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ORIGINAL ARTICLE

Comparison of blast percentage calculated based on bone marrow all nucleated cells and non-erythroid cells in myelodysplastic syndromes with erythroid hyperplasia

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Abstract

It is controversial whether blast percentage based on all nucleated cells (ANC) or non-erythroid cells (NEC) more accurately reflects the prognosis of patients with myelodysplastic syndromes (MDS). We considered that the impact of blast percentage on survival should be similar in MDS with erythroid hyperplasia (MDS-E) and MDS with no erythroid hyperplasia (MDS-NE), and from this perspective, we retrospectively analyzed 322 patients, including 44 with MDS-E and 278 with MDS-NE. Overall survival was similar between the MDS-E and MDS-NE groups ($P = 0.94$). In a subgroup of patients with bone marrow (BM) blasts of < 5%, no difference in survival was found between MDS-E and MDS-NE by either calculation method. However, in patients with a blast percentage between 5 and 10%, a significant difference in survival was observed only when the blast percentage in MDS-E was calculated from ANC ($P < 0.001$ by ANC and $P = 0.66$ by NEC). A similar result was observed when we analyzed the remaining patients with higher blasts together with those with blasts between 5 and 10%. These results suggest that the calculation of the BM blast percentage based on NEC in MDS-E provides a blast percentage value with a clinical impact consistent with that in MDS-NE.

Keywords Myelodysplastic syndromes (MDS) · Erythroid hyperplasia · Non-erythroid cells (NEC) · Acute myeloid leukemia (AML)

Introduction

Myelodysplastic syndromes (MDS) are heterogeneous hematopoietic stem cell disorders characterized by ineffective hematopoiesis resulting in cytopenia and the risk of progression to acute myeloid leukemia (AML) [1, 2]. Treatment strategies for MDS are usually decided upon based on a prognostic scoring system that includes the percentage of bone marrow (BM) blasts, genetic abnormalities, and peripheral cytopenia [3–6]. Therefore, the accurate estimation of BM blasts is important.

In the new World Health Organization (WHO) 2016 classification, the bone marrow blast percentage in MDS is calculated based on all nucleated cells (ANC) regardless of the percentage of erythroid cells. However, some recent reports have suggested that calculation of the blast percentage based on non-erythroid cells (NEC) more accurately reflects the prognosis of MDS with erythroid hyperplasia (MDS-E) than that based on ANC [7, 8]. Thus, the method for calculating BM blasts is still controversial.

We considered that the impact of blast percentage on prognosis should be similar in MDS-E and MDS with no erythroid hyperplasia (MDS-NE). From this perspective, in this study, we classified patients according to the

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percentage of BM blasts calculated from both ANC and NEC and compared the prognosis of MDS-E versus MDS-NE in each classification.

Patients and methods

This study was a retrospective analysis of 322 patients with MDS diagnosed at a single center, Jichi Medical University (Tochigi, Japan), during the period 2006–2016. All patients were diagnosed as MDS according to the World Health Organization (WHO) 2008 classification. Therefore, patients with 50% or more BM erythroid cells who had 20% or more myeloblasts calculated from NEC were diagnosed as AML. Also, cases with 80% or more BM erythroid cells were defined as pure erythroid leukemia. Patients with chronic myelomonocytic leukemia or myelodysplastic/myeloproliferative neoplasms were excluded from this study. BM with 50% or more erythroid cells was defined as MDS-E. Otherwise, the patients were defined as MDS-NE. This study was approved by the ethics committee of Jichi Medical University.

Statistical considerations

Differences between the two populations were evaluated by Fisher's exact test or the chi-square test, as appropriate. Overall survival and time to leukemic transformation were analyzed by a Kaplan-Meier analysis. A P value of < 0.05 was considered to be statistically significant. All statistical tests were performed with EZR (Saitama Medical Center, Jichi Medical University, Saitama, Japan) [9].

Result

Characteristics of the patients

This study included 322 patients, aged between 19 and 107. A total of 44 patients (13.7%) had MDS-E and 278 had MDS-NE. The characteristics of the patients at diagnosis are shown in Table 1. The patients were classified into four risk groups according to the International Prognostic Scoring System (IPSS) [3]. The median percentage of blasts calculated from ANC was 2.2% in patients with MDS-E and 3.2% in those with MDS-NE ($P = 0.02$). Median hemoglobin at diagnosis in the MDS-E group was lower than that in the MDS-NE group (7.6 vs 8.5, $P = 0.02$).

Survival and time to AML transformation of MDS-E and MDS-NE

As shown in Fig. 1a, no survival difference was found between the MDS-E and MDS-NE groups (median OS, 75.5 months vs 52.6 months; $P = 0.57$). Also, the time to AML transformation was similar between the MDS-E and MDS-NE groups (median time to 25% AML transformation, 19.2 months vs 14.8 months; $P = 0.94$; Fig. 1b).

Comparison of BM blast percentage calculated from ANC and from NEC

The BM blast percentages for all the patients were calculated from both ANC and NEC. The number of patients classified according to both the BM blast percentage and IPSS calculated using these two different methods is shown in Table 2. According to the NEC method, a total of 18 patients in MDS-NE group were calculated to have 20% or more BM blasts (considered as AML).

Comparison of the impact of BM blast percentage calculated from ANC versus NEC on survival and time to AML transformation in MDS

We considered that the impact of BM blasts on survival should be similar in the MDS-E and MDS-NE groups. From this perspective, we classified the patients according to the percentage of BM blasts and compared the prognosis of MDS-E versus MDS-NE in each classification.

When the BM blasts were calculated from NEC, there were fewer patients with < 5% BM blasts compared to the value calculated using ANC in both MDS-E and MDS-NE patients. Regardless of the method used to calculate blasts, no statistically significant difference in survival was found between the MDS-E and MDS-NE groups among patients with low blast levels of < 5% (median OS, 121 months vs 171 months, $P = 0.60$ by the ANC method; 121 months vs 171 months, $P = 0.45$ by the NEC method; Fig. S1) (Supplementary material).

In patients with BM blast percentages between 5 and 10%, MDS-E patients had survival similar to that in MDS-NE patients when blasts percentages were calculated from NEC (median OS, 39.7 months vs 39.5 months; $P = 0.93$; Fig. 2b). However, when blast percentages were calculated from ANC, the survival of MDS-E patients was significantly inferior to that of MDS-NE patients (median OS, 7.66 months vs 24.5 months; $P < 0.001$; Fig. 2a).

As shown in Table 2, none of the MDS-E patients had 10% or more blasts calculated from ANC, and none of the MDS-E patients had 20% or more blasts calculated from NEC, because all of the included patients were diagnosed according to the WHO 2008 classification. Therefore, we analyzed all the patients with 10% or more blasts together with those with 5 to < 10% BM blasts. When all the patients with high blasts (>5%) were analyzed altogether, even though the MDS-NE group had 51 additional patients with higher blasts, the

Table 1 Characteristics of the patients

Total, n	Group	MDS-NE 278	MDS-E 44	*P* value
Median age [range], years		69 [19–90]	63.00 [35–107]	0.02
Gender, *n* (%)	Female	79 (28.5)	9 (20.5)	0.36
	Male	198 (71.5)	35 (79.5)	
Median BM erythroid cells (%)		23.8 [0.0–49.8]	56.2 [50.0–76.6]	< 0.001
Median BM non-erythroid cells (%)		76.2 [50.2, 100.0]	43.8 [23.4, 50.0]	< 0.001
Median BM blasts calculated from ANC (%) [range]		3.2 [0.0–18.8]	2.2 [0.2–9.0]	0.02
	< 5 (%)	175 (62.9)	39 (88.6)	0.001
	≤ 5, < 10 (%)	60 (21.6)	5 (11.4)	
	≤ 10 (%)	43 (15.5)	0 (0.0)	
Median hemoglobin [range] (g/dL)		8.5 [2.6–16.2]	7.6 [3.0–15.6]	0.02
	≥ 10 (%)	83 (29.9)	7 (15.9)	0.07
	> 10 (%)	195 (70.1)	37 (84.1)	
Median neutrophil count [range] (/μL)		1415 [0–15,732]	1198 [0–11,470]	0.19
	1800 ≤ (%)	95 (34.2)	12 (27.3)	0.40
	1800 > (%)	183 (65.8)	32 (72.7)	
Median platelet count [range] ($\times 10^9$/μL)		8.05 [0.2, 105.6]	6.95 [1.0, 50.6]	0.29
	≥ 100 (%)	117 (42.1)	15 (34.1)	0.41
	< 100 (%)	161 (57.9)	29 (65.9)	
WHO 2008 categories				
	5q-	1 (0.4)	0 (0.0)	NA
	RA	93 (33.5)	11 (25.0)	
	RAEB1	58 (20.9)	7 (15.9)	
	RAEB2	46 (16.5)	0 (0.0)	
	RARS	8 (2.9)	1 (2.3)	
	RCMD	57 (20.5)	21 (47.7)	
	RCUD	7 (2.5)	1 (2.3)	
	MDS-U	8 (2.9)	3 (6.8)	
IPSS cytogenetic group, *n* (%)	Low (%)	205 (74.0)	27 (61.4)	0.17
	Intermediate (%)	39 (14.1)	8 (18.2)	
	High (%)	33 (11.9)	9 (20.5)	

survival of MDS-E patients using the ANC method was still inferior to that of MDS-NE patients using the ANC method (median OS, 7.66 months vs 18.6 months; $P < 0.01$; Fig. 2c). Similar to patients with a BM blast percentage between 5 and 10%, the survival of MDS-E with blasts based on the NEC method was not significantly different from that of MDS-NE with blasts calculated from the NEC method (median OS, 34.4 months vs 23.6 months; $P = 0.97$; Fig. 2d).

We also assessed time to AML transformation in our cohort. The same as OS analysis, there was no statistically significant difference in time to AML transformation between the MDS-E and MDS-NE groups among the patients with low blast percentage regardless of blast calculating methods, whereas it was significant among the patients with higher blasts (Fig. S2a–f).

Impact of the two different methods for calculating blasts on the risk scoring system

Next, we stratified the patients into two risk groups according to IPSS (low risk; IPSS low and intermediate-1, high risk; IPSS intermediate-2 and high) using the two different methods for calculating BM blasts, based on ANC and based on NEC, and evaluated which method was more appropriate for use in the risk scoring system. Table 2 shows the number of patients in each IPSS risk group. Five patients with MDS-E and 13 patients with MDS-NE who were at low risk using the BM blast percentage calculated from ANC were reclassified as high risk when their blasts were calculated based on NEC. When MDS-E and MDS-NE were analyzed together, there were statistically significant differences in survival between patients at low risk and high risk regardless of blast calculating

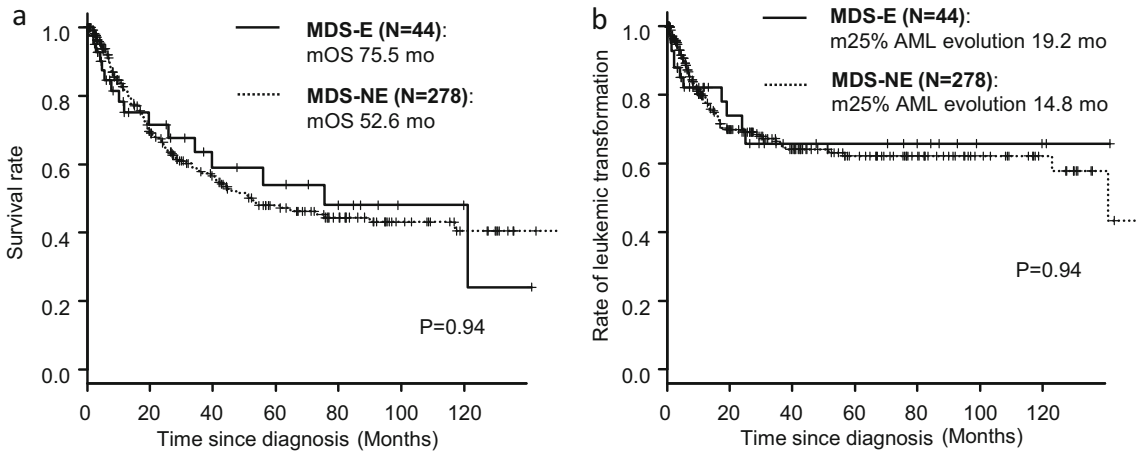

Fig. 1 Comparisons of survival (**a**) and time to AML transformation (**b**) between all MDS-E and MDS-NE

methods (median OS, low risk vs high risk from ANC method, 117 months vs 14.0 months; $P < 0.001$; Fig. S3, median OS, low risk vs high risk from NEC method, 117 months vs 17.1 months; $P < 0.001$).

Next, since we considered that the impact of BM blasts on survival should be similar in MDS-E and MDS-NE, we compared the survival of patients in these two groups for each risk classification. Among the patients at low risk, no difference in survival was found between MDS-E and MDS-NE regardless of the method used to calculate BM blasts (ANC method: median OS, 121 months vs 117 months; $P = 0.34$; Fig. S4a, NEC method: median OS, 121 months vs 117 months; $P = 0.21$; Fig. S4b). In contrast, in patients at high risk, patients with MDS-E had significantly inferior survival compared to those with MDS-NE when BM blasts were calculated from ANC (MDS-E vs MDS-NE: median OS, 7.66 months vs 14.9 months; $P = 0.029$; Fig. 3a). No statistically difference in survival was detected between MDS-NE and MDS-E based on BM blasts calculated from NEC (MDS-E vs MDS-NE: median OS, 7.66 months vs 17.8 months; $P = 0.11$; Fig. 3b).

Discussion

This study focused on the methods used to calculate the percentage of BM blasts in MDS patients. Our previous study proved that peripheral WT1 showed a better correlation with blasts calculated based on NEC than with blasts calculated based on ANC in MDS with erythroid hyperplasia. This might indicate that blasts calculated from NEC reflect the tumor burden in MDS more accurately than those based on ANC [10]. In this study, we evaluated whether blasts based on NEC may reflect the prognosis of MDS more accurately than those based on ANC. We considered that the impact of BM blasts on survival should be similar in MDS-E and MDS-NE. As shown by our results, MDS-E with the blast percentage calculated from NEC, rather than ANC, had a prognosis similar to MDS-NE. Notably, the survival of MDS-E patients with a blast percentage calculated from ANC was significantly inferior to that of MDS-NE patients with blasts calculated from ANC, especially among patients

Table 2 Classification of MDS patients according to blast percentage and IPSS based on ANC and NEC

	ANC method		NEC method	
	MDS-E, *n* (%)	MDS-NE, *n* (%)	MDS-E, *n* (%)	MDS-NE, *n* (%)
Blast (%)				
< 5	39 (12)	175 (54)	19 (6.0)	153 (48)
5 to < 10	5 (1.5)	52 (16)	18 (5.6)	53 (16)
10 to < 20	– (0)	51 (16)	7 (2.2)	54 (17)
≤ 20 (AML)	– (0)	– (0)	– (0)	18 (5.6)
IPSS				
Low	6 (1.9)	47 (15)	4 (1.2)	40 (12)
Int-1	29 (9.0)	157 (49)	26 (8.1)	151 (47)
Int-2	9 (2.8)	64 (20)	10 (3.1)	65 (20)
High	0 (0)	10 (3.1)	4 (1.2)	22 (6.8)

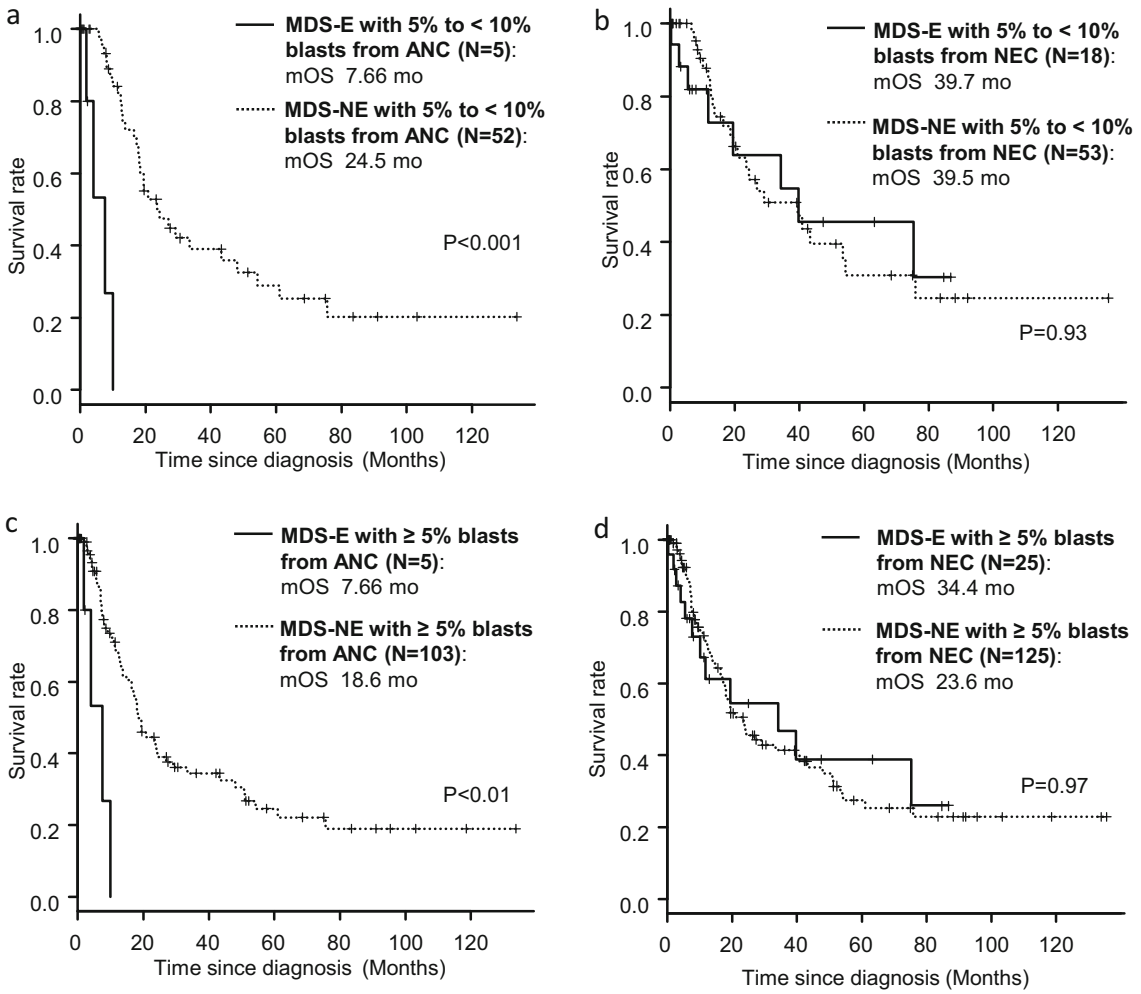

Fig. 2 Comparison of survival between MDS-E and MDS-NE calculated from ANC (**a**) (median OS, 7.66 months vs 24.5 months; $P < 0.001$) and that calculated from NEC (**b**) (median OS, 39.7 months vs 39.5 months; $P = 0.93$) in patients with a blast percentage between 5 and 10%. Comparison of survival between MDS-E and MDS-NE calculated from ANC (**c**) (median OS, 7.66 months vs 18.6 months; $P < 0.01$) and that calculated from NEC (**d**) (median OS, 34.4 months vs 23.6 months; $P = 0.97$) in patients with a high blast percentage ($\leq 5\%$)

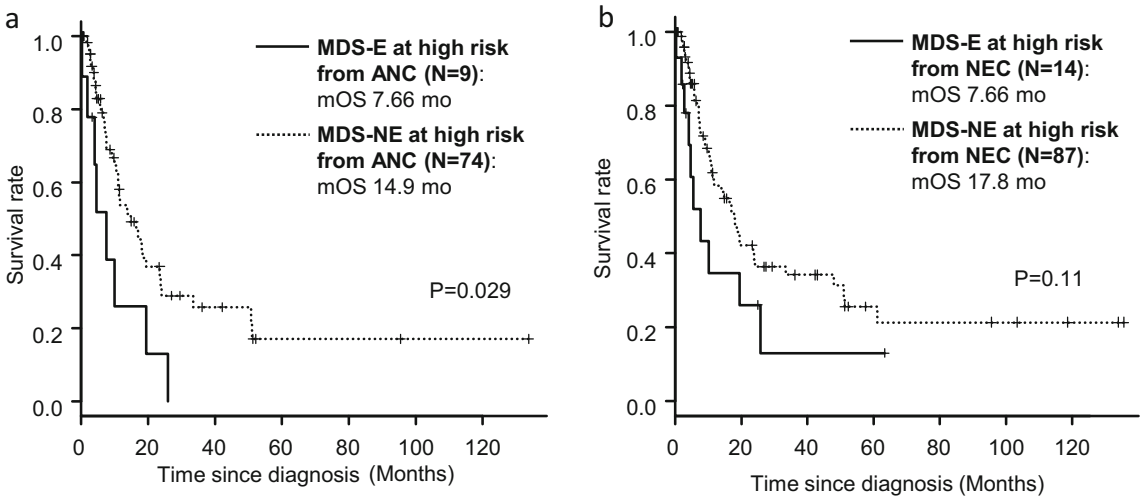

Fig. 3 Comparison of survival between MDS-E and MDS-NE calculated from ANC (**a**) (median OS, 7.66 months vs 14.9 months; $P = 0.029$) and that calculated from NEC (b) (median OS, 7.66 months vs 17.8 months; $P = 0.11$) in IPSS high-risk patients

with a worse prognosis, suggesting that the blast count based on ANC might underestimate the tumor burden. Moreover, the blast percentage calculated from NEC might be more appropriate than that calculated from ANC for IPSS risk classification in patients with MDS-E.

A recent study showed that patients with MDS whose blast categories were upgraded when blasts calculated from ANC were recalculated from NEC (i.e., blasts of < 5 to ≥ 5%, RAEB-1 to RAEB-2 or IPSS low to IPSS high) had significantly inferior survival compared to those who remained in their original blast category [7]. IPSS-R scores using blasts calculated from NEC divided patients with MDS into each risk group more precisely than those calculated from ANC [8], whereas another report showed opposite results [11]. Wang also indicated that MDS-E patients with < 5% blasts calculated from NEC had survival similar to that of MDS-NE patients with < 5% blasts calculated from ANC, but had inferior survival when blasts were calculated from ANC [12]. Although we did not detect a significant difference in survival between MDS-E and MDS-NE patients with < 5% blasts regardless of the method used to calculate blasts, the survival of MDS-E patients with ≥ 5% blasts was inferior to that of MDS-NE patients when blasts were calculated from ANC. These results suggested that the blast percentage based on NEC rather than ANC might have a consistent impact on survival between MDS-E and MDS-NE patients.

Since this study only included patients with MDS according to the WHO 2008 classification, the patients with erythroblastosis and with < 20% of blasts calculated from ANC were excluded when the blasts calculated from NEC exceeded 20%. During 2006 and 2016, there were 13 patients who were diagnosed as AML with erythroblastosis according to the WHO 2008 classification, but were recategorized as MDS according to the WHO 2016 classification. Even when the reclassified MDS patients were included in this study, we obtained almost the same results as former analysis (Fig. S5). However, the results would be changed if these AML patients had been treated as MDS according to the WHO 2016 classification,because the patients diagnosed as AML according to the WHO 2008 classification initially received inductionchemotherapy for AML soon after diagnosis.

This study had several limitations. First, this was a retrospective study in a limited number of patients. Second, since all the MDS patients included in this study were diagnosed according to the WHO 2008 classification, patients diagnosed as AML by WHO 2008 but as MDS by WHO 2016 (BM blast of ≤ 20% by NEC but > 20% by ANC) were not included in the main analysis.

In evaluating the prognosis of MDS patients, the bone marrow blast percentage is an important parameter, and thus, its impact on survival should be consistent between MDS-E and MDS-NE patients. This study indicated that the blast count based on NEC showed a more consistent impact than that based on ANC. This might indicate that BM blasts calculated from NEC reflect the prognosis of MDS-E more accurately than those calculated from ANC.

Author's contribution KM performed the research, collected, and analyzed the data. YK performed the research and analyzed the data. All the authors wrote the paper and approved the final version.

Compliance with ethical standards

Conflict of interest The authors declare that they have no conflict of interest.

Ethical approval For this retrospective study, formal informed consent is not required. This study was approved by the ethics committee of Jichi Medical University.

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