Molecular Therapy
Oncolytics
Review

Defueling the cancer: ATP synthase as an emerging target in cancer therapy

Ting Wang,[1,3] Fei Ma,[2] and Hai-li Qian[1]

[1]State Key Laboratory of Molecular Oncology, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100021, China; [2]Department of Medical Oncology, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100021, China; [3]Key Laboratory of Carcinogenesis and Translational Research (Ministry of Education/Beijing), Laboratory of Molecular Oncology, Peking University Cancer Hospital & Institute, Beijing 100021, China

Reprogramming of cellular metabolism is a hallmark of cancer. Mitochondrial ATP synthase (MAS) produces most of the ATP that drives the cell. High expression of the MAS-composing proteins is found during cancer and is linked to a poor prognosis in glioblastoma, ovarian cancer, prostate cancer, breast cancer, and clear cell renal cell carcinoma. Cell surface-expressed ATP synthase, translocated from mitochondrion to cell membrane, involves the angiogenesis, tumorigenesis, and metastasis of cancer. ATP synthase has therefore been considered a therapeutic target. We review recent various ATP synthase inhibitors that suppress tumor growth and are being tested for the clinic.

INTRODUCTION

A century ago, Warburg reported that the metabolism in cancer has changed.[1] Reprogramming of metabolism is one of the hallmarks of cancer.[2] Drugs that target this reprogramming are emerging as novel drug families, and some are being evaluated in clinical trials (Figure 1).[3] The mitochondrion plays a key role in the metabolism reprogrammed in cancer.[3–5] For example, cancer cells without mitochondria can only form tumors if they reconstitute oxidative phosphorylation (OXPHOS) from host stroma.[6] The mitochondrion is also linked to the resistance of tumor stem cells to treatment and in pancreatic cancer to *KRAS*-driven signaling inhibition.[7–9]

Mitochondrial ATP synthase (MAS), an enzyme that produces ATP, also known as complex V, is the final and pivotal enzyme of OXPHOS in the mitochondrion.[10] In 1994, ectopically expressed ATP synthase (ectopic ATP synthase [EAS]) on the cell surface of vascular endothelial cells and tumor cells was found.[11] The existence of EAS is reportedly related to tumor angiogenesis, metastasis, and drug resistance.[12–15]

We review the ongoing development of ATP synthase inhibitors that have shown effectiveness in the treatment of cancer.[16]

THE STRUCTURE OF MAS

MAS is the primary generator of ATP and is widely found in the cell membranes of bacteria, the thylakoid membranes of chloroplasts in algae, and the inner membranes of the mitochondria of eukaryotic cells.[17] MAS transduces the electrochemical gradient of the proton motive force across these membranes into ATP, which is generated during respiration (Figure 2).

Its complex structure has been elucidated by electron microscopy.[18–20] The mammalian MAS consists of 28 subunits from 17 polypeptides, assembled into (1) a membrane-bound rotor and (2) a catalytic, ATP-generating center in the cytoplasm, linked by a central stalk and a peripheral stalk (PS).[18] Protons traverse the membrane inside the rotor, which combined with the PS is called the Fo region; the F1 region consists of the ATP-generating catalytic crown and its asymmetrical central stalk (Figure 3).

The rotor consists of a ring of eight c-subunits, clung by an ATP6 subunit. The PS part includes the subunits of the oligomycin sensitivity conferring protein (OSCP), b, d, and F6. Subunits e, f, g, ATP8, and 6.8 proteolipid are the accessory subunits of the PS.

The catalytic crown harbors three β subunits, the catalytic sites, and three noncatalytic α subunits. These β subunits can undergo three different conformations: (1) an open state with no nucleotide bound (E), (2) the loose state with ADP and Pi bound (D), and (3) the tight state with synthesized or hydrolyzed bound ATP (T). With the rotation of the c-ring, the conformation of catalytic sites alternates from E to T and D states. The asymmetrical stalk, composed of subunits γ, δ, and ε, connects the Fo inner membrane domain and the F1 catalytic domain.

The monomeric MASs dimerize via interaction between ATP6 subunits and between 6.8 proteolipids, and then the dimers are linked together by a DAPIT subunit along the edge of the cristae.[20,21]

https://doi.org/10.1016/j.omto.2021.08.015.

Correspondence: Hai-li Qian, PhD, State Key Laboratory of Molecular Oncology, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100021, China.
E-mail: qianhaili001@163.com
Correspondence: Fei Ma, PhD, Department of Medical Oncology, National Cancer Center/National Clinical Research Center for Cancer/Cancer Hospital, Chinese Academy of Medical Sciences and Peking Union Medical College, Beijing 100021, China.
E-mail: mafei2011@139.com

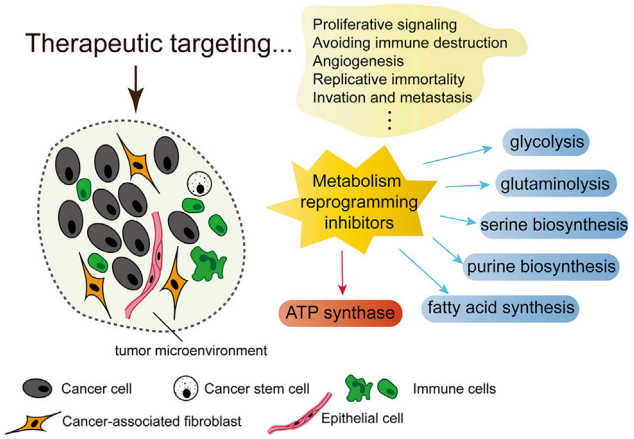

Figure 1. Different therapeutic strategies in cancer treatment
ATP synthase as a novel target among different therapeutic strategies of cancer.

ATP SYNTHASE IS REQUIRED FOR MALIGNANT TUMOR GROWTH

MAS in tumor

MAS relates to cancer through mutation of the genes coding for its subunits and also through abnormal expression of genes coding for these subunits.[14,22–25] In glioblastoma, the mRNA levels of the MAS F1 subunits α and β (*ATP5A1*, and *ATP5B*) are significantly higher in tumor cells and endothelial cell of microvascular proliferation, attributed to the downregulation of microRNAs (miRNAs) that target *ATP5A1* and *ATP5B*.[26]

ATP5B

In ovarian cancer, downregulated miR-450a that targets *ATP5B* expression increases metastasis, indicating that ATP5B might support the invasiveness of ovarian cancer.[27] In prostate cancer, breast cancer, and thyroid cancer,[28,29] overexpressed *ATP5B* negatively correlates with metastasis-free and overall survival. $HER2^+$ breast tumors that had acquired resistance to HER2-targeted therapies showed increased expression of *ATP5B* and ATP synthase PS subunit F6 (*ATP5J*).[26]

ATP5A1

In clear cell renal cell carcinoma and in breast cancer progression and metastasis, *ATP5A1*'s overexpression is associated with progression, a potential biomarker for diagnosis and prognosis and the treatment response.[30,31] In prostate cancer, ATP5A1 levels positively correlate with the early onset of tumor.[32] In colorectal cancer, high expression of *ATP5A1* is associated with SNPs, *TP53* mutation, and chromosomal instability to facilitate tumor development.[33] In hepatocellular cancer, *ATP5A1* also fell in the ingredient-target network of dihydroartemisinin and was responsible for the anti-hepatocellular carcinoma effect.[34] In lung cancer, periplocin inhibits growth of lung cancer by downregulating ATP5A1 and other key proteins.[35]

ATP level alteration also reflects the development of carcinogenesis,[36] marking the tumor region and its microenvironment.[37,38] Moreover, MAS is able to modulate the astrocyte inflammatory response, which is related to the initiation of glioma.[39,40]

Additionally, mutations of MAS-coding genes are accumulated with the carcinogenesis process, suggesting the participation of MAS in the biological process of cancer cells. Two mutations in *MT-ATP6*, which encodes subunit ATP6 of MAS, affect the calcium homeostasis and activation of the permeability transition pore (PTP) in yeast,[23] the latter of which is prominent for cancer cells to escape from apoptosis.

EAS in tumor

EAS is located not only on the mitochondrial inner membrane, but also on the plasma membrane of tumor cells and human vascular endothelial cells.[11,41] EAS is the receptor of angiostatin in endothelial cells, and it might be involved in maintaining the normal intracellular pH of cancer cells in an acidic extracellular microenvironment.[12] It is translocated from mitochondria to cell surface, and mitochondrial

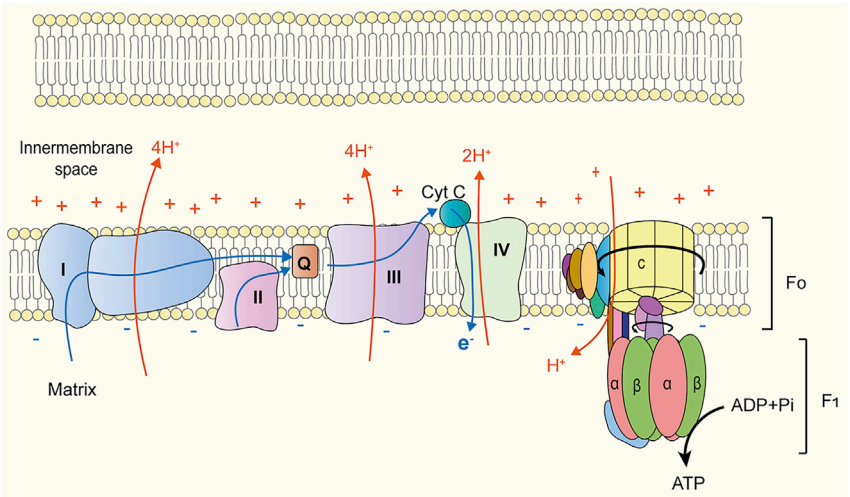

Figure 2. Schematic diagram of the how MAS works in mitochondrion
The respiratory chain builds up proton motive force, and ATP is produced by MAS. I, complex I; II, complex II; III, complex III; IV, complex IV; Q, coenzyme Q.

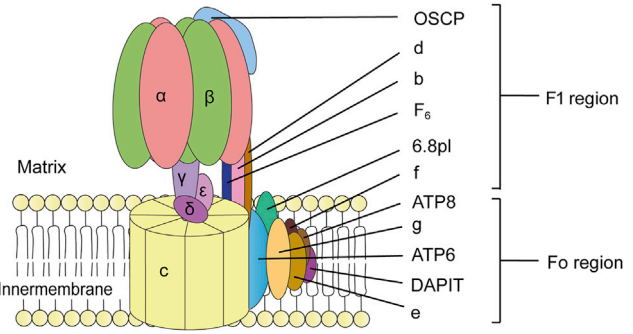

Figure 3. Current view of the structure of ATP synthase
Different subunits are shown by different colors, and their names are on the right linked by black lines.

transit peptide is essential for ATP synthase localization on the cell surface and mitochondria.[13] In hepatocellular carcinoma (HCC), blocking EAS could inhibit extracellular ATP synthesis and have anti-angiogenic and antitumorigenic activities.[14] In lung cancer, the level of EAS is correlated with gefitinib sensitivity and cell survival.[15] In leukemia, ATP5B of EAS on the human leukemia cell surface interacts with the platelet GPIIb epitope, causing platelet-cancer aggregation, with the latter serving as a target for a mimetic cytotoxic peptide to be specifically delivered to ectoATP5B-positive cells, contributing to targeted cancer cell killing.[42]

Inhibiting the activity of ATP synthase can effectively impair tumor metabolism reprogramming and tumor growth.[43] ATP synthase is essential for tumor progression and has potential as a target in cancer treatment.

CANCER THERAPY BASED ON TARGETING ATP SYNTHASE

Given the role of ATP synthase in fueling cancer, ATP synthase inhibitors have been explored as a new cancer therapy.[44,45] Categories that have been developed and investigated are polyketide inhibitors, polyphenolic phytochemicals, estrogens, polyenic α-pyrone derivatives, synthetic compounds, and protein peptides,[46,47] some of which were earlier used as antibiotics to treat infectious diseases or anti-obesity drugs and later showed antiangiogenetic and anticancer activity.[48–50]

AS inhibitor development is at the start of its debut in cancer treatment. None has become commercially available for clinical cancer treatment. Table 1 summarizes current inhibitors under investigation, sources, inhibitory sites, and related studies in tumor treatment (see Figure 4 and Table 1). Figure 5 displays the structure of AS inhibitors.

Polyketide inhibitors

Polyketides are bioactive natural products isolated from diverse microorganisms, polymers of two ketides synthesized by polyketide synthase.[104] Macrolides such as oligomycin, apoptolidin, and cytovaricin are polyketides and are MAS inhibitors with an anti-tumor activity.

Oligomycin

This compound, produced by *Streptomyces* strains, is a specific ATP synthase inhibitor that binds to the c subunit of Fo, as well at high concentrations to the F1 region,[51,105] inhibiting ATP synthesis. The subunit oligomycin sensitivity-conferring protein (OSCP) is required for MAS sensitivity to oligomycin.[106] Oligomycin can suppress cell viability in multiple cancers. It reduces the viability of hepatocellular carcinoma and non-small cell lung cancer cells with upregulated OXPHOS-like high-level SALL4 expression *in vitro* and *in vivo*.[52] In colon cancer and triple-negative breast carcinoma it decreases cell proliferation, migration capacity, and invasiveness, revealing the potential for therapy.[53,107] Oligomycin also significantly suppressed the survival and metastasis of microsatellite-stable colorectal cancer cells with increased mitochondrial DNA copy number.[108]

Moreover, oligomycin exhibits antitumor efficiency, particularly in tumors showing resistance to corresponding target therapies. It reverses the acquired resistance of $HER2^+$ breast tumors to HER2-targeted therapies and enhances the antitumor efficacy of c-MET inhibition in glioblastoma cells; it also prevents recurrence in ovarian cancer.[25,109,110] Inhibitors of OXPHOS, such as oligomycin, might be used for combined treatments.

Oligomycin, ossamycin, apoptolidin, and cytovaricin belong to the class of polyketide inhibitors of MAS with antifungal activities.[46] The former three macrolide inhibitors belong to the top 0.1% cytotoxic agents in 37,000 small molecules tested against 60 human cancer cell lines.[111]

Apoptolidin

This compound, including isoforms A–D, are microbial secondary metabolites that highly selectivity induce apoptosis in some cancer cell lines.[54,55,112] MAS is one of the biological targets for apoptolidins to exert antiproliferative activity.[113] In a study by Ishmael and colleagues,[114] apoptolidins A and C acted as AMPK activators and triggered autophagy in sensitive cell types without significant inhibition of mTORC1, showing that all macrolides do not function similarly.

Cytovaricin B

This compound was screened as an inhibitor of the JAK-STAT signaling pathway 20 years ago, and extended effects of this compound on tumor cells are being explored.[115]

Polyphenolic phytochemicals and estrogen

Polyphenolic phytochemicals are natural compounds from plants that contain phenol groups and show antitumor properties.[116] Many inhibit ATP synthase. The number of phenol groups and the position of the hydroxyl groups affect the strength of the inhibition.[117]

Resveratrol and piceatannol

These two polyphenolic compounds are naturally found in diverse plants, mainly such as grapes, berries, and white tea. They are phytoestrogens that possess antioxidant, anti-inflammatory, cardioprotective,

Table 1. ATP synthase inhibitors as anticancer drugs

Name	Source	Mode of action	Diseases	Research stage	Refs.
Polyketides					
Oligomycin	*Streptomyces*	binding to the Fo region	liver cancer, non-small cell lung cancer, colon cancer, triple-negative breast cancer, ovarian cancer	preclinical	51–53
Apoptolidin	*Nocardiopsis* spp.	binding to the Fo region	lung cancer	preclinical	54,55
Polyphenolics					
Resveratrol	grapes and berries	binding a hydrophobic pocket between the C-terminal region of the γ subunit and the hydrophobic inside of the β subunit	pheochromocytoma colon cancer	phase I in colon cancer (completed): ClinicalTrials.gov: NCT00256334, NCT00920803, and NCT00433576; phase I in unspecified adult solid tumor (completed): ClinicalTrials.gov: NCT00098969; phase II in multiple myeloma (terminated): Clincial Trials.gov: NCT00920556; phase I in polycystic ovary syndrome (terminated): ClinicalTrials.gov: NCT01489319	56–58
EGCG	green tea	inhibiting the ATP hydrolysis activity of ATP synthase	hepatocellular carcinoma, malignant pleural mesothelioma, colon cancer	early phase I in colon cancer (recruiting): ClinicalTrials.gov: NCT02891538; phase I & II in prostate cancer: ClinicalTrials.gov: NCT00459407 (completed) and NCT04300855 (recruiting); phase I & II in breast cancer: ClinicalTrials.gov: NCT00516243 (completed), NCT02580279 (unknown), and NCT04597359 (not yet recruiting); phase II in lung cancer (enrolling by invitation): ClinicalTrials.gov: NCT02577393; phase II in nonmetastatic bladder cancer (completed): ClinicalTrials.gov: NCT00666562.	59–61
Curcumin	the roots and stalks of Zingiberaceae	inhibiting the activity of ATP synthase	liver cancer, breast cancer, cervical carcinoma, prostatic cancer, lung cancer, leukemia, melanoma, colon cancer	phase II in breast cancer (completed): ClinicalTrials.gov: NCT01042938; phase III in prostate cancer (recruiting): ClinicalTrials.gov: NCT03769766; phase II in colorectal cancer (completed): ClinicalTrials.gov: NCT02439385, and so on	43,62–68
17β-estradiol	*Homo sapiens*	binding to the OSCP subunit of Fo region	liver tissue, rat brain	phase II in breast cancer: ClinicalTrials.gov: NCT01083641(terminated, has result), NCT00324259 (completed), and NCT01385280 (completed); phase II in prostate cancer (terminated, has result): ClinicalTrials.gov: NCT00459810; phase I in solid cancers (completed): ClinicalTrials.gov: NCT01209143; phase I in hematologic malignancies (recruiting): ClinicalTrials.gov: NCT03557619, and so on	69–74

(*Continued on next page*)

Table 1. *Continued*

Name	Source	Mode of action	Diseases	Research stage	Refs.
Genistein	Soybean	targeting polyphenol binding pocket of ATP synthase and blocking the rotation of the γ subunit	breast cancer	phase II in breast cancer (completed): ClinicalTrials.gov: NCT00244933 and NCT00290758; phase II & III in prostate cancer (completed): ClinicalTrials.gov: NCT00584532 and NCT01325311; phase I & II in non-small cell lung cancer (completed): ClinicalTrials.gov: NCT01628471 and NCT02567799; phase I & II in colon cancer (completed): ClinicalTrials.gov: NCT01985763; phase I in breast cancer & endometrial cancer (completed): ClinicalTrials.gov: NCT00099008; phase II in bladder cancer (recruiting): ClinicalTrials.gov: NCT01489813 and NCT00118040; phase II in pancreatic cancer (completed): ClinicalTrials.gov: NCT00376948; phase I & II in pediatric relapsed or refractory malignancies (completed): ClinicalTrials.gov: NCT02499861; early phase I in kidney cancer & melanoma (completed): ClinicalTrials.gov: NCT00276835; phase I in cancer of head and neck (completed): ClinicalTrials.gov: NCT02075112	75,76
Polyenic a-pyrones					
Citreoviridin	*Penicillium*, *Aspergillus*	binding to the β subunit of the F1 region	lung adenocarcinoma, breast cancer	preclinical	41,77–79
Aurovertin B	*Calcarisporium arbuscular*	binding to the β subunit of the F1 region	breast cancer, colon cancer	preclinical	79–81
Asteltoxin	*Aspergillus stellatus* Curzi and *Emericella variecolor*	binding to the β subunit of the F1 region	lung cancer, breast cancer	preclinical	82,83
Synthetics					
Bedaquiline	synthetic	binding to the c subunit rotor and subunit ε of the ATP synthase	tuberculosis, lung cancer, glioblastoma, breast cancer	preclinical	84–90
Gboxin	synthetic	its positive charge influences the proton gradient of the inner mitochondrial membrane	glioblastoma	preclinical	5
Pd(II) COS@GbA	synthetic	inducing mitochondrial fragmentation and disrupting ATP synthase action	prostate cancer	preclinical	91
Edelfosine	synthetic	dissipation of the mitochondrial membrane potential	cervix epithelioid carcinoma, hematopoietic cancer	preclinical	92–95
Miltefosine	synthetic	dissipation of the mitochondrial membrane potential	breast cancer	approved in the topical treatment of metastatic skin lesions in breast cancer	96
Perifosine	synthetic	dissipation of the mitochondrial membrane potential	advanced solid and leukemic tumors, brain tumors	phase I in advanced solid and leukemic tumors	97,98
Proteins					
IF1	*Homo sapiens*	interacting with the F1 domain at the catalytic surface between the α and β subunit of ATP synthase, blocking the rotation of the complex	bladder carcinoma, glioma, metastatic colon cancer, triple-negative breast cancer	preclinical	10,99–101

(*Continued on next page*)

Table 1. *Continued*

Name	Source	Mode of action	Diseases	Research stage	Refs.
Angiostatin	*Homo sapiens*	binding to α/β subunits of ATP synthase	colon carcinoma	phase I in unspecified adult solid tumor (completed): ClinicalTrials.gov: NCT00086723; phase II in non-small cell lung cancer (completed): ClinicalTrials.gov: NCT00049790	102
Hai178	humanized antibody	targeting ATP synthase	breast cancer, prostate cancer, lung cancer	preclinical	103

EGCG, epigallocatechin gallate; OSCP, oligomycin sensitivity conferring protein; Pd(II)COS@GbA, Pd(II) complex of Gboxin analog-chitooligosaccharides conjugate; IF1, ATP synthase inhibitory factor 1.

and anti-cancer properties.[56,57] Resveratrol and piceatannol provide a wide range of preventive and therapeutic options against different types of cancer.[57] They bind to a hydrophobic pocket between the hydrophobic C terminal region of the γ subunit and the hydrophobic inside of the β subunit.[46] In this way, resveratrol promotes human pheochromocytoma cell death.[58]

EGCG

One of the catechins, epigallocatechin gallate (EGCG), is abundant in green tea and inhibits ATP hydrolysis.[46] EGCG facilitates apoptosis as well as cell cycle arrest, growth inhibition in hepatocellular carcinoma, malignant pleural mesothelioma cells, and bladder urothelial cells.[59–61] EGCG can prevent various chronic diseases, including neurodegenerative disorders, through several molecular signaling pathways including MAS inhibition.[118]

Curcumin

This MAS inhibitor is a natural polyphenol extracted from the roots and stalks of Zingiberaceae *Curcuma longa*, an herbal medicine with antitumor activity.[62,119] It inhibits many cancers, including liver cancer, breast cancer, cervical carcinoma, prostatic cancer, and lung cancer. It affects several signaling pathways, with nuclear factor κB (NF-κB) signaling being prominently inhibited.[63–68] Recently MAS inhibition independent of NF-κB signaling was reported, which lowered ATP levels concomitantly with oxygen consumption, both *in vivo* and *in vitro*.

The energetic impairment by curcumin of L1210 murine lymphocytic leukemia, 4T1 murine breast, B16 murine melanoma, and CT26 murine colon tumor cell lines translated, also both *in vitro* and *in vivo*, into decreased tumor growth.[44] According to ClinicalTrials.gov, 69 clinical studies have been done on curcumin, with 28 completed clinical trials, including 14 phase II trials. Curcumin on its own or in combination has been applied in studies of primary or metastatic breast cancer, colorectal cancer, pancreatic cancer, and endometrial carcinoma.

17β-estradiol (E2)

This compound plays a well-known role in breast cancer, binding to the estrogen receptor. In liver cells it also binds to the OSCP protein of MAS, promoting uncoupling.[69–72,120]

Genistein

This isoflavone phytoestrogen from soybeans noncompetitively inhibits MAS. It targets the polyphenol binding pocket and blocks the rotation of the γ subunit.[75] Both genistein and estradiol induce

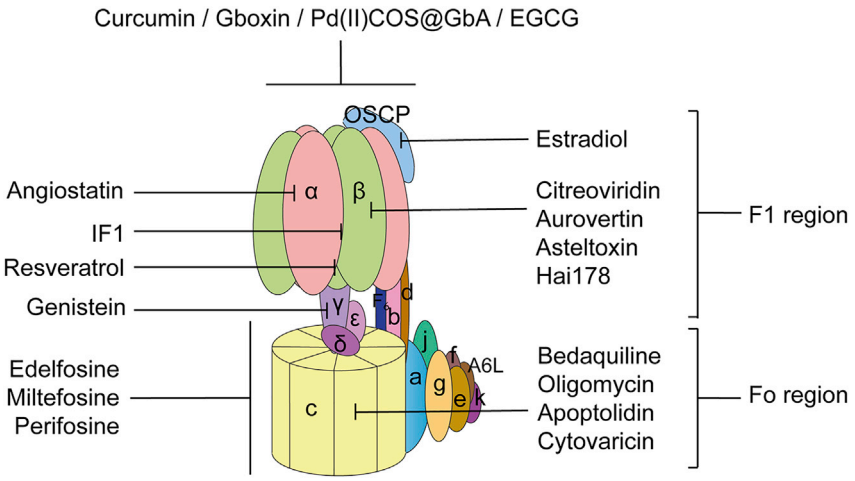

Figure 4. Inhibitory sites of ATP synthase
Different ATP synthase inhibitors targets different subunits, and the inhibitory sites are indicated by black lines.

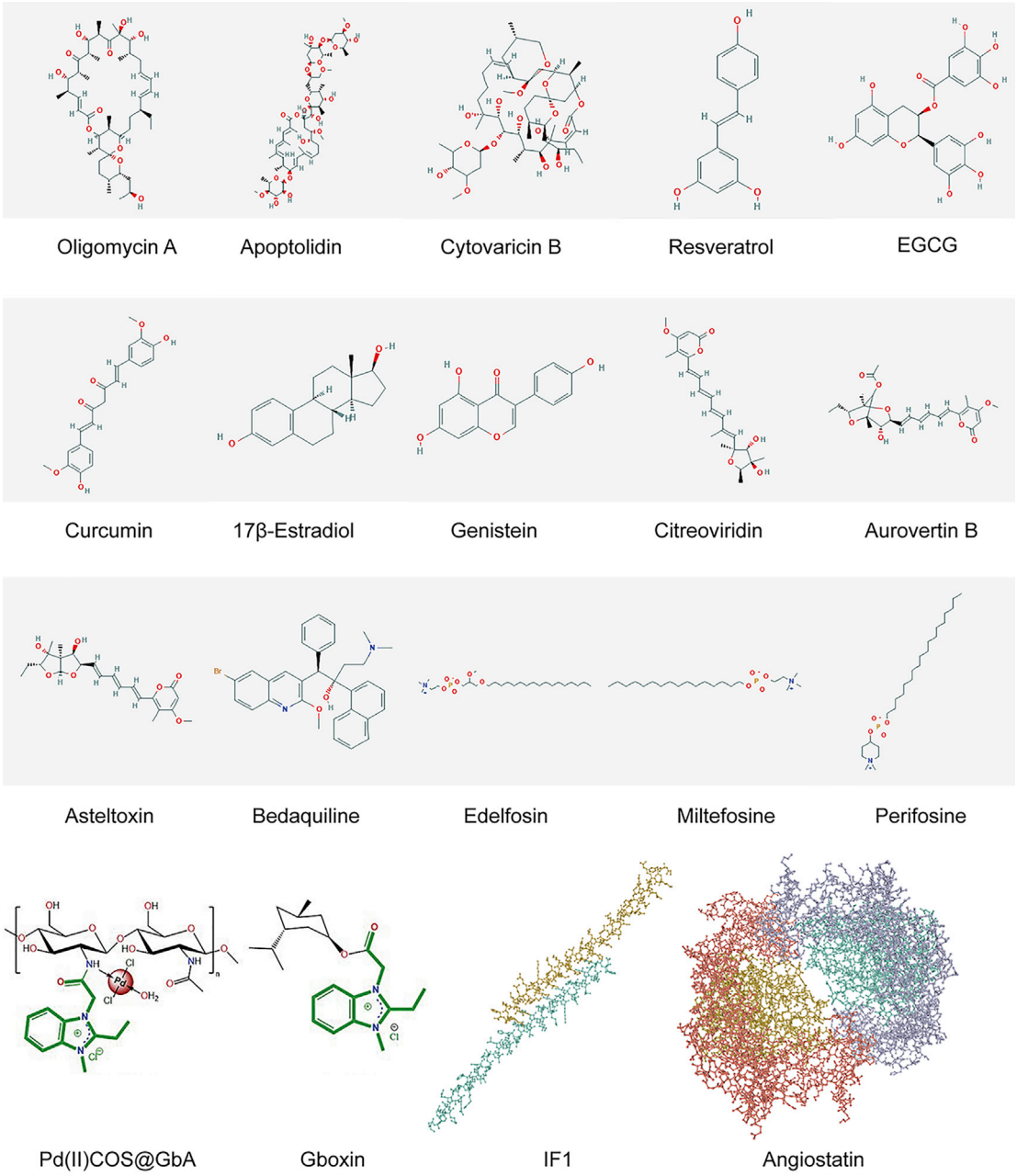

Figure 5. Structure of MAS inhibitors

EGCG, epigallocatechin gallate; Pd(II)COS@GbA, Pd(II) complex of Gboxin analog-chitooligosaccharides conjugate; IF1, ATP synthase inhibitory factor 1. The structures of inhibitors were obtained from the ChemSpider database.

cell proliferation and apoptosis inhibition, cell cycle arrest and mitochondrial function, and dynamics damage of breast cancer cell lines.[76]

At present, 353 clinical studies of estradiol have been published. 61 clinical trials have been completed with results, and three clinical trials (phase I and phase II) have been completed. The drug was applied in the treatment of estrogen receptor-positive breast cancer, recurrent breast cancer, or solid cancers (ClinicalTrials.gov: NCT00324259, NCT01385280, and NCT01209143).

Polyenic α-pyrones

A polyenic α-pyrone is a six-membered cyclic unsaturated ester present in many natural products. Citreoviridin, aurovertin B and

asteltoxin are all polyenic α-pyrone, and they target the β subunit of F1 to inhibit.[121]

Citreoviridin

This compound consists of an α-pyrone ring conjugated to a furan ring.[73] It inhibits MAS by binding to the β subunit of F1, inhibiting in turn the proliferation and growth of lung adenocarcinoma cell lines. The unfolded protein response is activated without affecting healthy cells.[41,77,122]

Aurovertin B

This naturally occurring antibiotic is isolated from *Calcarisporium arbuscular*. It also interacts with F1's β subunit and inhibits ATP synthesis.[80] Aurovertin B strongly inhibits the proliferation of several breast cancer cell lines by suppressing the enhanced ATP synthesis in breast carcinoma.[50] Aurovertin B, D, and E exhibit potent antiproliferative activity, particularly against triple-negative breast cancer *in vivo* and *in vitro*.[123,124] They also facilitate the recognition and lysis of colon cancer cells by natural killer (NK) cells, by a mechanism that may involve MAS inhibition.[81]

Asteltoxin

This α-pyrone-containing mycotoxin is produced in *Aspergillus stellatus* Curzi and *Emericella variecolor*. Asteltoxin and its three dimers exert inhibitory effects on the lung cancer cell line H1299 and breast cancer line MCF7.[82] Asteltoxin and asteltoxin B are moderately cytotoxic against several tumor cell lines.[83]

Synthetic compounds

In addition to the mentioned natural compounds, several synthetic MAS inhibitors have been developed for treating infectious diseases such as tuberculosis by inhibiting ATP synthase.[125] Some were developed using computational science, with some working against cancer.[5]

Bedaquiline

This compound, developed by Janssen Pharmaceuticals in 2005, was approved by the US Food and Drug Administration (FDA) in 2012 for treating multidrug-resistant tuberculosis.[125,126] It is a synthetic heterocyclic MAS synthase inhibitor that targets both the c-subunit rotor and subunit ε.[84] Recently, Wu et al.[85] suggested that bedaquiline blocks the growth, survival, and tumor angiogenesis of lung cancer, stem-like cancer cells, and glioblastoma.[86–88] Similar to other ATP synthase inhibitors, it diminishes mitochondrial oxygen consumption by depleting and blocking the proliferative expansion of breast cancer stem cells.[86]

Gboxin

This small molecule was found by a cell-based high-throughput chemical screen. It inhibits the growth of mouse and human glioblastoma cells.[5] A Pd(II) complex of a Gboxin analog-chitooligosaccharides conjugate (Pd(II)COS@GbA) was found to disrupt ATP synthase rotation, diminish ATP synthesis, and show anti-prostate cancer activity.[91]

Alkylphospholipid analogs (APLs) are developed as anticancer drugs, with amphiphilic structures similar to natural phospholipids, and they incorporate preferentially in the membranes of tumor cells.[127] Structurally related anti-tumor APLs include a number of clinically promising drugs, such as edelfosine, miltefosine, perifosine, and erucylphosphocholine.[92,96,97]

Edelfosine, with antileishmanial and anticancer activity, specifically accumulated in the mitochondria or the lipid raft of tumor cells, leading to dissipation of the mitochondrial membrane potential and reactive oxygen species (ROS) generation by ATP synthase inhibition and raft disruption, eventually inducing apoptosis-like cell death.[92,93]

Miltefosine, also belonging to the APLs family and trademarked as Miltex, is the only APL that has been approved as an antitumor drug in the topical treatment of metastatic skin lesions in breast cancer.[96]

Perifosine and erucylphosphocholine are the new molecules synthesized as miltefosine variants.[97] Perifosine has entered many phase I clinical trials in treating plenty of advanced solid and leukemic tumors, and many preclinical studies are being carried out with erucylphosphocholine in brain tumors.[98]

Peptide inhibitors

Several peptide inhibitors, including the endogenous MAS inhibitory factor 1 (IF1), angiostatin, and some chimeric antibodies, target MAS or ectopic ATP synthase, showing an anti-tumor activity in preclinical studies.

IF1

This protein interacts with the F1 domain when dephosphorylated at the catalytic surface between the α and β subunit of MAS, blocking the rotation of the complex and inhibiting the synthesis and hydrolysis of ATP.[99] Its phosphorylation status is regulated by 5′-AMP-activated protein kinase (AMPK). Increased cyclic AMP (cAMP) levels activate AMPK: the phosphorylation of S39 in IF1 and the activation of MAS thus facilitate ATP synthesis.[128] High expression of IF1 in non-small cell lung cancer, bladder carcinoma, and gliomas has a bad prognosis.[10] A recent study demonstrated that IF1 modulates the interplay between ROS and hypoxia in cancer cells, which might be associated with the cell survival supports, tumor progression, and anticancer drug resistance.[10] In contrast, a high expression level of IF1 in breast and colon cancer predicts a better prognosis, especially in triple-negative breast cancer and metastatic colorectal cancer patients.[10,100] Thus, the comprehensive role of IF1 in malignancy needs subtle stratification to help anticancer therapeutic strategies.

Angiostatin

This protein is an internal proteolytic fragment of plasminogen, dramatically inhibiting primary and metastatic tumor growth by blocking tumor angiogenesis.[129] EAS is relevant to angiogenesis and the direct target of angiostatin.[130] Angiostatin binds the α/β subunits of EAS, mediating the downregulation of endothelial cell proliferation

and migration, and these effects will be depleted by the presence of the anti-α-subunit ATP synthase antibody. Hence, inhibition of EAS has been suggested for the antiangiogenetic therapeutic strategy by blocking tumor angiogenesis.[130,131] Moreover, radiolabeled angiostatin may be a powerful tool for specific imaging or targeted radiotherapy of tumors due to the specific interaction.[102]

Other chimeric antibodies have been developed that target the β subunit of EAS, inhibiting its activity.[132] A strain of murine monoclonal antibody (mAb) mAb6F2C4 showed the angiostatin-like property to block extracellular ATP generation under extracellular acidic conditions and interrupt the angiogenetic process.[14] Another murine mAb targeting the same site, McAb178-5G10, showed antitumor activity in multiple cancers.[12,133] The humanized antibody Hai178 against the β subunit of EAS produced by the same team also had an anti-tumor effect in tumor xenografts,[103] which provides a possibility for clinical application. This evidence ensures the potential of antibodies blocking EAS activity in cancer treatment.

DISCUSSION

MAS is upregulated in many cancers, and evidence is accumulating that MAS may serve as a potent anticancer target, especially in glioblastoma, ovarian cancer, prostate cancer, breast cancer, and clear cell renal cell carcinoma.[26–30,36,134,135] However, there remain several challenges in translating preclinical MAS-targeting drugs to the clinic.

First, MAS universally exists in tumor cells and normal cells, therefore raising the concern about the potential of systematic toxicity to patients. According to ClinicalTrials.gov (https://www.clinicaltrials.gov/), the potential side effects caused by various MAS inhibitors are different. There are five MAS inhibitors that have reported clinical trial results. Resveratrol was safe and lacked serious adverse reactions, with mild gastrointestinal symptoms, including nausea, flatulence, abdominal discomfort, and diarrhea (ClinicalTrials.gov: NCT00098969). EGCG was well tolerated with minimal adverse events, including nausea, diarrhea, headache. These side effects were all grade 1 or 2 based on the National Cancer Institute (NCI) Common Terminology Criteria for Adverse Events (CTCAE) version 3.0 (ClinicalTrials.gov: NCT00666562). Curcumin was safe, and no adverse events occurred during the study (ClinicalTrials.gov: NCT01042938). The incidence rate of serious adverse events caused by using estradiol was 23.53%, which included abdominal pain, diarrhea, nausea, and vomiting. All patients reported mild adverse events in the study (ClinicalTrials.gov: NCT00324259). The incidence rate of serious adverse events by using genistein was 9.68%, including dyspnea and ulceration. 80.65% of the patients reported mild adverse events in this study (ClinicalTrials.gov: NCT00290758).

To overcome the shortcomings of potential toxicity, (1) antibody drug conjugates (ADCs) can be a strategy to enhance the specificity of MAS-targeting drugs. ADCs represent a new class of cancer-targeting drugs that endow the specificity to cytotoxic chemicals by conjugating them to cancer antigen-specific antibodies. Antibody-assisted drug delivery dramatically reduces the systematic toxicity to cancer patients (e.g., T-DM1). T-DM1 significantly improved overall survival of patients with previously treated $HER2^+$ metastatic breast cancer,[136] and the rates of adverse events of grade 3 or above were reduced from 57% to 41%.[137] (2) VDAC1 is a voltage-dependent anion channel protein that is a major component of the outer mitochondrial membrane, often overexpressed in many cancers.[138] Studies have shown that silencing VDAC1 expression in cancer cells leads to metabolic reprogramming, reducing the expression of ATP5F1A.[139–142] Therefore, silencing VDAC1 may decrease the level of ATP synthase without affecting healthy cells. (3) Another strategy to improve the efficacy/toxicity balance is to combine the energy supply blockers from MAS inhibitors with agents of other anti-cancer mechanisms. For instance, combination of the MAS inhibitor citreoviridin and the 26S proteasome inhibitor bortezomib improved anticancer activity in breast cancer cells.[78] Combinatory use of multi-mechanism anti-cancer drugs is the basic and mostly chosen regimen for cancers at advanced stages.

Patel et al.[47] provided an update on recently discovered ATP synthase modulators. A more comprehensive view of ATP synthase-targeting inhibitors was covered in their study. Because we have concentrated preferentially on the indications of ATP synthase-targeting inhibitors in cancer treatment, some natural inhibitors of the complex that have systemic toxicity or about which there is almost no relevant research in cancer treatment are not included in our review. We have elaborated the structure of the complex, detailed its significance in different types of malignant tumors, and reviewed the basic and clinical research progress of ATP synthase-targeting inhibitors in cancer treatment. A larger-scale and comprehensive screening of MAS inhibitors should be performed with the current high-throughput chemical screening technology to find more potential effective and specific anti-cancer drugs.

Second, the expression levels of the MAS-composing genes, the status of OXPHOS, and mitochondrial metabolism vary in different types of cancer.[143,144] High expression of ATP5B predicts better local recurrence-free survival in nasopharyngeal carcinoma.[134] In papillary thyroid cancer, *ATP5A1* mRNA expression levels are downregulated in tumorous tissue with more lymph node metastases.[145] Similarly, ATP5B is downregulated in breast, gastric, lung, and esophageal cancer.[135] Thus, it is necessary to measure the status and inhibitory contribution of MAS before using a MAS inhibitor on tumor or in combination with other anticancer drugs.

Additionally, the challenge faced to make the MAS a dominant antitumor target should be ideally resolved. Only a certain part of tumors, which are still under to-be-defined conditions, are potentially sensitive to MAS inhibitors. For instance, mitoxantrone significantly benefits the progression-free and overall survival of cancer patients. However, in metastatic breast cancer, the response rate of mitoxantrone is only 20%.[146] The expression of MAS was increased in mitoxantrone-resistant cells, so MAS might be a key target to reverse the resistance of metastatic breast cancer patients to this drug.[31] Therefore, in certain cancers, the value of this complex as a target to

suppress primary cancer or reverse the resistance of tumor cell to the existing treatment should be determined.

Third, MAS is mutated in some primary human cancers. A total of 19 genes are involved in encoding ATP synthase. Except for MT-ATP6 and MT-ATP8, which are mitochondrial genes, the others are nuclear genes. According to The Cancer Genome Atlas (TCGA) database, the variation frequencies of these genes in pan cancers range from 0.5% to 2%, including structure variation, amplification, deep deletion, and other point mutations. Although not many relevant results have been reported, the abnormality of MAS composing genes on the binding sites of ATP inhibitors in about 2% of tumor patients may lead to a less sensitivity to the drugs. Given that condition, we may consider the use of other inhibitors that target different sites of ATP synthase, or redesign the inhibitor to specifically bind the mutated ATP synthase, hence inhibiting the activity of the enzyme.

Fourth, metabolism reprogramming as a process that occurred in cancer cells influences not only the cancer cells but also the microenvironment.[147] For instance, lipid accumulation in myeloid-derived suppressor cells and tumor-associated macrophages, which may derive from adjacent cancer cells with enhanced fatty acid synthesis, has been verified to promote metabolic reprogramming and assisted these cells changing into immunosuppressive phenotypes.[148] Additionally, tumor cells consume more glucose, which results in a glucose limitation for T cells, leading to a defective antitumor response.[149] Metabolite imbalance aroused by enhanced MAS in tumor cells may provide an advantageous immune microenvironment for tumor progression. Interfering with the MAS-related metabolite deregulation may be a breakthrough auxiliary approach to phenotypically renormalize the tumors to a sluggish status.

Additionally, previous studies have shown that estrogen supported the growth of tumors by activating a series of signaling pathways, and sustained exposure to exogenous estrogen was a risk factor for various cancers.[150] Anti-estrogen therapies are important for cancers, especially for estrogen receptor-positive breast cancer.[151] However, the function of estrogen as an ATP synthase inhibitor suppressing the activity of MAS and inducing mitochondrial functional damage and cell apoptosis have been discussed in this review. The distribution of the binding of estrogen on (1) MAS and on (2) the estrogen receptor has in general not been determined in the past, and it should be measured in future studies. This binding should also be considered and measured when using endocrine therapy on cancers such as breast cancer, ovarian cancer, and endometrial cancers.

In conclusion, MAS-targeted drugs represent an emerging family of anti-cancer agents in active development and should be identified and tested in further clinical trials to improve cancer patient outcomes in the future.

ACKNOWLEDGMENTS

This work was supported by grants from the National Natural Science Foundation of China (nos. 81874122, 81872280, and 82073094), the Open Issue of State Key Laboratory of Molecular Oncology (no. SKL-KF-2017-16), and the Independent Issue of State Key Laboratory of Molecular Oncology (no. SKL-2017-16).

AUTHOR CONTRIBUTIONS

All listed authors contributed to the writing of this review. All authors read and provided critical revision of the manuscript and approved the final version.

DECLARATION OF INTERESTS

The authors declare no competing interest.

REFERENCES

1. Liberti, M.V., and Locasale, J.W. (2016). The Warburg effect: How does it benefit cancer cells? Trends Biochem. Sci *41*, 211–218.
2. Hanahan, D., and Weinberg, R.A. (2011). Hallmarks of cancer: The next generation. Cell *144*, 646–674.
3. Pavlova, N.N., and Thompson, C.B. (2016). The emerging hallmarks of cancer metabolism. Cell Metab *23*, 27–47.
4. Caro, P., Kishan, A.U., Norberg, E., Stanley, I.A., Chapuy, B., Ficarro, S.B., Polak, K., Tondera, D., Gounarides, J., Yin, H., et al. (2012). Metabolic signatures uncover distinct targets in molecular subsets of diffuse large B cell lymphoma. Cancer Cell *22*, 547–560.
5. Shi, Y., Lim, S.K., Liang, Q., Iyer, S.V., Wang, H.Y., Wang, Z., Xie, X., Sun, D., Chen, Y.J., Tabar, V., et al. (2019). Gboxin is an oxidative phosphorylation inhibitor that targets glioblastoma. Nature *567*, 341–346.
6. Bajzikova, M., Kovarova, J., Coelho, A.R., Boukalova, S., Oh, S., Rohlenova, K., Svec, D., Hubackova, S., Endaya, B., Judasova, K., et al. (2019). Reactivation of dihydroorotate dehydrogenase-driven pyrimidine biosynthesis restores tumor growth of respiration-deficient cancer cells. Cell Metab *29*, 399–416, e10.
7. Viale, A., and Draetta, G.F. (2016). Metabolic features of cancer treatment resistance. Recent Results Cancer Res. *207*, 135–156.
8. Bosc, C., Selak, M.A., and Sarry, J.E. (2017). Resistance is futile: Targeting mitochondrial energetics and metabolism to overcome drug resistance in cancer treatment. Cell Metab *26*, 705–707.
9. Viale, A., Pettazzoni, P., Lyssiotis, C.A., Ying, H., Sánchez, N., Marchesini, M., Carugo, A., Green, T., Seth, S., Giuliani, V., et al. (2014). Oncogene ablation-resistant pancreatic cancer cells depend on mitochondrial function. Nature *514*, 628–632.
10. Esparza-Moltó, P.B., and Cuezva, J.M. (2018). The role of mitochondrial H^+-ATP synthase in cancer. Front. Oncol. *8*, 53.
11. Das, B., Mondragon, M.O., Sadeghian, M., Hatcher, V.B., and Norin, A.J. (1994). A novel ligand in lymphocyte-mediated cytotoxicity: Expression of the beta subunit of H+ transporting ATP synthase on the surface of tumor cell lines. J. Exp. Med. *180*, 273–281.
12. Wang, W.J., Shi, X.X., Liu, Y.W., He, Y.Q., Wang, Y.Z., Yang, C.X., and Gao, F. (2013). The mechanism underlying the effects of the cell surface ATP synthase on the regulation of intracellular acidification during acidosis. J. Cell. Biochem *114*, 1695–1703.
13. Ma, Z., Cao, M., Liu, Y., He, Y., Wang, Y., Yang, C., Wang, W., Du, Y., Zhou, M., and Gao, F. (2010). Mitochondrial F1Fo-ATP synthase translocates to cell surface in hepatocytes and has high activity in tumor-like acidic and hypoxic environment. Acta Biochim. Biophys. Sin. (Shanghai) *42*, 530–537.
14. Wang, J., Han, Y., Liang, J., Cheng, X., Yan, L., Wang, Y., Liu, J., Luo, G., Chen, X., Zhao, L., et al. (2008). Effect of a novel inhibitory mAb against β-subunit of F1F0 ATPase on HCC. Cancer Biol. Ther. *7*, 1829–1835.
15. Chang, Y.W., Hsu, C.L., Tang, C.W., Chen, X.J., Huang, H.C., and Juan, H.F. (2020). Multiomics reveals ectopic ATP synthase blockade induces cancer cell death via a lncRNA-mediated phospho-signaling network. Mol. Cell. Proteomics *19*, 1805–1825.

16. Kim, M.S., Gernapudi, R., Cedeno, Y.C., Polster, B.M., Martinez, R., Shapiro, P., Kesari, S., Nurmemmedov, E., and Passaniti, A. (2020). Targeting breast cancer metabolism with a novel inhibitor of mitochondrial ATP synthesis. Oncotarget *11*, 3863–3885.

17. Okuno, D., Iino, R., and Noji, H. (2011). Rotation and structure of FoF1-ATP synthase. J. Biochem. *149*, 655–664.

18. Pinke, G., Zhou, L., and Sazanov, L.A. (2020). Cryo-EM structure of the entire mammalian F-type ATP synthase. Nat. Struct. Mol. Biol. *27*, 1077–1085.

19. Srivastava, A.P., Luo, M., Zhou, W., Symersky, J., Bai, D., Chambers, M.G., Faraldo-Gomez, J.D., Liao, M., and Mueller, D.M. (2018). High-resolution cryo-EM analysis of the yeast ATP synthase in a lipid membrane. Science *360*, eaas9699.

20. Spikes, T.E., Montgomery, M.G., and Walker, J.E. (2020). Structure of the dimeric ATP synthase from bovine mitochondria. Proc. Natl. Acad. Sci. USA *117*, 23519–23526.

21. He, J., Ford, H.C., Carroll, J., Douglas, C., Gonzales, E., Ding, S., Fearnley, I.M., and Walker, J.E. (2018). Assembly of the membrane domain of ATP synthase in human mitochondria. Proc. Natl. Acad. Sci. USA *115*, 2988–2993.

22. Lieberthal, W., Menza, S.A., and Levine, J.S. (1998). Graded ATP depletion can cause necrosis or apoptosis of cultured mouse proximal tubular cells. Am. J. Physiol. *274*, F315–F327.

23. Niedzwiecka, K., Tisi, R., Penna, S., Lichocka, M., Plochocka, D., and Kucharczyk, R. (2018). Two mutations in mitochondrial ATP6 gene of ATP synthase, related to human cancer, affect ROS, calcium homeostasis and mitochondrial permeability transition in yeast. Biochim. Biophys. Acta Mol. Cell. Res *1865*, 117–131.

24. Li, J., Agarwal, E., Bertolini, I., Seo, J.H., Caino, M.C., Ghosh, J.C., Kossenkov, A.V., Liu, Q., Tang, H.Y., Goldman, A.R., et al. (2020). The mitophagy effector FUNDC1 controls mitochondrial reprogramming and cellular plasticity in cancer cells. Sci. Signal *13*, eaaz8240.

25. Gale, M., Li, Y., Cao, J., Liu, Z.Z., Holmbeck, M.A., Zhang, M., Lang, S.M., Wu, L., Do Carmo, M., Gupta, S., et al. (2020). Acquired resistance to HER2-targeted therapies creates vulnerability to ATP synthase inhibition. Cancer Res. *80*, 524–535.

26. Xu, G., and Li, J.Y. (2016). ATP5A1 and ATP5B are highly expressed in glioblastoma tumor cells and endothelial cells of microvascular proliferation. J. Neurooncol. *126*, 405–413.

27. Muys, B.R., Sousa, J.F., Plaça, J.R., de Araújo, L.F., Sarshad, A.A., Anastasakis, D.G., Wang, X., Li, X.L., de Molfetta, G.A., Ramão, A., et al. (2019). miR-450a acts as a tumor suppressor in ovarian cancer by regulating energy metabolism. Cancer Res. *79*, 3294–3305.

28. Speransky, S., Serafini, P., Caroli, J., Bicciato, S., Lippman, M.E., and Bishopric, N.H. (2019). A novel RNA aptamer identifies plasma membrane ATP synthase beta subunit as an early marker and therapeutic target in aggressive cancer. Breast Cancer Res. Treat. *176*, 271–289.

29. Bikas, A., Jensen, K., Patel, A., Costello, J., Kaltsas, G., Hoperia, V., Wartofsky, L., Burman, K., and Vasko, V. (2019). Mitotane induces mitochondrial membrane depolarization and apoptosis in thyroid cancer cells. Int. J. Oncol. *55*, 7–20.

30. Yuan, L., Chen, L., Qian, K., Wang, G., Lu, M., Qian, G., Cao, X., Jiang, W., Xiao, Y., and Wang, X. (2018). A novel correlation between ATP5A1 gene expression and progression of human clear cell renal cell carcinoma identified by co-expression analysis. Oncol. Rep. *39*, 525–536.

31. Chang, F.W., Fan, H.C., Liu, J.M., Fan, T.P., Jing, J., Yang, C.L., and Hsu, R.J. (2017). Estrogen enhances the expression of the multidrug transporter gene *ABCG2*—Increasing drug resistance of breast cancer cells through estrogen receptors. Int. J. Mol. Sci. *18*, E163.

32. Feichtinger, R.G., Schäfer, G., Seifarth, C., Mayr, J.A., Kofler, B., and Klocker, H. (2018). Reduced levels of ATP synthase subunit ATP5F1A correlate with earlier-onset prostate cancer. Oxid. Med. Cell. Longev. *2018*, 1347174.

33. Seth, R., Keeley, J., Abu-Ali, G., Crook, S., Jackson, D., and Ilyas, M. (2009). The putative tumour modifier gene ATP5A1 is not mutated in human colorectal cancer cell lines but expression levels correlate with TP53 mutations and chromosomal instability. J. Clin. Pathol. *62*, 598–603.

34. Liu, T., Guo, J., Wang, T., Zhang, S., Yu, X., Hou, C., and Guo, D. (2019). Network pharmacology-based analysis of mechanisms of the anti-hepatocellular carcinoma effect by dihydroartemisinin. Discov. Med. *28*, 139–147.

35. Lu, Z., Song, Q., Yang, J., Zhao, X., Zhang, X., Yang, P., and Kang, J. (2014). Comparative proteomic analysis of anti-cancer mechanism by periplocin treatment in lung cancer cells. Cell. Physiol. Biochem. *33*, 859–868.

36. Ratajczak, K., Lukasiak, A., Grel, H., Dworakowska, B., Jakiela, S., and Stobiecka, M. (2019). Monitoring of dynamic ATP level changes by oligomycin-modulated ATP synthase inhibition in SW480 cancer cells using fluorescent "On-Off" switching DNA aptamer. Anal. Bioanal. Chem. *411*, 6899–6911.

37. Di Virgilio, F., Sarti, A.C., Falzoni, S., De Marchi, E., and Adinolfi, E. (2018). Extracellular ATP and P2 purinergic signalling in the tumour microenvironment. Nat. Rev. Cancer *18*, 601–618.

38. Pellegatti, P., Raffaghello, L., Bianchi, G., Piccardi, F., Pistoia, V., and Di Virgilio, F. (2008). Increased level of extracellular ATP at tumor sites: In vivo imaging with plasma membrane luciferase. PLoS ONE *3*, e2599.

39. Astakhova, A., Chistyakov, D., Thomas, D., Geisslinger, G., Brüne, B., Sergeeva, M., and Namgaladze, D. (2019). Inhibitors of oxidative phosphorylation modulate astrocyte inflammatory responses through AMPK-dependent Ptgs2 mRNA stabilization. Cells *8*, E1185.

40. Park, B.N., Kim, G.H., Ko, S.A., Shin, G.H., Lee, S.J., An, Y.S., and Yoon, J.K. (2019). Zr-89 immuno-PET targeting ectopic ATP synthase enables in-vivo imaging of tumor angiogenesis. Int. J. Mol. Sci. *20*, E3928.

41. Chang, H.Y., Huang, H.C., Huang, T.C., Yang, P.C., Wang, Y.C., and Juan, H.F. (2012). Ectopic ATP synthase blockade suppresses lung adenocarcinoma growth by activating the unfolded protein response. Cancer Res. *72*, 4696–4706.

42. Wang, T., Shen, Y., Li, Y., Wang, B., Wang, B., Wu, D., Ruan, C., and Wang, Y. (2019). Ectopic ATP synthase β subunit proteins on human leukemia cell surface interact with platelets by binding glycoprotein IIb. Haematologica *104*, e364–e368.

43. Bianchi, G., Ravera, S., Traverso, C., Amaro, A., Piaggio, F., Emionite, L., Bachetti, T., Pfeffer, U., and Raffaghello, L. (2018). Curcumin induces a fatal energetic impairment in tumor cells in vitro and in vivo by inhibiting ATP-synthase activity. Carcinogenesis *39*, 1141–1150.

44. Salomon, A.R., Voehringer, D.W., Herzenberg, L.A., and Khosla, C. (2001). Apoptolidin, a selective cytotoxic agent, is an inhibitor of F0F1-ATPase. Chem. Biol. *8*, 71–80.

45. Mowery, Y.M., and Pizzo, S.V. (2008). Targeting cell surface F1F0 ATP synthase in cancer therapy. Cancer Biol. Ther. *7*, 1836–1838.

46. Hong, S., and Pedersen, P.L. (2008). ATP synthase and the actions of inhibitors utilized to study its roles in human health, disease, and other scientific areas. Microbiol. Mol. Biol. Rev *72*, 590–641.

47. Patel, B.A., D'Amico, T.L., and Blagg, B. (2020). Natural products and other inhibitors of F1FO ATP synthase. Eur. J. Med. Chem *207*, 112779.

48. Narang, R., Kumar, R., Kalra, S., Nayak, S.K., Khatik, G.L., Kumar, G.N., Sudhakar, K., and Singh, S.K. (2019). Recent advancements in mechanistic studies and structure activity relationship of FoF1 ATP synthase inhibitor as antimicrobial agent. Eur. J. Med. Chem *182*, 111644.

49. Arakaki, N., Kita, T., Shibata, H., and Higuti, T. (2007). Cell-surface H^+-ATP synthase as a potential molecular target for anti-obesity drugs. FEBS Lett. *581*, 3405–3409.

50. Huang, T.C., Chang, H.Y., Hsu, C.H., Kuo, W.H., Chang, K.J., and Juan, H.F. (2008). Targeting therapy for breast carcinoma by ATP synthase inhibitor aurovertin B. J. Proteome Res. *7*, 1433–1444.

51. Devenish, R.J., Prescott, M., Boyle, G.M., and Nagley, P. (2000). The oligomycin axis of mitochondrial ATP synthase: OSCP and the proton channel. J. Bioenerg. Biomembr. *32*, 507–515.

52. Tan, J.L., Li, F., Yeo, J.Z., Yong, K.J., Bassal, M.A., Ng, G.H., Lee, M.Y., Leong, C.Y., Tan, H.K., Wu, C.S., et al. (2019). New high-throughput screening identifies compounds that reduce viability specifically in liver cancer cells that express high levels of SALL4 by inhibiting oxidative phosphorylation. Gastroenterology *157*, 1615–1629.e17.

53. Lin, C.S., Liu, L.T., Ou, L.H., Pan, S.C., Lin, C.I., and Wei, Y.H. (2018). Role of mitochondrial function in the invasiveness of human colon cancer cells. Oncol. Rep. *39*, 316–330.

54. Wender, P.A., and Longcore, K.E. (2007). Isolation, structure determination, and anti-cancer activity of apoptolidin D. Org. Lett. *9*, 691–694.

55. Wender, P.A., Sukopp, M., and Longcore, K. (2005). Apoptolidins B and C: Isolation, structure determination, and biological activity. Org. Lett. *7*, 3025–3028.

56. Wang, S., Moustaid-Moussa, N., Chen, L., Mo, H., Shastri, A., Su, R., Bapat, P., Kwun, I., and Shen, C.L. (2014). Novel insights of dietary polyphenols and obesity. J. Nutr. Biochem. *25*, 1–18.

57. Rauf, A., Imran, M., Butt, M.S., Nadeem, M., Peters, D.G., and Mubarak, M.S. (2018). Resveratrol as an anti-cancer agent: A review. Crit. Rev. Food Sci. Nutr. *58*, 1428–1447.

58. Fliedner, S.M., Yang, C., Thompson, E., Abu-Asab, M., Hsu, C.M., Lampert, G., Eiden, L., Tischler, A.S., Wesley, R., Zhuang, Z., et al. (2015). Potential therapeutic target for malignant paragangliomas: ATP synthase on the surface of paraganglioma cells. Am. J. Cancer Res. *5*, 1558–1570.

59. Valenti, D., de Bari, L., Manente, G.A., Rossi, L., Mutti, L., Moro, L., and Vacca, R.A. (2013). Negative modulation of mitochondrial oxidative phosphorylation by epigallocatechin-3 gallate leads to growth arrest and apoptosis in human malignant pleural mesothelioma cells. Biochim. Biophys. Acta *1832*, 2085–2096.

60. Khiewkamrop, P., Phunsomboon, P., Richert, L., Pekthong, D., and Srisawang, P. (2018). Epistructured catechins, EGCG and EC facilitate apoptosis induction through targeting de novo lipogenesis pathway in HepG2 cells. Cancer Cell Int. *18*, 46.

61. Liu, M., Xu, Y.F., Feng, Y., Yang, F.Q., Luo, J., Zhai, W., Che, J.P., Wang, G.C., and Zheng, J.H. (2013). Epigallocatechin gallate attenuates interstitial cystitis in human bladder urothelium cells by modulating purinergic receptors. J. Surg. Res *183*, 397–404.

62. Nelson, K.M., Dahlin, J.L., Bisson, J., Graham, J., Pauli, G.F., and Walters, M.A. (2017). The essential medicinal chemistry of curcumin. J. Med. Chem. *60*, 1620–1637.

63. Rawat, D., Shrivastava, S., Naik, R.A., Chhonker, S.K., Mehrotra, A., and Koiri, R.K. (2018). An overview of natural plant products in the treatment of hepatocellular carcinoma. Anticancer. Agents Med. Chem. *18*, 1838–1859.

64. Dang, Y.P., Yuan, X.Y., Tian, R., Li, D.G., and Liu, W. (2015). Curcumin improves the paclitaxel-induced apoptosis of HPV-positive human cervical cancer cells via the NF-κB-p53-caspase-3 pathway. Exp. Ther. Med. *9*, 1470–1476.

65. Faião-Flores, F., Suarez, J.A., Pardi, P.C., and Maria, D.A. (2012). DM-1, sodium 4-[5-(4-hydroxy-3-methoxyphenyl)-3-oxo-penta-1,4-dienyl]-2-methoxy-phenolate: A curcumin analog with a synergic effect in combination with paclitaxel in breast cancer treatment. Tumour Biol. *33*, 775–785.

66. Chen, Q.H. (2015). Curcumin-based anti-prostate cancer agents. Anticancer. Agents Med. Chem. *15*, 138–156.

67. Wan Mohd Tajuddin, W.N.B., Lajis, N.H., Abas, F., Othman, I., and Naidu, R. (2019). Mechanistic understanding of curcumin's therapeutic effects in lung cancer. Nutrients *11*, E2989.

68. Deguchi, A. (2015). Curcumin targets in inflammation and cancer. Endocr. Metab. Immune Disord. Drug Targets *15*, 88–96.

69. Alvarez-Delgado, C., Mendoza-Rodriguez, C.A., Picazo, O., and Cerbon, M. (2010). Different expression of alpha and beta mitochondrial estrogen receptors in the aging rat brain: Interaction with respiratory complex V. Exp. Gerontol *45*, 580–585.

70. Massart, F., Paolini, S., Piscitelli, E., Brandi, M.L., and Solaini, G. (2002). Dose-dependent inhibition of mitochondrial ATP synthase by 17 beta-estradiol. Gynecol. Endocrinol. *16*, 373–377.

71. Zheng, J., and Ramirez, V.D. (1999). Purification and identification of an estrogen binding protein from rat brain: Oligomycin sensitivity-conferring protein (OSCP), a subunit of mitochondrial F0F1-ATP synthase/ATPase. J. Steroid Biochem. Mol. Biol. *68*, 65–75.

72. Zheng, J., and Ramirez, V.D. (1999). Rapid inhibition of rat brain mitochondrial proton F0F1-ATPase activity by estrogens: comparison with Na^+, K^+-ATPase of porcine cortex. Eur. J. Pharmacol. *368*, 95–102.

73. Pitt, J.I., and Miller, J.D. (2017). A concise history of mycotoxin research. J. Agric. Food Chem. *65*, 7021–7033.

74. Kumar, A., Banerjee, A., Singh, D., Thakur, G., Kasarpalkar, N., Gavali, S., Gadkar, S., Madan, T., Mahale, S.D., Balasinor, N.H., and Sachdeva, G. (2018). Estradiol: A steroid with multiple facets. Horm. Metab. Res. *50*, 359–374.

75. Chinnam, N., Dadi, P.K., Sabri, S.A., Ahmad, M., Kabir, M.A., and Ahmad, Z. (2010). Dietary bioflavonoids inhibit *Escherichia coli* ATP synthase in a differential manner. Int. J. Biol. Macromol. *46*, 478–486.

76. Pons, D.G., Nadal-Serrano, M., Blanquer-Rossello, M.M., Sastre-Serra, J., Oliver, J., and Roca, P. (2014). Genistein modulates proliferation and mitochondrial functionality in breast cancer cells depending on ERalpha/ERbeta ratio. J. Cell. Biochem *115*, 949–958.

77. Gause, E.M., Buck, M.A., and Douglas, M.G. (1981). Binding of citreoviridin to the β subunit of the yeast F_1-ATPase. J. Biol. Chem. *256*, 557–559.

78. Chang, H.Y., Huang, T.C., Chen, N.N., Huang, H.C., and Juan, H.F. (2014). Combination therapy targeting ectopic ATP synthase and 26S proteasome induces ER stress in breast cancer cells. Cell Death Dis. *5*, e1540.

79. Pohland, A.E. (1993). Mycotoxins in review. Food Addit. Contam. *10*, 17–28.

80. Matsuno-Yagi, A., Yagi, T., and Hatefi, Y. (1985). Studies on the mechanism of oxidative phosphorylation: Effects of specific F0 modifiers on ligand-induced conformation changes of F1. Proc. Natl. Acad. Sci. USA *82*, 7550–7554.

81. Zhu, H., Wang, F., Ju, X., Kong, L., An, T., Zhao, Z., Liu, J., and Li, Y. (2018). Aurovertin B sensitizes colorectal cancer cells to NK cell recognition and lysis. Biochem. Biophys. Res. Commun *503*, 3057–3063.

82. Long, H., Cheng, Z., Huang, W., Wu, Q., Li, X., Cui, J., Proksch, P., and Lin, W. (2016). Diasteltoxins A–C, asteltoxin-based dimers from a mutant of the sponge-associated *Emericella variecolor* fungus. Org. Lett. *18*, 4678–4681.

83. Wu, Q., Long, H.L., Liu, D., Proksch, P., and Lin, W.H. (2015). Varioxiranols I-L, new lactones from a sponge-associated *Emericella variecolor* fungus. J. Asian Nat. Prod. Res. *17*, 1137–1145.

84. Preiss, L., Langer, J.D., Yildiz, Ö., Eckhardt-Strelau, L., Guillemont, J.E., Koul, A., and Meier, T. (2015). Structure of the mycobacterial ATP synthase Fo rotor ring in complex with the anti-TB drug bedaquiline. Sci. Adv. *1*, e1500106.

85. Wu, X., Li, F., Wang, X., Li, C., Meng, Q., Wang, C., Huang, J., Chen, S., and Zhu, Z. (2018). Antibiotic bedaquiline effectively targets growth, survival and tumor angiogenesis of lung cancer through suppressing energy metabolism. Biochem. Biophys. Res. Commun *495*, 267–272.

86. Fiorillo, M., Lamb, R., Tanowitz, H.B., Cappello, A.R., Martinez-Outschoorn, U.E., Sotgia, F., and Lisanti, M.P. (2016). Bedaquiline, an FDA-approved antibiotic, inhibits mitochondrial function and potently blocks the proliferative expansion of stem-like cancer cells (CSCs). Aging (Albany NY) *8*, 1593–1607.

87. Liu, Y., Fang, S., Sun, Q., and Liu, B. (2016). Anthelmintic drug ivermectin inhibits angiogenesis, growth and survival of glioblastoma through inducing mitochondrial dysfunction and oxidative stress. Biochem. Biophys. Res. Commun *480*, 415–421.

88. Skoda, J., Borankova, K., Jansson, P.J., Huang, M.L., Veselska, R., and Richardson, D.R. (2019). Pharmacological targeting of mitochondria in cancer stem cells: An ancient organelle at the crossroad of novel anti-cancer therapies. Pharmacol. Res *139*, 298–313.

89. Cox, E., and Laessig, K. (2014). FDA approval of bedaquiline—The benefit-risk balance for drug-resistant tuberculosis. N. Engl. J. Med. *371*, 689–691.

90. Hards, K., and Cook, G.M. (2018). Targeting bacterial energetics to produce new antimicrobials. Drug Resist. Updat *36*, 1–12.

91. Elbehairi, S.E.I., Alfaifi, M.Y., Shati, A.A., Alshehri, M.A., Elshaarawy, R.F.M., and Hafez, H.S. (2020). Role of Pd(II)-chitooligosaccharides-Gboxin analog in oxidative phosphorylation inhibition and energy depletion: Targeting mitochondrial dynamics. Chem. Biol. Drug Des. *96*, 1148–1161.

92. Gajate, C., and Mollinedo, F. (2014). Lipid rafts, endoplasmic reticulum and mitochondria in the antitumor action of the alkylphospholipid analog edelfosine. Anticancer. Agents Med. Chem. *14*, 509–527.

93. Villa-Pulgarín, J.A., Gajate, C., Botet, J., Jimenez, A., Justies, N., Varela-M, R.E., Cuesta-Marbán, Á., Müller, I., Modolell, M., Revuelta, J.L., and Mollinedo, F. (2017). Mitochondria and lipid raft-located FOF1-ATP synthase as major

therapeutic targets in the antileishmanial and anticancer activities of ether lipid edelfosine. PLoS Negl. Trop. Dis. *11*, e0005805.

94. van Blitterswijk, W.J., and Verheij, M. (2013). Anticancer mechanisms and clinical application of alkylphospholipids. Biochim. Biophys. Acta *1831*, 663–674.

95. Mollinedo, F., Fernández, M., Hornillos, V., Delgado, J., Amat-Guerri, F., Acuña, A.U., Nieto-Miguel, T., Villa-Pulgarín, J.A., González-García, C., Ceña, V., and Gajate, C. (2011). Involvement of lipid rafts in the localization and dysfunction effect of the antitumor ether phospholipid edelfosine in mitochondria. Cell Death Dis. *2*, e158.

96. Dorlo, T.P., Balasegaram, M., Beijnen, J.H., and de Vries, P.J. (2012). Miltefosine: A review of its pharmacology and therapeutic efficacy in the treatment of leishmaniasis. J. Antimicrob. Chemother. *67*, 2576–2597.

97. Mollinedo, F. (2014). Editorial: Antitumor alkylphospholipid analogs: A promising and growing family of synthetic cell membrane-targeting molecules for cancer treatment). Anticancer. Agents Med. Chem. *14*, 495–498.

98. Zaremberg, V., Ganesan, S., and Mahadeo, M. (2020). Lipids and membrane microdomains: The glycerolipid and alkylphosphocholine class of cancer chemotherapeutic drugs. Handb. Exp. Pharmacol. *259*, 261–288.

99. Esparza-Moltó, P.B., Nuevo-Tapioles, C., and Cuezva, J.M. (2017). Regulation of the H^+-ATP synthase by IF1: A role in mitohormesis. Cell. Mol. Life Sci. *74*, 2151–2166.

100. González-Llorente, L., Santacatterina, F., García-Aguilar, A., Nuevo-Tapioles, C., González-García, S., Tirpakova, Z., Toribio, M.L., and Cuezva, J.M. (2019). Overexpression of mitochondrial IF1 prevents metastatic disease of colorectal cancer by enhancing anoikis and tumor infiltration of NK cells. Cancers (Basel) *12*, E22.

101. Sgarbi, G., Gorini, G., Liuzzi, F., Solaini, G., and Baracca, A. (2018). Hypoxia and IF_1 expression promote ROS decrease in cancer cells. Cells *7*, E64.

102. Jung, K.H., Song, S.H., Paik, J.Y., Koh, B.H., Choe, Y.S., Lee, E.J., Kim, B.T., and Lee, K.H. (2007). Direct targeting of tumor cell F_1F_0 ATP-synthase by radioiodine angiostatin in vitro and in vivo. Cancer Biother. Radiopharm. *22*, 704–712.

103. Chen, C., Liang, H., Liao, X., Pan, J., Chen, J., Zhao, S., Xu, Y., Wu, Y., and Ni, J. (2016). A humanized chimeric antibody Hai178 targeted to the β subunit of F1F0 ATP synthase. Tumour Biol. *37*, 15903–15912.

104. Ridley, C.P., and Khosla, C. (2009). Polyketides. In Encyclopedia of Microbiology, Third Edition, M. Schaechter, ed. (Academic Press), pp. 472–481.

105. Penefsky, H.S. (1985). Mechanism of inhibition of mitochondrial adenosine triphosphatase by dicyclohexylcarbodiimide and oligomycin: Relationship to ATP synthesis. Proc. Natl. Acad. Sci. USA *82*, 1589–1593.

106. Giorgio, V., Fogolari, F., Lippe, G., and Bernardi, P. (2019). OSCP subunit of mitochondrial ATP synthase: Role in regulation of enzyme function and of its transition to a pore. Br. J. Pharmacol *176*, 4247–4257.

107. Pacheco-Velázquez, S.C., Robledo-Cadena, D.X., Hernández-Reséndiz, I., Gallardo-Pérez, J.C., Moreno-Sánchez, R., and Rodríguez-Enríquez, S. (2018). Energy metabolism drugs block triple negative breast metastatic cancer cell phenotype. Mol. Pharm. *15*, 2151–2164.

108. Sun, X., Zhan, L., Chen, Y., Wang, G., He, L., Wang, Q., Zhou, F., Yang, F., Wu, J., Wu, Y., et al. (2018). Increased mtDNA copy number promotes cancer progression by enhancing mitochondrial oxidative phosphorylation in microsatellite-stable colorectal cancer. Signal Transduct. Target. Ther. *3*, 8.

109. Zhang, Y., Nguyen, T.T.T., Shang, E., Mela, A., Humala, N., Mahajan, A., Zhao, J., Shu, C., Torrini, C., Sanchez-Quintero, M.J., et al. (2020). MET inhibition elicits PGC1α-dependent metabolic reprogramming in glioblastoma. Cancer Res. *80*, 30–43.

110. Huang, Z., Kondoh, E., Visco, Z.R., Baba, T., Matsumura, N., Dolan, E., Whitaker, R.S., Konishi, I., Fujii, S., Berchuck, A., and Murphy, S.K. (2021). Targeting dormant ovarian cancer cells *in vitro* and in an *in vivo* mouse model of platinum resistance. Mol. Cancer Ther. *20*, 85–95.

111. Salomon, A.R., Voehringer, D.W., Herzenberg, L.A., and Khosla, C. (2000). Understanding and exploiting the mechanistic basis for selectivity of polyketide inhibitors of F_0F_1-ATPase. Proc. Natl. Acad. Sci. USA *97*, 14766–14771.

112. Daniel, P.T., Koert, U., and Schuppan, J. (2006). Apoptolidin: Induction of apoptosis by a natural product. Angew. Chem. Int. Ed. Engl. *45*, 872–893.

113. Wender, P.A., Jankowski, O.D., Longcore, K., Tabet, E.A., Seto, H., and Tomikawa, T. (2006). Correlation of F_0F_1-ATPase inhibition and antiproliferative activity of apoptolidin analogues. Org. Lett. *8*, 589–592.

114. Serrill, J.D., Tan, M., Fotso, S., Sikorska, J., Kasanah, N., Hau, A.M., McPhail, K.L., Santosa, D.A., Zabriskie, T.M., Mahmud, T., et al. (2015). Apoptolidins A and C activate AMPK in metabolically sensitive cell types and are mechanistically distinct from oligomycin A. Biochem. Pharmacol *93*, 251–265.

115. Yamashita, N., Kazuo, S.Y., Kitamura, M., Wakao, H., Furihata, K., Furihata, K., Hayakawa, Y., Miyajima, A., and Seto, H. (1997). Cytovaricin B, a new inhibitor of JAK-STAT signal transduction produced by *Streptomyces torulosus*. J. Antibiot. (Tokyo) *50*, 440–442.

116. Estrela, J.M., Mena, S., Obrador, E., Benlloch, M., Castellano, G., Salvador, R., and Dellinger, R.W. (2017). Polyphenolic phytochemicals in cancer prevention and therapy: Bioavailability versus bioefficacy. J. Med. Chem. *60*, 9413–9436.

117. Genestie, C., Leary, A., Devouassoux, M., and Auguste, A. (2017). [Histological and molecular classification of endometrial carcinoma and therapeutical implications]. Bull. Cancer *104*, 1001–1012.

118. Weinreb, O., Amit, T., and Youdim, M.B. (2008). The application of proteomics for studying the neurorescue activity of the polyphenol (−)-epigallocatechin-3-gallate. Arch. Biochem. Biophys. *476*, 152–160.

119. Kunnumakkara, A.B., Bordoloi, D., Harsha, C., Banik, K., Gupta, S.C., and Aggarwal, B.B. (2017). Curcumin mediates anticancer effects by modulating multiple cell signaling pathways. Clin. Sci. (Lond.) *131*, 1781–1799.

120. Moreno, A.J., Moreira, P.I., Custódio, J.B., and Santos, M.S. (2013). Mechanism of inhibition of mitochondrial ATP synthase by 17β-estradiol. J. Bioenerg. Biomembr. *45*, 261–270.

121. Schäberle, T.F. (2016). Biosynthesis of α-pyrones. Beilstein J. Org. Chem. *12*, 571–588.

122. Hu, C.W., Hsu, C.L., Wang, Y.C., Ishihama, Y., Ku, W.C., Huang, H.C., and Juan, H.F. (2015). Temporal phosphoproteome dynamics induced by an ATP synthase inhibitor citreoviridin. Mol. Cell. Proteomics *14*, 3284–3298.

123. Zhao, H., Wu, R., Ma, L.-F., Wo, L.-K., Hu, Y.-Y., Chen, C., and Zhan, Z.-J. (2016). Aurovertin-type polyketides from Calcarisporium arbuscula with potent cytotoxic activities against triple-negative breast cancer. Helvetica Chimica Acta *99*, 543–546.

124. Wu, R., Yang, X., Zhou, Q., Yu, W., Li, M., Wo, J., Shan, W., Zhao, H., Chen, Y., and Zhan, Z. (2020). Aurovertin B exerts potent antitumor activity against triple-negative breast cancer *in vivo* and *in vitro via* regulating ATP synthase activity and DUSP1 expression. Pharmazie *75*, 261–265.

125. Calvert, M.B., Furkert, D.P., Cooper, C.B., and Brimble, M.A. (2020). Synthetic approaches towards bedaquiline and its derivatives. Bioorg. Med. Chem. Lett *30*, 127172.

126. Nieto Ramirez, L.M., Quintero Vargas, K., and Diaz, G. (2020). Whole genome sequencing for the analysis of drug resistant strains of *Mycobacterium tuberculosis*: A systematic review for bedaquiline and delamanid. Antibiotics (Basel) *9*, E133.

127. Rios-Marco, P., Marco, C., Galvez, X., Jimenez-Lopez, J.M., and Carrasco, M.P. (2017). Alkylphospholipids: An update on molecular mechanisms and clinical relevance. Biochim. Biophys. Acta Biomembr *1859*, 1657–1667.

128. Garcia-Bermudez, J., Sanchez-Arago, M., Soldevilla, B., Del, A.A., Nuevo-Tapioles, C., and Cuezva, J.M. (2015). PKA phosphorylates the ATPase inhibitory factor 1 and inactivates its capacity to bind and inhibit the mitochondrial H+-ATP synthase. Cell. Rep *12*, 2143–2155.

129. Cao, Y., and Xue, L. (2004). Angiostatin. Semin. Thromb. Hemost. *30*, 83–93.

130. Kenan, D.J., and Wahl, M.L. (2005). Ectopic localization of mitochondrial ATP synthase: A target for anti-angiogenesis intervention? J. Bioenerg. Biomembr. *37*, 461–465.

131. Moser, T.L., Stack, M.S., Asplin, I., Enghild, J.J., Højrup, P., Everitt, L., Hubchak, S., Schnaper, H.W., and Pizzo, S.V. (1999). Angiostatin binds ATP synthase on the surface of human endothelial cells. Proc. Natl. Acad. Sci. USA *96*, 2811–2816.

132. Yuan, J., Zhang, C., Fang, S., Zhuang, Z., Ling, S., and Wang, S. (2012). A monoclonal antibody against F1-F0 ATP synthase beta subunit. Hybridoma (Larchmt.) *31*, 352–357.

133. Wen-Li, Z., Jian, W., Yan-Fang, T., Xing, F., Yan-Hong, L., Xue-Ming, Z., Min, Z., Jian, N., and Jian, P. (2012). Inhibition of the ecto-beta subunit of F1F0-ATPase inhibits proliferation and induces apoptosis in acute myeloid leukemia cell lines. J. Exp. Clin. Cancer Res. *31*, 92.

134. Chung, I.C., Chen, L.C., Tsang, N.M., Chuang, W.Y., Liao, T.C., Yuan, S.N., OuYang, C.N., Ojcius, D.M., Wu, C.C., and Chang, Y.S. (2020). Mitochondrial oxidative phosphorylation complex regulates NLRP3 inflammasome activation and predicts patient survival in nasopharyngeal carcinoma. Mol. Cell. Proteomics *19*, 142–154.

135. Isidoro, A., Martínez, M., Fernández, P.L., Ortega, A.D., Santamaría, G., Chamorro, M., Reed, J.C., and Cuezva, J.M. (2004). Alteration of the bioenergetic phenotype of mitochondria is a hallmark of breast, gastric, lung and oesophageal cancer. Biochem. J. *378*, 17–20.

136. Dieras, V., Miles, D., Verma, S., Pegram, M., Welslau, M., Baselga, J., Krop, I.E., Blackwell, K., Hoersch, S., Xu, J., et al. (2017). Trastuzumab emtansine versus capecitabine plus lapatinib in patients with previously treated HER2-positive advanced breast cancer (EMILIA): a descriptive analysis of final overall survival results from a randomised, open-label, phase 3 trial. Lancet Oncol *18*, 732–742.

137. Verma, S., Miles, D., Gianni, L., Krop, I.E., Welslau, M., Baselga, J., Pegram, M., Oh, D.Y., Diéras, V., Guardino, E., et al.; EMILIA Study Group (2012). Trastuzumab emtansine for HER2-positive advanced breast cancer. N. Engl. J. Med. *367*, 1783–1791.

138. Arif, T., Paul, A., Krelin, Y., Shteinfer-Kuzmine, A., and Shoshan-Barmatz, V. (2018). Mitochondrial VDAC1 silencing leads to metabolic rewiring and the reprogramming of tumour cells into advanced differentiated states. Cancers (Basel) *10*, E499.

139. Arif, T., Amsalem, Z., and Shoshan-Barmatz, V. (2019). Metabolic reprograming via silencing of mitochondrial VDAC1 expression encourages differentiation of cancer cells. Mol. Ther. Nucleic Acids *17*, 24–37.

140. Arif, T., Stern, O., Pittala, S., Chalifa-Caspi, V., and Shoshan-Barmatz, V. (2019). Rewiring of cancer cell metabolism by mitochondrial VDAC1 depletion results in time-dependent tumor reprogramming: Glioblastoma as a proof of concept. Cells *8*, E1330.

141. Amsalem, Z., Arif, T., Shteinfer-Kuzmine, A., Chalifa-Caspi, V., and Shoshan-Barmatz, V. (2020). The mitochondrial protein VDAC1 at the crossroads of cancer cell metabolism: the epigenetic link. Cancers (Basel) *12*, E1031.

142. Zerbib, E., Arif, T., Shteinfer-Kuzmine, A., Chalifa-Caspi, V., and Shoshan-Barmatz, V. (2021). VDAC1 silencing in cancer cells leads to metabolic reprogramming that modulates tumor microenvironment. Cancers (Basel) *13*, 2850.

143. Vyas, S., Zaganjor, E., and Haigis, M.C. (2016). Mitochondria and cancer. Cell *166*, 555–566.

144. Weinberg, F., Hamanaka, R., Wheaton, W.W., Weinberg, S., Joseph, J., Lopez, M., Kalyanaraman, B., Mutlu, G.M., Budinger, G.R., and Chandel, N.S. (2010). Mitochondrial metabolism and ROS generation are essential for Kras-mediated tumorigenicity. Proc. Natl. Acad. Sci. USA *107*, 8788–8793.

145. Zhan, S., Wang, T., Wang, M., Li, J., and Ge, W. (2019). In-depth proteomics analysis to identify biomarkers of papillary thyroid cancer patients older than 45 years with different degrees of lymph node metastases. Proteomics Clin. Appl. *13*, e1900030.

146. Henderson, I.C., Allegra, J.C., Woodcock, T., Wolff, S., Bryan, S., Cartwright, K., Dukart, G., and Henry, D. (1989). Randomized clinical trial comparing mitoxantrone with doxorubicin in previously treated patients with metastatic breast cancer. J. Clin. Oncol. *7*, 560–571.

147. Allison, K.E., Coomber, B.L., and Bridle, B.W. (2017). Metabolic reprogramming in the tumour microenvironment: A hallmark shared by cancer cells and T lymphocytes. Immunology *152*, 175–184.

148. Li, F., and Simon, M.C. (2020). Cancer cells don't live alone: Metabolic communication within tumor microenvironments. Dev. Cell *54*, 183–195.

149. Lyssiotis, C.A., and Kimmelman, A.C. (2017). Metabolic interactions in the tumor microenvironment. Trends Cell Biol *27*, 863–875.

150. Liang, J., and Shang, Y. (2013). Estrogen and cancer. Annu. Rev. Physiol. *75*, 225–240.

151. Jameera, B.A., Jubie, S., and Nanjan, M.J. (2017). Estrogen receptor agonists/antagonists in breast cancer therapy: A critical review. Bioorg. Chem *71*, 257–274.