Review

Circular RNAs Activity in the Leukemic Bone Marrow Microenvironment

Francesca Liccardo [ID], Alessia Iaiza, Martyna Śniegocka, Silvia Masciarelli *[ID] and Francesco Fazi *[ID]

Department of Anatomical, Histological, Forensic & Orthopedic Sciences,
Section of Histology & Medical Embryology, Sapienza University of Rome, Via A. Scarpa, 14-16,
00161 Rome, Italy; francesca.liccardo@uniroma1.it (F.L.); alessia.iaiza@uniroma1.it (A.I.);
martyna.sniegocka@uniroma1.it (M.Ś.)
* Correspondence: silvia.masciarelli@uniroma1.it (S.M.); francesco.fazi@uniroma1.it (F.F.);
  Tel.: +39-06-49766575 (S.M. & F.F.); Fax: +39-06-4462854 (S.M. & F.F.)

Abstract: Acute myeloid leukemia (AML) is a hematological malignancy originating from defective hematopoietic stem cells in the bone marrow. In spite of the recent approval of several molecular targeted therapies for AML treatment, disease recurrence remains an issue. Interestingly, increasing evidence has pointed out the relevance of bone marrow (BM) niche remodeling during leukemia onset and progression. Complex crosstalk between AML cells and microenvironment components shapes the leukemic BM niche, consequently affecting therapy responsiveness. Notably, circular RNAs are a new class of RNAs found to be relevant in AML progression and chemoresistance. In this review, we provided an overview of AML-driven niche remodeling. In particular, we analyzed the role of circRNAs and their possible contribution to cell–cell communication within the leukemic BM microenvironment. Understanding these mechanisms will help develop a more effective treatment for AML.

Keywords: circRNA; acute myeloid leukemia; bone marrow niche; tumor microenvironment; non-coding RNA; hematopoietic stem cell; therapy resistance

1. Introduction

Acute myeloid leukemia (AML) is the most common form of acute leukemia in adults. It is aggressive cancer characterized by clonal expansion and progressive accumulation of incompletely differentiated myeloid cells within the bone marrow. AML is a heterogeneous disease because of the plethora of mutations responsible for its development. Thus it is defined by diverse symptoms, prognosis and treatment responsiveness [1]. In most patients, etiology remains unknown, with the exception of therapy-related leukemias due to the previous exposure to chemotherapy and/or radiation administered for a primary condition. Incidence is higher in elderly people than in younger, and males are 1.2–1.6 times more likely to develop AML than females. Furthermore, conventional frontline treatments such as intensive chemotherapy and allogeneic stem cell transplantation are reliable and more effective in young and fit patients than in old/unfit ones [1–3]. Treatment is mostly based on conventional chemotherapy with a generally dismal outcome, especially in the elderly, with a 5-year survival of 30–35% in younger patients (age < 60 years) and 10–15% in older ones (age $\geq$ 60 years) [4]. This large gap in therapy efficacy is beginning to close thanks to the approval of nine new drugs for targeted therapy of AML within just three years (2017–2020). An enormous effort in better understanding of AML genomic and molecular landscape has made this possible [5]. Although these different compounds are promising targeted therapies for many distinct AML subtypes [6,7], still primary and secondary resistance remains an issue [8]. Therefore, there is an extreme need to explore the mechanisms of resistance to treatments. In the last few years, many new evidences stressed the role of the bone marrow niche in sustaining leukemic cells' survival, progression and resistance to

chemotherapy. The maintenance of this integrated system occurs through the exchange of factors and signals, generating intense cell–cell crosstalk. Numerous studies found several non-coding RNAs (ncRNAs), such as long non-coding RNAs, microRNAs and circular RNAs, playing a prominent role in both AML leukemogenesis and chemoresistance [9–12]. In this review, we aimed to report new insights into the interplay between AML cells and the microenvironment in order to provide a comprehensive view of the role of niche components during leukemic progression. Moreover, since the clinical significance of microRNA and long non-coding RNA has been investigated for several years, we highlighted the very last insights on the role of circular RNAs (circRNAs) in AML pathogenesis, with a particular focus on their function as potential mediators of crosstalk within the bone marrow niche.

2. Hematopoietic Stem Cells and Their Niche

Hematopoiesis is a process starting from a small group of Hematopoietic Stem Cells (HSCs) which are multipotent precursors endowed with self-renewal capacity and the ability to generate all types of mature blood cells through multi-lineage differentiation programs [13]. HSCs are identified on the basis of the expression of specific surface markers. Indeed, murine HSCs are identified by a marker combination $Lin^{neg/low}$, $Thy1.1^{low}$, $c\text{-}Kit^{high}$, $Sca\text{-}1^+]$, and a similar combination is detected in human HSCs Lin^-, $Thy1^+$, $CD34^+$, $CD38^{neg/low}]$ [14]. Moreover, HSCs can be divided on the basis of their differentiation potential. Long-term HSCs (LT-HSCs) have an indefinite self-renewal capacity, while their short-term derivative HSCs (ST-HSCs) maintain self-renewal property for eight weeks, then giving rise to multipotent progenitors (MMPs). HSCs are rare cells located in a complex and heterogeneous bone marrow microenvironment composed of both hematopoietic and non-hematopoietic cells surrounded by an extracellular matrix, in a ratio of 1:5000 for LT-HSC and 1:1000 for ST-HSC and multipotent progenitors in the murine bone marrow. Other components of the bone marrow are mesenchymal stem and progenitor cells, osteoprogenitor cells, perivascular stromal cells, endothelial cells, adipocytes, unmyelinated Schwann cells and cells of the immune system. The spatial organization and composition of these populations play a pivotal role in the regulation of HSCs' maintenance and fate decisions. Although the specific location of HSCs still remains not fully clear, they preferentially localize in perisinusoidal areas [15,16]. Indeed, bone marrow is highly vascular, with arterioles close to the endosteum and sinusoids winding through a network of reticular stromal cells. Especially endothelial cells and perivascular stromal cells support HSCs maintenance and long-term repopulating activity by producing factors such as chemokine CXCL12, angiopoietin and stem cell factor (SCF) [17–19]. Additionally, HSC niche maintenance relies on osteolineage, adipocytes and macrophages [20–22]. A detailed characterization of the other bone marrow niche components can be found in dedicated reviews [23,24].

3. Circular RNAs and Their Role in Hematopoiesis

The hematopoietic process is tightly regulated through gene expression modulation in HSCs and progenitor cells. It is well known that diverse differentiation programs are triggered by both specific transcription factors and non-coding RNAs, such as long non-coding RNAs (lncRNAs) and microRNAs (miRNAs) [25–27]. However, there is a need to better understand the roles played by circular RNAs (circRNAs) in hematopoiesis. circRNAs are covalently closed single-stranded molecules derived from back-splicing, non-canonical splicing in which flanking regions of one or multiple exons are joined together. Previously thought to result from random splicing errors, circRNAs were proven to be conserved and associated with inverted repeated Alu sequences within the flanking introns [28]. RNA splicing occurs within the nucleus. During back splicing, the canonical spliceosomal machinery recognizes the splice sites, and circularization can be direct or preceded by the production of a long lariat. The entire process is usually regulated by RNA-binding proteins. For instance, during epithelial to mesenchymal transition, the RNA-binding protein Quaking induces the formation of numerous circRNAs [29]. Human circRNAs are co-transcriptionally or post-transcriptionally produced, most of them containing two or

three exons. The vast majority of circRNAs have no introns and undergo nuclear export (Figure 1A). Notably, circularization competes with linear splicing, strongly suggesting that circRNAs have a role in gene regulation [30]. Indeed, several biological functions of circRNAs have been identified so far: they can act as "sponges" for miRNA or RNA-binding proteins, activate gene transcription and, in some cases, they are also translated into proteins in a N 6-methyladenosine (m^{6}A)-dependent manner (Figure 1B) [31]. An interesting work by Pengyan Xia and colleagues showed that maintenance of LT-HSCs homeostasis is regulated by a circRNA named cia-cGAS. It binds the DNA sensor cGAS in the nucleus, thus blocking its synthase activity. Indeed, circRNA cia-cGAS deficiency leads to increased levels of type I IFNs and LT-HSCs cycling. Therefore, since cia-cGAS is highly expressed in dormant HSCs and protects them from c-GAS-mediated exhaustion, it is a key factor in keeping the balance between HSCs' quiescent and cycling state [32]. Nicolet and colleagues identified more than 59,000 circRNAs in hematopoietic cells, providing the first comprehensive analysis of circRNAs expression in these cells [33]. Importantly, they found that circRNAs are cell-type specific, and their expression levels are altered during terminal hematopoietic differentiation. Differentiated cells, especially erythrocytes and platelets, produce more circRNAs than progenitor cells. Just recently, this group also showed that there is limited correspondence between circRNAs expressed in mature cells and those found in their progenitor cells, suggesting that differential expression of circRNAs is a regulated process rather than mere accumulation. Importantly, by comparing the expression of circRNAs with the translation efficiency of the counterpart mRNA, they found a correlation only in a small percentage (0.04%) of cases. Hence, the ways through which these molecules are regulated still need to be elucidated [34]. Nevertheless, these findings indicate a remarkable role for circRNAs in the modulation of hematopoietic differentiation.
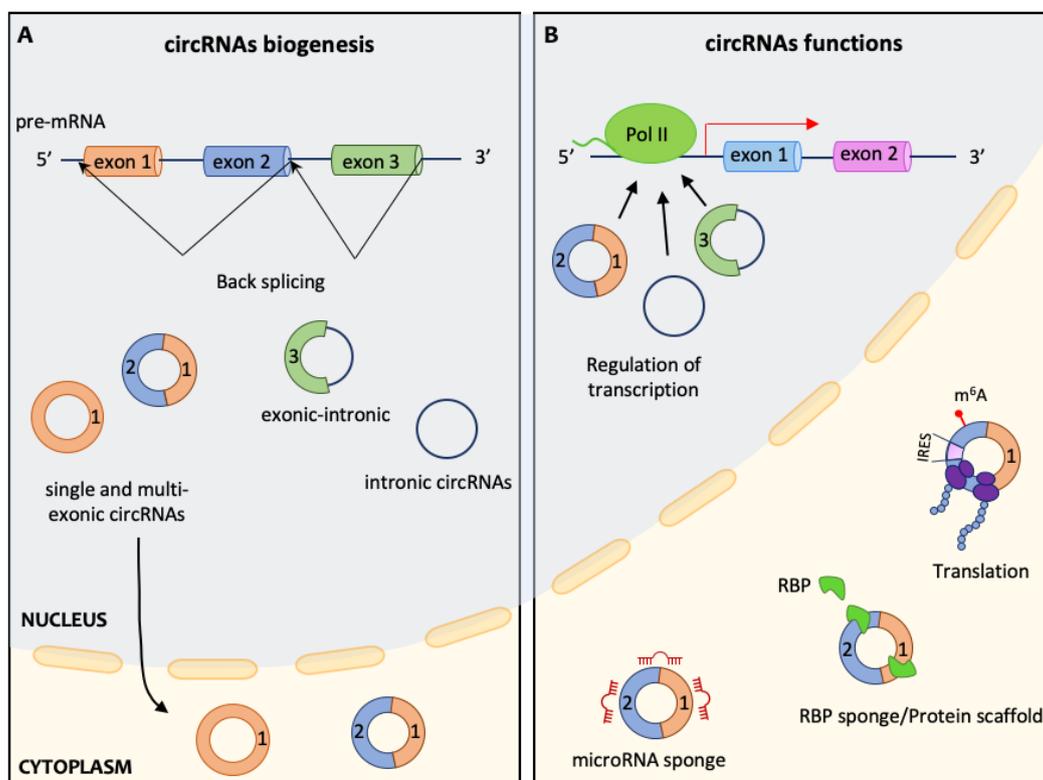

Figure 1. *Cont.*
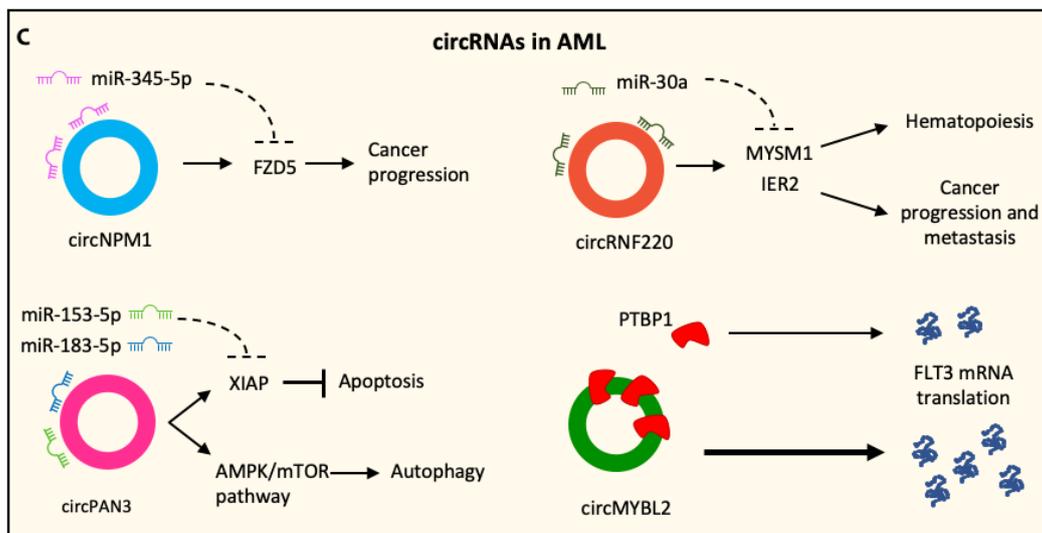
Figure 1. (**A**) circRNAs biogenesis. circRNAs are generated by unconventional splicing, named back-splicing, in which the 3′ end of an exon binds the 5′ end of an upstream exon, forming a covalently closed molecule. circRNAs can be divided into three main classes: single or multi-exonic circRNAs (on the left), which can translocate in the cytoplasm; exonic-intronic circRNAs (in the middle) and intronic-circRNAs (on the right), which are retained in the nucleus. (**B**) circRNAs functions. CircRNAs can regulate transcription, interacting with Polymerase-II (Pol II) in the nucleus. In the cytoplasm, they can act as microRNA sponges or RBP sponges, and they can also be translated thanks to the presence of an IRES or m6A modification. (**C**) circRNAs in AML. Representation of some circRNAs involved in AML progression: circNPM1 acts as miR-345–5p sponge, increasing FZD5 expression, a well-known oncogene; circPAN3, sponging miR-153-5p and miR-183-5p, leads to increased autophagy and inhibits apoptosis; circRNF220 induces MYSM1 and IER2, affecting hematopoiesis and promoting cancer progression and metastasis; circMYBL2 favors mRNA FLT3 translation by binding PTBP1.

4. AML and the Leukemic BM Niche

The starting point of AML onset is the progressive addition of several leukemia-associated mutations in HSCs, which become pre-leukemic HSCs (pre-LSCs) [35]. The conversion from pre-LSCs to fully transformed leukemic stem cells (LSCs) or leukemia-initiating cells (LICs) is a multistep process in which sequential aberrations in transcription, epigenetic regulation and expression of metabolic factors are acquired over the years [36]. While both pre-LSCs and LSCs maintain their self-renewal capacity, they also generate leukemic blasts, which form the bulk of the tumor [37]. In addition to the study of genetic alterations in HSCs resulting in leukemogenesis, a new perspective focusing on alterations of the bone marrow niche has been emerging in the last few years. Indeed, germline mutations in stromal cells can promote dysregulated hematopoiesis, and they are sufficient for the development of AML [38]. Although niche genetic alterations that predispose to hematological malignancies were identified, these studies have been held in mouse models, and the mechanisms occurring in patients need to be elucidated. On the other hand, both leukemia progression and development of therapy resistance occur through a deep remodeling of the bone marrow microenvironment (Figure 2). For instance, neo-angiogenesis is driven by enhanced expression of vascular endothelial growth factor (VEGF) and angiopoietin in both leukemic and bone marrow stromal cells [39,40]. Moreover, patient-derived xenografts (PDX) have a remarkable vascular leakiness, and this abnormal permeability strongly affects drug delivery, proving to be associated with poor prognosis in AML patients [41]. Another strategy used by leukemic cells to invade the bone marrow microenvironment is the disruption of healthy hematopoietic stem and progenitor cells reservoir [42]. Moreover, immune escape is a critical point for leukemia progression and therapy resistance. To this aim, AML blasts implement several strategies: they downregulate surface membrane MHC

class I and II molecules to avoid immune recognition, induce NK and T cells dysfunction, favor immunosuppressive Treg cells and alter cytokine milieu in a pro-leukemic way. Of note, mesenchymal stem cells secrete factors so as to foster immunosuppression and recruit Treg and M2 macrophages [43,44]. Compared to their normal counterpart, leukemic cells have an altered energy metabolism, with higher mitochondrial mass and oxygen consumption but no concomitant increase in respiratory chain complex activity. Since AML blasts strongly rely on fatty acid oxidation (FAO) and oxidative phosphorylation (OXPHOS), they are susceptible to oxidative metabolic stress [45,46]. Likewise, some solid tumors and AML cells exploit adipocytes as a source of fatty acids (FA) [47]. Moreover, it was demonstrated that an increase in AML mitochondrial mass is due to the direct uptake of functional mitochondria from bone marrow stromal cells. This mitochondria transfer, which increases ATP production in leukemic blasts, is remarkably enhanced during chemotherapy and confers chemoresistance [48–50]. Within the bone marrow, exosomes play a critical role in cell-to-cell communication and in AML-mediated microenvironment remodeling in order to create a self-strengthening leukemic niche [51,52]. These vesicles transfer different bioactive molecules such as proteins, lipids, DNA and RNA. Since hypoxia, nutrients deprivation and acidosis render bone marrow an extremely hostile environment even for leukemic blasts, they cope with ER stress conditions by activating the unfolded protein response (UPR) [53] and it was shown that AML extracellular vesicles (AML-EVs) transfer ER stress to bone marrow mesenchymal stem cells, thus creating a leukemia permissive microenvironment [54]. Ultimately, the anchorage of leukemic stem cells to the niche also plays an essential role in AML pathogenesis [55,56].
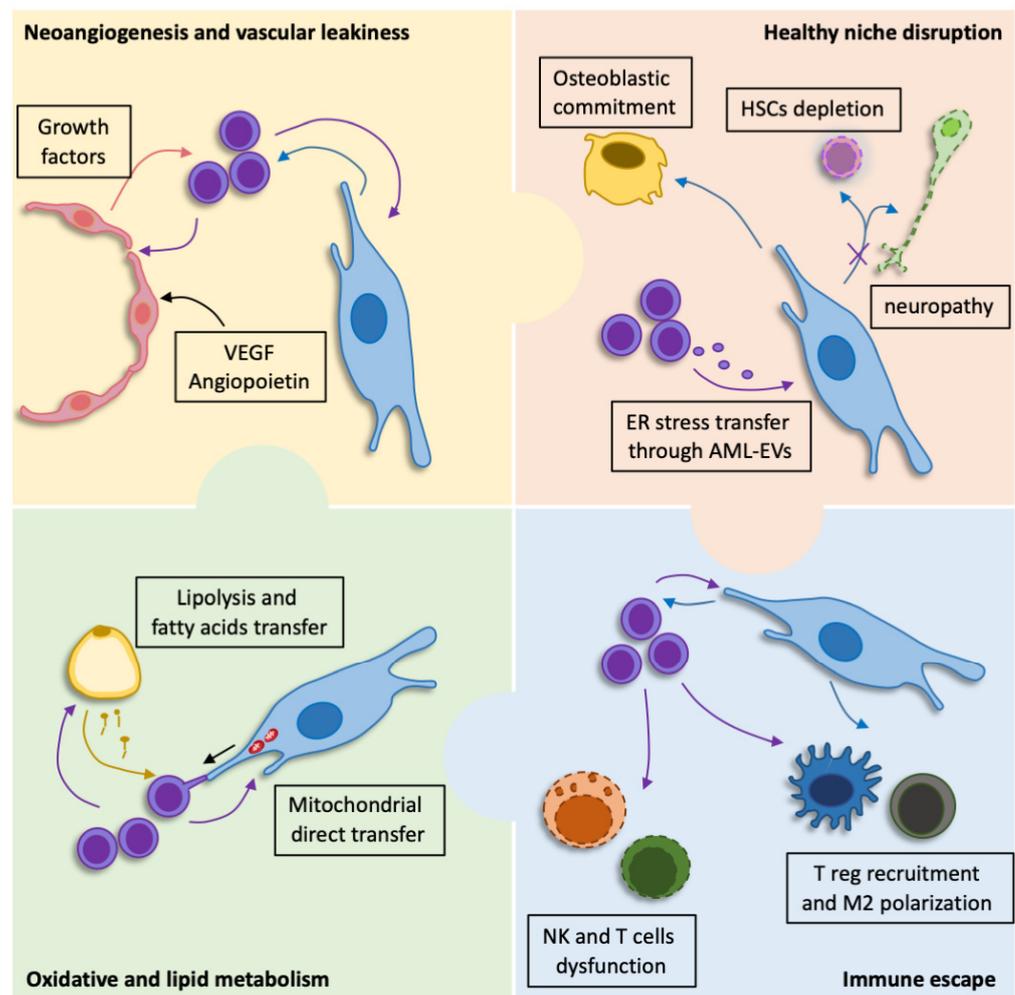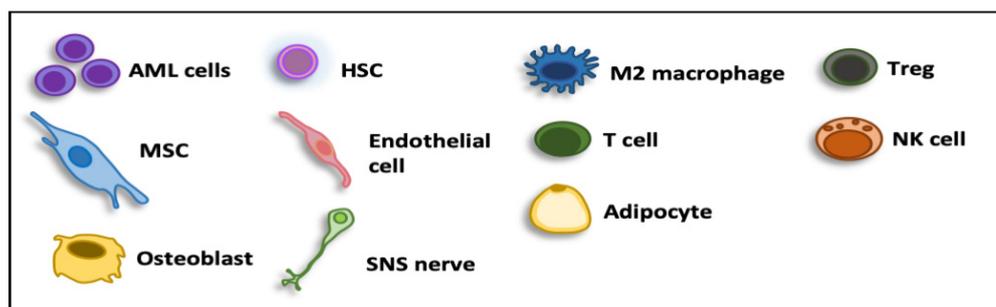

Figure 2. *Cont.*

Figure 2. Interaction between AML cells and the bone marrow microenvironment. Within the niche, AML cells communicate with mesenchymal stem cells and endothelial cells in order to increase bone marrow vascularization and alter vascular permeability (**yellow panel**); Moreover, AML cells strongly reprogram MSCs, leading to a self-reinforcing niche at the expense of normal hematopoiesis (**pink panel**); Leukemic cells escape the immune system and recruit anti-inflammatory components such as Treg and M2 macrophages (**light blue panel**); AML cells exploit stromal cells to enhance their antioxidant defenses and adipocytes as a source of cellular energy (**green panel**).

5. circRNA in AML Pathogenesis

Several circRNAs were found to play oncogenic or tumor-suppressive roles, and aberrant expression was detected in both solid and hematopoietic cancers [57], even though, in many cases, the underlying mechanisms and regulatory networks are still to be clarified. For example, it was shown that N6-methyladenosine (m^6A) modification has an important regulatory function for circRNAs in AML [58]. Table 1 shows the annotated circRNAs implicated in AML so far. The vast majority of these circRNAs act as miRNA sponges; they are defined as competing for endogenous RNAs (ceRNAs) that, by binding miRNAs, compete for their binding with the target mRNAs and hinder their regulatory function. Therefore, circRNAs can alter gene expression regulation, hence contributing to AML pathogenesis.

Table 1. Updated list of circRNAs involved in AML onset, progression and therapy resistance. Arrows ↑ and ↓ stand for upregulation and downregulation respectively.

Name	Gene of Origin	Levels	Pathway Targeted/ Mode of Action	Impact	Ref.
f-circPR	PML-RARA	de novo in AML	AKT signaling	Increased cell proliferation and chemotherapy resistance	[59]
f-circM9	MLL-AF9	de novo in AML	MAPK and AKT signalling	Increased cell proliferation and chemotherapy resistance	[59]
circ ANAPC7 hsa_circ_101141	ANAPC7	↑ in AML	miR181	Prognostic biomarker	[60]
circNPM1 hsa_circ_0075001	NPM1	↑ in AML	mir181 and TLR signalling miR-345-5p/FZD5	Hematopoietic differentiation Chemotherapy resistance	[61,62]
circDLEU hsa_circ_0000488	DLEU	↑ in AML	miR496/PRKACB	Increased cell proliferation and apoptosis inhibition	[63]
circANXA2 hsa_circ_0035559	Annexin A2	↑ in AML	miR-23a-5p and miR-503-3p	Prognostic biomarker and chemotherapy resistance	[64]
circVIM	Vimentin	↑ in AML	Unknown	Diagnostic and prognostic biomarker	[65]
circHIPK2	HIPK2	↓ in AML (APL)	miR-124-3p/CEBPA	Prognostic biomarker and ATRA-induced differentiation	[66]
circPAN3 hsa_circ_0100181	PAN3	↑ in AML ADM resistant	AMPK/mTOR miR-153-5p/XIAP	Chemotherapy resistance	[67,68]
hsa_circ_0004277	WDR7	↓ in AML	miR-134-5p/SSBP2	Diagnostic and prognostic biomarker	[69,70]
hsa_circ_0003602	SMARCC1	↑ in AML	miR-502-5p/IGF1R	Increased cell proliferation and apoptosis inhibition	[71]

Table 1. *Cont.*

Name	Gene of Origin	Levels	Pathway Targeted/ Mode of Action	Impact	Ref.
circMYBL2 hsa_circ_0006332	MYBL2	↑ in AML FLT3-ITD+	PTPB1/FLT3 translation	Increased cell proliferation and resistance to quizartinib	[72]
hsa_circ_0009910	MFN2	↑ in AML and AML exosomes	miR-5195-3p/GRB10 miR-20a-5p	Increased cell proliferation and apoptosis inhibition	[73,74]
hsa_circ_0121582	GSK3beta	↓ in AML	miR-224/GSK3β/Wnt/βcatenin	Inhibited cell proliferation	[75]
circFOXO3	FOXO3	↓ in AML	Apoptotic pathways	Increased apoptosis Diagnostic and prognostic biomarker	[76]
circRNF220 hsa_circ_0012152	RNF220	↑ in AML relapse	miR30a/MYSM1-IER2	Increased cell proliferation and apoptosis inhibition, biomarker to predict relapse	[77]
hsa_circ_100290	SLC30A7	↑ in AML	miR-203/Rab10	Increased cell proliferation and apoptosis inhibition	[78]
circRNF13 hsa_circ_0001346	RNF13	↑ in AML	miR-1224-5p	Increased cell proliferation and apoptosis inhibition	[79]
hsa_circ_104700	PTK2	↑ in AML	miR-330-5p/FOXM1	Increased cell proliferation and apoptosis inhibition	[80]
hsa_circ_002483	PTK2	↑ in AML	miR-758-3p/MYC	Increased cell proliferation and apoptosis inhibition	[81]
hsa_circ_0005774	CDK1	↑ in AML	miR192-5p/ULK1	Increased cell proliferation and apoptosis inhibition	[82]
circCRKL	CRKL	↓ in AML	miR-196a-5p/p27 miR-196b-5p/p27	Inhibited cell proliferation	[83]
circPOLA2	POLA2	↑ in AML	miR-34a	Increased cell proliferation	[84]
hsa_circ_0079480	ISPD	↑ in AML	miR-654-3p/HDGF	Increased cell proliferation and apoptosis inhibition Prognostic biomarker	[85,86]
circKLHL8	KLHL8	Associated with outcome	miR-155/CDKN1-CDKN2-BCL6-TLR4-CEBPD-CEBPB	Prognostic biomarker	[87]
circFBXW7	FBXW7	↓ in AML	Signal transduction Leukocyte differentiation	Tumor suppressor	[87]
circ_KCNQ5 hsa_circ_0004136	KCNQ5	↑ in AML	miR-142 miR-622/RAB10	Increased cell proliferation and apoptosis inhibition	[88,89]
hsa_circ_0004520	VAV2	↑ in AML	PLXNB2, VEGFA	Angiogenesis Prognostic biomarker for EMI	[90]
hsa_circ_0000370	FLI-1	↑ in AML FLT3-ITD+	miR-1299/S100A7A	Prognostic biomarker	[91]
circ_0040823	BANP	↓ in AML	miR-516b/PTEN	Inhibited cell proliferation and increased apoptosis	[92]
circPLXNB2	PLXNB2	↑ in AML	PLXNB2	Increased cell proliferation and migration, apoptosis inhibition Prognostic biomarker for EMI	[93]
circ_0002232	PTEN	↓ in AML	miR-92a-3p/PTEN	Diagnostic and prognostic biomarker	[94]
circ_0012152	RNF220	↑ in AML	miR-625-5p/SOX12	Increased cell proliferation and apoptosis inhibition	[95]
circ_SFMBT2 hsa_circ_0017639	SFMBT2	↑ in AML	miR-582-3p/ZBTB20	Increased cell proliferation, migration and invasion	[96]
circ-PVT1	PVT1	↑ in AML	c-Myc and BCL-2?	Prognostic biomarker	[97]
hsa_circ_0001947	AFF2	↓ in AML	miR-329-5p/CREBRF	Inhibited cell proliferation Prognostic biomarker	[98]
hsa_circ_0075451	GMDS	↑ in AML	miR-330-5p/PRDM16 miR-326/PRDM16	Prognostic biomarker	[99]

AML is characterized by both chromosomal rearrangements and gene mutations [100]. Translocations are very often present, leading to the production of mutant fusion proteins. An intriguing work by Guarnerio et al. showed that the well-established translocations forming the oncoproteins PML-RARA and MLL-AF9 also produce fusion circRNAs (f-circRNAs), named f-circPR and f-circM9, respectively [59]. These f-circRNAs, derived from the aberrant conjunction of exons from different genes, contribute to HSCs transformation, leukemia progression and therapy resistance. Regarding genetic mutations,

the most common genetic lesion in AML affects the gene encoding for nucleophosmin-1 (NPM1). Mutated NPM1 protein loses its nucleolar localization, and it is delocalized in the cytoplasm, where it is responsible for the block of myeloid cell differentiation [101]. There is a circRNA encoded by the NPM1 gene, circNPM1, that is highly expressed by AML patients and cell lines and is associated with lower expression of members of the Toll-Like Receptors family, which are involved in normal hematopoietic differentiation. The effects of circNPM1 on TLR pathway genes could be mediated through miR181 [61]. Moreover, another study showed that circNPM1 silencing counteracted AML chemoresistance to Adriamycin. circNPM1 is a ceRNA for miR-345-5p, leading to increased expression of the miR-345-5p target gene FZD. Notably, FZD5 is an oncogene in various cancers. Since circNPM1 serum levels are high in AML patients, it might be a potential biomarker for drug resistance in AML [62]. Another protein frequently mutated in AML is the tyrosine kinase receptor FLTInternal Tandem Duplications of its juxtamembrane domain generate the oncoprotein FLT3-ITD, which is associated with very poor prognosis [102]. It was demonstrated that circMYBL2—derived from cell cycle checkpoint gene MYBL2—is upregulated in AML patients carrying FLT3-ITD mutation. This circRNA is crucial in promoting FLT3 mRNA translation by recruiting the RNA binding protein PTBP. Moreover, circMYBL2 silencing impairs leukemic cells proliferation in vivo and overcomes acquired resistance to quizartinib. Therefore, circMYBL2 may be a potential therapeutic target for FLT3-ITD AML patients [72]. Another interesting example is circPAN3, deriving from the gene encoding for PAN3 exonuclease. This circRNA is highly expressed in both AML cell lines and primary blasts resistant to doxorubicin. It promotes autophagy through the AMPK/mTOR pathway, thus conferring drug resistance [67]. Autophagy was found to be a mechanism for resistance in a range of solid tumors. Indeed, circPAN3 silencing can reduce autophagy and restore drug sensitivity. Moreover, an additional study showed that circPAN3 action might depend on miR-153-5p/miR-183-5p-XIAP axis [68]. XIAP is an anti-apoptotic protein that binds caspases 3, 7 and 9, leading them to proteasome-mediated degradation. However, circPAN3 downregulation decreases XIAP levels, and this could be due to sponging activity on miR-153-5p and miR-183-5p. These findings confirm that circPAN3 could be a predictor for treatment efficacy and also a therapeutic target in chemoresistance. Another circRNA related to AML originates from the gene RNF220, encoding for a RING domain E3 ubiquitin ligase that mediates ubiquitination of multiple targets. circRNF220 is especially expressed in pediatric AML, and it is a predictor of relapse. It acts as a sponge for miR-30a, thus increasing levels of its target mRNAs, including MYSM1 and IER. Interestingly, MYSM1 is a key transcription factor in hematopoiesis, while IER2 is upregulated in many tumors and is associated with cancer progression and metastasis. Therefore, circRNF220 could be useful as a prognostic marker, particularly in terms of relapse prediction [77] (Figure 1C).

6. circRNA in Leukemic Bone Marrow Niche

Although some circRNAs identified so far definitely lack a full description of the molecular mechanisms that render them pro-oncogenic or tumor-suppressive molecules, their association with AML development, progression and relapse has been proven (see Table 1). It is currently not clear whether these circRNAs are directly involved in the interplay between AML and the bone marrow microenvironment, but since leukemic blasts deeply change the composition of the niche to their advantage, the idea that circRNAs may be a part of this cell–cell communication is definitely intriguing. In support of this, enrichment of circRNAs in exosomes was reported in various diseases, including cancer [103]. In different types of solid tumors, it was demonstrated that circRNAs have a prominent role in regulating cancer cell metabolism, in particular, glycolysis, fatty acid oxidation, oxidative respiration and glutamine production [104]. Moreover, it is known that some circRNAs strongly interact with the tumor microenvironment in order to promote different steps of metastasis, including cancer cell migration, invasion, intravasation and neo-angiogenesis [105]. There is a reason to think that circRNAs involved in these processes may be packaged in exosomes. In regard to AML, the key role of extracellular vesicles

in implementing bone marrow niche remodeling is well-established. It was shown that levels of plasma-derived exosomes were higher in newly diagnosed AML patients and that they contained a different cargo compared to normal cell-derived exosomes. Notably, the exosome amount was decreased during remission [106]. It is noteworthy that hsa_circ_0009910 was found upregulated in AML cells and especially in AML-derived exosomes. This circRNA exerts an oncogenic role by acting as a miRNA sponge in the miR-5195-3p-GRB10 axis, in which GRB10 is an adapter protein that is involved in aberrant proliferation in FLT3-ITD positive AML. The authors proposed that hsa_circ_0009910 may be shuttled via exosomes to surrounding AML cells in order to promote their malignant properties [73]. In view of the evidence concerning AML resistance mechanisms uncovered so far, the role of circRNAs in intercellular communication within the leukemic bone marrow niche needs to be further investigated. These studies could provide insights into new therapeutic opportunities aimed at avoiding relapse of the disease. Moreover, in addition to the accumulation within the bone marrow, in some cases, leukemic cells can infiltrate other organs. This phenomenon is called Extramedullary Infiltration (EMI), and it is quite common, with myeloid sarcoma and leukemia cutis appearing in 1.4–9% and 15% of AML patients, respectively [107]. It is known that EMI is often associated with poor prognosis and relapse/refractory AML. An interesting paper shows that EMI and non-EMI AML samples have a different circRNA/miRNA/gene regulatory network, with circRNAs in EMI involved in cell adhesion, migration, signal transduction and cell–cell communication [90]. In particular, hsa_circ_0004520, which increases PLXNB and VEGFA levels, could promote angiogenesis.

7. Conclusions

The list of circRNAs involved in AML has been increasing fast in the very last few years. This is promising in the view of new therapeutic approaches, but it is essential to deal with some critical limitations. A complete and updated database and a unified nomenclature for circRNAs should be generated to avoid confusion in their classification. CircBase is a useful tool in which merged data sets of circRNAs are freely accessible [108]. Other strong bioinformatics tools are circIMPACT and CRAFT, which are able to identify regulatory networks governed by circRNAs and produce functional predictions [109,110]. From present studies, several circRNAs could be potential diagnostic or prognostic biomarkers for AML. Their particular structure renders them much more stable than their linear cognate RNA. Their presence both in the bone marrow and in body fluids, such as blood and urine, is also a remarkable point, supporting their use as biomarkers. Regarding circRNAs mechanisms of action, the vast majority of them have been shown to act as miRNA sponges. However, the assessment of copy number and of the number of miRNA binding sites is essential for carrying out these functional studies. Indeed, a rare circRNA with many miRNA binding sites could be equally effective as miRNA sponges as an abundant one with few miRNA binding sites. In order to measure the copy number of circRNAs with high accuracy, quantitative PCR or digital PCR should be used. Moreover, the assessment of diagnostic potential through specific tests is another important point [111]. Regarding upregulated circRNAs in AML and their application as therapeutic targets, the use of antisense oligomers (ASOs) is a valuable option. However, ASOs need to target the junction sequence, specific for circRNA; otherwise, parental linear RNA silencing occurs as a side effect. An interesting alternative to avoid off-target effects could be the use of circular siRNAs [112]. For downregulated circRNAs, overexpression could be obtained through exogenous delivery methods such as nanocarriers and nanoparticles. A novel method based on ferritin nanoparticles, which deliver nucleic acids specifically into AML cells, was recently developed [113].

The study of pro-survival strategies implemented by AML during both progression and development of therapy resistance is tangled. Scientists have only recently approached the investigation of altered AML niche as a mine of information about leukemia necessities and vulnerabilities. Crosstalk between AML and the bone marrow microenvironment

occurs through a complex mutual exchange of molecular signals, hence establishing a symbiotic relationship that allows disease progression and chemoresistance. circRNAs are likely to be involved in this process. Deep knowledge of circRNAs transport within and outside the cell is necessary in order to clarify their involvement in cell–cell communication. Future evaluation of circRNAs biological mechanisms will shed light on new promising strategies for AML treatment.

References

1. Shallis, R.M.; Wang, R.; Davidoff, A.; Ma, X.; Zeidan, A.M. Epidemiology of Acute Myeloid Leukemia: Recent Progress and Enduring Challenges. *Blood Rev.* **2019**, *36*, 70–87. [CrossRef] [PubMed]
2. Löwenberg, B.; Ossenkoppele, G.J.; van Putten, W.; Schouten, H.C.; Graux, C.; Ferrant, A.; Sonneveld, P.; Maertens, J.; Jongen-Lavrencic, M.; von Lilienfeld-Toal, M.; et al. High-Dose Daunorubicin in Older Patients with Acute Myeloid Leukemia. *N. Engl. J. Med.* **2009**, *361*, 1235–1248. [CrossRef] [PubMed]
3. Cornelissen, J.J.; Blaise, D. Hematopoietic Stem Cell Transplantation for Patients with AML in First Complete Remission. *Blood* **2016**, *127*, 62–70. [CrossRef] [PubMed]
4. Döhner, H.; Weisdorf, D.J.; Bloomfield, C.D. Acute Myeloid Leukemia1. *N. Engl. J. Med.* **2015**, *373*, 1136–1152. [CrossRef] [PubMed]
5. Stanchina, M.; Soong, D.; Zheng-Lin, B.; Watts, J.M.; Taylor, J. Advances in Acute Myeloid Leukemia: Recently Approved Therapies and Drugs in Development. *Cancers* **2020**, *12*, 3225. [CrossRef]
6. Masciarelli, S.; Capuano, E.; Ottone, T.; Divona, M.; de Panfilis, S.; Banella, C.; Noguera, N.I.; Picardi, A.; Fontemaggi, G.; Blandino, G.; et al. Retinoic Acid and Arsenic Trioxide Sensitize Acute Promyelocytic Leukemia Cells to ER Stress. *Leukemia* **2018**, *32*, 285–294. [CrossRef]
7. Masciarelli, S.; Capuano, E.; Ottone, T.; Divona, M.; Lavorgna, S.; Liccardo, F.; Śniegocka, M.; Travaglini, S.; Noguera, N.I.; Picardi, A.; et al. Retinoic Acid Synergizes with the Unfolded Protein Response and Oxidative Stress to Induce Cell Death in FLT3-ITD1 AML. *Blood Adv.* **2019**, *3*, 4155–4160. [CrossRef]
8. Short, N.J.; Konopleva, M.; Kadia, T.M.; Borthakur, G.; Ravandi, F.; DiNardo, C.D.; Daver, N. Advances in the Treatment of Acute Myeloid Leukemia: New Drugs and New Challenges. *Cancer Discov.* **2020**, *10*, 506–525. [CrossRef]
9. Pagano, F.; de Marinis, E.; Grignani, F.; Nervi, C. Epigenetic Role of MiRNAs in Normal and Leukemic Hematopoiesis. *Epigenomics* **2013**, *5*, 539–552. [CrossRef]
10. Fatica, A.; Fazi, F. MicroRNA-Regulated Pathways in Hematological Malignancies: How to Avoid Cells Playing out of Tune. *Int. J. Mol. Sci.* **2013**, *14*, 20930–20953. [CrossRef]
11. Mangiavacchi, A.; Sorci, M.; Masciarelli, S.; Larivera, S.; Legnini, I.; Iosue, I.; Bozzoni, I.; Fazi, F.; Fatica, A. The MiR-223 Host Non-Coding Transcript Linc-223 Induces IRF4 Expression in Acute Myeloid Leukemia by Acting as a Competing Endogenous RNA. *Oncotarget* **2016**, *7*, 60155–60168. [CrossRef] [PubMed]
12. Di Agostino, S.; Riccioli, A.; de Cesaris, P.; Fontemaggi, G.; Blandino, G.; Filippini, A.; Fazi, F. Circular RNAs in Embryogenesis and Cell Differentiation With a Focus on Cancer Development. *Front. Cell Dev. Biol.* **2020**, *8*, 389. [CrossRef] [PubMed]
13. Till, J.E.; McCulloch, E.A. A Direct Measurement of the Radiation Sensitivity of Normal Mouse Bone Marrow Cells. *Radiat. Res.* **1961**, *14*, 213. [CrossRef] [PubMed]
14. Morrison, S.J.; Uchida, N.; Weissman, I.L. The Biology of Hematopoietic Stem Cells. *Annu. Rev. Cell Dev. Biol.* **1995**, *11*, 35–71. [CrossRef]
15. Acar, M.; Kocherlakota, K.S.; Murphy, M.M.; Peyer, J.G.; Oguro, H.; Inra, C.N.; Christabel, J.; Zhao, Z.; Luby-Phelps, K.; Morrison, S.J. Deep imaging of bone marrow shows non-dividing stem cells are mainly perisinusoidal. *Nature* **2015**, *526*, 126–130. [CrossRef]

16. Kokkaliaris, K.D.; Kunz, L.; Cabezas-Wallscheid, N.; Christodoulou, C.; Renders, S.; Camargo, F.; Trumpp, A.; Scadden, D.T.; Schroeder, T. Adult blood stem cell localization reflects the abundance of reported bone marrow niche cell types and their combinations. *Blood* **2020**, *136*, 2296–2307. [CrossRef]

17. Anthony, B.A.; Link, D.C. Regulation of hematopoietic stem cells by bone marrow stromal cells. *Trends Immunol.* **2013**, *35*, 32–37. [CrossRef]

18. Baccin, C.; Al-Sabah, J.; Velten, L.; Helbling, P.M.; Grünschläger, F.; Hernández-Malmierca, P.; Nombela-Arrieta, C.; Steinmetz, L.M.; Trumpp, A.; Haas, S. Combined single-cell and spatial transcriptomics reveal the molecular, cellular and spatial bone marrow niche organization. *Nat. Cell Biol.* **2020**, *22*, 38–48. [CrossRef]

19. Méndez-Ferrer, S.; Bonnet, D.; Steensma, D.P.; Hasserjian, R.P.; Ghobrial, I.M.; Gribben, J.G.; Andreeff, M.; Krause, D.S. Bone marrow niches in haematological malignancies. *Nat. Rev. Cancer* **2020**, *20*, 285–298. [CrossRef]

20. Galán-Díez, M.; Kousteni, S. The Osteoblastic Niche in Hematopoiesis and Hematological Myeloid Malignancies. *Curr. Mol. Biol. Rep.* **2017**, *3*, 53–62. [CrossRef]

21. Zhou, B.O.; Yu, H.; Yue, R.; Zhao, Z.; Rios, J.J.; Naveiras, O.; Morrison, S.J. Bone marrow adipocytes promote the regeneration of stem cells and haematopoiesis by secreting SCF. *Nat. Cell Biol.* **2017**, *19*, 891–903. [CrossRef] [PubMed]

22. Winkler, I.G.; Sims, N.A.; Pettit, A.R.; Barbier, V.; Nowlan, B.; Helwani, F.; Poulton, I.J.; Van Rooijen, N.; Alexander, K.; Raggatt, L.J.; et al. Bone marrow macrophages maintain hematopoietic stem cell (HSC) niches and their depletion mobilizes HSCs. *Blood* **2010**, *116*, 4815–4828. [CrossRef] [PubMed]

23. Comazzetto, S.; Shen, B.; Morrison, S.J. Niches that regulate stem cells and hematopoiesis in adult bone marrow. *Dev. Cell* **2021**, *56*, 1848–1860. [CrossRef] [PubMed]

24. Wei, Q.; Frenette, P.S. Niches for Hematopoietic Stem Cells and Their Progeny. *Immunity* **2018**, *48*, 632–648. [CrossRef] [PubMed]

25. Goode, D.K.; Obier, N.; Vijayabaskar, M.S.; Lie, M.; Lilly, A.J.; Hannah, R.; Lichtinger, M.; Batta, K.; Florkowska, M.; Patel, R.; et al. Dynamic Gene Regulatory Networks Drive Hematopoietic Specification and Differentiation. *Dev. Cell* **2016**, *36*, 572–587. [CrossRef] [PubMed]

26. Luo, M.; Jeong, M.; Sun, D.; Park, H.J.; Rodriguez, B.A.T.; Xia, Z.; Yang, L.; Zhang, X.; Sheng, K.; Darlington, G.J.; et al. Long Non-Coding RNAs Control Hematopoietic Stem Cell Function. *Cell Stem Cell* **2015**, *16*, 426–438. [CrossRef]

27. Bissels, U.; Bosio, A.; Wagner, W. MicroRNAs are shaping the hematopoietic landscape. *Haematologica* **2011**, *97*, 160–167. [CrossRef]

28. Zhang, X.O.; Wang, H.B.; Zhang, Y.; Lu, X.; Chen, L.L.; Yang, L. Complementary Sequence-Mediated Exon Circularization. *Cell* **2014**, *159*, 134–147. [CrossRef]

29. Conn, S.J.; Pillman, K.A.; Toubia, J.; Conn, V.M.; Salmanidis, M.; Phillips, C.A.; Roslan, S.; Schreiber, A.W.; Gregory, P.A.; Goodall, G.J. The RNA Binding Protein Quaking Regulates Formation of circRNAs. *Cell* **2015**, *160*, 1125–1134. [CrossRef]

30. Ashwal-Fluss, R.; Meyer, M.; Pamudurti, N.R.; Ivanov, A.; Bartok, O.; Hanan, M.; Evantal, N.; Memczak, S.; Rajewsky, N.; Kadener, S. circRNA Biogenesis Competes with Pre-mRNA Splicing. *Mol. Cell* **2014**, *56*, 626–639. [CrossRef]

31. Chen, L.-L. The expanding regulatory mechanisms and cellular functions of circular RNAs. *Nat. Rev. Mol. Cell Biol.* **2020**, *21*, 475–490. [CrossRef]

32. Xia, P.; Wang, S.; Ye, B.; Du, Y.; Li, C.; Xiong, Z.; Qu, Y.; Fan, Z. A Circular RNA Protects Dormant Hematopoietic Stem Cells from DNA Sensor cGAS-Mediated Exhaustion. *Immunity* **2018**, *48*, 688–701.e7. [CrossRef] [PubMed]

33. Nicolet, B.P.; Engels, S.; Aglialoro, F.; Van Den Akker, E.; Von Lindern, M.; Wolkers, M.C. Circular RNA expression in human hematopoietic cells is widespread and cell-type specific. *Nucleic Acids Res.* **2018**, *46*, 8168–8180. [CrossRef] [PubMed]

34. Nicolet, B.P.; Jansen, S.B.G.; Heideveld, E.; Ouwehand, W.H.; Akker, E.; von Linderen, M.; Wolkers, M.C. Circular RNAs exhibit limited evidence for translation, or translation regulation of the mRNA counterpart in terminal hematopoiesis. *RNA* **2022**, *28*, 194–209. [CrossRef] [PubMed]

35. Corces-Zimmerman, M.R.; Majeti, R. Pre-leukemic evolution of hematopoietic stem cells: The importance of early mutations in leukemogenesis. *Leukemia* **2014**, *28*, 2276–2282. [CrossRef] [PubMed]

36. Pandolfi, A.; Barreyro, L.; Steidl, U. Concise Review: Preleukemic Stem Cells: Molecular Biology and Clinical Implications of the Precursors to Leukemia Stem Cells. *Stem Cells Transl. Med.* **2013**, *2*, 143–150. [CrossRef]

37. Velten, L.; Story, B.A.; Hernández-Malmierca, P.; Raffel, S.; Leonce, D.R.; Milbank, J.; Paulsen, M.; Demir, A.; Szu-Tu, C.; Frömel, R.; et al. Identification of leukemic and pre-leukemic stem cells by clonal tracking from single-cell transcriptomics. *Nat. Commun.* **2021**, *12*, 1366. [CrossRef]

38. Miller, L.H.; Qu, C.K.; Pauly, M. Germline mutations in the bone marrow microenvironment and dysregulated hematopoiesis. *Exp. Hematol.* **2018**, *66*, 17–26. [CrossRef]

39. Dias, S.; Shmelkov, S.V.; Lam, G.; Rafii, S. VEGF165 promotes survival of leukemic cells by Hsp90-mediated induction of Bcl-2 expression and apoptosis inhibition. *Blood* **2002**, *99*, 2532–2540. [CrossRef]

40. Hatfield, K.; Ryningen, A.; Corbascio, M.; Bruserud, Ø. Microvascular endothelial cells increase proliferation and inhibit apoptosis of native human acute myelogenous leukemia blasts. *Int. J. Cancer* **2006**, *119*, 2313–2321. [CrossRef]

41. Passaro, D.; di Tullio, A.; Abarrategi, A.; Rouault-Pierre, K.; Foster, K.; Ariza-McNaughton, L.; Montaner, B.; Chakravarty, P.; Bhaw, L.; Diana, G.; et al. Increased Vascular Permeability in the Bone Marrow Microenvironment Contributes to Disease Progression and Drug Response in Acute Myeloid Leukemia. *Cancer Cell* **2017**, *32*, 324–341.e6. [CrossRef] [PubMed]

42. Hanoun, M.; Zhang, D.; Toshihide, M.; Piho, S.; Pierce, H.; Kunisaki, Y.; Lacombe, J.; Armstrong, S.A.; Dührsen, U.; Frenette, P.S. Acute Myelogenous Leukemia-Induced Sympathetic Neuropathy Promotes Malignancy in an Altered Hematopoietic Stem Cell Niche. *Cell Stem Cell* **2014**, *15*, 365–375. [CrossRef] [PubMed]

43. Tettamanti, S.; Pievani, A.; Biondi, A.; Dotti, G.; Serafini, M. Catch me if you can: How AML and its niche escape immunotherapy. *Leukemia* **2022**, *36*, 13–22. [CrossRef] [PubMed]

44. Al-Matary, Y.S.; Botezatu, L.; Opalka, B.; Hönes, J.M.; Lams, R.F.; Thivakaran, A.; Schütte, J.; Köster, R.; Lennartz, K.; Schroeder, T.; et al. Acute myeloid leukemia cells polarize macrophages towards a leukemia supporting state in a Growth factor independence 1 dependent manner. *Haematologica* **2016**, *101*, 1216–1227. [CrossRef]

45. Sriskanthadevan, S.; Jeyaraju, D.V.; Chung, T.E.; Prabha, S.; Xu, W.; Skrtic, M.; Jhas, B.; Hurren, R.; Gronda, M.; Wang, X.; et al. AML cells have low spare reserve capacity in their respiratory chain that renders them susceptible to oxidative metabolic stress. *Blood* **2015**, *125*, 2120–2130. [CrossRef]

46. Tabe, Y.; Konopleva, M. Break the lifeline of AML cells. *Blood* **2021**, *137*, 3465–3467. [CrossRef]

47. Shafat, M.S.; Oellerich, T.; Mohr, S.; Robinson, S.D.; Edwards, D.R.; Marlein, C.R.; Piddock, R.E.; Fenech, M.; Zaitseva, L.; Abdul-Aziz, A.; et al. Leukemic blasts program bone marrow adipocytes to generate a protumoral microenvironment. *Blood* **2017**, *129*, 1320–1332. [CrossRef]

48. Moschoi, R.; Imbert, V.; Nebout, M.; Chiche, J.; Mary, D.; Prebet, T.; Saland, E.; Castellano, R.; Pouyet, L.; Collette, Y.; et al. Protective mitochondrial transfer from bone marrow stromal cells to acute myeloid leukemic cells during chemotherapy. *Blood* **2016**, *128*, 253–264. [CrossRef]

49. Marlein, C.R.; Zaitseva, L.; Piddock, R.E.; Robinson, S.D.; Edwards, D.R.; Shafat, M.S.; Zhou, Z.; Lawes, M.; Bowles, K.M.; Rushworth, S. NADPH oxidase-2 derived superoxide drives mitochondrial transfer from bone marrow stromal cells to leukemic blasts. *Blood* **2017**, *130*, 1649–1660. [CrossRef]

50. Forte, D.; García-Fernández, M.; Sánchez-Aguilera, A.; Stavropoulou, V.; Fielding, C.; Martín-Pérez, D.; López, J.A.; Costa, A.S.; Tronci, L.; Nikitopoulou, E.; et al. Bone Marrow Mesenchymal Stem Cells Support Acute Myeloid Leukemia Bioenergetics and Enhance Antioxidant Defense and Escape from Chemotherapy. *Cell Metab.* **2020**, *32*, 829–843.e9. [CrossRef]

51. Kumar, B.; Garcia, M.; Weng, L.; Jung, X.; Murakami, J.L.; Hu, X.; McDonald, T.; Lin, A.; Kumar, A.R.; DiGiusto, D.L.; et al. Acute myeloid leukemia transforms the bone marrow niche into a leukemia-permissive microenvironment through exosome secretion. *Leukemia* **2018**, *32*, 575–587. [CrossRef] [PubMed]

52. Kumar, B.; Garcia, M.; Murakami, J.L.; Chen, C.C. Exosome-mediated microenvironment dysregulation in leukemia. *Biochim. Biophys. Acta* **2016**, *1863*, 464–470. [CrossRef] [PubMed]

53. Féral, K.; Jaud, M.; Philippe, C.; Bella, D.B.; Pyronnet, S.; Rouault-Pierre, K.; Mazzolini, L.; Touriol, C. ER Stress and Unfolded Protein Response in Leukemia: Friend, Foe, or Both? *Biomolecules* **2021**, *11*, 199. [CrossRef] [PubMed]

54. Doron, B.; Abdelhamed, S.; Butler, J.T.; Hashmi, S.K.; Horton, T.M.; Kurre, P. Transmissible ER stress reconfigures the AML bone marrow compartment. *Leukemia* **2019**, *33*, 918–930. [CrossRef]

55. Zeng, Z.; Shi, Y.X.; Samudio, I.J.; Wang, R.Y.; Ling, X.; Frolova, O.; Levis, M.; Rubin, J.B.; Negrin, R.R.; Estey, E.H.; et al. Targeting the leukemia microenvironment by CXCR4 inhibition overcomes resistance to kinase inhibitors and chemotherapy in AML. *Blood* **2009**, *113*, 6215–6224. [CrossRef]

56. Jacamo, R.; Chen, Y.; Wang, Z.; Ma, W.; Zhang, M.; Spaeth, E.L.; Wang, Y.; Battula, V.L.; Mak, P.Y.; Schallmoser, K.; et al. Reciprocal leukemia-stroma VCAM-1/VLA-4-dependent activation of NF-κB mediates chemoresistance. *Blood* **2014**, *123*, 2691–2702. [CrossRef]

57. Chen, L.; Shan, G. CircRNA in cancer: Fundamental mechanism and clinical potential. *Cancer Lett.* **2021**, *505*, 49–57. [CrossRef]

58. Issah, M.A.; Wu, D.; Zhang, F.; Zheng, W.; Liu, Y.; Chen, R.; Lai, G.; Shen, J. Expression profiling of N6-methyladenosine modified circRNAs in acute myeloid leukemia. *Biochem. Biophys. Res. Commun.* **2022**, *601*, 137–145. [CrossRef]

59. Guarnerio, J.; Bezzi, M.; Jeong, J.C.; Paffenholz, S.V.; Berry, K.; Naldini, M.M.; Lo-Coco, F.; Tay, Y.; Beck, A.H.; Pandolfi, P.P. Oncogenic Role of Fusion-circRNAs Derived from Cancer-Associated Chromosomal Translocations. *Cell* **2016**, *165*, 289–302. [CrossRef]

60. Chen, H.; Liu, T.; Liu, J.; Feng, Y.; Wang, B.; Wang, J.; Bai, J.; Zhao, W.; Shen, Y.; Wang, X.; et al. Circ-ANAPC7 is Upregulated in Acute Myeloid Leukemia and Appears to Target the MiR-181 Family. *Cell. Physiol. Biochem.* **2018**, *47*, 1998–2007. [CrossRef]

61. Hirsch, S.; Blätte, T.J.; Grasedieck, S.; Cocciardi, S.; Rouhi, A.; Jongen-Lavrencic, M.; Paschka, P.; Krönke, J.; Gaidzik, V.I.; Döhner, H.; et al. Circular RNAs of the nucleophosmin (NPM1) gene in acute myeloid leukemia. *Haematologica* **2017**, *102*, 2039–2047. [CrossRef] [PubMed]

62. Ding, J.; Zhang, X.; Xue, J.; Fang, L.; Ban, C.; Song, B.; Wu, L. CircNPM1 strengthens Adriamycin resistance in acute myeloid leukemia by mediating the miR-345-5p/FZD5 pathway. *Central Eur. J. Immunol.* **2021**, *46*, 162–182. [CrossRef] [PubMed]

63. Wu, D.-M.; Wen, X.; Han, X.-R.; Wang, S.; Wang, Y.-J.; Shen, M.; Fan, S.-H.; Zhang, Z.-F.; Shan, Q.; Li, M.-Q.; et al. Role of Circular RNA DLEU2 in Human Acute Myeloid Leukemia. *Mol. Cell. Biol.* **2018**, *38*, e00259-18. [CrossRef] [PubMed]

64. Ding, Y.; Dong, Y.; Lu, H.; Luo, X.; Fu, J.; Xiu, B.; Liang, A.; Zhang, W. Circular RNA profile of acute myeloid leukaemia indicates circular RNA annexin A2 as a potential biomarker and therapeutic target for acute myeloid leukaemia. *Am. J. Transl. Res.* **2020**, *12*, 1683–1699. [PubMed]

65. Yi, Y.Y.; Yi, J.; Zhu, X.; Zhang, J.; Zhou, J.; Tang, X.; Lin, J.; Wang, P.; Deng, Z.Q. Circular RNA of *vimentin* expression as a valuable predictor for acute myeloid leukemia development and prognosis. *J. Cell. Physiol.* **2019**, *234*, 3711–3719. [CrossRef] [PubMed]

66. Li, S.; Ma, Y.; Tan, Y.; Ma, X.; Zhao, M.; Chen, B.; Zhang, R.; Chen, Z.; Wang, K. Profiling and functional analysis of circular RNAs in acute promyelocytic leukemia and their dynamic regulation during all-trans retinoic acid treatment. *Cell Death Dis.* **2018**, *9*, 651. [CrossRef]

67. Shang, J.; Chen, W.M.; Liu, S.; Wang, Z.H.; Wei, T.N.; Chen, Z.Z.; Wu, W.B. CircPAN3 contributes to drug resistance in acute myeloid leukemia through regulation of autophagy. *Leuk. Res.* **2019**, *85*, 106198. [CrossRef]

68. Shang, J.; Chen, W.M.; Wang, Z.H.; Wei, T.N.; Chen, Z.Z.; Wu, W.B. CircPAN3 mediates drug resistance in acute myeloid leukemia through the miR-153-5p/miR-183-5p–XIAP axis. *Exp. Hematol.* **2019**, *70*, 42–54.e3. [CrossRef]

69. Li, W.; Zhong, C.; Jiao, J.; Li, P.; Cui, B.; Ji, C.; Ma, D. Characterization of hsa_circ_0004277 as a New Biomarker for Acute Myeloid Leukemia via Circular RNA Profile and Bioinformatics Analysis. *Int. J. Mol. Sci.* **2017**, *18*, 597. [CrossRef]

70. Liu, Y.; Chen, X.; Liu, J.; Jin, Y.; Wang, W. Circular RNA circ_0004277 Inhibits Acute Myeloid Leukemia Progression Through MicroRNA-134-5p/Single stranded DNA binding protein 2. *Bioengineered* **2022**, *13*, 9662–9673. [CrossRef]

71. Ye, Q.; Li, N.; Zhou, K.; Liao, C. Homo sapiens circular RNA 0003602 (Hsa_circ_0003602) accelerates the tumorigenicity of acute myeloid leukemia by modulating miR-502-5p/IGF1R axis. *Mol. Cell. Biochem.* **2022**, *477*, 635–644. [CrossRef] [PubMed]

72. Sun, Y.M.; Wang, W.T.; Zeng, Z.C.; Chen, T.Q.; Han, C.; Pan, Q.; Huang, W.; Fang, K.; Sun, L.Y.; Zhou, Y.F.; et al. circMYBL2, a circRNA from MYBL2, regulates FLT3 translation by recruiting PTBP1 to promote FLT3-ITD AML progression. *Blood* **2019**, *134*, 1533–1546. [CrossRef] [PubMed]

73. Wang, D.; Ming, X.; Xu, J.; Xiao, Y. Circ_0009910 shuttled by exosomes regulates proliferation, cell cycle and apoptosis of acute myeloid leukemia cells by regulating miR-5195-3p/GRB10 axis. *Hematol. Oncol.* **2021**, *39*, 390–400. [CrossRef] [PubMed]

74. Ping, L.; Jian-Jun, C.; Chu-Shu, L.; Guang-Hua, L.; Ming, Z. Silencing of circ_0009910 inhibits acute myeloid leukemia cell growth through increasing miR-20a-5p. *Blood Cells Mol. Dis.* **2018**, *75*, 41–47. [CrossRef]

75. Chen, J.J.; Lei, P.; Zhou, M. hsa_circ_0121582 inhibits leukemia growth by dampening Wnt/β-catenin signaling. *Clin. Transl. Oncol.* **2020**, *22*, 2293–2302. [CrossRef]

76. Zhou, J.; Zhou, L.Y.; Tang, X.; Zhang, J.; Zhai, L.-L.; Yi, Y.Y.; Yi, J.; Lin, J.; Qian, J.; Deng, Z.-Q. Circ-Foxo3 is positively associated with the Foxo3 gene and leads to better prognosis of acute myeloid leukemia patients. *BMC Cancer* **2019**, *19*, 930. [CrossRef]

77. Liu, X.; Liu, X.; Cai, M.; Luo, A.; He, Y.; Liu, S.; Zhang, X.; Yang, X.; Xu, L.; Jiang, H. CircRNF220, not its linear cognate gene RNF220, regulates cell growth and is associated with relapse in pediatric acute myeloid leukemia. *Mol. Cancer* **2021**, *20*, 139. [CrossRef]

78. Fan, H.; Li, Y.; Liu, C.; Liu, Y.; Bai, J.; Li, W. Circular RNA-100290 promotes cell proliferation and inhibits apoptosis in acute myeloid leukemia cells via sponging miR. *Biochem. Biophys. Res. Commun.* **2018**, *507*, 178–184. [CrossRef]

79. Zhang, R.; Li, Y.; Wang, H.; Zhu, K.; Zhang, G. The Regulation of circRNA RNF13/miRNA-1224-5p Axis Promotes the Malignant Evolution in Acute Myeloid Leukemia. *BioMed Res. Int.* **2020**, *2020*, 5654380. [CrossRef]

80. Yi, L.; Zhou, L.; Luo, J.; Yang, Q. Circ-PTK2 promotes the proliferation and suppressed the apoptosis of acute myeloid leukemia cells through targeting miR-330-5p/FOXM1 axis. *Blood Cells Mol. Dis.* **2021**, *86*, 102506. [CrossRef]

81. Xiao, Y.; Ming, X.; Wu, J. Hsa_circ_0002483 regulates miR-758-3p/MYC axis to promote acute myeloid leukemia progression. *Hematol. Oncol.* **2021**, *39*, 243–253. [CrossRef] [PubMed]

82. Li, Q.; Luan, Q.; Zhu, H.; Zhao, Y.; Ji, J.; Wu, F.; Yan, J. Circular RNA circ_0005774 contributes to proliferation and suppresses apoptosis of acute myeloid leukemia cells via circ_0005774/miR-192–5p/ULK1 ceRNA pathway. *Biochem. Biophys. Res. Commun.* **2021**, *551*, 78–85. [CrossRef] [PubMed]

83. Liu, W.; Cheng, F. Circular RNA circCRKL inhibits the proliferation of acute myeloid leukemia cells via the miR-196a-5p/miR-196b-5p/p27 axis. *Bioengineered* **2021**, *12*, 7704–7713. [CrossRef] [PubMed]

84. Li, H.; Bi, K.; Feng, S.; Wang, Y.; Zhu, C. CircRNA circ_POLA2 is Upregulated in Acute Myeloid Leukemia (AML) and Promotes Cell Proliferation by Suppressing the Production of Mature miR-34a. *Cancer Manag. Res.* **2021**, *13*, 3629–3637. [CrossRef] [PubMed]

85. Hu, Q.; Gu, Y.; Chen, S.; Tian, Y.; Yang, S. Hsa_circ_0079480 promotes tumor progression in acute myeloid leukemia via miR-654-3p/HDGF axis. *Aging* **2021**, *13*, 1120–1131. [CrossRef]

86. Guo, L.; Kou, R.; Song, Y.; Li, G.; Jia, X.; Li, Z.; Zhang, Y. Serum hsa_circ_0079480 is a novel prognostic marker for acute myeloid leukemia. *J. Clin. Lab. Anal.* **2022**, *36*, e24337. [CrossRef]

87. Papaioannou, D.; Volinia, S.; Nicolet, D.; Świerniak, M.; Petri, A.; Mrózek, K.; Bill, M.; Pepe, F.; Walker, C.J.; Walker, A.E.; et al. Clinical and functional significance of circular RNAs in cytogenetically normal AML. *Blood Adv.* **2020**, *4*, 239–251. [CrossRef]

88. Yuan, D.M.; Ma, J.; Fang, W.B. Identification of non-coding RNA regulatory networks in pediatric acute myeloid leukemia reveals circ-0004136 could promote cell proliferation by sponging miR-142. *Eur. Rev. Med. Pharmacol. Sci.* **2019**, *23*, 9251–9258.

89. Hu, X.; Yin, J.; He, R.; Chao, R.; Zhu, S. Circ_KCNQ5 participates in the progression of childhood acute myeloid leukemia by enhancing the expression of RAB10 via binding to miR. *Hematology* **2022**, *27*, 431–440. [CrossRef]

90. Lv, C.; Sun, L.; Guo, Z.; Li, H.; Kong, D.; Xu, B.; Lin, L.; Liu, T.; Guo, D.; Zhou, J.; et al. Circular RNA regulatory network reveals cell–cell crosstalk in acute myeloid leukemia extramedullary infiltration. *J. Transl. Med.* **2018**, *16*, 361. [CrossRef]

91. Zhang, L.; Bu, Z.; Shen, J.; Shang, L.; Chen, Y.; Wang, Y. A novel circular RNA (hsa_circ_0000370) increases cell viability and inhibits apoptosis of FLT3-ITD-positive acute myeloid leukemia cells by regulating miR-1299 and S100A7A. *Biomed. Pharmacother.* **2020**, *122*, 109619. [CrossRef]

92. Wang, N.; Yang, B.; Jin, J.; He, Y.; Wu, X.; Yang, Y.; Zhou, W.; He, Z. Circular RNA circ_0040823 inhibits the proliferation of acute myeloid leukemia cells and induces apoptosis by regulating miR-516b/PTEN. *J. Gene Med.* **2022**, *24*, e3404. [CrossRef]

93. Lin, L.; Wang, Y.; Bian, S.; Sun, L.; Guo, Z.; Kong, D.; Zhao, L.; Guo, D.; Li, Q.; Wu, M.; et al. A circular RNA derived from PLXNB2 as a valuable predictor of the prognosis of patients with acute myeloid leukaemia. *J. Transl. Med.* **2021**, *19*, 123. [CrossRef] [PubMed]

94. Su, X.Y.; Zhao, Q.; Ke, J.M.; Wu, D.H.; Zhu, X.; Lin, J.; Deng, Z.Q. Circ_0002232 Acts as a Potential Biomarker for AML and Reveals a Potential ceRNA Network of Circ_0002232/miR-92a-3p/PTEN. *Cancer Manag. Res.* **2020**, *12*, 11871–11881. [CrossRef] [PubMed]

95. Shang, Z.; Ming, X.; Wu, J.; Xiao, Y. Downregulation of circ_0012152 inhibits proliferation and induces apoptosis in acute myeloid leukemia cells through the miR-625-5p/SOX12 axis. *Hematol. Oncol.* **2021**, *39*, 539–548. [CrossRef] [PubMed]

96. Chang, W.; Shang, Z.; Ming, X.; Wu, J.; Xiao, Y. Circ-SFMBT2 Facilitates the Malignant Growth of Acute Myeloid Leukemia Cells by Modulating MiR-582-3p/ZBTB20 Pathway. *Histol. Histopathol.* **2022**, *37*, 137–149. [CrossRef]

97. Chen, T.; Chen, F. The role of circular RNA plasmacytoma variant translocation 1 as a biomarker for prognostication of acute myeloid leukemia. *Hematology* **2021**, *26*, 1018–1024. [CrossRef]

98. Han, F.; Zhong, C.; Li, W.; Wang, R.; Zhang, C.; Yang, X.; Ji, C.; Ma, D. *hsa_circ_0001947* suppresses acute myeloid leukemia progression via targeting *hsa-miR-329-5p/CREBRF* axis. *Epigenomics* **2020**, *12*, 935–953. [CrossRef]

99. Wang, J.; Pan, J.; Huang, S.; Li, F.; Huang, J.; Li, X.; Ling, Q.; Ye, W.; Wang, Y.; Yu, W.; et al. Development and validation of a novel circular RNA as an independent prognostic factor in acute myeloid leukemia. *BMC Med.* **2021**, *19*, 28. [CrossRef]

100. Bochtler, T.; Fröhling, S.; Krämer, A. Role of chromosomal aberrations in clonal diversity and progression of acute myeloid leukemia. *Leukemia* **2015**, *29*, 1243–1252. [CrossRef]

101. Kunchala, P.; Kuravi, S.; Jensen, R.; McGuirk, J.; Balusu, R. When the good go bad: Mutant NPM1 in acute myeloid leukemia. *Blood Rev.* **2018**, *32*, 167–183. [CrossRef] [PubMed]

102. Lagunas-Rangel, F.A.; Chávez-Valencia, V. FLT3–ITD and its current role in acute myeloid leukaemia. *Med. Oncol.* **2017**, *34*, 114. [CrossRef] [PubMed]

103. Wang, Y.; Liu, J.; Ma, J.; Sun, T.; Zhou, Q.; Wang, W.; Wang, G.; Wu, P.; Wang, H.; Jiang, L.; et al. Exosomal circRNAs: Biogenesis, effect and application in human diseases. *Mol. Cancer* **2019**, *18*, 116. [CrossRef] [PubMed]

104. Yu, T.; Wang, Y.; Fan, Y.; Fang, N.; Wang, T.; Xu, T.; Shu, Y. CircRNAs in cancer metabolism: A review. *J. Hematol. Oncol.* **2019**, *12*, 90. [CrossRef]

105. Shao, Y.; Lu, B. The crosstalk between circular RNAs and the tumor microenvironment in cancer metastasis. *Cancer Cell Int.* **2020**, *20*, 448. [CrossRef]

106. Boyiadzis, M.; Whiteside, T.L. Plasma-derived exosomes in acute myeloid leukemia for detection of minimal residual disease: Are we ready? *Expert Rev. Mol. Diagn.* **2016**, *16*, 623–629. [CrossRef]

107. Bakst, R.L.; Tallman, M.S.; Douer, D.; Yahalom, J. How I treat extramedullary acute myeloid leukemia. *Blood* **2011**, *118*, 3785–3793. [CrossRef]

108. Glažar, P.; Papavasileiou, P.; Rajewsky, N. circBase: A database for circular RNAs. *RNA* **2014**, *20*, 1666–1670. [CrossRef]

109. Buratin, A.; Gaffo, E.; Molin, A.D.; Bortoluzzi, S. CircIMPACT: An R Package to Explore Circular RNA Impact on Gene Expression and Pathways. *Genes* **2021**, *12*, 1044. [CrossRef]

110. Dal Molin, A.; Gaffo, E.; Difilippo, V.; Buratin, A.; Tretti Parenzan, C.; Bresolin, S.; Bortoluzzi, S. Correction to: CRAFT a bioinformatics software for custom prediction of circular RNA functions. *Briefings Bioinform.* **2022**, *23*, bbab601. [CrossRef]

111. Li, F.; Yang, Q.; He, A.T.; Yang, B.B. Circular RNAs in cancer: Limitations in functional studies and diagnostic potential. *Semin. Cancer Biol.* **2021**, *75*, 49–61. [CrossRef] [PubMed]

112. Zhang, L.; Liang, D.; Chen, C.; Wang, Y.; Amu, G.; Yang, J.; Yu, L.; Dmochowski, I.J.; Tang, X. Circular siRNAs for Reducing Off-Target Effects and Enhancing Long-Term Gene Silencing in Cells and Mice. *Mol. Ther.-Nucleic Acids* **2018**, *10*, 237–244. [CrossRef] [PubMed]

113. Palombarini, F.; Masciarelli, S.; Incocciati, A.; Liccardo, F.; Di Fabio, E.; Iazzetti, A.; Fabrizi, G.; Fazi, F.; Macone, A.; Bonamore, A.; et al. Self-Assembling Ferritin-Dendrimer Nanoparticles for Targeted Delivery of Nucleic Acids to Myeloid Leukemia Cells. *J. Nanobiotechnol.* **2021**, *19*, 172. [CrossRef] [PubMed]