferative neoplasms, but not in patients in chronic-phase polycythemia vera or essential thrombocythemia,[22] suggests that these mutations play an important role in leukemogenesis. Our results indicate that MDS patients with mutated *IDH1* undergo a high rate of leukemic transformation, which is in accordance with the data in myeloproliferative neoplasms.[22] Interestingly, no *IDH2* mutations were observed in MDS patients while the incidence of *IDH2* mutations in patients with AML arising from MDS was found to be 7.5%. Functional studies may clarify whether mutations in *IDH1* and *IDH2* have distinct effects on leukemic progression from MDS to AML.

In summary, we identified *IDH1* mutations of amino acid 132 in 3.6% of MDS patients, and found a strong correlation of mutated *IDH1* with unfavorable outcome in these patients. Because of the low frequency of *IDH1* mutations occurring in MDS the prognostic impact of the mutation should be confirmed in larger groups of uniformly treated MDS patients and put in context with other novel markers such as *TET2*, *ASXL1* and *RUNX1*. Our study also provides evidence that mutations of codons R140 and R172 of *IDH2* occur in patients with AML arising from MDS. Mutation analysis of *IDH1* in MDS patients may become useful for risk and treatment stratification in the future.

Authorship and Disclosures

The information provided by the authors about contributions from persons listed as authors and in acknowledgments is available with the full text of this paper at www.haematologica.org.

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