Chinese Journal of Integrative Medicine
Available online at link.springer.com/journal/11655
Journal homepage: www.cjim.cn/zxyjhen/zxyjhen/ch/index.aspx
E-mail: cjim_en@cjim.cn

Review

Research Progress on Artemisinin and Its Derivatives against Hematological Malignancies*

LI Ying, SHAN Ning-ning, and SUI Xiao-hui

ABSTRACT Although current therapeutic methods against hematological malignancies are effective in the early stage, they usually lose their effectiveness because of the development of drug resistances. Seeking new drugs with significant therapeutic effects is one of the current research hotspots. Artemisinin, an extract from the plant *Artemisia annua* Linne, and its derivatives have excellent antimalarial effects in clinical applications as well as excellent safety. Recent studies have documented that artemisinin and its derivatives (ARTs) also have significant effects against multiple types of tumours, including hematological malignancies. This review focuses on the latest research achievements of ARTs in the treatment of hematological malignancies as well as its mechanisms and future applications. The mechanisms of ARTs against different types of hematological malignancies mainly include cell cycle arrest, induction autophagy and apoptosis, inhibition of angiogenesis, production of reactive oxygen species, and induction of differentiation. Additionally, the review also summarizes the anticancer effects of ARTs in many drug-resistant hematological malignancies and its synergistic effects with other drugs.

KEYWORDS artemisinin, artesunate, dihydroartemisinin, hematological malignancies, review

Hematological malignancies are a collective term for neoplastic diseases of hematopoietic and lymphoid tissues. According to the classification of hematopoietic and lymphoid tumours by the World Health Organization (WHO) in 2016, hematological malignancies are classified into two major categories: myeloid neoplasms and lymphoid neoplasms. Common hematological malignancies include leukaemia, lymphoma, and multiple myeloma (MM).[1] The causes of most hematological malignancies remain unclear. Related possible factors include genetics, immunity, pollution, physical, chemical, and biological factors. In recent years, the incidence of hematologic diseases has increased.

Currently, the main therapeutic methods against hematological malignancies are chemotherapy, targeted therapy, cell therapy, and hematopoietic stem cell transplantation. Although these methods are effective in the early stage, they usually lose their effectiveness because of the development of cancer cell resistance. Therefore, seeking new drugs with significant therapeutic effects against hematological malignancies and without the problems of drug resistance is particularly important. Indeed, there is a long history of Chinese medicine (CM) used to treat malignant tumours. CM treatment of leukaemia not only has a positive therapeutic effect but also demonstrates fewer side effects.[2] Chinese herbal medicine is becoming increasingly influential in world medicine, and developing effective natural antitumour drugs is one of the hotspots in tumour therapy. *Artemisia annua*, also called *Artemisia annua* Linné, has excellent antimalarial effects. It has been used in medicine historically and was first discovered in "*Fifty-two Prescriptions* (Wu Shi Er Bing Fang)", the silk books unearthed from the Han Dynasty tombs in No. 3 Mawangdui (approximately 168 BC). Later, it was documented in the books "*Shen Nong's Herbal Classics* (Shen Nong Ben Cao Jing)", "*Da Guan Materia Medica*" and "*Compendium of Materia Medica*".

*Supported by the National Natural Science Foundation of China (No. 81570104), Key Research and Development Project of Shandong Province (No. 2015GSF118025, 2015GSF118058, 2016GSF201026)
Department of Hematology, Shandong Provincial Hospital Affiliated to Shandong University, Jinan (250021), China
Correspondence to: Prof. LI Ying, E-mail: yingren10301@sina.com
DOI: https://doi.org/10.1007/s11655-019-3207-3

Aside from antimalarial effects,[3,4] ARTs have also been reported to have anti-schistosomiasis,[5,6] antiviral,[7,8] antibacterial,[9,10] antifungal,[11] and anti-tumour[12-19] effects. The anti-tumour effects of ARTs have begun attracting increasing attention. In this review, we summarize recent published literature regarding effects and mechanisms of ARTs on haematological malignancies.

Type and Structure of ARTs

In the 1960s, the development of malaria strains resistant to chloroquine in endemic areas, including China, called for the discovery of new and more efficient antimalarial drugs. In 1967, Chinese scientists screened a series of traditional remedies and found that the extracts of *Artemisia annua* L. had potent antimalarial activity. In 1972, the active component of the plant, named Qinghaosu or artemisinin, was purified,[20] which is presented in the leaf and stem parts at approximately 0.01%–0.8% of the dry weight of the plant.[21] In the first clinical trial to investigate the effect of artemisinin against malaria, thirty people infected with malaria were cured, and no malaria parasite was subsequently detected in their blood. Due to this pioneering work, the Chinese scientist TU You-you was awarded the Nobel Prize for physiology and medicine in 2015.[22-24]

The chemical structure of artemisinin was determined in 1979.[20] Chemically, artemisinin is a sesquiterpene trioxane lactone with the empirical formula of $C_{15}H_{22}O_5$. It contains a peroxide bridge that is essential for its activity, three methyl groups (one tertiary and two secondary), and several other aliphatic carbon atoms. The lactone in artemisinin can easily be reduced, resulting in the formation of dihydroartemisinin (DHA), which can be used to synthesize many other derivatives, including artesunate (ART), artemether, and arteether. The chemical structures of artemisinin and its common derivatives are shown in Figure 1.[12,13,25,26] ARTs can induce a very rapid reduction of parasitaemia, starting almost immediately after administration. The derivatives even have better bioavailability, efficacy and tolerability than artemisinin, as well as fewer side effects.[27] The mechanisms of killing malaria parasites involve peroxide bridges from ARTs interacting with the haem in malaria parasites to produce carbon-centred free radicals that can alkylate proteins and damage the microorganelles and membranes of the parasites.[28]

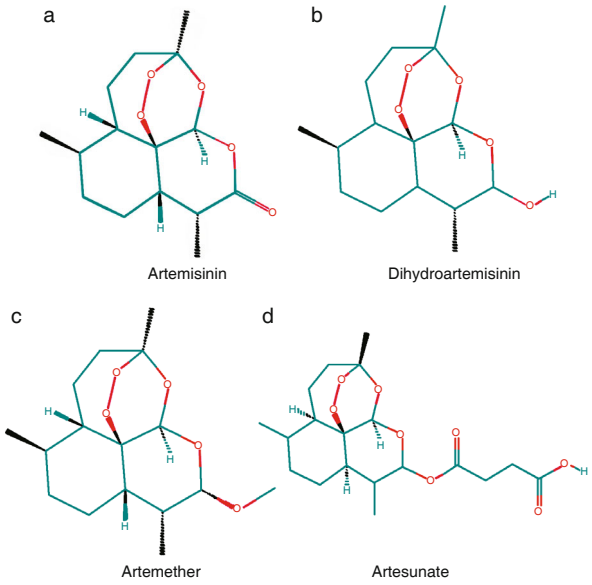

Figure 1. Chemical Structure of ARTs

Notes: (a) artemisinin, (b) dihydroartemisinin, (c) artemether, (d) artesunate

Effects of ARTs on Hematological Malignancies

Aside from antimalarial effects, ARTs have also exhibited anti-proliferative and pro-apoptotic effects against several types of cancer cells, including leukaemia, lymphoma, and multiple myeloma, the three common types of hematological malignancies. Table 1 summarizes the *in vitro* studies of ARTs on various hematological cancer cell lines. However, to date there has been no review on the mechanisms of artemisinin on hematological malignancies. Herein, we summarized related studies about the effects and mechanisms of ARTs against leukaemia, lymphoma, and multiple myeloma.

Inhibition of Proliferation and Arrest of Cell Cycle

Numerous studies have found that ARTs can inhibit the proliferation of haematological malignancy cells, although the stated mechanisms are not exactly the same across these studies. For example, Wang, et al[29] found that DHA can inhibit the growth of human myeloid leukaemia K562 cells. And the further investigations revealed the ROS dependency in DHA-induced autophagy of leukaemia K562 cells. LC3-Ⅱ protein expression and caspase-3 activation occurred following the autophagy processes. Kim, et al[30] investigated the effect of artesunate on various protein kinases, associated gene products, cellular response, and apoptosis and examined the *in vivo* effect of artesunate on the growth of human chronic myeloid leukemia (CML) xenograft tumours

Table 1. Summary of *In Vitro* Studies with ARTs on Various Haematological Cancer Cell Lines

Origin	Cell lines	Drug	Effects	Molecular mechanisms	Ref.
Leukaemia	KBM-5	ART	Causes the accumulation of cells in sub-G_1 phase of the cell cycle	Suppresses the phosphorylation of p38, ERK, CREB, CHK-2, STAT5, and RSK.	(66)
	HL-60, NB4, U937	DHA	Induces apoptosis	Induces apoptosis in acute myeloid leukaemia through the Noxa-mediated pathway requiring iron and endoperoxide moiety.	(67)
	MV-4-11, MOLM-13	ART	Inhibits MV4-11 and MOLM-13 cells with IC_{50} values of 1.1 and 0.82 μmol/L, respectively	Leads to cellular and mitochondrial ROS accumulation, decreases cellular BCL-2 level and double stranded DNA damage, causes mitochondrial membrane potential loss and induces the intrinsic mitochondrial apoptotic cascade in AML cell lines.	(68)
	AML and ALL primary human leukaemia cells	DHA	Induces apoptosis of primary human leukaemia cells	DHA-induced apoptosis was accompanied by caspase activation, cytochrome c release, Mcl-1 downregulation, and MEK/ERK inactivation.	(36)
	THP-1	ART	Induces apoptosis in THP-1 cells	Inhibition of STAT3 and activation of caspase-3 and caspase-8.	(69)
	THP-1	DHA	May induce apoptosis in the AMoL THP-1 cell line	Downregulation of ERK and Akt and activation of caspase-3.	(70)
	HL60	DHA	Induces HL60 cell apoptosis	Induces HL60 cell apoptosis via downregulation of TfR expression, resulting in an induction of apoptosis through the mitochondrial pathway.	(37)
	HL60	DHA	Induces apoptosis	Induces apoptosis is dependent on iron and p38 MAPK activation	(38)
	K562	DHA	Inhibits the proliferation of leukaemia K562 cells	Facilitates the induction of apoptosis by downregulating the expression of the BCR/ABL fusion gene.	(71)
	K562	DHA	Inhibits the proliferation of and induces apoptosis in cells. Effectively inhibits the VEGF expression and secretion in K562 cells	NA.	(50)
	K562	DHA	Inhibits the growth of CML K562 cells by its anti-proliferative and apoptotic effects	Influences BCR/ABL expression and its downstream signal factors.	(72)
	K562	DHA	Inhibits the growth of K562 cells, and induces autophagy	Mitochondrial pathway, related to ROS.	(29)
	K562	ART	Decreases the simulating angiogenic activity of K562 cells	Inhibits the VEGF expression.	(49)
	K562, U937	ART	Inhibits cell growth and induce the cell apoptosis	ART treatment increases Fas expression, while it decreases the c-Fos level in K562 cells.	(62)
Lymphoma	18 different B-cell lymphoma cell lines	ART	Reduces cell growth with IC_{50} values from 0.189 to 3.72 μmol/L; a high sensitivity to artesunate (IC_{50}<1 μmol/L) is observed in 11 out of 18 cell lines tested	Artesunate-induced expression of the ER stress markers ATF-4 and DDIT3 are specifically upregulated in malignant B-cells.	(41)
	Raji	ART	Inhibits the proliferation activity of Raji cells	Induces autophagy and apoptosis.	(73)
	Jurkat	DHA	Decreases Jurkat cell viability and induces apoptosis	Induces ROS generation, downregulates TfR, VEGF and hTERT mRNA expression, promotes apoptosis, and triggers cell cycle arrest.	(55)
	Karpas-422, sudhl-4, sudhl-6, OCI-Ly3, OCI-Ly10	ART	Inhibits cell growth and induces apoptosis	Downregulates MYC and anti-apoptotic Bcl-2 family proteins with cleavage of caspase-3.	(43)
	Raji, Jurkat	ART	Inhibits the proliferation of Raji and Jurkat cells	Mitochondria transmembrane potential collapse; increases caspase-3 activities.	(40)
	Jurkat	DHA	Induces apoptosis in Jurkat cells; relies on the intrinsic death pathway	Induces a breakdown of mitochondrial transmembrane potential, releases cytochrome c, and activates caspases.	(39)
	Ramos	ART	Induces apoptosis *in vitro*	Induces Fas/CD95 expression and the formation of ROS and results in a breakdown of mitochondrial membrane potential.	(65)
MM	ANBL-6; CAG; IH-1; OH-2; RPMI-8266; U266; VOLIN; INA-6; JJN-3;	ART	Inhibits cell growth and induces apoptosis	Downregulates MYC and anti-apoptotic Bcl-2 family proteins with cleavage of caspase-3.	(43)
	SP2/0	ART	Inhibits proliferation and induces apoptosis of cells	Decreases the level of NFkappaB p65 protein in the nucleus and increases the level of IkappaBalpha protein in the cytoplasm.	(34)
	U266	DHA	Decreases cell viability; increases the sub-G0/G1-phase cell population	Increases caspase-3 and c-Jun expression in U266 cells through activation of the JNK signalling pathway.	(35)
	RPMI8226	DHA	Reduces microvessel growth in chicken chorioallantoic membranes	Decreases VEGF secretion.	(51)
	RPMI8226	ART	Efficiently suppresses angiogenic ability	ART can block ERK1/2 activation and downregulate VEGF and Ang-1 expression.	(52)

Notes: ART: artesunate; DHA: dihydroartemisinin; MM: multiple myeloma; NA: not available

in athymic nu/nu mice. Their results suggest that artesunate exerts its anti-proliferative and pro-apoptotic effects through suppression of multiple signalling cascades in CML both *in vitro* and *in vivo*. Sun, et al[31] found that different concentrations of artesunate have different effects on the cell cycle of K562 cells. When the concentration of artesunate increased to 200 μmol/L, the proportion of cells at G_2/LM phase decreased, but the inactivation proportion increased to 39.65%. These results suggest that ARTs may act on the cell cycle to inhibit the proliferation of haematological malignancy cells.

In addition to leukaemia, ARTs can also inhibit the proliferation of lymphoma and multiple myeloma. Zhao and colleagues[32] used diffuse large B-cell lymphoma (DLBCL) cells to test the anti-lymphoma effects and mechanisms of artemether. They found that artemether inhibited the proliferation of cancer cells and caused them to stagnate in G_0/G_1 phase and induced apoptosis as drug concentrations increased. Toril, et al[33] also found that artesunate at the dose used for the treatment of malaria could efficiently inhibit the growth of both lymphoma and myeloma cells without serious adverse effects. Li, et al[34] reported that artesunate have an anti-myeloma effect by inhibiting proliferation and inducing apoptosis of SP2/0 cells. Wang, et al[35] observed that DHA can decrease cell viability and increase the sub-G_0/G_1-phase cell population. They also found that DHA treatment increased caspase-3 and c-Jun expression in U266 cells via JNK signalling pathway activation. These results showed that DHA can inhibit the proliferation of multiple myeloma cells by interfering with the JNK signalling pathway.

Induction of Apoptosis

Induction of apoptosis is an important mechanism for the anticancer effects of ARTs on haematological malignancies. Gao, et al[36] applied DHA to treat primary leukaemic cells from acute myeloid leukemia (AML) and acute lymphoblastic leukemia (ALL) patients. They found that DHA can induce apoptosis in both kinds of primary human leukaemia cells. Apoptosis of the HL60 cell line was also confirmed upon exposure to DHA. Zhou, et al[37] found that DHA can specifically reduce the mRNA and expression of TfR in HL60 cells, induce the activation of caspase-3, and affect the expression of Bcl-2 and Bax. Therefore, they believe that DHA can induce apoptosis of HL60 cells through the mitochondrial pathway by lowering the TfR expression. Lu, et al[38] also found that DHA can induce HL-60 cells apoptosis effectively. Mitochondrial dysfunction and cysteine protease activation were also found. Further studies demonstrated that the signalling pathway of DHA-induced cell apoptosis is dependent on iron and p38 MAPK activation.

In addition to leukaemia cells, ARTs can also induce the apoptosis of lymphoma cells. Handrick, et al[39] has found that DHA treatment can induce several established features of apoptosis in T-cell lymphoma Jurkat cells, such as the breakdown of the mitochondrial transmembrane potential, release of cytochrome c, activation of caspases, and production of DNA fragments. Zeng, et al[40] observed and confirmed that the proliferation of Raji and Jurkat cells was inhibited by artesunate. Mitochondrial transmembrane potential was collapsed once these cells were exposed to artesunate, thus activating caspase-3-dependent apoptosis in these cells. Vatsveen, et al[41] also evaluated the anti-lymphoma effect of artesunate both *in vitro* and *in vivo*. An invasive B-cell lymphoma cell xenograft model from NSG mice and 18 species of B-cell lymphoma cell lines were used as their subjects in the study. Their results demonstrate that artesunate has a strong apoptosis-inducing effect in a variety of B-cell lymphoma cell lines and has significant anti-lymphoma activity *in vivo*. The authors suggested artesunate as a drug for the treatment of B-cell lymphoma. Cheng, et al[42] synthesized a new water-soluble artemisinin derivative named SM1044, which can induce autophagy-dependent apoptosis in diffuse large B-cell lymphoma (DLBCL) cells. This was achieved by accelerating the degradation of the apoptosis inhibitory protein survivin through its acetylation-dependent interaction with the autophagy-related protein LC3-Ⅱ. Holien, et al[43] also confirmed that artesunate can effectively inhibit the growth of DLBCL cell line and induce cell apoptosis by downregulating MYC and BCL-2 family proteins and activating caspase-3.

To search for more effective therapies for the treatment of myeloma, Papanikolaou, et al[44] applied artesunate to treat 8 types of multiple myeloma cell lines. The results showed that artesunate can induce immature and drug-resistant MM cell death by the non-caspase-mediated pathway. Holien, et al[43] have also shown that artesunate treatment effectively inhibited the growth and induced apoptosis of myeloma cell

lines. The IC_{50} values of artesunate against the used cell lines varied from 0.3–16.6 μmol/L, which can be obtained in plasma after intravenous administration of artesunate in malaria treatment. Therefore, artesunate used in doses similar to current malaria treatments can potentially be used for multiple myeloma with no serious adverse effects.

Inhibition of Angiogenesis

Angiogenesis, the formation of new blood vessels, is an important component of the pathogenesis of haematologic malignancies, as it is in solid tumours.[45-48] A negative prognostic implication of increased angiogenesis has been established for several types of haematologic malignancy. Therefore, it is rational to seek out therapeutic uses of both old and new drugs that might affect new blood vessel formation in the treatment of haematologic malignancies. Several cytokines are involved in promoting tumour angiogenesis, vascular endothelial growth factor (VEGF) was one of the most important factors, which is a survival and a proliferation factor for human microvascular endothelial cells. Therefore, it is theoretically feasible to use VEGF inhibitors for the treatment of patients with haematologic malignancies.

Several studies have shown that ARTs can inhibit the expression of VEGF and thereby inhibiting angiogenesis. For example, Zhou, et al[49] found that artesunate can decrease the stimulating angiogenic activity in K562 cells, a model cell of CML. They compared the VEGF expression levels and VEGF mRNA in K562 cells with or without artesunate treatment. The results showed that artesunate is capable of inhibiting VEGF expression. A study by Lee, et al[50] also showed that DHA can inhibit VEGF expression and secretion in K562 cells obtained from CML patients. In addition to leukaemia, ARTs can also inhibit angiogenesis in other haematologic malignancies. Wu, et al[51] investigated the effect of DHA on multiple myeloma RPMI8226 cell lines. They found that DHA can inhibit angiogenesis induced by RPMI8226 cells under hypoxic conditions. Besides, DHA downregulated the expression of VEGF and significantly reduced its secretion, thereby reducing the generation of blood vessels. Chen, et al[52] evaluated the effect of artesunate on the angiogenesis induced by RPMI8226 cells. They found that artesunate can efficiently suppress the angiogenic ability of myeloma RPMI8226 in a dose-dependent manner. Further investigations showed that artesunate can block ERK1/2 activation, downregulate VEGF and ang-1 expression, and inhibit angiogenesis induced by human multiple myeloma RPMI8226 cells. All of these studies showed that ARTs can play a role in the treatment of haematologic malignancies by inhibiting abnormal angiogenesis.

Production of ROS

ARTs mediate antimalarial activity by generating ROS in the parasite facilitated by intraparasitic haem iron, which in turn induces DNA damage and death in the malaria parasite.[53] Indeed, ROS induction is also critical to the activity of ARTs against haematological malignancies. Kumar, et al[54] proved that both cellular and mitochondrial ROS in AML cells were induced by artesunate exposure, suggesting that the cytotoxicity of artesunate against AML is mediated by the induction of ROS. Wang, et al[55] also found that Jurkat cells (a T-cell lymphoma cell line) were sensitive to DHA treatment, which increased the production of intracellular ROS and further led to cell cycle arrest and apoptosis. Xenofon, et al[56] have found that the anti-multiple myeloma effects of artesunate are related to intracellular levels of bivalent iron, suggesting that the effects of artesunate are through iron, which further have many other effects, such as dysfunction of mitochondria, generation of ROS, and non-caspase-mediated apoptosis. They have also examined the effect of artesunate on mitochondrial membrane potential and ROS status and the effect of iron on artesunate's anti-MM efficacy. Their results proved that the antimyeloma activity of artesunate is carried out via affecting the mitochondria and the ROS status of myeloma cells. Additionally, its efficacy is dependent on intracellular bivalent iron levels.[57]

Induction of Differentiation

Differentiation block is a characteristic of leukaemia, and thus, induction of differentiation is a way to treat leukaemia differentiation. Studies have shown the effectiveness of ARTs in the induction of leukaemia cells. Bo, et al[58] found that artemisinin at a dose of 6 μmol/L can induce the apoptosis of U937 cells, while at 5 μmol/L it efficiently promotes the differentiation of U937 cells to mature granulocytes, suggesting that induction of differentiation plays an important role in the anti-leukaemia effects of ARTs. Kim, et al[59] found that artemisinin markedly increased the degree of HL-60 leukaemia cell differentiation when simultaneously combined with low doses of

1α, 25-dihydoxyvitamin D3 [1,25-$(OH)_2D_3$] or all-trans retinoic acid (all-trans RA). They also found that the combination of IFN-α and ART synergistically induced the leukaemia cell differentiation, although IFN-α alone did not affect cell proliferation and differentiation.[60]

Action on Drug-Resistant Hematological Malignancies

Although novel drugs have contributed immensely to improve the outcomes of patients with hematological malignancies, many patients develop drug resistance and ultimately die. It is especially important to develop novel anti-hematological malignancy drugs that would not lead to developing drug resistance. Indeed, ARTs are a potential set of drugs that could fill this role. Efferth, et al[61] tested 22 drugs for their activity towards sensitive and multidrug resistance 1 (MDR-1)- or multidrug resistance-related protein 1 (MRP1)-overexpressing multidrug-resistant human CCRF-CEM leukaemia cells. They found that artesunate and bufalin exhibit anti-leukaemia activity when used alone or in combination with daunorubicin in MDR. The two drugs may be suitable for a new combination of therapies to improve leukaemia cell killing. They also investigated the effects of artesunate on the doxorubicin-resistant leukaemia cell line and found that artesunate induces apoptosis in leukaemic T cells mainly through the mitochondrial pathway via generation of ROS.[25] Xenofon, et al[56] have shown that artesunate reliably induces cell death of drug-resistant multiple myeloma cells at a safe concentrations shown to be used in humans.

Cooperative Therapy

The current standard of care for cancer is the use of combination chemotherapy with the goal of maximizing efficacy and systemically minimizing toxicity. Unlike traditional chemotherapeutics, the multiple mechanisms of action of ARTs suggest a possible synergism between ARTs and chemotherapy. A previous study showed that low levels of artesunate or arsenic trioxide had no impact on cell growth. However, the combination of these two drugs, even at low dosage, drastically induced apoptotic and necrotic cell death, implying a synergistic effect of these two drugs.[62] Budhrjara, et al[63] used mouse and human leukaemic cell lines and primary patient-derived xenografts to test the effects of DHA and ABT-263 on survival. They found that the therapeutic response in BCR/ABL(+) B-ALL can be improved by combining DHA with ABT-263. Upon investigation of its mechanisms, they found DHA triggered a cellular stress response and repressed translation, ultimately causing downregulation of MCL-1 expression. Consequently, this caused leukaemia cells to be highly sensitive to ABT-263-induced synergistic cell death, both *in vitro* and *in vivo*. Kim, et al[64] demonstrated that IFN-α combined with artemisinin can not only inhibit the growth of HL-60 leukaemia cells but also increase differentiation. These effects were significantly suppressed by inhibitors of protein kinase C (PKC), extracellular signal-regulated kinase (ERK), and c-Jun N-terminal kinase (JNK). These results indicate that activation of the PKC α/ERK signalling pathway by IFN-α can enhance artemisinin-induced differentiation in HL-60 cells. This evidence suggests that the anti-leukaemia effects of artemisinin can be enhanced by acting synergistically with other drugs.

Synergistical effects of ARTs with other drugs are also observed in other haematological malignancies. Sieber, et al[65] used the anti-CD20 antibody rituximab and artesunate to treat malignant B cells, and a synergistic effect was observed. They found that artesunate induced apoptosis and Fas/CD95 expression, and resulted in a breakdown of mitochondrial membrane potential. This demonstrated that the receptor-driven extrinsic and mitochondria intrinsic routes are involved in cell apoptosis. Rituximab increased Fas/CD95 expression and decreased mitochondrial membrane potential, eventually resulting in an increase in the rate of apoptosis. This implies that rituximab may affect molecular mechanisms relevant for the action of artesunate. They have a synergistic effect, which might be explained by an interaction of both agents at the level of apoptosis or upstream. Future clinical trials will be anticipated, allowing further evaluation of combination therapy in relapsed and refractory hematological malignancies.

Conclusions and Perspectives

ARTs, known as effective antimalarial drugs, have excellent safety in clinical applications. There are already a large number of studies that have applied ARTs to various types of tumours treatment. This review summarizes the recent progress of ARTs in the study of hematological malignancies. Various studies have shown that ARTs can act via several mechanisms, inducing cell cycle arrest, induction autophagy and apoptosis, inhibition of angiogenesis, production of

ROS, and induction of differentiation. Additionally, ARTs also showed anticancer effects in many drug-resistant hematological malignancies and synergistic effects with other drugs. At present, clinical trials of patients with solid tumours have been completed, and the results have been published, while ongoing and unpublished clinical trials continue to be run. However, clinical trials currently registered on ClinicalTrialsgov have not yet been targeted at hematological malignancies; this will be the direction of our further efforts. After years of research, ARTs may become a new promising anti-hematological malignancy drug.

Conflict of Interests

The authors declare that they have no competing interests.

Authors Contributions

Li Y was responsible for reviewing the literature, summarizing data and was a major contributor in writing the manuscript. Shan NN and Sui XH conceptualized and developed an outline for the manuscript. All authors read and approved the final manuscript for publication.

REFERENCES

1. Swerdlow SH, Campo E, Pileri SA, Harris NL, Stein H, Siebert R, et al. The 2016 revision of the World Health Organization classification of lymphoid neoplasms. Blood 2016;127:2375-2390.
2. Lu A, Chen K. Integrative medicine in clinical practice: from pattern differentiation in traditional Chinese medicine to disease treatment. Chin J Integr Med 2009;15:152-152.
3. Tshabalalamsimang M. Guidelines for the treatment of malaria. World Health Organization 2008;6:632-646. http://www.indiaenvironmentportal.org.in/files/WHO_Mar2010.pdf
4. Adam I, Ibrahim Y, Gasim GI. Efficacy and safety of artemisinin-based combination therapy for uncomplicated Plasmodium falciparum malaria in Sudan: a systematic review and meta-analysis. Malar J 2018;17:110-117.
5. Wang W, Li HJ, Qu GL, Xing YT, Yang ZK, Dai JR, et al. Is there a reduced sensitivity of dihydroartemisinin against praziquantel-resistant *Schistosoma japonicum*? Parasitol Res 2014;113:223-228.
6. Wang W, Li TY, Ji Y, Qu GL, Qian YL, Li HJ, et al. Efficacy of artemether and artesunate in mice infected with praziquantel non-susceptible isolate of *Schistosoma japonicum*. Parasitol Res 2014;113:925-931.
7. Efferth T. Beyond malaria: the inhibition of viruses by artemisinin-type compounds. Biotechnol Adv 2018; 36:1730-1737.
8. Efferth T, Marschall M, Wang X, Huong SM, Hauber I, Olbrich A, et al. Antiviral activity of artesunate towards wild-type, recombinant, and ganciclovir-resistant human cytomegaloviruses. J Mol Med (Berl) 2002;80:233-242.
9. Juteau F, Masotti V, Bessière JM, Dherbomez M, Viano J. Antibacterial and antioxidant activities of *Artemisia annua* essential oil. Fitoterapia 2002;73:532-535.
10. Ćavar S, Maksimović M, Vidic D, Parić A. Chemical composition and antioxidant and antimicrobial activity of essential oil of *Artemisia annua* L. from Bosnia. Ind Crops Prod 2012;37:479-485.
11. Algamal MA, Marei GIK, Saad MM, Abdelgaleil SA. Antimicrobial and phytotoxic properties of artemisinin and related derivatives. World Appl Sci J 2013;28:1382-1388.
12. Efferth T, Dunstan HA, Miyachi H, Chitambar C. The anti-malarial artesunate is also active against cancer. Int J Oncol 2001;18:767-773.
13. Paiboon R, Samlee M. Modulation of multidrug resistance by artemisinin, artesunate and dihydroartemisinin in K562/ADR and GLC4/ADR resistant cell lines. Biol Pharm Bull 2002;25:1555-1561.
14. Efferth T. Molecular pharmacology and pharmacogenomics of artemisinin and its derivatives in cancer cells. Curr Drug Targets 2006;7:407-421.
15. Junmei H, Disong W, Ruiwen Z, Hui W. Experimental therapy of hepatoma with artemisinin and its derivatives: *in vitro* and *in vivo* activity, chemosensitization, and mechanisms of action. Clin Cancer Res 2008;14:5519.
16. Nam W, Tak J, Ryu JK, Jung M, Yook JI, Kim HJ, et al. Effects of artemisinin and its derivatives on growth inhibition and apoptosis of oral cancer cells. Head Neck 2007;29:335-340.
17. Oranuch T, Pornthip W, Hiroaki S, Ikuo S. Artesunate enhances TRAIL-induced apoptosis in human cervical carcinoma cells through inhibition of the NF-κB and PI3K/Akt signaling pathways. Int J Oncol 2011;39:279-285.
18. Liu L, Zuo LF, Zuo J, Wang J. Artesunate induces apoptosis and inhibits growth of Eca109 and Ec9706 human esophageal cancer cell lines *in vitro* and *in vivo*. Mol Med Rep 2015;12:1465-1472.
19. Zhou X, Sun WJ, Wang WM, Chen K, Zheng JH, Lu MD, et al. Artesunate inhibits the growth of gastric cancer cells through the mechanism of promoting oncosis both *in vitro* and *in vivo*. Anticancer Drugs 2013;24:920-927.
20. Klayman DL. Qinghaosu (artemisinin): an antimalarial drug from China. Science 1985;228:1049-1055.
21. Agtmael MAV, Eggelte TA, Boxtel CJV. Artemisinin drugs in the treatment of malaria: from medicinal herb to registered medication. Trends Pharmacol Sci 1999;20:199.
22. Tu Y. The discovery of artemisinin (qinghaosu) and gifts from Chinese medicine. Nat Med 2011;17:1217.

23. Tu Y. Artemisinin—a gift from traditional Chinese medicine to the world (Nobel Lecture). Angewandte Chemie 2016;47:10210-10226.
24. Kong LY, Tan RX. Artemisinin, a miracle of traditional Chinese medicine. Nat Prod Rep 2015;32:1617.
25. Efferth T, Giaisi M, Merling A, Krammer PH, Li-Weber M. Artesunate induces ROS-mediated apoptosis in doxorubicin-resistant T leukemia cells. PLoS One 2007;2:e693.
26. Hou L, Block KE, Huang H. Artesunate abolishes germinal center B cells and inhibits autoimmune arthritis. PLoS One 2014;9:e104762.
27. Chen PQ, Li GQ, Guo XB, He KR, Fu YX, Fu LC, et al. The infectivity of gametocytes of *Plasmodium falciparum* from patients treated with artemisinin. Chin Med J (Engl) 1994;107:709-711.
28. De Vries PJ, Dien T K. Clinical pharmacology and therapeutic potential of artemisinin and its derivatives in the treatment of malaria. Drugs 1996;52:818-836.
29. Wang Z, Hu W, Zhang JL, Wu XH, Zhou HJ. Dihydroartemisinin induces autophagy and inhibits the growth of iron-loaded human myeloid leukemia K562 cells via ROS toxicity. FEBS Open Bio 2012;2:103-112.
30. Kim C, Lee JH, Kim SH, Sethi G, Ahn KS. Artesunate suppresses tumor growth and induces apoptosis through the modulation of multiple oncogenic cascades in a chronic myeloid leukemia xenograft mouse model. Oncotarget 2015;6:4020.
31. Sun YM, Zhao LZ, Li Q, Zhong Y, Li Y. The effects of cell proliferation and apoptosis induced by artesunate on leukemia cell. Chin J Gerontol (Chin) 2015;35:6647-6648.
32. Zhao X, Guo X, Yue W, Wang J, Yang J, Chen J. Artemether suppresses cell proliferation and induces apoptosis in diffuse large B cell lymphoma cells. Exp Ther Med 2017;14:4083-4090.
33. Toril H, Olsen OE, Kristine M, Hanne H, Anders W, Torstein BR, et al. Lymphoma and myeloma cells are highly sensitive to growth arrest and apoptosis induced by artesunate. Eur J Haematol 2013;91:339-346.
34. Li S, Xue F, Cheng Z, Yang X, Wang S, Geng F, et al. Effect of artesunate on inhibiting proliferation and inducing apoptosis of SP2/0 myeloma cells through affecting NFkappaB p65. Int J Hematol 2009;90:513-521.
35. Wang Y, Xu X, Wu X, Chen W, Huang F, Gui X. Dihydroartemisinin treatment of multiple myeloma cells causes activation of c-Jun leading to cell apoptosis. Oncol Lett 2018;15:2562-2566.
36. Gao N, Budhraja A, Cheng S, Liu EH, Huang C, Chen J, et al. Interruption of the MEK/ERK signaling cascade promotes dihydroartemisinin-induced apoptosis *in vitro* and *in vivo*. Apoptosis 2011;16:511-523.
37. Zhou HJ, Wang Z, Li A. Dihydroartemisinin induces apoptosis in human leukemia cells HL60 via downregulation of transferrin receptor expression. Anticancer Drugs 2008;19:247-255.
38. Lu JJ, Meng LH, Cai YJ, Chen Q, Tong LJ, Lin LP, et al. Dihydroartemisinin induces apoptosis in HL-60 leukemia cells dependent of iron and p38 mitogen-activated protein kinase activation but independent of reactive oxygen species. Cancer Biol Ther 2008;7:1017-1023.
39. Handrick R, Ontikatze T, Bauer KD, Freier F, Rubel A, Durig J, et al. Dihydroartemisinin induces apoptosis by a Bak-dependent intrinsic pathway. Mol Cancer Ther 2010;9:2497-2510.
40. Zeng Y, Ni X, Meng WT, Wen Q, Jia YQ. Inhibitive effect of artesunate on human lymphoblastic leukemia/lymphoma cells. J Sichuan Univ (Med Sci, Chin) 2009;40:1038-1043.
41. Vatsveen TK, Myhre MR, Steen CB, Walchli S, Lingjaerde OC, Bai B, et al. Artesunate shows potent anti-tumor activity in B-cell lymphoma. J Hematol Oncol 2018;11:23.
42. Cheng C, Wang T, Song Z, Peng L, Gao M, Hermine O, et al. Induction of autophagy and autophagy-dependent apoptosis in diffuse large B-cell lymphoma by a new antimalarial artemisinin derivative, SM1044. Cancer Med 2018;7:380-396.
43. Holien T, Olsen OE, Misund K, Hella H, Waage A, Ro TB, et al. Lymphoma and myeloma cells are highly sensitive to growth arrest and apoptosis induced by artesunate. Eur J Haematol 2013;91:339-346.
44. Papanikolaou X, Johnson S, Garg T, Tian E, Tytarenko R, Zhang Q, et al. Artesunate overcomes drug resistance in multiple myeloma by inducing mitochondrial stress and non-caspase apoptosis. Oncotarget 2014;5:4118-4128.
45. Markovic O, Marisavljevic D, Cemerikic V, Vidovic A, Perunicic M, Todorovic M, et al. Expression of VEGF and microvessel density in patients with multiple myeloma: clinical and prognostic significance. Med Oncol 2008;25:451-457.
46. Rajkumar SV, Fonseca R, Witzig TE, Gertz MA, Greipp PR. Bone marrow angiogenesis in patients achieving complete response after stem cell transplantation for multiple myeloma. Leukemias 1999;13:469-472.
47. Perez-Atayde AR, Sallan SE, Tedrow U, Connors S, Allred E, Folkman J. Spectrum of tumor angiogenesis in the bone marrow of children with acute lymphoblastic leukemia. Am J Pathol 1997;150:815-821.
48. Rajkumar SV, Witzig TE. A review of angiogenesis and antiangiogenic therapy with thalidomide in multiple myeloma. Cancer Treatment Rev 2000;26:351-362.
49. Zhou HJ, Wang WQ, Wu GD, Lee J, Li A. Artesunate inhibits angiogenesis and downregulates vascular endothelial growth factor expression in chronic myeloid leukemia K562 cells. Vascul Pharmacol 2007;47:131-138.
50. Lee J, Zhou HJ, Wu XH. Dihydroartemisinin downregulates vascular endothelial growth factor expression and induces

apoptosis in chronic myeloid leukemia K562 cells. Cancer Chemother Pharmacol 2006;57:213-220.
51. Wu XH, Zhou HJ, Lee J. Dihydroartemisinin inhibits angiogenesis induced by multiple myeloma RPMI8226 cells under hypoxic conditions via downregulation of vascular endothelial growth factor expression and suppression of vascular endothelial growth factor secretion. Anticancer Drugs 2006;17:839-848.
52. Chen H, Shi L, Yang X, Li S, Guo X, Pan L. Artesunate inhibiting angiogenesis induced by human myeloma RPMI8226 cells. Int J Hematol 2010;92:587-597.
53. Gopalakrishnan AM, Nirbhay K. Antimalarial action of artesunate involves DNA damage mediated by reactive oxygen species. Antimicrob Agents Chemother 2015;59:317-325.
54. Kumar B, Kalvala A, Chu S, Rosen S, Forman SJ, Marcucci G, et al. Antileukemic activity and cellular effects of the antimalarial agent artesunate in acute myeloid leukemia. Leuk Res 2017;59:124-135.
55. Wang Q, Wu S, Zhao X, Zhao C, Zhao H, Huo L. Mechanisms of dihydroartemisinin and dihydroartemisinin/holotransferrin cytotoxicity in T-cell lymphoma cells. PLoS One 2015;10:e0137331.
56. Xenofon P, Sarah J, Tarun G, Erming T, Ruslana T, Qing Z, et al. Artesunate overcomes drug resistance in multiple myeloma by inducing mitochondrial stress and non-caspase apoptosis. Oncotarget 2014;5:4118-4128.
57. Johnson SK, Garg TK, Tian E, Tytarenko R, Barlogie B, Epstein J, et al. The antimalarial agent artesunate exerts its antimyeloma activity by affecting the mitochondria and the reactive oxygen status of the myeloma cells and its efficacy depends on intracellular bivalent iron levels. Blood 2013;122:4444.
58. Bo J, Wang W, Wang Q, LI H, Zhao Y, Wu X, et al. Effects of artemisinin on apoptosis and differentiation of human leukemia U937 cells. J Fourth Milit Med Univ (Chin) 2008;29:634-637.
59. Kim SH, Kim HJ, Kim TS. Differential involvement of protein kinase C in human promyelocytic leukemia cell differentiation enhanced by artemisinin. Eur J Pharmacol 2003;482:67-76.
60. Kim SH, Chun SY, Kim TS. Interferon-α enhances artemisinin-induced differentiation of HL-60 leukemia cells via a PKCα/ERK pathway. Eur J Pharmacol 2008;587:65-72.
61. Efferth T, Davey M, Olbrich A, Rucker G, Gebhart E, Davey R. Activity of drugs from traditional Chinese medicine toward sensitive and MDR1- or MRP1-overexpressing multidrug-resistant human CCRF-CEM leukemia cells. Blood Cells Mol Dis 2002;28:160-168.
62. Rubini G, Altini C, Notaristefano A, Merenda N, Rubini D, Ianora AAS, et al. Role of 18F-FDG PET/CT in diagnosing peritoneal carcinomatosis in the restaging of patient with ovarian cancer as compared to contrast enhanced CT and tumor marker CA-125. Rev Esp Med Nucl Imag Mol 2014;33:22-27.
63. Budhraja A, Turnis ME, Churchman ML, Kothari A, Yang X, Xu H, et al. Modulation of navitoclax sensitivity by dihydroartemisinin-mediated MCL-1 repression in BCR-ABL(+) B-lineage acute lymphoblastic leukemia. Clin Cancer Res 2017;23:7558-7568.
64. Kim SH, Chun SY, Kim TS. Interferon-alpha enhances artemisinin-induced differentiation of HL-60 leukemia cells via a PKC alpha/ERK pathway. Eur J Pharmacol 2008;587:65-72.
65. Sieber S, Gdynia G, Roth W, Bonavida B, Efferth T. Combination treatment of malignant B cells using the anti-CD20 antibody rituximab and the anti-malarial artesunate. Int J Oncol 2009;35:149-158.
66. Kim C, Lee JH, Kim SH, Sethi G, Ahn KS. Artesunate suppresses tumor growth and induces apoptosis through the modulation of multiple oncogenic cascades in a chronic myeloid leukemia xenograft mouse model. Oncotarget 2015;6:4020-4035.
67. Zhao X, Zhong H, Wang R, Liu D, Waxman S, Zhao L, et al. Dihydroartemisinin and its derivative induce apoptosis in acute myeloid leukemia through Noxa-mediated pathway requiring iron and endoperoxide moiety. Oncotarget 2015;6:5582-5596.
68. Kumar B, Kalvala A, Chu S, Rosen S, Forman SJ, Marcucci G, et al. Antileukemic activity and cellular effects of the antimalarial agent artesunate in acute myeloid leukemia. Leuk Res 2017;59:124-135.
69. Tan M, Rong Y, Su Q, Chen Y. Artesunate induces apoptosis via inhibition of STAT3 in THP-1 cells. Leuk Res 2017;62:98-103.
70. Cao JT, Mo HM, Wang Y, Zhao K, Zhang TT, Wang CQ, et al. Dihydroartemisinin-induced apoptosis in human acute monocytic leukemia cells. Oncol Lett 2018;15:3178-3184.
71. Gao JL, Ding XP, Li QJ, Xia ZL, Xia QJ. Effect of dihydroartemisinin on the expression of BCR/ABL fusion gene in leukemia K562 cells. Chin J Med Genet (Chin) 2012;29:19-22.
72. Lee J, Zhang G, Wu X, Xu F, Zhou J, Zhang X. Growth inhibitory effect of dihydroartemisinin on Bcr/Abl+ chronic myeloid leukemia K562 cells involve AKT, ERK and NF-kappaB modulation. J Cancer Res Clin Oncol 2012;138:2095-2102.
73. Wang ZC, Liu Y, Wang H, Han QK, Lu C. Research on the relationship between artesunate and Raji cell autophagy and apoptosis of Burkitt's lymphoma and its mechanism. Eur Rev Med Pharmacol Sci 2017;21:2238-2243.

(Accepted May 22, 2019; First Online February 11, 2020)
Edited by WANG Wei-xia