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Continuity and efficacy of real-world use of azacitidine

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Long-term azacitidine (AZA) treatment is necessary for its maximal therapeutic effect. This study examined the continuity and efficacy of AZA treatment in real-world use. We conducted a retrospective study in 38 patients who had completed AZA treatment at the Ogaki Municipal Hospital between April 2011 and August 2019. The median number of AZA received cycles was 4. The number of AZA treatment cycles received was 1-3 cycles in 15 (39.5%), 4-6 cycles in 15 (39.5%), and ≥ 7 cycles in 8 (21.1%). The most common reason for discontinued AZA treatment was infection. Overall response rate was 33.3% in patients with discontinued AZA use (< 4 cycles) and 56.5% in patients with continued AZA (≥ 4). Median overall survival (OS) was 124 (15-529) days and 391 (132-2,825) days in the respective groups (p<0.01). The presence of peripheral blood blasts (PBs) was a prognostic factor for continuation of treatment (p=0.03). Discontinued AZA treatment due to infection (p<0.01), and PBs (p=0.03) were unfavourable prognostic factors for OS. Long-term AZA use is beneficial for improvement and survival. Infection control and presence of PBs were important factors for continuing AZA. These data support the idea of long-term continued treatment with AZA for optimal benefit to patients.

1. Introduction

Myelodysplastic syndromes (MDSs) are a group of diseases with unexplained ineffective hematopoiesis and high risk of progression to acute myeloid leukemia (AML) (Tefferi et al. 2009). Prognosis is predicted using clinical factors, and MDS patients are treated based on their individual risk scores (i.e. low risk or high risk). The high-risk patients with a poor prognosis are treated aggressively. Allogeneic hematopoietic stem cell transplantation is the only way to cure MDSs. However, many patients with MDS are older and azacitidine (AZA) treatment is regarded as the first-line therapy in patients who cannot undergo transplantation (Silverman et al. 2002; Fenaux et al. 2009). In recent years, the effectiveness of AZA has been reported in elderly AML patients, and its frequency of use has increased (Dombret et al. 2015; Yamamoto et al. 2014). However, effectiveness of AZA may appear in late treatment phases. Therefore, assessments of effectiveness should be made following at least 4-6 treatment cycles (Silverman et al. 2006; Götze et al. 2010). It is necessary to determine whether AZA treatment can be used as a long-term treatment to ensure its maximum therapeutic effect. In a Japanese clinical trial for MDS patients, 79.2% of the patients were able to continue treatment for more than 4 cycles (Uchida et al. 2011). However, many patients have difficulty in continuing AZA treatment. Real-world data on long-term AZA treatment are scarce. Additionally, clinical trial data may not always reflect real-world clinical practice and patient outcomes. Thus, a retrospective analysis of AZA therapy was conducted to determine its continuity and efficacy in real-world use.

2. Investigations and results

2.1. Patient characteristics

The baseline characteristics of 38 patients who received AZA are listed in Table 1. A total of 191 cycles of AZA were administered. Median age (range) was 78 (55-88) years. Diagnosis included 23 (60.5%) cases of refractory anemia with excess blasts in transformation (RAEB-T), 7 (18.4%) RAEB, and 6 (15.8%) AML. IPSS-R indicated very high-risk in 18 (47.4%), high or intermediate in 6 (15.8%), and very low in 2 (5.3%). Cytogenetic risk was good in 17 (44.7%), intermediate in 3 (7.9%), and poor in 2 (5.3%), and very poor in 16 (42.1%). RBC transfusion dependency was 4 RBC Units/8weeks in 22 (57.9%), 1-3 RBC Units/8weeks in 9 (23.7%), and no RBC in 7 (18.4%). Peripheral blood blasts were present in 26 (68.4%), and absent in 14 (36.8%). Median neutrophil, platelets, and bone marrow blasts were 765.5/μL, $4.6x10^4$/ μL, and 9.7%, respectively.

2.2. Continuity of AZA treatment

The median number of AZA treatment cycles received among patients was 4 (1-28; Table 2). The number of AZA treatment cycles received was 1-3 cycles in 15 (39.5%), 4-6 cycles in 15 (39.5%), and ≥ 7 cycles in 8 (21.1%). The reasons of discontinued AZA are summarized in Table 3. Infection was the most common reason for AZA treatment termination (8 patients, 53.3%) in the discontinued AZA group and PD (7 patients, 30.4%) in the continued AZA group. Dose reduction or delays in treatment cycles of AZA are summarized in Table 4. Dose of AZA was reduced in 1 (6.7%) in the discontinued AZA group and 7 (30.4%) in the continued AZA group. The median treatment interval was 43 (28-82) and 32 (27-120) days in each group, respectively. There was postponement over 60 days in 3 (11.5%) and 12 (7.3%) cycles of treatment in each group, respectively.

2.3. AZA efficacy

The best response to AZA is summarized in Table 5. The overall response rate (CR+PR+mCR+SD with HI) was 5 (33.3%) patients responded to AZA in the discontinued AZA group, while 13 (56.5%) responded in the continued AZA group, though the patient improvements were not statistically significant. CR and PD rates were significantly different between the two groups (p=0.02, p<0.01). After a median follow-up of 37 months, 27 deaths had occurred, and median OS was 124 (15-529) days and 391 (132-2,825) days in each group, respectively with significant difference (p<0.01) (Fig.).

Table 1: Baseline patient characteristics

	N or medium	% or range
No. of patients	38	
No. of total AZA treatment (cycles)	191	
Age (year), median (range)	78	55-88
Sex		
Male	29	76.3
Female	9	23.7
Diagnosis		
RA/RARS	1	2.6
RAEB	7	18.4
RAEB-T	23	60.5
CMMoL	1	2.6
AML	6	15.8
IPSS-R		
Very low	2	5.3
Low	0	0.0
Intermediate	6	15.8
High	6	15.8
Very high	18	47.4
Cytogenetic risk (IPSS-R)		
Very good	0	0.0
Good	17	44.7
Intermediate	3	7.9
Poor	2	5.3
Very poor	16	42.1
ECOG PS		
0-1	33	86.8
≥ 2	5	13.2
RBC transfusion dependence		
4 RBC Units/8w	22	57.9
1-3 RBC Units/8w	9	23.7
No RBC	7	18.4
Neutrophil (per µl), median (range)	765.5	10-30,787
Platelets (x10^4 per µl), median (range)	4.6	0.9-44.9
Peripheral blood blasts		
Present	24	63.2
Absent	14	36.8
Bone marrow blasts (%), median (range)	9.7	0.6-64.8
Prior treatment regimens		
AML intensive treatment	6	15.8
AML salvage treatment	5	13.2
Other hematological cancer regimen	1	2.6
Solid tumor cancer regimens	4	10.5
Comorbidities		
Diabetes	7	18.4
Hypertension	11	28.9
Cardiovascular events	11	28.9
Chronic obstructive pulmonary disease	3	7.9
Hyperlipemia	8	21.1
Other cancer	8	21.1

Data are presented as number (%) or median (range). RA/ RARS, refractory anemia/refractory anemia with ringed sideroblasts-CB; RAEB, refractory anemia with excess blasts; RAEB-T, refractory anemia with excess blasts in transformation; CMMoL, chronic myelomonocytic leukemia; AML, acute myeloid leukemia; IPSS-R, Revised-International Prognostic Scoring System; ECOG PS, Eastern Cooperative Oncology Group performance status; RBC, red blood cell.

2.4. Investigation of the factors affecting continuation and OS

To determine baseline characteristics that could predict the need for early termination of AZA treatment (less than 4 cycles), the 38 patients were dichotomized and the results are shown in Table 6. In a univariate analysis, discontinued AZA treatment was observed in patients with infection (p=0.04) and PB (p=0.01). In a multivariate analysis with all factors found significant in the univariate analysis, only PB was an independent prognostic factor for less than 4 cycles of AZA treatment (odds ratio [OR]=7.67 [95% confidence interval: 1.49-63.11]; p=0.03). In a univariate analysis, shorter survival was observed in patients with discontinued AZA due to infection (p<0.01), cytogenetic risk (≥Poor) (p=0.04), and PB (p=0.02). In a multivariate analysis, only discontinued AZA treatment due to infection (hazard ratio [HR]=1.88 [1.23-2.86]; p<0.01), and PBs (HR=1.65 [1.06-2.80]; p=0.03) were unfavourable prognostic factors for OS (Table 7).

Table 2: Continuity of AZA treatment

	No. of patient	% or range
The number of AZA treatment cycles		
1	6	15.8
2	7	18.4
3	2	5.3
4	5	13.2
5	5	13.2
6	5	13.2
7	2	5.3
8	2	5.3
9	2	5.3
≥ 10	2	5.3
1-3 cycles	15	39.5
4-6 cycles	15	39.5
≥ 7 cycles	8	21.1
AZA treatment cycles, median (range)	4	1-28

Data are presented as number (%) or median (range). AZA, azacitidine.

Table 3: Reasons of discontinued AZA treatment

	discontinued AZA (n=15)	%	continued AZA (n=23)	%	p-Value
Infection complication	8	53.3	5	21.7	0.03
Progressive disease	1	6.7	7	30.4	0.11
Patient's request	1	6.7	3	13.0	1.00
Achieve Complete response	0	0.0	3	13.0	0.26
Allogeneic hematopoietic stem-cell transplantation	1	6.7	2	8.7	1.00
No therapeutic effect	0	0.0	2	8.7	0.51
Deterioration in performance status	1	6.7	1	4.3	0.55
Deterioration of comorbidities	2	13.3	0	0.0	0.14
Other	1	6.7	0	0.0	0.39

Data are presented as number (%). AZA, azacitidine.

Table 4: Dose reduction or delays in treatment cycles of AZA

	discontinued AZA (n=15)		continued AZA (n=23)		p-Value
	N or median	% or range	N or median	% or range	
Total AZA treatment (cycles)	26		165		
Dose reduction of AZA					0.06
Number of patients	1	6.7	7	30.4	
> 75%dose	1	6.7	1	4.3	
75%-50% dose	0	0.0	5	21.7	
< 50%	0	0.0	1	4.3	
Treatment Interval of AZA, median (range) (days)	43	28-82	32	27-120	0.06
≥ 35 days interval (cycles)	8	72.7	58	40.6	0.04
≥ 60 days interval (cycles)	3	11.5	12	7.3	0.07

Data are presented as number (%) or median (range). AZA, azacitidine.

Table 5: Best response to AZA according to IWG 2006 criteria

	Discontinued AZA (n=15)	%	continued AZA (n=23)	%	p-Value
Complete response (CR)	0	0.0	5	21.7	0.02
Partial response (PR)	2	13.3	3	13.0	0.98
Marrow CR (mCR)	1	6.7	1	4.3	0.76
Stable disease (SD) with hematological improvement (HI)	2	13.3	4	17.4	0.74
SD without HI	3	20.0	8	34.8	0.33
Progressive disease	7	46.7	2	8.7	< 0.01
Overall response rate (CR+PR+mCR+SD with HI)	5	33.3	13	56.5	0.17

Data are presented as number (%). AZA, azacitidine.

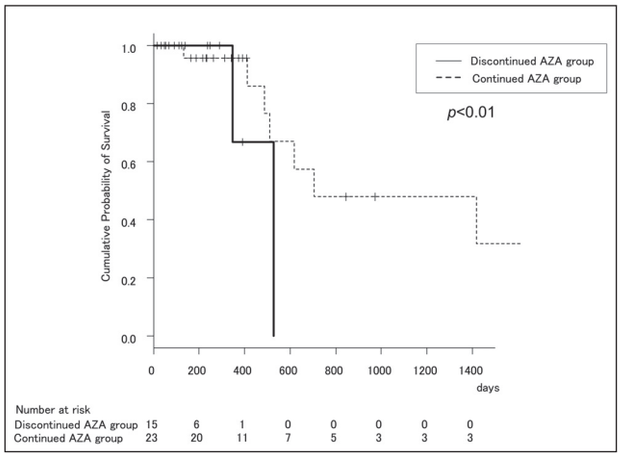

Fig.: Overall survival probability for Ogaki Municipal Hospital survivors with AZA treatment (n=38). Discontinued AZA group, continuous line; Continued AZA group, dotted line.

3. Discussion

AZA is a cytidine nucleoside analog, the activity of which against abnormal hematopoietic cells may be suppressed by demethylation of DNA and cytotoxic effects. The effectiveness of AZA has been reported for high-risk MDS and elderly AML patients. AZA use in clinical settings is expanding (Dombret et al. 2015; Yamamoto et al. 2014; Huls et al. 2019). A characteristic of AZA is its increased efficacy with long-term treatment. Once AZA treatment is stopped, aberrant promoter methylation and gene silencing return. Therefore, long-term treatment of AZA will be needed to maintain inhibition of DNA methylation (Herman et al. 2003). Although clinical data show the importance of continuing AZA treatment (Silverman et al. 2006; Götze et al. 2010), there are some patients who are unable to continue treatment in real-world use. Therefore, the purpose of this study was to clarify the factors affecting the tolerability of AZA treatment.

AZA was given to patients for a median of 9 cycles in a Phase 3 clinical trial (AZA-001). Patients with therapeutic effects could be treated for as long as 14 cycles, suggesting the importance of treatment continuation (Fenaux et al. 2009). In a Phase 2 study in a Japanese population, 79.2% of the patients were able to continue treatment for more than 4 cycles (Uchida et al. 2011). However, in this study, the median number of treatments was 4 cycles, and only 23 (60.5%) could continue treatment for more than 4 cycles. The number of cycles of AZA received was 1 cycles in 6 patients (15.8%), 2 cycles in 7 (18.4%), and less than 4 cycles in 15 (39.5%). As a reason for not being able to continue treatment, 13 (34.2%) patients were discontinued due to infections. There were significantly more patients who discontinued AZA treatment than those who continued it (p=0.03). In order to determine the effectiveness of AZA, it is important to prevent discontinuation of treatment by infectious complications. It was reported that low Hb level, low PLT below 20×10^9/L, and unfavourable cytogenetics are significant risk factors for infections in MDS patients treated with AZA (Merkel et al. 2013). In this study, none of these factors were identified as a significant risk factor for infectious complications. However, they are important factors in MDS risk classification, and infection control is especially important in the high-risk MDS group.

The median treatment interval in the AZA-001 trial was 28 days, with an interval of only 19% over 35 days. However, in this study, the median treatment interval for patients who were treated with more than 2 cycles was longer: 43 days in the discontinued AZA group and 32 days in the continued AZA group. Treatment intervals of more than 35 days were required in 72.7% and 40.6%, and more than 60 days in 11.5% and 7.3% of cases in each group, respectively. This was because 23 (60.5%) patients developed an infection during the treatment and needed to be given antibiotics. Preventing infectious will shorten treatment intervals.

Table 6: Factors related to the early termination of AZA treatment (less than 4 cycles)

	Univariate analysis			Multivariate analysis	
Factor	early termination of AZA treatment (< 4 cycles)	%	p-Value	OR (95% CI)	p-Value
Age			0.38		
< 75 y	5/16	31.3			
≥ 75 y	10/22	45.5			
ECOG PS			0.31		
0-1	12/33	36.4			
≥ 2	3/5	60.0			
Incidence of infection			0.25		
Yes	10/21	47.6			
No	5/17	29.4			
Discontinued AZA treatment due to infection			0.04	4.52 (0.99-25.39)	0.06
Yes	8/13	66.7			
No	7/25	28.0			
Cytogenetic risk (≥Poor)			0.21		
Yes	9/18	50.0			
No	6/20	30.0			
IPSS-R (≥very high)			0.52		
Yes	8/19	42.1			
No	4/13	30.8			
RBC transfusion dependence			0.38		
≥ 4 RBC Units/8w	10/11	45.5			
0-3 RBC Units/8w	5/16	31.3			
Neutrophil count of first AZA treatment			0.72		
≥ 1,000 per μl	5/14	35.7			
< 1,000 per μl	10/24	41.7			
Platelets count of first AZA treatment			0.85		
≥ 5.0 x10^4 per μl	7/17	41.2			
< 5.0 x10^4 per μl	8/21	38.1			
Peripheral blood blasts			0.01	7.67 (1.49-63.11)	0.03
Present	13/24	54.2			
Absent	2/14	14.3			
Marrow blasts			0.46		
≥ 15%	7/15	46.7			
< 15%	8/23	34.8			
Comorbidities			0.74		
Present	13/32	40.6			
Absent	2/6	33.3			

Data are presented as number (%). AZA, azacitidine; OR, odds ratio; CI, confidence interval; ECOG PS, Eastern Cooperative Oncology Group performance status; IPSS-R, Revised-International Prognostic Scoring System; RBC, red blood cell.

In the AZA-001 trial, 50.8% of the patients exhibited a favourable response (CR, PR, marrow CR or SD with HI) (Silverman et al. 2011). Itzykson et al. (2011) reported that 43.3% of the patients with higher-risk MDS treated with AZA showed clinical improve-

Table 7: Prognostic factors of overall survival

	Univariate analysis		Multivariate analysis	
Factor	Median OS	*p*-Value	HR (95% CI)	*p*-Value
Age		0.76		
< 75 y	343			
≥ 75 y	313			
ECOG PS		0.65		
0-1	342			
≥ 2	236			
Incidence of infection		0.16		
Yes	249			
No	390			
Discontinued AZA treatment due to infection		< 0.01		<0.01
Yes	215		1.88 (1.23-2.86)	
No	391		1	
Cytogenetic risk (≥Poor)		0.04		0.14
Yes	249		1.35 (0.91-2.04)	
No	391		1	
IPSS-R (≥very high)		0.11		
Yes	249			
No	845			
RBC transfusion dependence		0.26		
≥ 4 RBC Units/8w	236			
0-3 RBC Units/8w	374			
Neutrophil count of first AZA treatment		0.44		
≥ 1,000 per µl	313			
< 1,000 per µl	342			
Platelets count of first AZA treatment		0.99		
≥ 5.0 x10^4 per µl	344			
< 5.0 x10^4 per µl	290			
Peripheral blood blasts		0.02		0.03
Present	263		1.65 (1.06-2.80)	
Absent	974		1	
Marrow blasts		0.30		
≥ 15%	290			
< 15%	342			
Comorbidities		0.18		
Present	263			
Absent	436			

Data are presented as number (%). OS, overall survival; HR, hazard ratio; CI, confidence interval; ECOG PS, Eastern Cooperative Oncology Group performance status; AZA, azacitidine; IPSS-R, Revised-International Prognostic Scoring System; RBC, red blood cell.

ment. In this study, 47.4% of the patients showed a favourable response. Although there are differences in patient characteristics in each study, there is no difference in the overall response rate. There were no significant differences in response rate (≥ SD with HI) between the discontinued AZA group and continued AZA group in this study. However, there was a significant difference in CR and PD rate (p=0.02, p<0.01), and OS (p<0.01). It was clear that treatment continuation improved the effect of AZA.

Factors affecting treatment continuation and OS were investigated. The results of our study indicate that the presence of PBs was an independent factor affecting OS. Knipp et al. (2008) reported that the survival and risk of AML evolution in untreated lower risk MDS patients depended on the presence of PBs. Itzykson et al. (2011) reported that the presence of PBs was associated with shorter survival in high-risk MDS patients. The presence of circulating blasts is an independently predictive factor of OS without a relationship with MDS risk. These findings were consistent with our results. The presence of PBs may be an important factor in AZA treatment.

The limitations of this study include the fact that it was a retrospective study conducted at a single general hospital, as well as the fact that the number of patients included was small. We believe that the results would be useful as an exploratory study, but further investigations should be conducted in a larger number of patients to clarify the influence of the continued and efficacy of real-world use of AZA.

In conclusion, our results suggest that continued treatment of AZA is beneficial, and many patients were forced to discontinue AZA treatment due to infectious complications. Although there was no significant difference in the number of responding patients (those who achieved CR, PR, hematological improvement) between the discontinued AZA group and continued AZA group, there was a significant difference in OS (p <0.01). Infection control and the presence of PBs are important markers to identify patients who may or may not continue AZA treatment. These data support the idea of long-term continued treatment with AZA for optimal benefit to MDS patients.

4. Experimental

4.1. Patient characteristics

The subjects of this study were patients who had completed AZA treatment at the Ogaki Municipal Hospital (Ogaki, Japan) between April 2011 and August 2019. AZA was administered at 75 mg/m^2 once daily for 7 days on a 28-day cycle *via* either subcutaneous or intravenous injection (10 min infusion). Patients treated with AZA were assessed for their age, sex, diagnosis, international prognostic scoring system-revised (IPSS-R), cytogenetic risk, performance status (ECOG), red blood cell (RBC) transfusion dependence, laboratory data at initiation of AZA treatment, presence of peripheral blood blasts (PBs), and bone marrow blasts, prior treatment, and comorbidities.

4.2. Continuity of AZA treatment

The AZA dosing information (the number of AZA treatment cycles, reason of failure to continue AZA treatment, and dose reduction or interruption of AZA) was examined. Patients were divided into two groups: discontinued AZA group (< 4 cycles treatment) and continued AZA group (≥ 4 cycles treatment).

4.3. AZA efficacy

The AZA efficacy (best response and overall survival (OS)) was examined within the two groups described above. The response to AZA was defined according to IWG 2006 criteria (complete response: CR, partial response: PR, marrow CR, stable disease with hematological improvement: SD with HI, or progressive disease: PD) (Cheson et al. 2006). OS was measured from the onset of AZA treatment.

4.4. Investigation of the factors affecting continuation and OS

Factors related to the early termination of AZA treatment (less than 4 cycles) and prognostic factors of OS were analysed.

4.5. Statistical analysis

Statistical analysis was performed using JMP software, version 5.0.1J (SAS Institute Japan Ltd., Tokyo, Japan). The Mann-Whitney U test or Fisher's exact test were used for comparison between the two patient groups. Predictive factors for continuity and efficacy were analysed using the Fisher's exact test or Mann-Whitney rank-sum test for univariate comparisons. Significantly changed variables in univariate analysis were included in a multivariate logistic regression analysis. The Kaplan-Meier log-rank test was used to compare overall survival. The recorded p-Values were two-sided and values of p<0.05 were considered statistically significant difference.

4.6. Ethical considerations

The present study was reviewed and approved by the Ethics Committee at Ogaki Municipal Hospital (20191024-7).

Conflict of interest: None declared.

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