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Review Article

microRNAs in T-Cell Acute Lymphoblastic Leukemia: Roles and Contributions to Treatment Response

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T-cell acute lymphoblastic leukemia (T-ALL) is an aggressive hematologic malignancy induced by the proliferation of immature T-cell precursors. Even with the development of multi-agent chemotherapy, treatment failure, and relapse remain the most important challenges because of drug resistance. miRNAs are a class of small non-coding RNAs that modulate the expression of target mRNAs at the post-transcription level. They play significant roles in many biological processes, including tumorigenesis, differentiation, and apoptosis. Recent research has underlined the contribution of specific miRNAs to the pathogenesis of T-ALL and drug resistance. In the present review, the therapeutic potential of miRNA modulation in T-ALL disease will be discussed according to their role in disease biology, mechanisms of resistance, and possible strategies for clinical application.

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Introduction

T-cell acute lymphoblastic leukemia (T-ALL) is a hematological neoplasm caused by the progressive development of tumor immature T-cell precursors in the bone marrow and peripheral blood. This kind of ALL comprises about 15-20% of all cases and strikingly prevails among children and young adults [1, 2]. The pathogenesis of T-ALL is complex, with the involvement of genetic, epigenetic, and environmental factors. The common cytogenetic abnormalities include chromosomal rearrangements, notably involving the T-cell receptor loci, while mutations in the critical signaling pathways include *NOTCH1*, *PTEN*, and *FLT3*. The *NOTCH1* mutation is a particularly critical gene, as it is implicated in T-cell differentiation and survival; hence, this takes a leading role in disease pathogenesis. The malignant transformation present in T-ALL may thus be further contributed to by additional disrupted oncogenes and tumor suppressor genes [3, 4].

T-ALL can present with lymphadenopathy, splenomegaly, and bone pain. The most common symptoms of cytopenias caused by bone marrow infiltration are fatigue, increased risk of infection, and bleeding problems. There are several diagnostic requirements for T-ALL: morphological assessment, immunophenotyping, and cytogenetic studies. Immunophenotyping provides the necessary means for distinguishing T-ALL from other leukemias; it determines specific T-cell antigens, such as CD2, CD3, CD4, and CD8, expressed by the leukemic cells [5, 6].

The T-ALL treatment landscape has drastically changed in the last couple of decades; current regimens are dominated by multi-agent chemotherapy. The standard approach generally includes an induction phase followed by consolidation and maintenance treatments aimed at attaining complete remission and preventing relapse. Allogeneic stem cell transplantation is often pursued in high-risk patients due to the potential of eradicating residual disease. Also, the other presented novel developments include targeted therapies and immunotherapies that hold promise for improved patient outcomes in relapsed/ refractory T-ALL, including chimeric antigen receptor (CAR)-T cell therapy [7, 8].

Prognostic factors in T-ALL include age of diagnosis, initial white blood cell count, genetic abnormalities, and response to initial treatment. In general, younger patients tend to have a better prognosis, while specific genetic alterations, such as those involving the *NOTCH1* or *TLX* gene, carry an especially poor prognosis. Despite improvements in therapeutic modalities, relapse remains a significant threat in T-ALL patients, especially among the high-risk group; hence, further studies are needed to devise therapeutic strategies for a better understanding of disease biology [9].

miRNAs are tiny, non-coding RNA molecules that usually range between 19 and

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25 nucleotides in length but have become indispensable in regulating gene expression at the post-transcriptional level. Since their discovery, miRNAs have emerged as critical regulators of many cellular processes, such as differentiation, development, proliferation, and programmed cell death. Their importance in cancer biology is due to their widespread dysregulation in cancers, where they play critical roles in cancer initiation, progression, metastasis, and resistance to therapy. miRNAs function as oncogenes and tumor suppressors, controlling the expression of genes involved in major cancer-related pathways [10].

miRNA synthesis begins with the transcription of *miRNA* genes, which can be located within the introns of protein-coding genes or in non-coding regions. Initially, pri-miRNAs are transcribed by RNA polymerase II or III, and these pri-miRNAs are then processed into pre-miRNAs by the microprocessor complex, consisting of the RNase III enzyme Drosha and its cofactor DGCR8. These pre-miRNAs are transported to the cytoplasm, where they are cleaved by the Dicer enzyme into mature miRNA duplexes [11].

The guide strand of the miRNA duplex is incorporated into the RISC, guiding the complex to cognate target mRNAs through complementary base pairing. The cognate targets of miRNAs are mainly their 3' UTRs, and their interaction with the miRNA normally leads to degradation of the mRNA or inhibition of translation. This tight regulation of gene expression allows miRNAs to control a wide range of cellular activities [12].

In cancer, miRNAs may function as oncogenes or tumor suppressors depending on the context of their gene targets. The miRNAs that support tumorigenesis by repressing genes involved in cell cycle regulation or apoptosis are called oncomiRs, whereas those repressing oncogenes to reduce tumor growth are known as tumor-suppressor miRNAs [13].

Importance of studying miRNAs in T-ALL

microRNAs have become important regulatory molecules in many biological processes, and their involvement in the biology of cancer, especially in T-ALL, has become a focus of interest. T-ALL is an aggressive leukemia that results from the malignant transformation of T-cell precursors. Despite the progress of the therapeutic approach, the disease often features a high risk of relapse and drug resistance; hence, the extreme necessity of further understanding its molecular mechanisms. The investigation of miRNAs in the context of T-ALL is an important issue because they regulate gene expression at the post-transcriptional level; thus, they regulate several cellular processes involved in leukemogenesis, disease progression, and response to therapy (Table 1). This section explains the various reasons why miRNAs hold a central place regarding both understanding and treating T-ALL [14, 15].

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Table 1. A summary of miRNAs involved in the biological processes related to the progression and prognosis of T-ALL

miRNA	Role in T-ALL	Targets	Reference
miR-17-92 cluster (miR-19b, miR-17, etc.)	Promotes cell proliferation and survival, part of an oncogenic cluster.	PTEN, BIM	[17]
miR-21	Functions as an oncogene; enhances proliferation and inhibits apoptosis.	PTEN, PDCD4	[23]
miR-26a	Acts as a tumor suppressor; inhibits cell growth and promotes apoptosis.	EZH2	[36]
miR-34a	Induces apoptosis and cell cycle arrest; often downregulated in T-ALL.	NOTCH1, BCL2	[32]
miR-128b	Impairs cell viability and proliferation.	MYC, BMI1	[48]
miR-19b	Promotes cell proliferation and invasion; associated with poor prognosis.	FOXO1	[27]
miR-223	Serves as a tumor suppressor by inhibiting cell cycle progression and proliferation.	FBXW7, RASA1	[26]
miR-326	Influences drug sensitivity; potentially impacts chemotherapy resistance.	SMO, NOTCH1	[49]
miR-210 NF-kB	Supports cell survival in hypoxic conditions; involved in response to hypoxia.	ISCU, E2F3	[33]
miR-155	Overexpressed in several lymphoid malignancies; promotes proliferation and survival.	SHIP1, SOCS1	[42]
miR-181	Influences T-cell differentiation and can enhance sensitivity to apoptosis.	BCL-2, TCL1	[29]
miR-150	Acts as a tumor suppressor; involved in cell differentiation and apoptosis.	c-MYB, DNMT3B	[31]

1. miRNAs as key regulators of gene expression

miRNAs are powerful regulators of gene expression, acting at the level of mRNA stability and translation. They bind to the 3' untranslated regions of target mRNAs to inhibit expression. This post-transcriptional regulation allows miRNAs to fine-tune various cellular pathways, such as those involved in cell proliferation, differentiation, apoptosis, and immune responses. In T-ALL, dysregulation of certain miRNAs may alter the expression levels of critical oncogenes and tumor suppressor genes that result in uncontrollable cell growth and survival of malignant T-cell precursors. Their identification and study are crucial for the detailed elucidation of molecular mechanisms driving T-ALL and providing potential therapeutic insights [16].

2. miRNAs as drivers of leukemogenesis in T-ALL

The pathogenesis of T-ALL is driven by multiple genetic and epigenetic changes disrupting normal T-cell development. miRNAs lie centrally in driving this process through their regulation of genes important for T-cell differentiation and maturation. For

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example, miR-17-92 and miR-19b are overexpressed in T-ALL and function to drive leukemogenesis through the direct targeting of tumor suppressor pathways such as PTEN and BIM that enhance cell proliferation and survival. Moreover, downregulation of tumor-suppressive miRNAs, such as miR-34a, equally contributes to leukemic development by enabling the cells to escape from apoptotic signals. The way in which miRNAs orchestrate such oncogenic pathways is an important aspect of unraveling the molecular underpinnings of T-ALL [17].

3. miRNAs and the maintenance of leukemia stem cells

A critical role played by the leukemia stem cells (LSCs) partly depends on the initiation, maintenance, and recurrence of T-ALL. These cells possess the capacity for self-renewal and differentiation into various cell types that drive the growth and progression of leukemia. In T-ALL, the self-renewal and differentiation of LSCs are partly modulated at the molecular level by miRNAs. For instance, miR-128 has been identified as one of the main regulators in LSCs maintenance; its dysregulation has been associated with an increased ability of self-renewal and resistance to differentiation. The study of the role that miRNAs play in the biology of LSC can establish new therapeutic strategies that attack the very roots of leukemia and prevent relapse [18, 19].

4. miRNAs as biomarkers for diagnosis and prognosis

Disease heterogeneity is one of the big challenges in trying to treat T-ALL and leads to discrepancies in the patient's response to therapy and prognosis. This provides a premise for miRNAs as great diagnostic and prognostic biomarkers based on their stability in tissues and body fluids, including blood and serum. Specific expression profiles of miRNAs have been associated with different molecular subtypes in T-ALL, which can be used to risk stratify patients for relapse or resistance to therapy. For example, high levels of miR-155 expression have been associated with poor prognosis in patients with T-ALL. Moreover, changes in miRNA expression will indicate early treatment response or recurrence of disease and are, therefore, important for personalized treatment approaches [20].

Mechanisms of miRNA dysregulation in T-ALL

Dysregulation of miRNAs in T-ALL is multifactorial and thus complex, with each contributing differently to leukemogenesis and disease development. This includes the following: One such factor is genomic alterations; genetic alterations in miRNA genes themselves are one of the leading causes of the dysregulation of miRNAs. This can be understood from the fact that one cluster of well-known oncogenic miRNAs, namely miR-17-92, is frequently amplified in T-ALL. This amplification leads to the overexpression of several miRNAs that support cell proliferation and survival through the repression of tumor suppressors, including PTEN [20]. Another example is Transcriptional regulation by oncogenes. Some oncogenes and transcription factors, such as NOTCH1 and MYC, have been playing a critical role in regulating miRNA expression. NOTCH1 mutations are

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frequent in T-ALL and lead to perturbation of several miRNAs. Among them, miR-34a, a tumor suppressor miRNA that is normally downregulated in T-ALL, is included. Another potent oncogene, MYC, is often upregulated in T-ALL and modulates miRNA expression profiles by promoting oncomiRs while repressing tumor-suppressive miRNAs.

Additionally, these aberrant regulations of miRNAs are further facilitated by other epigenetic modifications, including DNA methylation and histone modifications [21]. For example, in T-ALL cells, the promoter of the tumor-suppressive miRNA miR-34a can be hypermethylated, resulting in its silencing. The loss of miR-34a allows for uncontrolled cell proliferation and escape from apoptosis, contributing to disease aggressiveness. Also, miRNA expression can be further influenced by the tumor microenvironment and external signals. Through the action of inflammatory cytokines and other microenvironmental factors, specific signaling pathways may be initiated that will lead to the upregulation or downregulation of the expression of certain miRNAs. This dynamic regulation further complicates the involvement of miRNAs in the pathogenesis of T-ALL [22].

miRNAs as regulators of oncogenes and tumor suppressors in T-ALL

1. OncomiRs: miRNAs promoting T-ALL progression

1.1- miR-21: A powerful modulator of tumor suppression and cell

miR-21 is one of the most well-acknowledged oncomiRs; it plays a major role in T-ALL through the repression of several tumor suppressor genes. Its overexpression in T-ALL has led to reduced quantities of many important suppressors, including PDCD4 and PTEN. Both these genes have central roles in inducing apoptosis and cell cycle regulation. miR-21 down-regulates PDCD4, driving unchecked cellular proliferation in T-ALL cells. PDCD4 is a recognized tumor suppressor; it inhibits translation and cell growth. Overexpression of miR-21 leads to the loss of function in PTEN, causing further activation of this pathway and promoting survival and growth in leukemic cells. PTEN is a well-recognized brake on the PI3K/AKT pathway, which is crucial for cell survival and cellular proliferation [23, 24]. It is, therefore, at the center of T-ALL cell survival, providing resistance to apoptosis and enabling unlimited growth, a characteristic feature of cancer cells. Its very potent effects have also meant that a promising approach to targeted therapy in T-ALL is the use of antagomirs-small anti-miRNA oligonucleotides or small-molecule inhibitors to sensitize the cells to chemotherapy [25].

1.2- miR-223: Modulator of NOTCH signaling

Another critical oncomiR, miR-223, has been similarly implicated in T-ALL development, significantly impacting the NOTCH signaling pathway, one of the key oncogenic pathways in this kind of leukemia. NOTCH signaling controls T-cell development, and its dysregulation represents a hallmark of T-ALL . By maintaining high levels of NOTCH activation, miR-223 ensures the continuous proliferation of T-ALL cells unchecked. Thus, miR-223 can be regarded as an important oncomiR in sustaining a leukemic state [26].

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1.3- miR-19b: Part of the miR-17-92 cluster

miR-19b belongs to the miR-17-92 cluster and is involved in cell survival and proliferation regulation in T-ALL. Its target is the PI3K/AKT pathway. The miR-17-92 cluster has been sometimes referred to as the "oncomiR cluster" as it has a very complex and multifaceted role in cancer- each component of miRNA regulates many facets of cell cycle progression and apoptosis. Of these, miR-19b is of special importance in T-ALL for its strong impact on survival pathways [27].

1.4- miR-181a: Enhancing leukemic proliferation

miR-181a has been overexpressed frequently in T-ALL and drives its aggressiveness. It basically works through the repression of pro-apoptotic genes and the induction of T-cell proliferation. The targets of miR-181a include a host of genes responsible for the induction of apoptosis, which it down-regulates, including *BIM* and *BAX*, critical for the mitochondrial pathway of apoptosis. Suppression helps leukemic cells to escape cell death due to the suppression of these apoptotic regulators [28]. miR-181a also modulates leukemic cell proliferation by targeting transcription programs that control cell cycle progression. This propensity to act dually in both repressing apoptosis and promoting proliferation places miR-181a as a central oncomiR in T-ALL [29].

1.5- miR-150: A context-dependent oncomiR and tumor suppressor

miR-150 displays a unique position in T-ALL, working in a context-dependent way. Whereas in some contexts, it acts like a tumor suppressor, in other contexts, it acts more like a promoter of oncogenic processes. miR-150 has been reported to regulate differentiation and proliferation during normal T-cell development [30]. The loss of miR-150 in T-ALL marks its tumor-suppressive role because of the interdependence of dysregulated differentiation and unchecked proliferation. On the other hand, miR-150 has been shown to drive leukemic transformation through its impact on a discrete differentiation stage of T-cells. Therefore, miR-150 can be considered as an agent in multiple functions within the pathogenesis of T-ALL [31].

2. Tumor-suppressive miRNAs in T-ALL

2.1- miR-15a and miR-16-1

Both of these miRNAs target the *BCL2* gene, known to be a crucial regulator of the mitochondrial pathway of apoptosis. *BCL2* functions as an anti-apoptotic member that prevents pro-apoptotic proteins involved in the release of cytochrome c from the mitochondria, an obligatory step in caspase activation, enzymes responsible for cellular destruction. In those cancers where the expression level of miR-15a/16-1 is low, *BCL2* is expressed too high and inhibits apoptosis, allowing the leukemic cells to live longer than they normally would, thus perpetuating tumor growth. Apoptosis could be reinstated by reconstituting the levels or function of miR-15a/16-1, which in turn would cause the cancerous cells to die [32].

2.2- miR-34a

This miRNA is part of the p53 tumor suppressor pathway. p53 plays a crucial role in the induction of cell cycle arrest or apoptosis

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in response to DNA damage and cellular stress. miR-34a increases apoptosis through the downregulation of SIRT1, a deacetylase that deactivates p53. In this way, miR-34a enhances indirectly the pro-apoptotic function of p53. Besides, miR-34a targets *BCL2*, enhancing apoptosis. This dual action makes miR-34a a very potent tumor suppressor, and its downregulation in T-ALL might be one of the ways through which malignant cells try to escape the resistance to apoptosis as an important defense mechanism [32].

3. Cell cycle regulation and proliferation control

3.1- miR-223

MiR-223 targets the transcription factor E2F1 in T-ALL, which has a critical role in the cell cycle at the transition between G1 to the S phase. E2F1 promotes genes responsible for DNA synthesis and replication. Such reduction of E2F1 by miR-223 leads to a block in the ability of cells to enter the S phase, a critical requirement for uncontrollable cell division. In the context of leukemia, loss of miR-223 results in unrestricted E2F1 activity, enabling T-ALL cells to proliferate uncontrollably. It targets the transcription factor E2F1, which is responsible for advancing the cell cycle from the G1 to the S phase. E2F1 initiates the transcription of genes whose products are required during DNA synthesis and replication. With reduced levels of E2F1 triggered by miR-223, a cell cannot progress into the S phase, resulting in the inhibition of excessive cell division. In the case of leukemia, downregulation of miR-223 led to unabated action of E2F1, thus enabling T-ALL cells to proliferate uncontrollably [33].

3.2- miR-193b

It targets the CCND1 regulatory protein, which advances the cell cycle through the G1 phase by activating CDKs. The complex of CDK4/6 and Cyclin D1 phosphorylates the Rb protein, releasing E2F transcription factors to initiate DNA synthesis. miR-193b suppresses this through direct inhibition of CCND1. Without this suppression, T-ALL cells surge through the G1/S checkpoint with uncontrolled proliferation. Therefore, one possible therapeutic avenue for treatment in T-ALL involves restoring the functionality of miR-193b to impede or arrest tumor cell growth [34].

4- Epigenetic regulation and oncogene suppression

4.1- miR-101

It represses EZH2, the core component of PRC2 that catalyzes the trimethylation of histone H3 on lysine 27 (H3K27me3), leading to the transcriptional repression of various tumor suppressor genes. Overexpression of EZH2 has been commonly observed in many cancers, including T-ALL, where it facilitates oncogenesis via silencing genes involved in the regulation of cell differentiation and proliferation [35]. By repressing EZH2, miR-101 maintains a more open chromatin conformation and the expression of tumor suppressor genes. Loss of miR-101 results in excessive repression of tumor suppressor pathways, sustaining the oncogenic state of the T-ALL cells [36].

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4.2- miR-29 family

The miR-29 family, including miR-29a, miR-29b, and miR-29c, targets several epigenetic regulators, such as DNMT3A, a DNA methyltransferase, and TET2, a demethylase involved in DNA hydroxymethylation. These enzymes regulate gene expression by adding and removing methyl groups to DNA, respectively. In T-ALL, overexpression of DNMT3A can hypermethylate and silence tumor suppressor genes, while loss-of-function mutations of TET2 prevent the removal of these repressive marks [36]. The miR-29 family acts as tumor suppressors by knocking down both DNMT3A and TET2 to maintain a balanced epigenetic state toward normal cell function. Loss of expression of miR-29 is linked with the hypermethylation of tumor suppressor genes, facilitating the development of T-ALL [37].

5. Metabolic reprogramming and tumor suppression

Metabolic reprogramming is one of the features shared among diverse types of cancers, including T-ALL, which enables its rapid proliferation. On the other hand, tumor-suppressive miRNAs may target genes coding for proteins that are part of crucial metabolic pathways, hence limiting the growth of cancer cells. The miR-125b modulates cellular metabolism by directly targeting HK2, one of the key enzymes involved in glycolysis. Glycolysis is the process in which glucose is metabolized to pyruvate, thereby producing energy in the form of ATP and giving precursors of biomolecules for cell growth. Cancer cells, including those in T-ALL, often upregulate glycolysis even when oxygen is in its normal condition. This is called the Warburg effect. miR-125b directly targets HK2, reducing the glycolytic capacity in T-ALL cells, which in turn impairs energy and biomass production necessary to support their growth and survival. Thus, the loss of miR-125b promotes glycolytic flux, enabling leukemic cells to support their aggressive growth. The re-establishment of miR-125b expression could counteract the metabolic advantage of such tumor cells [38].

Role of miRNAs in leukemia stem cell maintenance

Whereas a subpopulation of cells within the leukemic population, similar to normal hematopoietic stem cells, possesses the capabilities of self-renewal and differentiation into myeloid and lymphatic lineages, in T-ALL the self-renewing process becomes dysregulated, leading to uncontrolled proliferation combined with disturbed differentiation [39]. Dysregulation often results from genetic mutations and epigenetic alterations involving aberrant expression of miRNAs. A number of miRNAs have been identified that either promote or suppress these stem cell-like properties. When tumor suppressor miRNAs, normally acting to inhibit self-renewal, are lost, or when oncogenic miRNAs (oncomiRs) that drive self-renewal are overexpressed, LSCs can persist to promote treatment resistance and relapse.

1. miR-34a: Suppressing self-renewal and inducing differentiation

MiR-34a is among the best-characterized tumor-suppressive miRNAs, usually down

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regulated in a wide range of cancers, including T-ALL. It acts as a critical effector of the p53 tumor suppressor pathway and elicits cell cycle arrest and apoptosis following DNA damage or oncogenic stress. Thus, in the context of T-ALL, miR-34a has been illustrated to be an essential regulator that restricts the self-renewal capability and enforces differentiation of leukemic stem cells [40]. miR-34a directly targets *BCL2* and *SIRT1*, crucial for cell survival and resistant to apoptosis. By down-regulating these proteins, miR-34a reconstitutes the apoptotic machinery in T-ALL stem cells and dampens their self-renewal capacity. In addition, miR-34a increases the differentiation of such cells by inhibiting other stem cell-associated pathways. It reduces the pool of LSCs and enhances the efficacy of differentiation therapies [41].

2. miR-155: A potent oncomir promoting LSC self-renewal

MiR-155 is a highly oncogenic miRNA whose overexpression is detected in the majority of hematological malignancies, including T-ALL. Overexpression is directly related to the maintenance of self-renewal capacity and inhibition of differentiation in T-ALL stem cells. Indeed, by targeting transcription factors critical for hematopoietic differentiation, such as PU.1 and C/EBPβ, miR-155 exerts pro-leukemic effects. In this way, repression of these targets by miR-155 suppresses differentiation signals and maintains the undifferentiated, stem-cell-like phenotype of the LSCs. Further, miR-155 is involved in promoting the survival of LSCs through controlling anti-apoptotic pathways and establishing a cytokine-rich milieu favorable for the expansion of LSCs [42].

3. miR-29 family: Balancing self-renewal and differentiation

The miR-29 family, which includes miR-29a, miR-29b, and miR-29c, plays an important role in regulating hematopoietic differentiation. In T-ALL, when miR-29 is downregulated, this leads to the perpetuation of a self-renewing, undifferentiated population. miR-29 targets a range of epigenetic regulators, including the DNA methyltransferase *DNMT3A* and the DNA demethylase TET2. Both of these enzymes are involved in establishing proper DNA methylation patterns that define gene expression. Overexpression of *DNMT3A* and *TET2*, because of reduced levels of miR-29, results in unusual patterns of DNA methylation, which support self-renewal and impede differentiation of LSCs. Normalization of the miR-29 level might reinstall normal methylation patterns, allowing differentiation and reducing the self-renewal capacity of leukemic cells [42].

4. miR-181: Regulating apoptosis and stemness

miR-181 plays a role in regulating T-cell development as well as the course of T-ALL. Its dysregulation leads to increased self-renewal and survival of T-ALL stem cells. One target through which miR-181 induces apoptosis is *BCL2L11*. As mentioned previously, *BCL2L11* controls the switch between apoptosis and survival in LSCs. miR-181 suppresses apoptosis and thereby mediates LSC survival to maintain them within the leukemic population. Added to that, miR-181

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modulates pathways responsible for the differentiation of progenitor T-cells and thus further supports the maintenance of the stem-like phenotype of T-ALL [43].

miRNAs and chemoresistance in T-ALL

1. miR-125b: A key player in glucocorticoid resistance

MiR-125b has now been established as the key player in regulating glucocorticoid resistance in T-ALL. Glucocorticoids, such as dexamethasone, are among the mainstays in treating T-ALL due to their capability of inducing apoptosis in lymphoid cells. However, glucocorticoid resistance is the major hindrance to effective treatment in a set of patients. miR-125b modulates glucocorticoid sensitivity by targeting several components of the glucocorticoid receptor pathway. It directly downregulates the expression of GRα, the receptor through which glucocorticoids exert their apoptotic effects [44]. Lower levels of GRα translate to impaired glucocorticoid signaling and diminished induction of pro-apoptotic genes. Besides, miR-125b represses pro-apoptotic genes such as *BAX* and *PUMA*, thereby promoting the survival of leukemic cells in the presence of glucocorticoid treatment. High expression levels of miR-125b have been associated with glucocorticoid resistance in patients with T-ALL. Therapy targeting the inhibition of miR-125b may be able to restore glucocorticoid sensitivity and improve the possibility of better treatment outcomes [45].

2. miR-19b: Targeting PI3K/Akt pathway and multidrug resistance

It has also been documented that miR-19b, part of the oncogenic miR-17-92 cluster, can induce multidrug resistance in T-ALL via PI3K/Akt signaling. resistance to chemotherapy agents is mediated by miR-19b through its target, tumor suppressor PTEN, which prevents stimulation of the PI3K/Akt pathway. Loss of PTEN function leads to activation of the PI3K/Akt pathway, which promotes survival and drug resistance of the cell. This also results in the upregulation of anti-apoptotic proteins, including BCL2 and MCL1, further contributing to drug resistance through the evasion of apoptosis in response to chemotherapy. Restoration of PTEN expression by inhibition of miR-19b could reactivate the apoptotic response to chemotherapy and decrease the survival advantage provided by Akt activation. The miR-19b inhibitors can be combined with PI3K/Akt inhibitors in an attempt to improve the efficacy of conventional chemotherapy for resistant T-ALL cases [46].

3. miR-21: A mediator of chemoresistance via anti-apoptotic pathways

It is one of the most studied oncogenic miRNAs, implicating it in the regulation of drug resistance in many cancers, including T-ALL. miR-21 induces resistance to chemotherapeutic agents by repressing PDCD4, a tumor suppressor and critical regulator of apoptosis. The downregulation of PDCD4 by miR-21 increases cell survival and makes cells resistant to apoptosis. Moreover, miR-21 targets PTEN, causing a further activation of the PI3K/Akt pathway and leading to the development of resistance against a wide range of chemotherapeutic agents. Overexpression of miR-21 in T-ALL is associated with poor prognosis and reduced

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sensitivity to treatment. Therapeutic inhibition of miR-21 may enhance apoptosis and sensitize T-ALL cells to chemotherapy, thereby improving patient outcomes [47].

4. miR-223: Modulating chemotherapy response and cell cycle progression

miR-223 had previously been identified to modulate drug resistance in T-ALL by targeting the critical regulators of cell-cycle progression and apoptosis. Mechanism of resistance: miR-223 downregulates FBXW7, an E3 ubiquitin ligase targeting numerous oncoproteins such as c-Myc, NOTCH1, and Cyclin E for degradation. Loss of FBXW7 function due to this downregulation results in the accumulation of these proteins, promoting cell proliferation and resistance to chemotherapy. miR-223 also targets E2F1, a critical regulator of G1/S cell cycle transition, further reinforcing its message of cell survival and drug resistance against agents disrupting cell cycle progression. Such strategies as the reconstitution of miR-223 expression or the suppression of its oncogenic targets are promising for the inhibition of T-ALL cell proliferation and improving their chemosensitivity. MiR-223 is probably a target for improving the efficiency of cell cycle inhibitors in T-ALL treatment [48].

5. miR-326: Modulating resistance to asparaginase

MiR-326 was implicated in resistance to asparaginase, an important chemotherapeutic agent in the treatment of T-ALL. miR-326 targets E2F1, a transcription factor involved in regulating the asparagine synthetase gene *ASNS*. Asparaginase works by depleting the extracellular levels of asparagine that are required for survival in leukemic cells. Overexpression of miR-326 reduces the E2F1 expression level, which in turn upregulates *ASNS*, enabling the T-ALL cells to synthesize asparagine themselves and thus confer resistance against the asparaginases. The suppression of *ASNS* expression by targeting miR-326 may overcome asparaginase resistance and enhance the therapeutic outcome. A combination of asparaginase and miR-326 inhibitors thus leads to better responses in resistant patients with T-ALL [49].

6. miR-708: A tumor suppressor involved in apoptosis and drug sensitivity

miR-708 is a tumor-suppressive miRNA that is downregulated in drug-resistant T-ALL. miR-708 targets *DDIT4*, encoding a critical regulator of the mTOR pathway. The mTOR pathway is involved in cell growth, metabolism, and survival, and its dysregulation contributes to drug resistance. Downregulation of miR-708 leads to higher *DDIT4* expression, causing activation of the mTOR pathway that results in enhanced survival of cells under chemotherapeutic stress. Downregulation of miR-708 attenuates its pro-apoptotic activity, further contributing to drug resistance. The reconstitution of miR-708 may inhibit mTOR signaling and sensitize T-ALL cells to chemotherapy. Targeting miR-708 therapy could be a very valuable strategy for overcoming chemoresistance, as the mTOR pathway is very frequently activated in drug-resistant tumors [50, 51].

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Therapeutic strategies for miRNA modulation in T-ALL

The manipulation of miRNAs offers a promising approach to improve therapeutic outcomes in T-ALL, particularly in overcoming drug resistance. Here, we will delve into several key strategies for miRNA modulation, focusing on their mechanisms, advantages, and challenges.

1. miRNA mimics

miRNA mimics are small synthetic molecules designed to mimic the function of naturally occurring miRNAs expressed in T-ALL. The introduction of such mimics into the cells aims at the restoration of expression of tumor-suppressive miRNAs, enhancing the pathways of apoptosis and inhibiting oncogenic signaling. Mimics selectively restore miRNA function by promoting the degradation of target oncogenes. Preclinical models have established that miR-34a mimics induce apoptosis with efficiency and increase sensitivity to chemotherapy. Delivery systems: Due to the fact that miRNAs are rapidly degraded when in circulation, delivery matters a great deal for the exercise of its efficacy. This needs the formulation of sophisticated delivery systems, such as lipid nanoparticles or viral vectors, that ensure stability within the bloodstream and further into cells. There is the potential to inadvertently modulate genes other than targets, requiring careful design and testing [52].

2. Antagomirs

Antagomirs are chemically modified oligonucleotides that target the inhibition of specific miRNAs overexpressed in T-ALL, such as miR-21 or miR-125b, and thereby contribute to drug resistance. By inhibiting these miRNAs, the antagomirs restore the expression of their target genes, which may reactivate the apoptotic pathways and enhance drug sensitivity.

Reversal of drug resistance: Targeting of oncogenic miRNAs by antagomirs has been demonstrated to improve the efficacy of existing therapies significantly. For instance, antagomirs against miR-21 have shown promise in improving the apoptotic response of T-ALL-resistant cells.

Specificity: Antagomirs can be designed to be specific for a particular miRNA in order to limit off-target effects.

Stability/Bioavailability: The antagomirs will have to be modified to avoid degradation and guarantee adequate delivery for therapeutic potential.

Possible immune response: The introduction of foreign nucleic acids may cause an immune response; thus, changes might be made in order to diminish such potential risks [53].

3. miRNA sponges

miRNA sponges are constructs harboring multiple binding sites for specific miRNAs, thereby effectively sequestering them and preventing their interaction with target mRNAs. This may be particularly useful for miRNAs, such as miR-19b, whose activities modulate a wide array of oncogenic pathways. Sponges provide a continuous approach to the inhibition of miRNAs that might lead to long-term therapeutic efficacy. Thus, the sponges target a single miRNA that might have several targets, therefore affecting several pathways with one target, making them more therapeutically effective. For the

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development of functional sponge constructs, great detail has to be known on the binding sites of the miRNA and their interaction with targets. Like mimics and antagomirs, sponges also present challenges in delivery and bioavailability [54].

4. Combination therapies

As a potential tool, miRNA modulation may be combined with traditional chemotherapy to enhance efficacy. For example, the administration of antagomirs in conjunction with chemotherapeutic agents may inhibit drug resistance pathways while killing leukemic cells directly. This approach has been shown to be effective in preclinical settings and, therefore, might translate into improved clinical benefits for patients with T-ALL by modulating miRNAs [55].

Conclusion

miRNAs play a crucial role in the pathogenesis and treatment response of T-ALL, as they fine-tune gene expression and regulate cellular processes like proliferation, apoptosis, and differentiation. Dysregulation of miRNAs in T-ALL leads to leukemogenesis and drug resistance, a major challenge in managing the aggressive malignancy. MiRNA mimics, antagomirs, and sponges offer new therapeutic strategies for improving treatment outcomes and reversing drug resistance. However, effective translation into the clinic faces challenges such as delivery systems, off-target effects, and regulatory issues. Further investigation into miRNA expression profiling in T-ALL is needed to identify new therapeutic targets and define complex networks.

Ethical Considerations

The authors state that there are no ethical issues related to this review. All data and literature are properly cited, with no involvement of human or animal subjects. Ethical guidelines for literature reviews have been followed, ensuring transparency and integrity in the manuscript preparation.

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Conflict of Interest

The authors declare no conflict of interest.

Authors' Contributions

K.T: Conceptualization, data curation, formal analysis, investigation, visualization, and writing the original draft; P.V.A: Conducted literature searches and contributed to analysis; N.G.M: Identified key themes and wrote specific sections; A.D: Assisted in organizing the manuscript; S.A: Oversaw conceptualization, funding acquisition, methodology, supervision, and editing of the manuscript. All authors have read and approved the final version and are accountable for the integrity of the work.

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