





Article

The Absolute Monocyte Count at Diagnosis Affects Prognosis in Myelodysplastic Syndromes Independently of the IPSS-R Risk Score

Tobias Silzle ^{1,*}, Sabine Blum ², Annika Kasprzak ³, Kathrin Nachtkamp ³, Martina Rudelius ⁴, Barbara Hildebrandt ⁵, Katharina S. Götze ⁶, Norbert Gattermann ³, Michael Lauseker ^{7,†} and Ulrich Germing ^{3,†}

¹ Department of Medical Oncology and Hematology, Cantonal Hospital St. Gallen, 9007 St. Gallen, Switzerland

² Service and Central Laboratory of Hematology, University Hospital of Lausanne and Lausanne University, 1011 Lausanne, Switzerland

³ Department of Hematology, Oncology, and Clinical Immunology, Heinrich Heine University Düsseldorf, 40225 Düsseldorf, Germany

⁴ Institute of Pathology, Faculty of Medicine, LMU Munich, 80337 Munich, Germany

⁵ Department of Human Genetics, Heinrich Heine University Düsseldorf, 40225 Düsseldorf, Germany

⁶ Department of Internal Medicine III, Klinikum Rechts der Isar, Technical University of Munich, 81675 Munich, Germany

⁷ Institute for Medical Information Processing, Biometry and Epidemiology, Faculty of Medicine, LMU Munich, 81377 Munich, Germany

* Correspondence: tobias.silzle@kssg.ch; Tel.: +41-71-494-2922

† These authors contributed equally to this work.



Citation: Silzle, T.; Blum, S.; Kasprzak, A.; Nachtkamp, K.; Rudelius, M.; Hildebrandt, B.; Götze, K.S.; Gattermann, N.; Lauseker, M.; Germing, U. The Absolute Monocyte Count at Diagnosis Affects Prognosis in Myelodysplastic Syndromes Independently of the IPSS-R Risk Score.

Cancers **2023**, *15*, 3572. <https://doi.org/10.3390/cancers15143572>

Academic Editor: Massimo Breccia

Received: 28 May 2023

Revised: 4 July 2023

Accepted: 9 July 2023

Published: 11 July 2023



Copyright: © 2023 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license (<https://creativecommons.org/licenses/by/4.0/>).

Simple Summary: The revised international prognostic scoring system (IPSS-R) represents the standard tool for prognostication in myelodysplastic syndromes (MDSs). It considers the degree of cytopenias together with the bone marrow blast count and the results of metaphase cytogenetics. Monocytes are a subgroup of white blood cells involved in host defense and tissue repair or remodelling. The goal of our study was to assess the prognostic impact of the absolute monocyte count (AMC) at the time point of diagnosis in patients with MDS. We found an IPSS-R-independent prognostic impact of the AMC, both when assessed as a continuous variable and when MDS patients with a low ($<0.2 \times 10^9/L$) or a higher ($>0.4 \times 10^9$) AMC were compared to MDS patients with an AMC of $0.2\text{--}0.4 \times 10^9/L$. A low AMC was associated with a higher risk of transformation to acute myeloid leukemia. Hence, considering the AMC might help to identify MDS patients who could benefit from more intense treatment strategies.

Abstract: The absolute monocyte count (AMC) is associated with mortality in a variety of medical conditions. Its prognostic impact in myelodysplastic syndromes (MDSs) is less well studied. Therefore, we investigated its potential prognostic value in a cohort from the Düsseldorf MDS registry in relationship to the revised international prognostic scoring system (IPSS-R). An AMC below the population's median ($<0.2 \times 10^9/L$) was associated with several adverse disease features such as lower haemoglobin levels, lower count of neutrophils and platelets, and a higher percentage of blasts in the bone marrow. MDS patients with an AMC $< 0.2 \times 10^9/L$ had a significantly higher risk of progression into acute myeloid leukemia (AML). In a univariate, proportional hazards model the effect of the AMC as a continuous variable was modelled via p-splines. We found a U-shaped effect with the lowest hazard around $0.3 \times 10^9/L$. Accordingly, an AMC within the last quartile of the population ($0.4 \times 10^9/L$) was associated with a reduced overall survival independently of IPSS-R, but not with the risk of secondary AML. Considering monocytopenia as a risk factor for AML progression in MDS may provide an additional argument for allogeneic transplantation or the use of hypomethylating agents in patients who are not clear candidates for those treatments according to current prognostic scoring systems and/or recommendations. Further studies are needed to assess the prognostic impact of the AMC in the context of prognostic scoring systems, considering the

molecular risk profile, and to identify the mechanisms responsible for the higher mortality in MDS patients with a subtle monocytosis.

Keywords: myelodysplastic syndrome; prognostication; absolute monocyte count; revised international prognostic scoring system

1. Introduction

The myelodysplastic syndromes (MDSs) are a heterogeneous group of clonal myeloid haemopathies, primarily observed in the elderly population. Hallmarks of these diseases include cytopenias due to an ineffective haematopoiesis and a variable risk of leukemic transformation due to genetic and epigenetic changes in haematopoietic stem cells and alterations within the haematopoietic niche [1,2]. Their classification is based on morphology (number of dysplastic cell lines, number of blasts, presence or absence of ringed sideroblasts) and the presence of certain cytogenetic or molecular aberrations [3–5].

Risk stratification with regard to the probability of leukemic transformation and death related to bone marrow failure is essential for newly diagnosed MDS patients [6]. The degree of cytopenias, the bone marrow blast count, and the presence and type of cytogenetic changes determined by conventional metaphase cytogenetics are factors taken into account by the standard tools used for prognostication, the international prognostic scoring system (IPSS) [7] and its revised version (IPSS-R) [8]. Recently, the addition of the molecular risk profile, as determined by next generation sequencing (NGS), has been shown to allow a refined prognostication in addition to the conventional factors. The IPSS-molecular takes into account mutations of 31 different genes, with *TP53*^{multihit}, *MLL*^{PTD}, and *FLT3* mutations as top genetic predictors of adverse outcomes, and mutations in genes such as *ASXL1*, *BCOR*, *EZH2*, *NRAS*, *RUNX1*, *STAG2*, and *U2AF1* being significantly associated with an adverse risk [9].

Patients with higher-risk MDSs are candidates for allogeneic haematopoietic stem cell transplantation (HSCT) since the risk of leukemic transformation is high and overall survival (OS) is short even without transformation. Best supportive care and improvement of cytopenias are the cornerstones of therapy for lower-risk MDS patients [1].

Monocytes are a functionally heterogeneous group of mononuclear phagocytic cells that contribute in many ways to host defense and tissue repair or remodelling [10]. A low absolute monocyte count (AMC) has been shown to be of prognostic value in MDSs in two studies [11,12]. In other chronic myeloid diseases, such as Philadelphia chromosome-negative myeloproliferative neoplasms, a higher AMC is associated with adverse disease features and inferior prognosis [13,14]. In MDSs, data on the prognostic impact of a subtle monocytosis below the threshold of $1.0 \times 10^9/L$, defining chronic-myelomonocytic leukemia (CMML) according to the WHO 2016 classification, are scarce.

We, therefore, intended to examine the distribution of the AMC in the different MDS sub- and risk groups according to WHO 2016 and IPSS-R and to analyse its impact on overall and leukemia-free survival in a cohort of primary MDS patients, both as a continuous variable and according to different cut-offs.

2. Material and Methods

We screened the Düsseldorf MDS registry for patients with information on the AMC and the IPSS-R category at diagnosis. The Düsseldorf MDS registry covers virtually all newly diagnosed MDS cases in the Greater Düsseldorf area since 1982. The structure of the registry and the diagnostic criteria for inclusion of cases have been described earlier in detail [15]. Monocyte counts were obtained from manual differentials and promonocytes and monoblasts were considered as blast equivalents.

Patients with an AMC $> 1.0 \times 10^9/L$ or a white blood cell count $>13.0 \times 10^9/L$ were excluded, as they were likely to suffer from CMML or myelodysplastic/myeloproliferative neoplasms.

For comparative analyses, patients were stratified in three groups according to the rounded quartiles of the AMC (first and second quartile: AMC $< 0.2 \times 10^9/L$, third quartile: AMC $0.2\text{--}0.4 \times 10^9/L$, and fourth quartile: AMC $> 0.4 \times 10^9/L$).

Categorical variables were analysed by frequency tables and compared using the χ^2 -test, continuous variables were described by median (range) and compared between different groups using the Mann–Whitney (comparison of two groups) or the Kruskal–Wallis (comparison of ≥ 3 groups) test. Overall and leukemia-free survival were calculated in months from the date of diagnosis to the respective event date. Time-to-event curves were calculated by the Kaplan–Meier method and the log-rank test was used for univariate comparison. Cox proportional hazard regression models were applied for multivariate analysis. A p -value < 0.05 was considered statistically significant.

To assess the effect of AMC on survival as a continuous variable, p-splines were estimated, both in a univariate as well as in multivariate settings. Four degrees of freedom were chosen. However, sensitivity analyses showed that this choice had only little influence on the results. Progression to AML was analysed via the competing risks methodology, with “death without progression” being the competing event. The Gray test based on the model of Fine and Gray was used to compare variables with regard to this endpoint. All analyses were performed with IBM SPSS Statistics, version 25 or R, version 4.2.1.

3. Results

3.1. Study Population

We identified a cohort of 993 patients (male $n = 576$ (58%), female $n = 417$ (42%)) with a median age of 66 years (IQR 58–73). A total of 112 cases (11.3%) were known to be therapy-related following cytotoxic treatment or radiation and seven (7%) cases represented secondary MDS evolving from another haematological neoplasm.

During follow-up (median 29 months, IQR 10–61), 633 patients (63.7%) died and 247 patients (24.9%) experienced a leukemic transformation.

Stem cell transplantation was performed in 132/993 patients (13.3%; allogeneic $n = 129$, autologous $n = 3$) and 77/993 (7.8%) patients received at least one cycle of an intensive induction chemotherapy regimen. Hypomethylating agents (HMA) were used in 85/993 patients (8.6%) and immune modulatory imide drugs (IMiD) in 69/993 (6.9%). The remaining patients (630/993; 63.4%) received low-intensity treatments such as erythropoiesis stimulating agents or immunosuppression (cyclosporine \pm anti-thymocyte globulin) or best supportive care, including transfusion and iron chelation. Details regarding MDS subtypes according to the WHO 2016 classification, cytogenetic risk groups according to IPSS-R and peripheral blood values, blast counts in bone marrow and peripheral blood, and additional parameters (LDH, presence or absence of bone marrow fibrosis, transfusion dependency) at the time point of diagnosis are shown in Table 1.

Table 1. Patient characteristics in the whole cohort and according to the absolute monocyte count (group A $< 0.2 \times 10^9/L$, group B $0.2\text{--}0.4 \times 10^9/L$, group C $> 0.4 \times 10^9/L$).

		Group A	Group B	Group C	<i>p</i>	
	All	AMC $< 0.2 \times 10^9/L$	AMC $0.2\text{--}0.4 \times 10^9/L$	AMC $> 0.4 \times 10^9/L$	A vs. B	B vs. C
WHO 2016, <i>n</i> (%)	993	529	241	223		
MDS-SLD	71 (7.2)	38 (7.2)	18 (7.5)	15 (6.7)		
MDS-MLD	302 (30.4)	157 (29.7)	70 (29)	75 (33.6)		
MDS-RS-SLD	46 (4.6)	15 (2.8)	17 (7.1)	14 (6.3)		
MDS-RS-MLD	92 (9.3)	37 (7)	24 (10)	31 (13.9)		

Table 1. Cont.

	All	Group A	Group B	Group C	<i>p</i>	
		AMC <0.2 × 10 ⁹ /L	AMC 0.2–0.4 × 10 ⁹ /L	AMC > 0.4 × 10 ⁹ /L	A vs. B	B vs. C
MDS(del5q)	115 (11.6)	54 (10.2)	46 (19.1)	15 (6.7)		
MDS-EB-1	162 (16.3)	86 (16.3)	34 (14.1)	42 (18.8)		
MDS-EB-2	194 (19.5)	138 (26.1)	29 (12)	27 (12.1)		
MDS-U	11 (1.1)	4 (0.8)	3 (1.2)	4 (1.8)		
IPSS-R, <i>n</i> (%)						
Very low	54 (5.4)	19 (3.6)	18 (7.5)	17 (7.6)		
Low	317 (31.9)	141 (26.7)	93 (38.6)	83 (37.2)		
Intermediate	258 (26.0)	146 (27.6)	62 (25.7)	50 (22.4)		
High	184 (18.5)	104 (19.7)	35 (14.5)	45 (20.2)		
Very high	180 (18.1)	119 (22.5)	33 (13.7)	28 (12.6)		
Cytogenetic risk groups (IPSS-R)						
Very good	34 (3.4)	14 (2.6)	5 (2.1)	15 (6.7)		
Good	609 (61.3)	321 (60.7)	153 (63.5)	135 (60.5)		
Intermediate	165 (16.6)	95 (18)	42 (17.4)	28 (12.6)		
Poor	72 (7.3)	33 (6.2)	17 (7.1)	22 (9.9)		
Very poor	113 (11.4)	66 (12.5)	24 (10)	23 (10.3)		
Demographics						
Age, median (years), IQR	66 (58–73)	66 (58–73)	66 (58–73)	67 (59–73)	0.782	0.220
Male, <i>n</i> (%)	576 (58)	285 (53.9)	138 (57.3)	153 (68.6)	0.381	0.012
Leukemic transformation, <i>n</i> (%)	247 (24.9)	145 (27.4)	55 (22.8)	47 (21.1)		
Deaths, <i>n</i> (%)	633 (63.7)	359 (67.9)	134 (55.6)	140 (62.8)		
Lost to follow-up, <i>n</i> (%)	35 (3.5)	20 (3.8)	9 (3.7)	6 (2.7)		
Peripheral blood values						
Haemoglobin, median (g/L), IQR	95 (83–110)	93 (82–109)	99 (84–113)	98 (85–112)	0.018	0.79
Neutrophil count, median (×10 ⁹ /L), IQR	1.59 (0.82–2.66)	1.12 (0.63–2.05)	1.84 (1.20–2.66)	2.54 (1.48–3.75)	<0.001	<0.001
Lymphocyte count, median (×10 ⁹ /L), IQR	1.21 (0.79–1.68)	1.1 (0.70–1.54)	1.26 (0.83–1.73)	1.47 (0.99–2.05)	0.006	<0.001
Platelet count, median (×10 ⁹ /L), IQR	115 (58–232)	97 (53–190)	151 (74–266)	128 (65–263)	<0.001	0.227
LDH						
Data available, <i>n</i> (%)	851 (85.7)	461 (87.1)	204 (84.6)	186 (83.4)		
LDH, median (U/L)	199 (172–256)	203 (170–257)	198 (177–243)	198 (172–268)	0.798	0.917
LDH > ULN (240 U/L), <i>n</i> (%)	259 (26.1)	149 (28.2)	51 (16.2)	59 (31.7)	0.058	0.141

Table 1. Cont.

	All	Group A	Group B	Group C	<i>p</i>	
		AMC <0.2 × 10 ⁹ /L	AMC 0.2–0.4 × 10 ⁹ /L	AMC > 0.4 × 10 ⁹ /L	A vs. B	B vs. C
Blasts						
Blasts bone marrow, median (%), IQR	3 (1–8)	4 (2–10)	2 (1–5)	3 (1–7)	<0.001	0.394
Blasts peripheral blood, median (%), IQR	0 (0–0)	0 (0–0)	0 (0–0)	0 (0–0)	0.484	0.382
Range	(0–19)	(0–19)	(0–18)	(0–19)		
Bone marrow fibrosis						
Data available, <i>n</i> (%)	539 (54.3)	299 (56.5)	117 (48.5)	123(55.2)		
With fibrosis, <i>n</i> (%)	75 (12.6)	39 (13.0)	19 (16.2)	17 (13.8)	0.398	0.600
Transfusion dependent						
Data available, <i>n</i> (%)	415 (41.8)	229 (43.7)	94 (39)	92 (41.3)		
Transfusion dependent, <i>n</i> (%)	218 (51.8)	129 (56.3)	45 (47.9)	44 (47.8)	0.166	0.995

MDS: myelodysplastic syndrome; SLD: single lineage dysplasia; MLD: multi-lineage dysplasia; RS: ringed sideroblasts; EB: excess blasts; MDS-U: myelodysplastic syndrome unclassifiable; IQR: inter-quartile range, LDH: lactate dehydroxygenase.

3.2. Absolute Monocyte Counts in the Study Population, Influence of Sex and Age

The median AMC of the whole population was $0.19 \times 10^9/L$ (IQR 0.07–0.374). It was higher in male patients compared to female patients (median $0.20 \times 10^9/L$ (IQR 0.09–0.42) versus $0.16 \times 10^9/L$ (IQR 0.07–0.32), $p = 0.002$), but not significantly affected by age $</\geq 65$ years (median $0.18 \times 10^9/L$ (IQR 0.07–0.36) versus $0.19 \times 10^9/L$ (IQR 0.08–0.39), $p = 0.159$).

3.3. Monocyte Counts in Different MDS Subgroups

In MDSs with excess blasts ($n = 356$) the AMC was significantly lower compared to MDSs without excess blasts ($n = 637$; median 0.12 vs. 0.21, $p < 0.001$). For MDSs with uni- or multi-lineage dysplasia ($n = 511$), a ring-sideroblastic phenotype ($n = 138$) was associated with a higher AMC (median 0.27 vs. $0.19 \times 10^9/L$, $p = 0.002$). MDSs with del(5q) did not differ with regard to the AMC compared to MDSs non-del(5q) (median 0.18 vs. $0.21 \times 10^9/L$, $p = 0.910$). There was no significant difference between primary and therapy-related MDSs (median AMC 0.19 vs. $0.19 \times 10^9/L$, $p = 0.608$) and the presence or absence of fibrosis (median 0.20 vs. $0.17 \times 10^9/L$, $p = 0.351$) or transfusion dependency (median 0.16 vs. $0.20 \times 10^9/L$, $p = 0.05$). Details are shown in Table 2.

Following stratification according to IPSS-R, a continuous decline in the AMC with rising IPSS-R categories was noted from a median of $0.30 \times 10^9/L$ in very low-risk patients to a median of $0.12 \times 10^9/L$ in very high-risk patients, with the differences between the single IPSS-R groups being highly significant (see Figure 1 and Table 2).

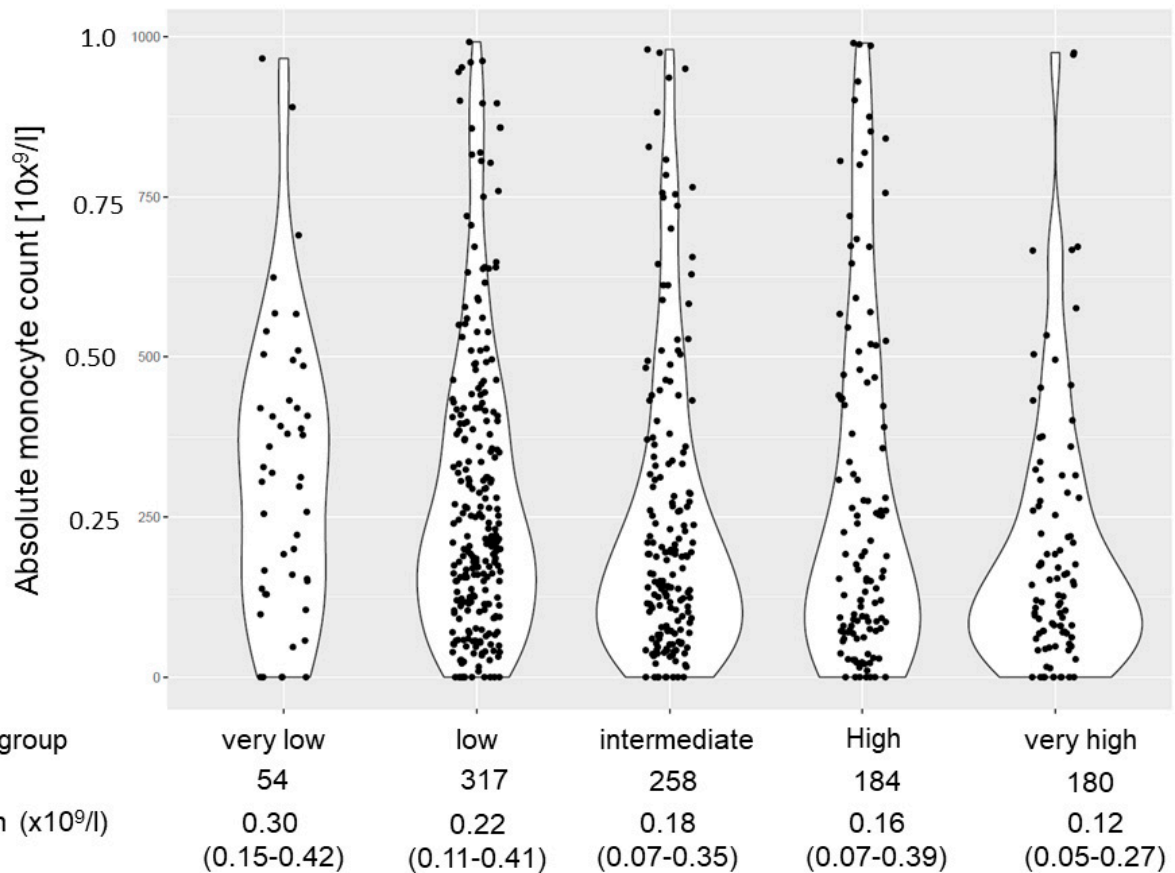


Figure 1. Absolute monocyte count in the single risk groups according to IPSS-R.

Table 2. Absolute monocyte count in different MDS subgroups.

	<i>n</i>	AMC (×10 ⁹) Median (IQR)	<i>p</i>
Total	993	0.19 (0.07–0.37)	
WHO 2016			
MDS-SLD	71	0.19 (0.11–0.33)	<0.001
MDS-MLD	302	0.19 (0.09–0.40)	
MDS-RS-SLD	46	0.31 (0.13–0.52)	
MDS-RS-MLD	92	0.26 (0.15–0.50)	
MDS(del5q)	115	0.21 (0.10–0.32)	
MDS-EB-1	162	0.16 (0.06–0.45)	
MDS-EB-2	194	0.10 (0.04–0.26)	
MDS-U	11	0.32 (0.09–0.59)	
IPSS-R			
Very low	54	0.30 (0.15–0.42)	<0.001
Low	317	0.22 (0.11–0.41)	
Intermediate	258	0.18 (0.07–0.35)	
High	184	0.16 (0.07–0.39)	
Very high	180	0.12 (0.05–0.27)	
MDS with excess blasts	356	0.12 (0.05–0.32)	<0.001
MDS without excess blasts	637	0.21 (0.11–0.40)	
MDS del(5q)	115	0.21 (0.09–0.32)	0.910
MDS non-del(q)	878	0.18 (0.07–0.39)	
MDS-SLD/MLD	373	0.19 (0.10–0.39)	0.002
MDS-RS-SLD/MLD	138	0.27 (0.13–0.50)	

Table 2. Cont.

	<i>n</i>	AMC ($\times 10^9$) Median (IQR)	<i>p</i>
Total	993	0.19 (0.07–0.37)	
Lower-risk MDSs (IPSS-R very low/low)	371	0.24 (0.11–0.42)	<0.001
Higher-risk MDSs (IPSS-R intermediate/high/very high)	622	0.15 (0.06–0.33)	
Therapy-related MDSs	112	0.19 (0.09–0.39)	0.608
Primary MDSs	881	0.19 (0.07–0.37)	
Transfusion dependent	218	0.16 (0.06–0.33)	0.05
Transfusion independent	197	0.20 (0.09–0.40)	
Without bone marrow fibrosis	464	0.17 (0.07–0.38)	0.351
With bone marrow fibrosis	75	0.20 (0.07.0.38)	

IQR: inter-quartile range; MDS: myelodysplastic syndrome; SLD: single lineage dysplasia; MLD: multi-lineage dysplasia; RS: ringed sideroblasts; EB: excess blasts; MDS-U: myelodysplastic syndrome unclassifiable.

3.4. Association of the Absolute Monocyte Count with Peripheral Blood Values and Other Disease Characteristics

An AMC $< 0.2 \times 10^9/L$ was associated with a significantly lower haemoglobin concentration and lower numbers of neutrophils, lymphocytes, and platelets and a higher count of bone marrow blasts. In patients with a monocyte count $>0.4 \times 10^9/L$, higher numbers of neutrophils and lymphocytes were noted, but there was no difference with regard to haemoglobin concentrations and counts of neutrophils, platelets, and bone marrow blasts (for details, see Table 1).

3.5. Impact of Monocyte Count on Overall Survival

3.5.1. Monocyte Count Divided According to Quartiles of the Population

Within the whole population ($n = 993$), both an AMC $< 0.2 \times 10^9/L$ (representing the first and second quartile of the population) and $>0.4 \times 10^9/L$ (representing the fourth quartile) were associated with a significantly shorter median overall survival compared to an AMC between 0.2 and $0.4 \times 10^9/L$ (median 34 or 37 versus 70 months, $p < 0.001$). However, for patients suffering from therapy-related MDSs, OS was not influenced by the AMC (median 22 or 18 versus 42 months, $p = 0.312$). The same was true for MDS patients having received HSCT or induction chemotherapy as part of their treatment (median OS 32 or 40 versus 40 months, $p = 0.759$). Therefore, further survival analyses were restricted to patients with primary MDSs, who did not receive intensive chemotherapy or stem cell transplantation ($n = 689$).

For this subgroup, an AMC $< 0.2 \times 10^9/L$ or $>0.4 \times 10^9/L$ was associated with a significantly shorter median overall survival compared to an AMC between 0.2 and $0.4 \times 10^9/L$ (median 39 or 40 versus 77 months, $p < 0.001$), with the survival curves of patients with an AMC $< 0.2 \times 10^9/L$ and an AMC $> 0.4 \times 10^9/L$ being superimposable, as shown in Figure 2A.

In a univariate Cox regression model, an AMC $< 0.2 \times 10^9/L$ or $> 0.4 \times 10^9/L$ was associated with a higher risk of death compared to an AMC between 0.2 – $0.4 \times 10^9/L$ (HR 1.61; 95% CI 1.28–2.02, $p < 0.001$; see Table 3, row 1).

After stratification according to IPSS-R, the OS according to these AMC subgroups differed significantly for intermediate-risk patients ($n = 179$; median OS 42 or 31 vs. 82 months, $p = 0.025$, see Figure 2B). Similar differences were noted for low-risk patients ($n = 258$; median OS 64 or 54 vs. 107 months) and high-risk patients ($n = 119$; median OS 25 and 18 months vs. 41 months). However, these differences were not statistically significant (low risk: $p = 0.103$; high risk $p = 0.413$). For very low-risk and very high-risk patients, no difference was observed, but patient numbers were low ($n = 49$ and $n = 93$, respectively).

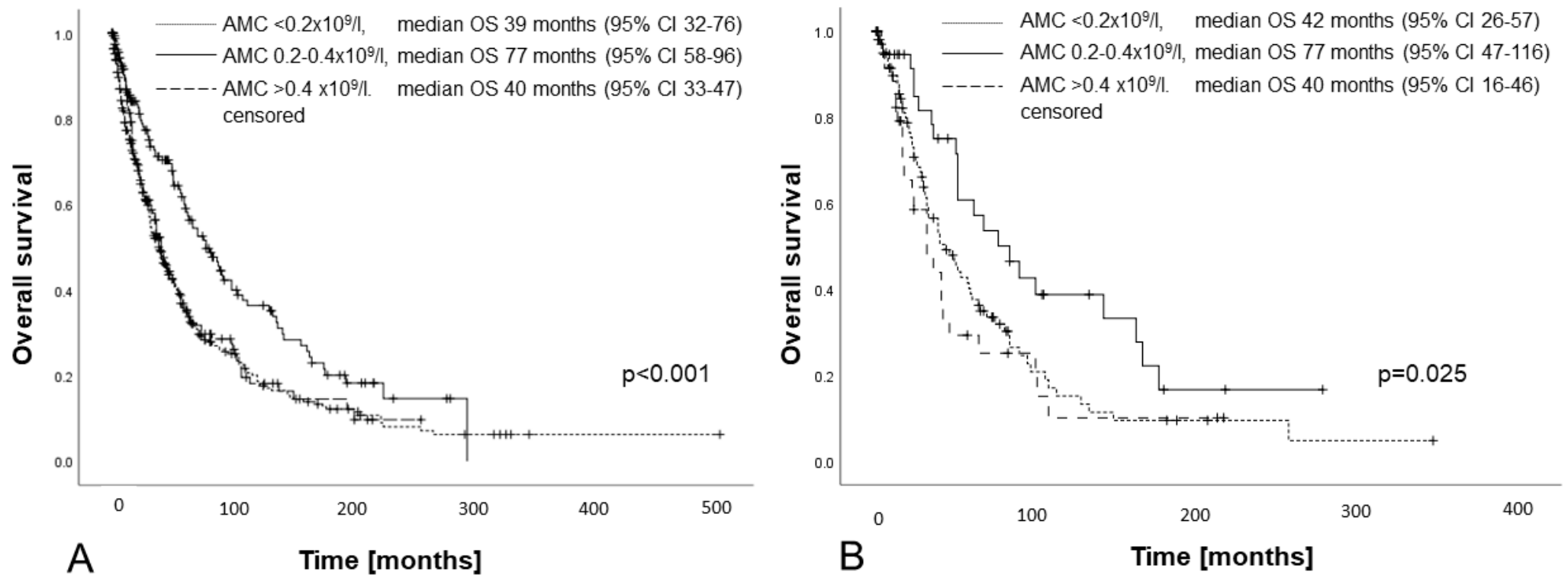


Figure 2. Survival of patients with primary MDSs, without induction chemotherapy or stem cell transplantation, stratified by the absolute monocyte count ($< 0.2 \times 10^9/L$, $0.2-0.4 \times 10^9/L$, and $> 0.4 \times 10^9/L$). (A) Whole cohort ($n = 689$); (B) IPSS-R intermediate risk ($n = 258$).

Table 3. Factors affecting prognosis in MDS patients in univariate analysis with the absolute monocyte count (AMC) as a dichotomous variable ($<0.2 \times 10^9/L$ or $>0.4 \times 10^9/L$ versus $0.2\text{--}0.4 \times 10^9/L$) and three multivariate models (multivariate I: AMC together with the degree of cytopenias, bone marrow blast count, and cytogenetic risk groups according to IPSS-R; multivariate II: AMC together with the single IPSS-R risk groups; multivariate III: AMC together with the single IPSS-R risk groups, age > 65 and male sex).

	Univariate			Multivariate I			Multivariate II			Multivariate III		
	HR	95% CI	<i>p</i>	HR	95% CI	<i>p</i>						
Age > 65	2.37	1.92; 2.94	<0.001							2.45	1.97; 3.04	<0.001
Male sex	1.31	1.08; 1.58	0.006							1.10	0.91; 1.34	0.332
Hb < 100 g/L	1.48	1.23; 1.79	<0.001	1.59	1.31; 1.94	<0.001						
Neutrophils $< 0.8 \times 10^9/L$	1.74	1.32; 2.30	<0.001	0.92	0.69; 1.24	0.582						
Platelets $< 100 \times 10^9/L$	1.56	1.23; 1.99	<0.001	1.59	1.30; 1.94	<0.001						
Bone marrow blasts $>5\%$	2.81	2.29; 3.45	<0.001	2.16	1.72; 2.70	<0.001						
IPSS-R cytogenetic risk category												
good vs. very good	1.67	0.86; 3.25	0.130	2.13	1.09; 4.15	0.027						
intermediate vs. very good	2.48	1.24; 4.96	0.010	2.80	1.40; 5.61	0.004						
poor vs. very good	2.55	1.22; 5.32	0.014	2.61	1.25; 5.48	0.011						
very poor vs. very good	8.15	4.01; 16.55	<0.01	7.18	3.49; 14.78	<0.001						
IPSS-R category												
low vs. very low	1.44	0.92; 2.25	0.113				1.43	0.91; 2.24	0.119	1.57	1.00; 2.46	0.050
intermediate vs. very low	2.16	1.37; 3.41	<0.001				2.13	1.35; 3.36	0.001	2.22	1.40; 3.50	0.001
high vs. very low	4.03	2.51; 6.46	<0.001				3.90	2.43; 6.26	<0.001	3.85	2.34; 6.19	<0.001
very high vs. very low	7.10	4.41; 11.43	<0.001				6.81	4.23; 10.97	<0.001	7.35	4.56; 11.85	<0.001
Monocyte count $<0.2 \times 10^9/L$ or $>0.4 \times 10^9/L$	1.61	1.28–2.02	<0.001	1.32	1.04–1.67	0.021	1.47	1.16; 1.85	0.001	1.63	1.29; 2.05	<0.001

In a multivariate Cox regression model together with the cytogenetic risk groups according to IPSS-R, the bone marrow blast count ($<$ or $\geq 5\%$) and levels of cytope-nias (haemoglobin $<$ or ≥ 10 g/dL, absolute neutrophil count $<$ or $\geq 0.8 \times 10^9/L$, and platelets $<$ or $\geq 100 \times 10^9/L$), the AMC remained an independent prognostic factor. Finally, the AMC retained its independent prognostic value in a multivariate model together with the single IPSS-R risk groups and age $</\geq 65$ years. For details, see Table 3.

3.5.2. Monocyte Count as a Continuous Variable

In a univariate proportional hazards model the effect of the AMC was analysed via p-splines. Here, we found a U-shaped effect with the lowest hazard around $0.3 \times 10^9/L$ (see Figure 3A). The prognostic impact of the AMC remained largely unchanged in multivariate proportional hazards models either together with the single factors of the IPSS-R (see Figure 3B and Table 4, row 2) or together with the single IPSS-R risk groups (see Figure 3C and Table 4, row 3).

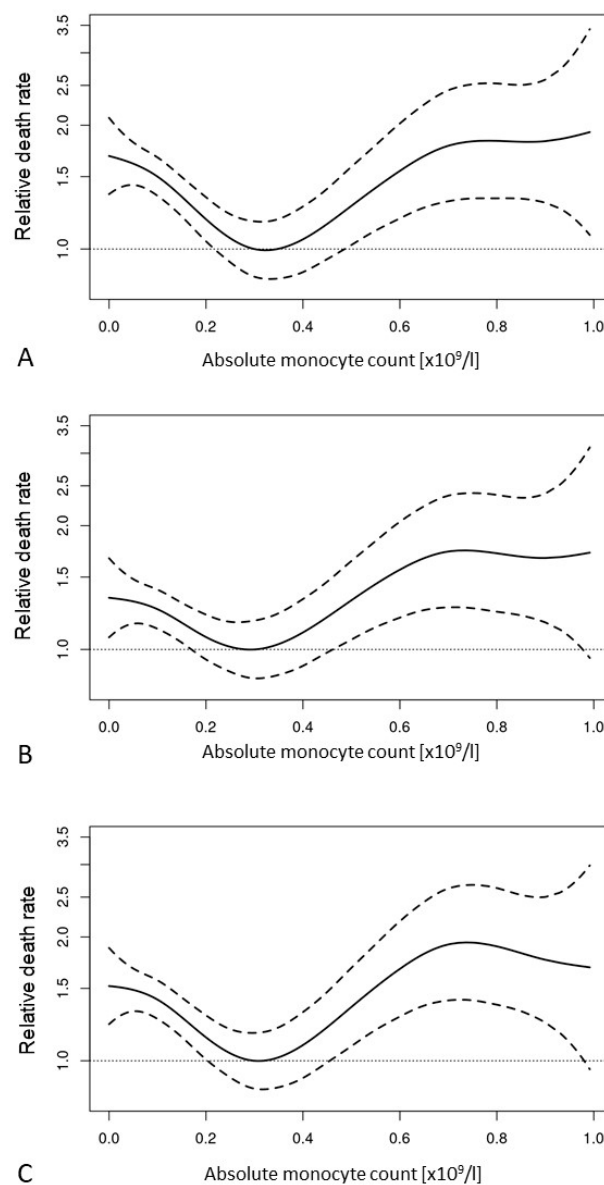


Figure 3. P-splines modelling the prognostic impact of the absolute monocyte count in a univariate proportional hazards model (A) and two multivariate proportional hazards models together with the single factors of the IPSS-R (B), see also Table 4, row 2 or the single IPSS-R risk groups (C), see also Table 4, row 3.

Table 4. Factors affecting prognosis in MDS patients in univariate analysis with the absolute monocyte count as a continuous variable (see Figure 3A–C) and two multivariate models (multivariate I: AMC together with the degree of cytopenias, bone marrow blast count, and cytogenetic risk groups according to IPSS-R; multivariate II: AMC together with the single IPSS-R risk groups).

	Univariate			Multivariate I			Multivariate II		
	HR	95% CI	<i>p</i>	HR	95% CI	<i>p</i>	HR	95% CI	<i>p</i>
Hb < 10 g/dL	1.48	1.23; 1.79	<0.001	1.59	1.30; 1.94	<0.001			
Neutrophil count < $0.5 \times 10^9/L$	1.74	1.32; 2.30	<0.001	0.94	0.70; 1.26	0.684			
Platelet count < $100 \times 10^9/L$	1.56	1.23; 1.99	<0.001	1.67	1.36; 2.05	<0.001			
Bone marrow blasts > 5%	2.81	2.29; 3.45	<0.001	2.09	1.66; 2.63	<0.001			
IPSS-R cytogenetic risk category									
good vs. very good	1.67	0.86; 3.25	0.130	2.13	1.09; 4.17	0.027			
intermediate vs. very good	2.48	1.24; 4.96	0.010	2.81	1.40; 5.66	<0.001			
poor vs. very good	2.55	1.22; 5.32	0.014	2.68	1.28; 5.63	<0.001			
very poor vs. very good	8.15	4.01; 16.55	<0.01	7.45	3.61; 15.37	<0.001			
IPSS-R category									
low vs. very low	1.44	0.92; 2.25	0.113				1.43	0.91; 2.25	0.119
intermediate vs. very low	2.16	1.37; 3.41	<0.001				2.14	1.35; 3.39	0.001
high vs. very low	4.03	2.51; 6.46	<0.001				3.82	2.37; 6.14	<0.001
very high vs. very low	7.10	4.41; 11.43	<0.001				7.23	4.48; 11.67	<0.001
Absolute monocyte count as a continuous variable	See Figure 3A			See Figure 3B			See Figure 3C		

3.5.3. Absolute Monocyte Count and Risk of Transformation to AML

Compared to patients with an AMC between 0.2 and $0.4 \times 10^9/L$, only monocytopenic patients ($AMC < 0.2 \times 10^9/L$) had a significantly higher risk of transformation to acute myeloid leukemia (AML) in a Fine and Gray model (HR 2.0; 95% CI 1.219–3.28, $p = 0.006$). An $AMC > 0.4 \times 10^9/L$ was not associated with a significantly higher risk of progression to AML (HR 1.38; 95% CI 0.766–2.48, $p = 0.280$). Cumulative incidence curves depicting the probability of progression to AML with “death without AML” as competing risk are shown in Figure 4.

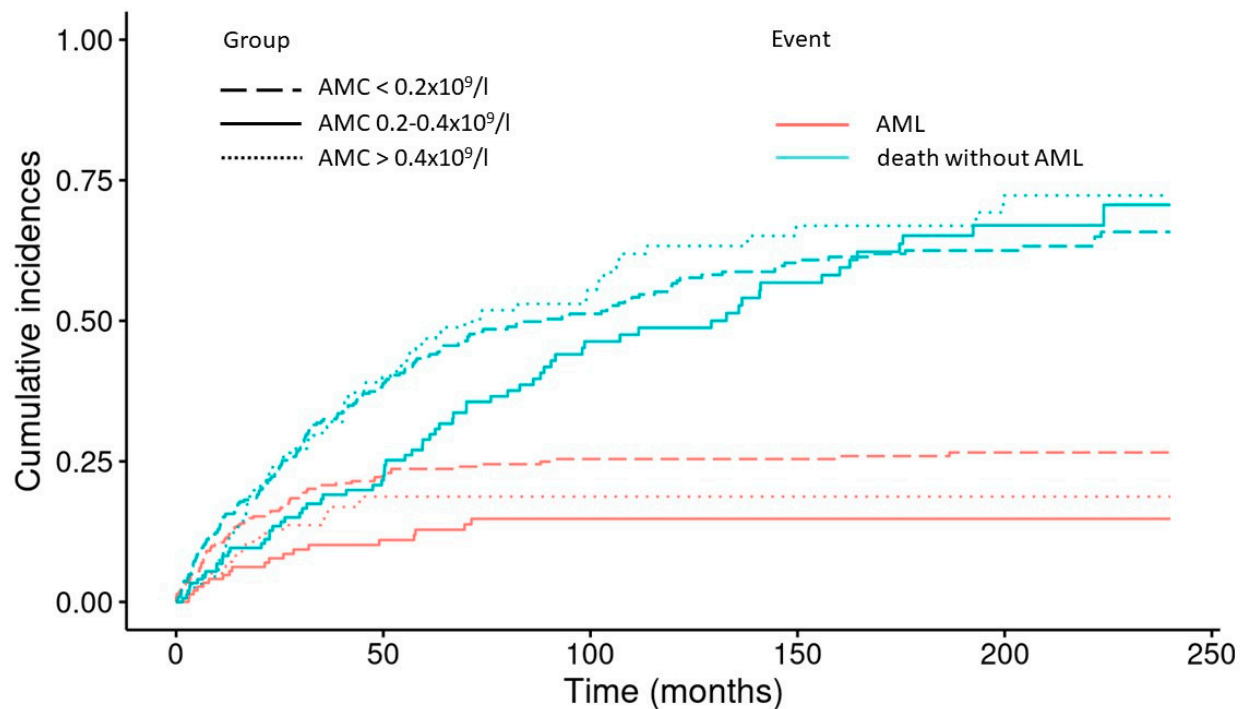


Figure 4. Cumulative incidence curves depicting the probability of progression into AML of patients with primary MDSs, without induction chemotherapy or stem cell transplantation, stratified by the absolute monocyte count ($<0.2 \times 10^9/L$ ($n = 529$), $0.2\text{--}0.4 \times 10^9/L$ ($n = 241$), and $>0.4 \times 10^9/L$ ($n = 223$)) and “death without progression” as the competing event.

4. Discussion

Our analysis of a cohort of 993 uniformly characterized patients reveals a high prevalence of monocytopenia in MDSs. In patients with primary MDSs, who did not receive allogeneic stem cell transplantation or intensive induction chemotherapy ($n = 689$), the AMC affects prognosis independently of the IPSS-R.

Whereas in an early study, a negative impact of monocytosis was reported in MDSs [16], the current data indicate clearly a negative impact of monocytopenia [11,12]. This discrepancy can be explained by the fact that in the early study [16], which was published before the introduction of the FAB classification, 11/37 patients had an $AMC > 1.5 \times 10^9/L$. Those cases would now be classified as chronic myelomonocytic leukemia, making a direct comparison of the results impossible. Currently, the largest body of evidence regarding the prognostic impact of monocytopenia comes from an analysis of the Greek MDS registry [12]. In this analysis of 1719 patients, monocytopenia, defined as an $AMC < 0.2 \times 10^9/L$, was associated with an inferior leukemia-free survival and was described as an adverse risk factor independently from the IPSS-R. Our results from the Düsseldorf MDS registry are fully in line with these observations. A large monocentric study in the USA ($n = 889$) found a negative prognostic impact of monocytopenia, however, only in a univariate analysis and

not within the context of the IPSS-R. This may be explained by a higher cut-off used to define monocytopenia ($AMC < 0.3 \times 10^9/L$).

According to our results, monocytopoiesis seems to be impaired in a considerable proportion of MDS patients. With a median of $0.198 \times 10^9/L$, the AMC in our cohort is considerably lower than the AMCs that have been reported for healthy Caucasian adult men ($0.34 \times 10^9/L$) and women ($0.30 \times 10^9/L$) [17]. There is no general consensus about the definition of monocytopenia, but with regard to a commonly used cut-off of $0.2 \times 10^9/L$ [18] nearly 50 percent of our cohort has to be considered monocytopenic.

Currently, the degree of cytopenias is assessed using binary cut-offs, which are by nature always somewhat arbitrary. Some information may, therefore, be lost. Hence, in the newest version of the IPSS (IPSS-M), cytopenias are for the first time taken into account as continuous variables. If analysed as a continuous variable, the AMC showed a U-shaped curve with the lowest hazard around $0.3 \times 10^9/L$, which corresponds to the median AMC reported for healthy Caucasians [17].

We observed significant differences with regard to the AMC between different MDS subtypes, for example, a higher AMC in MDSs without excess blasts compared to MDSs with excess blasts and in MDSs with ringed sideroblasts (MDS-RS-SLD/MLC) compared to MDSs without ringed sideroblasts (MDS-SLD/MLD). In addition, there was a significant difference regarding the AMC between the IPSS-R subcategories, with the AMC falling continuously with rising IPSS categories. Furthermore, MDS patients with an $AMC < 0.2 \times 10^9/L$ had significantly more severe cytopenias as well. Finally, yet importantly, monocytopenia was associated with a considerably higher risk of transition to acute myeloid leukemia. These observations hint at a direct relationship between MDS pathophysiology and monocytopenia in MDSs.

Systematic studies on the clonal involvement of the monocytic compartment in MDS patients are scarce and mainly report the results of restriction length fragment analyses of X-linked genes. In the small cohorts analysed, monocytes have been consistently shown to be clonal [19–21]. In addition, in MDS cases with monosomy 7, the respective cytogenetic abnormality has been described in both granulocytes and monocytes [22]. It is, therefore, likely that monocytopenia in MDS is due to clonal and ineffective monocytopoiesis. However, given the many changes in bone marrow microenvironment described in MDSs [23], a non-clonal suppression of monocytopoiesis may contribute to monocytopenia as well [24]. Even if monocytopenia in MDSs somehow reflects ineffective haematopoiesis, it represents an additional risk factor independent of the “classical” cytopenias (anemia, thrombocytopenia, and neutropenia), which are routinely taken into account by the IPSS-based scoring systems.

Monocytopenia is a hallmark of some rare inherited myeloid disorders with GATA2-deficiency, such as the MonoMAC (monocytopenia and mycobacterium avium complex infections) syndrome and the dendritic cell, monocyte, and lymphocyte deficiency, DCML [25]. Somatic GATA-2 mutations occur in sporadic forms of myeloid neoplasms as well and have been shown to be associated with monocytopenia, especially if the C-terminal zinc-finger domain is involved [26]. However, the frequency of GATA2 mutations in MDSs is low, with a frequency in the range of 1–2% [27,28]. Hence, GATA2 mutations cannot explain the high prevalence of monocytopenia in MDS patients. Whether monocytopenic MDS patients differ from non-monocytopenic MDS patients with regard to their mutational profile is currently unknown.

As phagocytes do, monocytes and neutrophils share some overlapping functions in host defence, especially against bacterial and fungal infections [29]. Recent reports imply that monocytes from MDS patients exhibit relatively normal innate immune functions compared to monocytes from healthy controls, at least when LPS-induced cytokine production and phagocytosis are assessed *in vitro* [30,31]. In low- and intermediate-risk MDS patients, an increased TNF- α production upon stimulation with LPS has been reported as well, together with an increased proportion of intermediate monocytes ($CD14^{bright}/CD16^{+}$) and a lower proportion of classical monocytes ($CD14^{bright}/CD16^{-}$) [32]. It is, therefore, likely

that monocytes can in part substitute for reduced or defective neutrophils. Hence, monocytopenia in MDS probably goes along with the loss of an additional line of antimicrobial defence and confers an increased risk of infections. However, reduced effector functions of monocytes [33] and monocyte-derived macrophages [34] have been reported as well in MDSs, indicating that more research is needed to clarify the functional capacity of cells of the mononuclear phagocyte system in MDSs, especially since existing data point towards a considerable heterogeneity between certain MDS subtypes [30,33].

Surprisingly, our analysis showed that not only is monocytopenia associated with an increased mortality, but a subtle monocytosis ($>0.4 \times 10^9/L$) as well. The majority of the respective MDS cases would now be classified as CMML according to both the recently revised WHO classification [4] and the newly proposed “international consensus classification” (ICC) [5], since “oligomonocytic” CMML with an AMC $> 0.5 \times 10^9/L$ and at least 10% monocytes in the peripheral blood with a clonal marker and “classical” CMML share largely overlapping features [35–37]. Mutations in genes involved in RNA splicing and epigenetic modification (e.g., *SRSF2*, *U2AF1*, *TET2*, and/or *ASXL1*) are a frequent event in CMML [38]. According to IPSS-M, mutations in *SRSF2*, *U2AF1*, and *ASXL1* are associated with an adverse prognosis in MDSs [9]. The negative prognostic impact of a subtle monocytosis in MDSs could, therefore, at least partially be due to an accumulation of adverse mutations in this patient group. However, an elevated risk of AML transformation was associated only with monocytopenia, but not with subtle monocytosis.

Cells of the monocyte/macrophage system play a crucial role in atherosclerosis and heart failure [39,40]. A higher AMC is associated with an increased cardiovascular risk both in the general population [41,42] and several medical conditions, such as end stage kidney disease [43] and chronic HIV infection [44], as shown recently. Therefore, a higher monocyte count could be an indicator of an increased cardiovascular risk in MDSs. Within this context, observations regarding clonal haematopoiesis of undetermined potential (CHIP) are of importance, which demonstrate an increased expression of inflammatory genes in innate immune cells, such as monocytes and macrophages, in this condition, which provides a potential explanation for the epidemiological link between mutations present in myeloid stem cells and an increased cardiovascular risk [45]. However, further studies are necessary to prove this hypothesis and to assess other potential sequelae of a raised AMC in MDSs.

From a clinical point of view, considering the AMC in MDS care seems to be of value in two ways. For transplant-eligible patients, especially in the intermediate-risk group according to IPSS-R, a low AMC could provide an additional argument for allogeneic stem cell transplantation, especially in combination with other risk factors such as elevated LDH [46], bone marrow fibrosis [47], or transfusion dependency [48]. Given its association with AML transformation, a low AMC could support the use of hypomethylating agents as well, as already proposed [12]. Especially for lower-risk MDS patients with a higher AMC, proper attention should be paid to modifiable cardiovascular risk factors.

Limitations

One major drawback of our study is the lack of molecular data, which prevents a study of the association of the mutational profile with the AMC, and assessing whether the AMC still provides prognostic information when the molecular risk profile is already considered. Furthermore, given the long history of the Düsseldorf MDS registry, the vast majority of non-transplant-eligible patients were treated with best supportive care, and only a minority received modern agents such as IMiDs or HMA. This fact prevents the assessment of a potential impact of these common treatments on the prognostic role of the AMC.

5. Conclusions

Our data clearly demonstrate a prognostic impact of the AMC in MDSs, which is independent from the IPSS-R. Surprisingly, we could demonstrate that both monocytopenia and a subtle monocytosis affect overall survival. Whereas monocytopenia was clearly associated with established adverse prognostic features and risk of AML progression, this

was not the case for a subtle monocytosis. Hence, the factors mediating the negative prognostic impact of a higher AMC in MDSs remain to be elucidated.

Author Contributions: Conceptualization, T.S., M.L. and U.G.; data curation, A.K. and K.N.; formal analysis, T.S. and M.L.; funding acquisition, U.G.; investigation, T.S. and M.L.; methodology, M.L. and U.G.; project administration, U.G.; resources, S.B., A.K., K.N., M.R., B.H., K.S.G. and N.G.; supervision, U.G.; visualization, T.S. and M.L.; writing—original draft, T.S.; writing—review and editing, S.B., A.K., K.N., M.R., B.H., K.S.G., N.G., M.L. and U.G. All authors have read and agreed to the published version of the manuscript.

Funding: Deutsche Krebshilfe (German Cancer Aid), project ID 70113711.

Institutional Review Board Statement: The study was conducted according to the guidelines of the Declaration of Helsinki and was continuously approved by the Ethics Committee of the University Hospital Düsseldorf (project ID 3768, date of current approval 4 March 2022).

Informed Consent Statement: Written informed consent was obtained from each patient before inclusion into the study.

Data Availability Statement: The datasets used and/or analysed during the current study available from the corresponding author on reasonable request.

Conflicts of Interest: No potential conflicts of interest relevant to this article were reported.

References

1. Volpe, V.O.; Garcia-Manero, G.; Komrokji, R.S. SOHO State of the Art & Next Questions: Myelodysplastic Syndromes: A New Decade. *Clin. Lymphoma Myeloma Leuk.* **2021**, *22*, 1–16. [[CrossRef](#)] [[PubMed](#)]
2. Hellström-Lindberg, E.; Tobiasson, M.; Greenberg, P. Myelodysplastic syndromes: Moving towards personalized management. *Haematologica* **2020**, *105*, 1765–1779. [[CrossRef](#)] [[PubMed](#)]
3. Arber, D.A.; Orazi, A.; Hasserjian, R.; Thiele, J.; Borowitz, M.J.; Le Beau, M.M.; Bloomfield, C.D.; Cazzola, M.; Vardiman, J.W. The 2016 revision to the World Health Organization classification of myeloid neoplasms and acute leukemia. *Blood* **2016**, *127*, 2391–2405. [[CrossRef](#)] [[PubMed](#)]
4. Khoury, J.D.; Solary, E.; Abla, O.; Akkari, Y.; Alaggio, R.; Apperley, J.F.; Bejar, R.; Berti, E.; Busque, L.; Chan, J.K.C.; et al. The 5th edition of the World Health Organization Classification of Haematolymphoid Tumours: Myeloid and Histiocytic/Dendritic Neoplasms. *Leukemia* **2022**, *36*, 1703–1719. [[CrossRef](#)] [[PubMed](#)]
5. Arber, D.A.; Orazi, A.; Hasserjian, R.P.; Borowitz, M.J.; Calvo, K.R.; Kvasnicka, H.-M.; Wang, S.A.; Bagg, A.; Barbui, T.; Branford, S.; et al. International Consensus Classification of Myeloid Neoplasms and Acute Leukemias: Integrating morphologic, clinical, and genomic data. *Blood* **2022**, *140*, 1200–1228. [[CrossRef](#)]
6. Kasprzak, A.; Nachtkamp, K.; Gattermann, N.; Germing, U. Assessing the Prognosis of Patients with Myelodysplastic Syndromes (MDS). *Cancers* **2022**, *14*, 1941. [[CrossRef](#)]
7. Greenberg, P.; Cox, C.; Lebeau, M.M.; Fenaux, P.; Morel, P.; Sanz, G.; Sanz, M.; Vallespi, T.; Hamblin, T.; Oscier, D.; et al. International Scoring System for Evaluating Prognosis in Myelodysplastic Syndromes. *Blood* **1997**, *89*, 2079–2088. [[CrossRef](#)]
8. Greenberg, P.L.; Tuechler, H.; Schanz, J.; Sanz, G.; Garcia-Manero, G.; Solé, F.; Bennett, J.M.; Bowen, D.; Fenaux, P.; Dreyfus, F.; et al. Revised International Prognostic Scoring System for Myelodysplastic Syndromes. *Blood* **2012**, *120*, 2454–2465. [[CrossRef](#)]
9. Bernard, E.; Tuechler, H.; Greenberg, P.L.; Hasserjian, R.P.; Ossa, J.E.A.; Nannya, Y.; Devlin, S.M.; Creignou, M.; Pinel, P.; Monnier, L.; et al. Molecular International Prognostic Scoring System for Myelodysplastic Syndromes. *NEJM Evid.* **2022**, *1*, EVIDoa2200008. [[CrossRef](#)]
10. Williams, M.; Mildner, A.; Yona, S. Developmental and Functional Heterogeneity of Monocytes. *Immunity* **2018**, *49*, 595–613. [[CrossRef](#)]
11. Saeed, L.; Patnaik, M.M.; Begna, K.H.; Al-Kali, A.; Litzow, M.R.; Hanson, C.A.; Ketterling, R.P.; Porrata, L.F.; Pardanani, A.; Gangat, N.; et al. Prognostic relevance of lymphocytopenia, monocytopenia and lymphocyte-to-monocyte ratio in primary myelodysplastic syndromes: A single center experience in 889 patients. *Blood Cancer J.* **2017**, *7*, e550. [[CrossRef](#)] [[PubMed](#)]
12. Diamantopoulos, P.T.; Charakopoulos, E.; Symeonidis, A.; Kotsianidis, I.; Viniou, N.-A.; Pappa, V.; Pontikoglou, C.; Tsokanas, D.; Drakos, G.; Kourakli, A.; et al. Real world data on the prognostic significance of monocytopenia in myelodysplastic syndrome. *Sci. Rep.* **2022**, *12*, 17914. [[CrossRef](#)] [[PubMed](#)]
13. Barraco, D.; Cerquozzi, S.; Gangat, N.; Patnaik, M.M.; Lasho, T.; Finke, C.; Hanson, C.A.; Ketterling, R.P.; Pardanani, A.; Tefferi, A. Monocytosis in polycythemia vera: Clinical and molecular correlates. *Am. J. Hematol.* **2017**, *92*, 640–645. [[CrossRef](#)]
14. Tefferi, A.; Shah, S.; Mudireddy, M.; Lasho, T.L.; Barraco, D.; Hanson, C.A.; Ketterling, R.P.; Elliott, M.A.; Patnaik, M.S.; Pardanani, A.; et al. Monocytosis is a powerful and independent predictor of inferior survival in primary myelofibrosis. *Br. J. Haematol.* **2017**, *183*, 835–838. [[CrossRef](#)] [[PubMed](#)]

15. Germing, U.; Strupp, C.; Kuendgen, A.; Isa, S.; Knipp, S.; Hildebrandt, B.; Giagounidis, A.; Aul, C.; Gattermann, N.; Haas, R. Prospective validation of the WHO proposals for the classification of myelodysplastic syndromes. *Haematologica* **2006**, *91*, 1596–1604.
16. Economopoulos, T.; Stathakis, N.; Maragoyannis, Z.; Gardikas, E.; Dervenoulas, J. Myelodysplastic Syndrome. Clinical and Prognostic Significance of Monocyte Count, Degree of Blastic Infiltration, and Ring Sideroblasts. *Acta Haematol.* **1981**, *65*, 97–102. [[CrossRef](#)]
17. Bain, B.J. Ethnic and sex differences in the total and differential white cell count and platelet count. *J. Clin. Pathol.* **1996**, *49*, 664–666. [[CrossRef](#)]
18. Zini, G. CHAPTER 16—Abnormalities in Leukocyte Morphology and Number. In *Blood and Bone Marrow Pathology*, 2nd ed.; Porwit, A., McCullough, J., Erber, W.N., Eds.; Churchill Livingstone: Edinburgh, UK, 2011; pp. 247–261, ISBN 978-0-7020-3147-2.
19. Janssen, J.W.; Buschle, M.; Layton, M.; Drexler, H.G.; Lyons, J.; van den Berghe, H.; Heimpel, H.; Kubanek, B.; Kleihauer, E.; Mufti, G.J. Clonal Analysis of Myelodysplastic Syndromes: Evidence of Multipotent Stem Cell Origin. *Blood* **1989**, *73*, 248–254. [[CrossRef](#)]
20. Tefferi, A.; Thibodeau, S.N.; Solberg, L.A. Clonal Studies in the Myelodysplastic Syndrome Using X-Linked Restriction Fragment Length Polymorphisms. *Blood* **1990**, *75*, 1770–1773. [[CrossRef](#)]
21. Van Kamp, H.; Fibbe, W.E.; Jansen, R.P.; van der Keur, M.; de Graaff, E.; Willemze, R.; Landegent, J.E. Clonal Involvement of Granulocytes and Monocytes, but Not of T and B Lymphocytes and Natural Killer Cells in Patients with Myelodysplasia: Analysis by X-Linked Restriction Fragment Length Polymorphisms and Polymerase Chain Reaction of the Phosphoglycerate Kinase Gene. *Blood* **1992**, *80*, 1774–1780.
22. Kere, J.; Ruutu, T.; de la Chapelle, A. Monosomy 7 in Granulocytes and Monocytes in Myelodysplastic Syndrome. *N. Engl. J. Med.* **1987**, *316*, 499–503. [[CrossRef](#)] [[PubMed](#)]
23. Calvi, L.M.; Li, A.J.; Becker, M.W. What is the role of the microenvironment in MDS? *Best Pract. Res. Clin. Haematol.* **2019**, *32*, 101113. [[CrossRef](#)] [[PubMed](#)]
24. Emoto, T.; Lu, J.; Sivasubramaniyam, T.; Maan, H.; Khan, A.B.; Abow, A.A.; Schroer, S.A.; Hyduk, S.J.; Althagafi, M.G.; McKee, T.D.; et al. Colony stimulating factor-1 producing endothelial cells and mesenchymal stromal cells maintain monocytes within a perivascular bone marrow niche. *Immunity* **2022**, *55*, 862–878.e8. [[CrossRef](#)] [[PubMed](#)]
25. Mir, M.A.; Kochuparambil, S.T.; Abraham, R.S.; Rodriguez, V.; Howard, M.; Hsu, A.P.; Jackson, A.E.; Holland, S.M.; Patnaik, M.M. Spectrum of myeloid neoplasms and immune deficiency associated with germline *GATA 2* mutations. *Cancer Med.* **2015**, *4*, 490–499. [[CrossRef](#)] [[PubMed](#)]
26. Crispino, J.D.; Horwitz, M.S. *GATA* factor mutations in hematologic disease. *Blood* **2017**, *129*, 2103–2110. [[CrossRef](#)]
27. Papaemmanuil, E.; Gerstung, M.; Malcovati, L.; Tauro, S.; Gundem, G.; Van Loo, P.; Yoon, C.J.; Ellis, P.; Wedge, D.C.; Pellagatti, A.; et al. Clinical and biological implications of driver mutations in myelodysplastic syndromes. *Blood* **2013**, *122*, 3616–3627. [[CrossRef](#)] [[PubMed](#)]
28. Haferlach, T.; Nagata, Y.; Grossmann, V.; Okuno, Y.; Bacher, U.; Nagae, G.; Schnittger, S.; Sanada, M.; Kon, A.; Alpermann, T.; et al. Landscape of genetic lesions in 944 patients with myelodysplastic syndromes. *Leukemia* **2014**, *28*, 241–247. [[CrossRef](#)]
29. Soehnlein, O.; Lindbom, L. Phagocyte partnership during the onset and resolution of inflammation. *Nat. Rev. Immunol.* **2010**, *10*, 427–439. [[CrossRef](#)]
30. Schmidt, C.S.; Lopez, P.A.; Dopheide, J.F.; Schmidt, F.; Theobald, M.; Schild, H.; Lauinger-Lörsch, E.; Nolte, F.; Radsak, M.P. Phenotypic and functional characterization of neutrophils and monocytes from patients with myelodysplastic syndrome by flow cytometry. *Cell. Immunol.* **2016**, *308*, 19–26. [[CrossRef](#)]
31. Pollyea, D.A.; Hedin, B.R.; O'Connor, B.P.; Alper, S. Monocyte function in patients with myelodysplastic syndrome. *J. Leukoc. Biol.* **2018**, *104*, 641–647. [[CrossRef](#)]
32. Velegraki, M.; Papakonstantinou, N.; Kalaitzaki, L.; Ntoufa, S.; Laidou, S.; Tsagiopoulou, M.; Bizymi, N.; Damianaki, A.; Mavroudi, I.; Pontikoglou, C.; et al. Increased proportion and altered properties of intermediate monocytes in the peripheral blood of patients with lower risk Myelodysplastic Syndrome. *Blood Cells Mol. Dis.* **2020**, *86*, 102507. [[CrossRef](#)] [[PubMed](#)]
33. Prodan, M.; Tulissi, P.; Perticarari, S.; Presani, G.; Franzin, F.; Pussini, E.; Pozzato, G. Flow cytometric assay for the evaluation of phagocytosis and oxidative burst of polymorphonuclear leukocytes and monocytes in myelodysplastic disorders. *Haematologica* **1995**, *80*, 212–218. [[PubMed](#)]
34. Han, Y.; Wang, H.; Shao, Z. Monocyte-Derived Macrophages Are Impaired in Myelodysplastic Syndrome. *J. Immunol. Res.* **2016**, *2016*, 5479013. [[CrossRef](#)] [[PubMed](#)]
35. Geyer, J.T.; Tam, W.; Liu, Y.-C.; Chen, Z.; A Wang, S.; Bueso-Ramos, C.; Oak, J.; Arber, D.A.; Hsi, E.; Rogers, H.J.; et al. Oligomonocytic chronic myelomonocytic leukemia (chronic myelomonocytic leukemia without absolute monocytosis) displays a similar clinicopathologic and mutational profile to classical chronic myelomonocytic leukemia. *Mod. Pathol.* **2017**, *30*, 1213–1222. [[CrossRef](#)] [[PubMed](#)]
36. Schuler, E.; Frank, F.; Hildebrandt, B.; Betz, B.; Strupp, C.; Rudelius, M.; Aul, C.; Schroeder, T.; Gattermann, N.; Haas, R.; et al. Myelodysplastic syndromes without peripheral monocytosis but with evidence of marrow monocytosis share clinical and molecular characteristics with CMML. *Leuk. Res.* **2017**, *65*, 1–4. [[CrossRef](#)]
37. Calvo, X.; Garcia-Gisbert, N.; Parraga, I.; Gibert, J.; Florensa, L.; Andrade-Campos, M.; Merchan, B.; Garcia-Avila, S.; Montesdeoca, S.; Fernández-Rodríguez, C.; et al. Oligomonocytic and overt chronic myelomonocytic leukemia show similar clinical, genomic, and immunophenotypic features. *Blood Adv.* **2020**, *4*, 5285–5296. [[CrossRef](#)]

38. Patnaik, M.M.; Lasho, T.L. Genomics of myelodysplastic syndrome/myeloproliferative neoplasm overlap syndromes. *Hematol. Am. Soc. Hematol. Educ. Program* **2020**, *2020*, 450–459. [[CrossRef](#)]
39. Fayad, Z.A.; Swirski, F.K.; Calcagno, C.; Robbins, C.S.; Mulder, W.; Kovacic, J.C. Monocyte and Macrophage Dynamics in the Cardiovascular System: JACC Macrophage in CVD Series (Part 3). *J. Am. Coll. Cardiol.* **2018**, *72*, 2198–2212. [[CrossRef](#)]
40. Shahid, F.; Lip, G.Y.; Shantsila, E. Role of Monocytes in Heart Failure and Atrial Fibrillation. *J. Am. Heart Assoc.* **2018**, *7*, e007849. [[CrossRef](#)]
41. Waterhouse, D.F.; Cahill, R.A.; Sheehan, F.; McCreery, C. Prediction of calculated future cardiovascular disease by monocyte count in an asymptomatic population. *Vasc. Health Risk Manag.* **2008**, *4*, 177–187. [[CrossRef](#)]
42. Gupta, K.; Kalra, R.; Pate, M.; Nagalli, S.; Ather, S.; Rajapreyar, I.; Arora, P.; Gupta, A.; Zhou, W.; Estepar, R.S.J.; et al. Relative Predictive Value of Circulating Immune Markers in US Adults Without Cardiovascular Disease: Implications for Risk Reclassification. *Mayo Clin. Proc.* **2021**, *96*, 1812–1821. [[CrossRef](#)] [[PubMed](#)]
43. Oh, E.S.; You, Z.; Nowak, K.L.; Jovanovich, A.J. Association of Monocyte Count and Monocyte/Lymphocyte Ratio with the Risk of Cardiovascular Outcomes in Patients with CKD. *Kidney360* **2022**, *3*, 657–665. [[CrossRef](#)] [[PubMed](#)]
44. Bogorodskaya, M.; Lyass, A.; Mahoney, T.; Borowsky, L.; Sen, P.; Swirski, F.; Srinivasa, S.; Longenecker, C.; Massaro, J.; D'agostino, R.; et al. Utilization of absolute monocyte counts to predict cardiovascular events in people living with HIV. *HIV Med.* **2020**, *22*, 314–320. [[CrossRef](#)] [[PubMed](#)]
45. Jaiswal, S.; Libby, P. Clonal haematopoiesis: Connecting ageing and inflammation in cardiovascular disease. *Nat. Rev. Cardiol.* **2019**, *17*, 137–144. [[CrossRef](#)] [[PubMed](#)]
46. Germing, U.; Hildebrandt, B.; Pfeilstöcker, M.; Nösslinger, T.; Valent, P.; Fonatsch, C.; Lübbert, M.; Haase, D.; Steidl, C.; Krieger, O.; et al. Refinement of the international prognostic scoring system (IPSS) by including LDH as an additional prognostic variable to improve risk assessment in patients with primary myelodysplastic syndromes (MDS). *Leukemia* **2005**, *19*, 2223–2231. [[CrossRef](#)] [[PubMed](#)]
47. Jain, A.G.; Zhang, L.; Bennett, J.M.; Komrokji, R. Myelodysplastic Syndromes with Bone Marrow Fibrosis: An Update. *Ann. Lab. Med.* **2022**, *42*, 299–305. [[CrossRef](#)]
48. Benton, C.B.; Khan, M.; Sallman, D.; Nazha, A.; Nogueras González, G.M.; Piao, J.; Ning, J.; Aung, F.; Al Ali, N.; Jabbour, E.; et al. Prognosis of patients with intermediate risk IPSS-R myelodysplastic syndrome indicates variable outcomes and need for models beyond IPSS-R. *Am. J. Hematol.* **2018**, *93*, 1245–1253. [[CrossRef](#)]

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.