Seminars in Cancer Biology 46 (2017) 65–83

Contents lists available at ScienceDirect

Seminars in Cancer Biology

journal homepage: www.elsevier.com/locate/semcancer

Review

From ancient herb to modern drug: *Artemisia annua* and artemisinin for cancer therapy

Thomas Efferth*

Johannes Gutenberg University, Institute of Pharmacy and Biochemistry, Department of Pharmaceutical Biology, Staudinger Weg 5, 55128 Mainz, Germany

ARTICLE INFO

Article history:
Received 17 January 2017
Received in revised form 15 February 2017
Accepted 24 February 2017
Available online 28 February 2017

Keywords:
Artemisia annua
Artemisinin
Cancer
Chemotherapy
Drug repurposing
Qinhaosu
Malaria
Phytotherapy

ABSTRACT

Artemisia annua L. is used throughout Asia and Africa as tea and press juice to treat malaria and related symptomes (fever, chills). Its active ingredient, artemisinin (ARS), has been developed as antimalarial drug and is used worldwide. Interestingly, the bioactivity is not restricted to malaria treatment. We and others found that ARS-type drugs also reveal anticancer *in vitro* and *in vivo*. In this review, we give a systematic overview of the literature published over the past two decades until the end of 2016. Like other natural products, ARS acts in a multi-specific manner against tumors. The cellular response of ARS and its derivatives (dihydroartemisinin, artesunate, artemether, arteether) towards cancer cells include oxidative stress response by reactive oxygen species and nitric oxide, DNA damage and repair (base excision repair, homologous recombination, non-homologous end-joining), various cell death modes (apoptosis, autophagy, ferroptosis, necrosis, necroptosis, oncosis), inhibition of angiogenesis and tumor-related signal transduction pathways (*e.g.* Wnt/β-catenin pathway, AMPK pathway, metastatic pathways, and others) and signal transducers (NF-κB, MYC/MAX, AP-1, CREBP, mTOR *etc*). ARS-type drugs are at the stairways to the clinics. Several published case reports and pilot phase I/II trials indicate clinical anticancer activity of these compounds. Because of unexpected cases of hepatotoxicity, combinations of ARS-type drugs with complementary and alternative medicines are not recommended, until controlled clinical trials will prove the safety of non-approved combination treatments.

Abbreviations: ABCB6, ATP-binding cassette transporter B6; ABCG2, ATP binding cassette transporter G2; AIF, apoptosis inducing factor; AKT, *v-Akt* murine thymoma viral oncogene homologue; AMPK, AMP-activated protein kinase; Ang-1, angiotensin 1; ARE, arteether; ARM, artemether; ARS, artemisinin; ART, artesunate; ATF4, activating transcription factor 4; Bak, Bcl2 antagonist/killer 1; Bax, Bcl2-associated x protein, pro-apoptotic BH3-only Bcl-2 family member; Bcl-2, B-cell CLL/lymphoma 2; Bcl-xL, B-cell CLL/lymphoma-x long; BCR/ABL, breakpoint cluster region/Abl proto-oncogene; Bid, BH3-interacting domain death agonist; Bim, pro-apoptotic Bcl2-family member; BSO, buthionine sulfoximine; C/EBPβ, CCAAT/enhancer binding protein beta; CAM, chorioallantoic membrane; CD, cluster of differentiation; CDK, cyclin-dependent kinase; CHOP/DDIT, DNA damage-inducible transcript; CIP1/WAF1, CDK-interacting protein 1/wild-type p53-activated fragment 1; c-JUN, Jun proto-oncogene; COX2, cyclooxygenase 2; CREB, cyclic ATP responsive element binding protein; DHA, dihydroartesunate; DNA-PK, DNA-dependent protein kinase; DR5, death receptor 5; E2F1, E2F transcription factor 1; EA, ethacrynic acid; EGFR, epidermal growth factor receptor; EMT, epithelial to mesenchymal transition; EndoG, endonuclease G; ERK, extracellular signal-regulated kinase; FAK, focal adhesion kinase; FAS, Fas cell surface death receptor; Flt-1, Fms-related tyrosine kinase 1; GADD153, growth arrest and DNA damage-inducible 153; GRP78, glucose-regulated protein; GSK3 β, glycogen synthase kinase 3 beta; HIF-1α, hypoxia-inducible factor-1 α; HPV39, human papilloma virus 39; HR, homologous repair; hTERT, human telomerase reverse transcriptase; hTR, human telomerase; HUVEC, human umbilical vein endothelial cells; IFN, interferon; IL, interleukin; IκBβ, inhibitor of kappa B beta; JNK, c-Jun N-terminal kinase; KDR/flk-1, kinase insert domain receptor; LC3, microtubule-associated protein 1 light chain 3; MAPK, nitogen-activated protein kinase; MAX, MYC-associated factor X; Mcl-1, myeloid cell leukemia 1; MDM2, mouse double minute 2 homologue; MEK, also known as MAPKK, mitogen-activated protein kinase kinase; MMP, matrix metalloproteinase; MPNST, malignant peripheral nerve sheath tumor; mTOR, mammalian target of rapamycin; MYC, avian myelomastosis viral oncogene homologue; NAC, N-acetyl cysteine; NFκB, nuclear factor kappa B; NHEJ, non-homologous end-joining; NO, nitric oxide; NOXA, also known as PMA/P1, phorbol-12-myristate-13-acetate-induced protein 1; PARK7, Parkinson disease protein 7protein deglycase DJ-1; PARP, poly ADP ribose polymerase; PCNA, proliferating cell nuclear antigen; PGE2, prostaglandine E2; PI3-K, phospoinositide-3 kinase; PMA, phorbol-12-myristate-13-acetate; RAF, Ras-associated factor proto-oncogene; RAS, Rat sarcoma viral oncogene homologue; RKIP, Raf-1 kinase inhibitor protein; ROS, reactive oxygen species; SMAC/DIABLO, IAP-binding mitochondrial protein; TCTP, translationally controlled tumor protein; TF, transferrin; TFRC, transferrin receptor 1 gene; TGB1, triple gene block protein β; TGF-β, tumor growth factor beta; TIMP, tissue inhibitor of metalloproteinase; TNF-α, tumor necrosis factor α; TOPO2A, DNA topoisomerase 2 α; TRAIL, tumor necrosis factor-related apoptosis-inducing ligand; Treg, regulatory T cells; VDAC2, voltage-dependent anion channel 2; VEGF, vascular endothelial growth factor; VEGFR, vascular endothelial growth factor receptor; XIAP, X-linked inhibitor of apoptosis; YY1, yin yang.

* Corresponding author.
E-mail address: efferth@uni-mainz.de

http://dx.doi.org/10.1016/j.semcancer.2017.02.009
1044-579X/

Contents

1. Introduction

Artemisinin is a 1,2,-trioxane from the Chinese medicinal plant Sweet Wormwood (*Artemisia annua* L., Asteraceae). The plant was first mentioned by Hong Ge (葛洪, (281–340 B.C.) as remedy to treat fever and chills in the "Handbook of Prescriptions for Emergency Treatment" (*Hou Bei Ji Fang*, 肘后备急方). The fact that it was still listed in the "Compedium of Materia Medica" (*Ben Cao Gang Mu*, 本草纲目), by Li Shizen (李时珍) in the year 1596 and is even known nowadays may be taken as a clue for its usefulness and activity.

In 1967, China's former chairman Mao Zedong initiated a research project to search for malaria-active medicinal plants from Chinese medicine. During the Vietnam War, the Vietnamese government asked China for help, because numerous Vietnamese soldiers suffered from malaria. Among the 500 scholars, who screened traditional Chinese plants and remedies, was Tu Youyou. She observed that *A. annua* was among the most active herbs. However, her results were not always repeatable with sufficient reliability. Going back to the ancient textbooks, Tu Youyou recognized that the recommended preparation of *A. annua* was not a hot decoction, as most frequently used standard procedure for medicinal herbs. Rather, the historical text described the use of a pressed juice of *A. annua*. Taking these details seriously, Tu Youyou then found that *A. annua* was more effective against *Plasmodia* infections, if she prepared low temperature extractions of this plant [1–3]. Bioactivity-guided fractionation subsequently allowed structure elucidation of sesquiterpene lactones of the artemisinin-type [2,4]. Together with its derivatives, artemisinin reached worldwide attraction, and artemisinin-based combination therapies nowadays belong to the established standard treatments of malaria worldwide [5–11]. As appreciation that artemisinin (ARS) helped to save millions of lifes, Tu Youyou was honored with numerous awards, including the Lasker DeBakey Clinical Research Award in 2011 and the Nobel Prize for Medicine or Physiology 2015 [12–15].

2. The antimalarial activity of artemisinin

In erythrocytes, *Plasmodium* trophozoites and schizonts feed on hemoglobin as the source for amino acids. Hemoglobin is toxic for *Plasmodia*, since heme-iron generates reactive oxygen species (ROS). Therefore, the malaria parasites convert hemoglobin to the non-toxic hemozoin [16,17]. During this reaction, the released heme-iron cleaves the endoperoxide bridge of ARS by a Fe(II) Fenton-type reaction, and free radical intermediates kill the *Plasmodia* [18–20]. Other mechanisms of the antimalarial activity of ARS include

- the inhibition of redox cycling,
- the inhibition of a glutathione S-transferase termed *Plasmodium falciparum* exported protein 1 (EXP1),
- the inhibition of *Plasmodium falciparum* PfATP6, which represents a sarcoendoplasmatic reticulum Ca^{2+} ATPase (SERCA),
- the inhibition of digestive vacuole cysteine protease, as well as
- the alkylation of specific parasite proteins, including translationally controlled tumor protein (TCTP) [21–26]

DNA lesions have not been observed in *Plasmodia*, in contrast to cancer cells [27].

3. Beyond malaria: activity of artemisinin to other diseases

Interestingly, numerous hints were accumulated during the past years, that activity of ARS is not restricted to malaria and that it may also be of therapeutic interest for several other diseases (Fig. 1). It was Tu Youyou, who provided first data that dihydroartemisinin may be beneficial for the treatment of Lupus erythematosus-related nephritis by inhibiting the production of anti-ds-DNA antibodies, the secretion of TNF-α, and NF-κB signaling pathway [28]. ARS-type drugs also revealed bioactivity towards viruses (*e.g.* human cytomegalovirus, HCMV), schistosomiasis, trypanosomiasis, cancer *in vitro* and *in vivo*, and even against plant tumors [29–37]. Resent results indicated that *A. annua* and ARS may not only be active against infectious and malignant diseases, but also to reduce glucose and act against diabetes mellitus [38,39].

Here, we present a timely review on the anti-cancer activity *in vitro* and *in vivo* of ARS and its derivatives artesunate (ART), artemether (ARM), arteether (ARE), as well as the first metabolite, dihydroartemisinin (DHA). Furthermore, we report on clinical data in cancer patients. Non-approved second generation derivatives, nanotherapeutic strategies with artemisinin-type compounds, as well as combination therapies involving ARS-type drugs were not included in this review. We searched the PubMed and Google Scholar databases with the following search term combinations: 'artemisinin/artesunate' and 'cancer' plus (1) '*in vivo*/xenograft/mice/rat', (2) 'cell cycle arrest', (3) 'reactive oxygen species/oxidative stress', (4) 'iron/transferrin', (5) 'DNA damage/DNA repair', (5) 'apoptosis/autophagy/necroptosis/ferroptosis', (6) 'angiogene-