Life Sciences 234 (2019) 116781

Contents lists available at ScienceDirect

Life Sciences

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

Review article

Cancer stem cell (CSC) resistance drivers

Masoud Najafi[a], Keywan Mortezaee[b,c,*], Jamal Majidpoor[d]

[a] Radiology and Nuclear Medicine Department, School of Paramedical Sciences, Kermanshah University of Medical Sciences, Kermanshah, Iran
[b] Department of Anatomy, School of Medicine, Kurdistan University of Medical Sciences, Sanandaj, Iran
[c] Cancer and Immunology Research Center, Kurdistan University of Medical Sciences, Sanandaj, Iran
[d] Department of Anatomy, School of Medicine, Iran University of Medical Sciences, Tehran, Iran

ARTICLE INFO

Keywords:
Cancer stem cell (CSC)
Tumor microenvironment (TME)
Epithelial-mesenchymal transition (EMT)
Dedifferentiation
Reactive oxygen species (ROS)
Stemness
Resistance
Plasticity
Radiotherapy
Chemotherapy

ABSTRACT

Cancer stem cells (CSCs) are a population of self-renewal cells with high tumorigenic potency. CSCs can adopt easily with changes in the nearby milieu, and are more resistant to conventional therapies than other cells within a tumor. CSC resistance can be induced secondary to radio- and chemotherapy, or even after chemotherapy secession. A combination of both intrinsic and extrinsic factors is contributed to CSC-mediated therapy resistance. CSCs represent protective autophagy and efficient cell cycling, along with highly qualified epithelial-mesenchymal transition (EMT) regulators, reactive oxygen species (ROS) scavengers, drug transporters, and anti-apoptotic and DNA repairing systems. In addition, CSCs develop cross-talking and share some characteristics with other cells within the tumor microenvironment (TME) being more intense in higher stage tumors, and thereby sophisticating tumor-targeted therapies. TME, in fact, is a nest for aggravating resistance mechanisms in CSCs. TME is exposed constantly to the nutritional, metabolic and oxygen deprivation; these conditions promote CSC adaptation. This review is aimed to discuss main (intrinsic and extrinsic) mechanisms of CSC resistance and suggest some strategies to revoke this important promoter of therapy failure.

1. Introduction

Despite growing scientific breakthrough toward designing therapeutic schemes with high accuracy for targeting tumors, concerns still remain about how to breakdown tumor refractoriness to the targeted therapy, which is known as a leading cause of high mortality rates among patients with advanced tumors [1]. For some tumors, surgery is requested as an option. For metastatic tumors, however, this procedure generates a hypoxic microenvironment fostering aggressive tumor relapse [2]. Patients undergoing radio- and chemotherapy regimens often suffer from adverse effects imposed by these therapies but offering only palliation. Although shrinkage or even transient eradication of tumor occurs after chemotherapy [3], unfortunately, not for long patients encounter tumor resistance and recurrence, the two major reasons of therapy failure [4]. This indicates that conventional therapies not only are ineffective for targeting advanced tumors, they may even promote tumor regrowth, so extending the current knowledge about the behavior of tumor in response to therapy is an urgent need.

Following chemotherapy for any tumor type, there are residual bodies enriched in cancer stem cells (CSCs) [5,6]. CSCs and their relevance with therapy resistance has gained an intense attention recently with over 19,000 online papers in PubMed from the year 2017 to 2019, indicating the importance of their targeting for cancer therapy. CSCs have sphere formation [7] and self-renewing abilities. CSCs are stochastically distributed within a tumor [8], but reside predominantly within hypoxic, low pH and less nutrient niches [9] and take important roles for initiation, relapse and metastatic dissemination of tumors [3,10,11]. CSCs share many traits with regenerative stem cells, such as self-renewal and multipotency [1], and the reversibility of their quiescence state [12]. However, there are features specific for CSCs, including the intense tumorigenic potential, the capacity to grow as spheres under serum deprivation, high aldehyde dehydrogenase (ALDH) activity [1], and dysregulated cell cycle [13]. CSCs are identified as a rare population of cells within a tumor [7,14,15] constituting $< 1\%$ of the cellular population in the most solid tumors [16], around 0.1–1% in osteosarcoma [17]. For colon cancer this amount is about 2% [7,18]. However, upon tumor progression this fraction can increase to about 30% [19] and this increase correlates with therapy resistance [8]. CSCs are highly tumorigenic [14] being about 10-fold more tumorigenic than their non-CSC counterpart in tumors like pancreas [6]. The cells are described commonly using other terms, including tumor propagating cells, tumor progenitor cells and cancer-initiating cells (CICs). A side population is also used to describe the cells [20]. The logic behind this nomenclature is that the cells are able to efflux drugs, and so

* Corresponding author at: Department of Anatomy, School of Medicine, Kurdistan University of Medical Sciences, Sanandaj, Iran.
E-mail address: Keywan987@yahoo.com (K. Mortezaee).

https://doi.org/10.1016/j.lfs.2019.116781
Received 17 June 2019; Received in revised form 2 August 2019; Accepted 16 August 2019
Available online 17 August 2019

promoting resistance to therapy. CSCs play pivotal roles in therapy resistance, accelerating tumor regrowth after therapy [21]. Self-renewal dysregulation is possibly the first step in carcinogenesis [22]. CSCs have unlimited self-renewal capacity [23] so that presence of one CSC at the tumor site is able to promote tumor recurrence [8], and at a distant site is capable of re-growing a metastatic tumor [24,25], thus an increase in the CSC signature of a tumor indicates weaker prognosis than tumors exhibiting diminished CSC population [26]. Traditional chemotherapies are failed to eradicate tumoral cells in full [27] in which apoptotic sensitive cancer cells are the main target of radio- and chemotherapy, leaving behind tumor resistant CSC population prone to further expansion [27,28], so affected patients ultimately succumb to the tumor. Therefore, enriching our current understanding about mechanisms involved in the CSC-related recurrence crisis is essential to override therapy resistance, especially in unresectable tumors. Thus, this review we designed in order to discuss over major drivers of therapy resistance in relation with CSCs and finished with proposing some strategies to overcome this important mediator of therapy failure.

2. Chemo-radio resistant CSCs

CSCs are resistant inherently to radio- and chemotherapy [29], and it is known that they are more resistant to standard chemotherapies than bulk cancer cells [17]. A cancer-initiating cell (CIC) theory is proposed for describing resistance of metastatic tumors to traditional chemotherapy. According to this theory, resistance of CICs to traditional chemotherapy is reconsidered as the primary mechanism for therapy resistance in a metastatic disease. In fact, the primary targets for radiotherapy and most of the cytotoxic therapies are rapidly dividing (activated) apoptotic sensitive differentiated cells (stem cells and cancer cells) not CICs, thereby allowing enrichment of tumor resistant CSCs [3,27,28,30,31]. For tumors like glioblastoma multiform (GBM), harvested CSCs are identified to be the only population of tumorigenic cells, and thus representing the primary target in cancer therapy [32]. The inherent resistance of CSCs is a driving force behind cancer cell colonization at distant sites of metastasis despite application of adjuvant chemotherapy [33]. CSCs are highly dynamic and heterogeneous [34,35] and easily adaptive with the surrounding environment [36]. CSCs are slow cycling, and have active anti-apoptotic machinery (Bcl-2 [27] and Mcl-1 [33]), efficient DNA repairing systems (DNA checkpoint kinases) and sustained stemness features, all of which are account for their therapy resistance [18,28,37]. DNA damage to the CSCs occurring after radiotherapy induces cell cycle arrest attempting to repair the injured DNA, followed by a decision into either apoptosis or their reentry to the cell cycle [37]. Colon CSCs injected to immunodeficient mice can easily reproduce the original tumor [7], implying high potency of the cells in creating a bed for cancer growth.

CSC resistance is also defined by extrinsic factors. The well-known extrinsic factor influencing CSC resistance is tumor microenvironment (TME). TME is a heterocellular place [38] where critical interactions determine the tumor fate [39]. Tumors have distinct genetic and epigenetic features, which is due in part to the contribution of TME. In fact, invasive mesenchymal type CSCs are found in the regions of intense cellular density [7,40], namely at the tumor edge close to the other TME cells [41]. This indicates the importance of the TME in shaping the morphology and functional features of CSCs, and that the potency of the cells will be greatly reduced when they are excluded from this milieu. The diagram presented in the Fig. 1 depicts various intrinsic and extrinsic factors implicated in resistance of CSCs.

2.1. Mechanisms of therapy resistance in CSCs

2.1.1. EMT

EMT is linked tightly with the CSC biology [42], including stemness [43], immune escape and resistance to radio- and chemotherapy [44]. EMT is a dedifferentiation program that is for converting polarized epithelial cells into cells with migratory mesenchymal phenotype [21], thereby being more prone to represent in circulation and acquire a metastatic fate [26]. A part of this expansion is due to an influence imposed by CSCs on non-CSCs to recall an EMT program in the cells, and thus shifting them toward apoptosis resistance CSCs [45,46]. Diverse array of growth factors and cytokines are involved in regulation of EMT; this infers different levels of adaptability (plasticity) among tumoral cells [47], complexing targeted therapies.

2.1.2. Hypoxia

Hypoxia is a common phenomenon in tumor tissue [48]. Hypoxia strongly induces drug resistance in CSCs [49], and severely blunts tumor response to radio- [48] and chemotherapy [50]. Hypoxia promotes EMT associated CSC properties including self-renewal [49] in which CSC enrichment occurs upon exposure to hypoxia-inducible factor (HIF)-1α in vivo; this is for promoting resistance to chemotherapy [33]. Hypoxia promotes early EMT by suppressing E-cadherin (an epithelial marker) through twist activation, and by inducing snail via Notch activation [24]. Late migratory and invasiveness is also promoted by hypoxia [51]. Hypoxia induces unfolded protein response (UPR) signaling (indirectly through activation of endoplasmic reticulum [ER] stress) [52], which is for maintaining CSC reactive oxygen species (ROS) at low concentrations [53].

2.1.3. Oxidative modulation

ROS at both high and low concentrations can induce therapy resistance in CSCs. Lee and colleagues reported a link between an increase in the rate of mitochondrial oxidative phosphorylation (OXPHOS)-mediated ROS generation with enhanced HIF-1α accumulation and CSC resistance to chemotherapy [33]. Cannito and colleagues proved that the inducible ROS effect on HIF-1α favors promotion of early EMT-related events [51]. A level of ROS is important for fixing DNA breaks [54], while ROS overloading can induce DNA damage and sensitize cells to radiotherapy. CSCs have efficient DNA repairing systems [18,55–57] to combat potential damage to DNA upon ROS accumulation (high ROS levels). These DNA repairing systems and the preference of the repairing capacity over apoptosis have noticeable effects on responses from CSCs to radiotherapy [58]. Interestingly, there is a report that cancer cells upon receiving DNA damage signals can reprogram into CSCs, so DNA damage inducers like radiotherapy (if not being so strength) can secondarily contribute to drug resistance and tumor recurrence [48,59]. In addition, high activity of telomeric components such as TRF2 accounts for maintenance of telomeric length and resistance to radiotherapy [59]. Furthermore, CSCs exhibit enhanced mitochondrial respiratory capacity that prevents ROS overloading and the subsequent death of the cells upon mitochondrial respiration [60] (Fig. 2a).

CSCs in some tumors develop a highly compatible ROS scavenging system to keep ROS at low concentrations, thereby representing a quiescence nature [55]. CSCs exhibit a quiescence (or dormancy) state to resist against therapy [61] (Fig. 2a). CSCs have the capacity to synthesis glutathione (GSH) at high amounts, which is contributed to survival of the cells after radio- and chemotherapy [26,62]. GSH depletion can induce CSC ferroptosis, an iron-mediated cell death independent on caspase cascade [63]. Low ROS levels expand CSC population by regulation of extracellular-signal-regulated kinase (ERK) [1] and cyclooxygenase-2 (COX-2) [64]. In fact, there is a negative feedback interaction between ROS and COX-2 in CSCs. ROS is implicated in COX-2 induction [65]. COX-2 from TME, in turn, attenuates ROS to possibly enforce CSC enrichment [64] and metastasis [66], so CSCs are equipped with an efficient oxidant/antioxidant machinery acquiring a highly compatible redox system to habitat readily with the nearby milieu and to resist oxidative stress induced by radio- and chemotherapy [67] (Fig. 2b). Disruption of these adaptation mechanisms can be an effective strategy to break down CSC resistance.

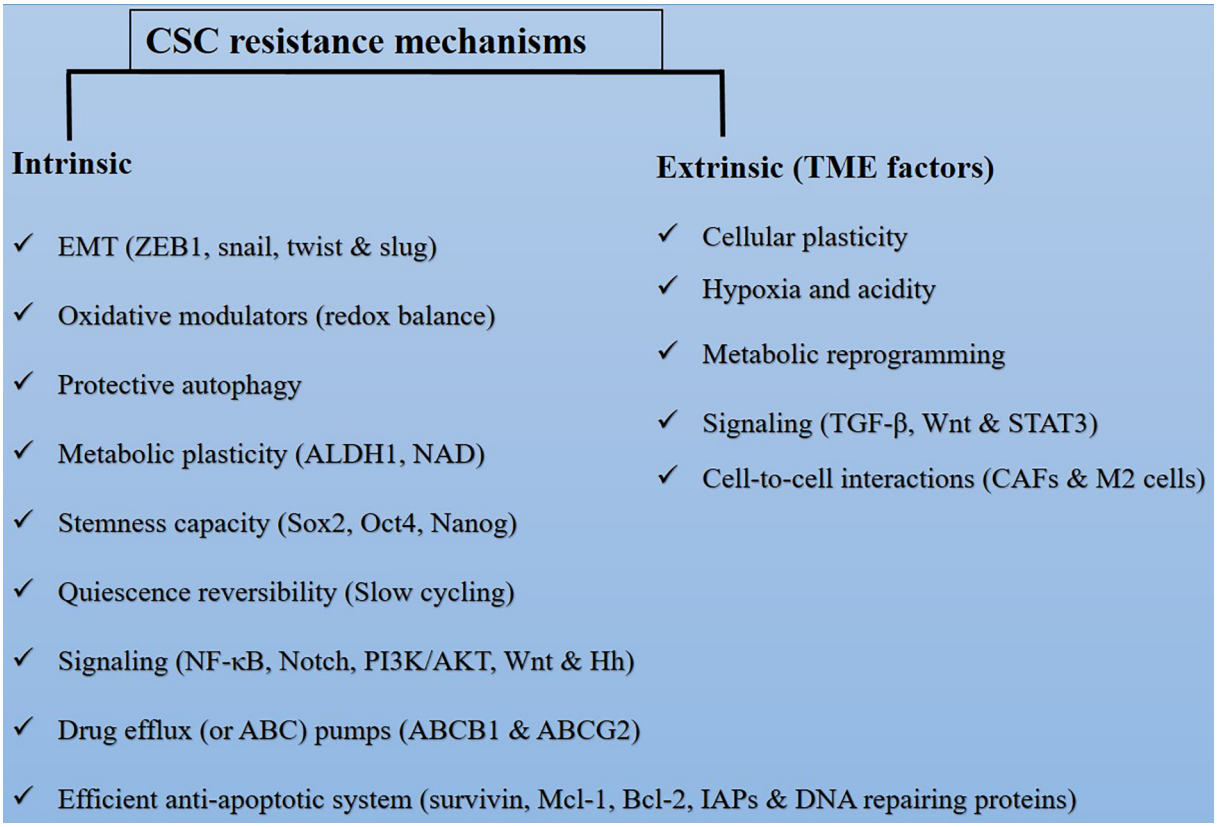

Fig. 1. Mechanisms of therapy resistance in cancer stem cells (CSCs). CSCs are inherently resistant to therapy. CSC resistance is also strengthened by signals from tumor microenvironment (TME). IAPs, inhibitor of apoptosis proteins; ALDH, aldehyde dehydrogenase; NAD, Nicotinamide adenine dinucleotide; NF-κB, nuclear factor-κB; PI3K, phosphoinositide 3-kinase; Hh, hedgehog; ABC, ATP-binding cassette; TGF-β, transforming growth factor-β; STAT3, signal transducer and activator of transcription3; CAF, cancer associated fibroblast; and M2, macrophage type 2.

2.1.4. Autophagy

CSCs are equipped with a protective autophagy machinery to maintain their stemness [68], keep them resistant to anoikis [69], and to help maintaining ROS at low concentrations [70]. Anoikis is a type of cell death resulted from inappropriate interactions between a cell type with its nearby extracellular matrix (ECM) [71]. Autophagy is protective upon cellular exposure to stressors (hypoxia and nutritional deprivation) [28]. CSCs maintain autophagy in an equilibrium in order to be able to promote resistance, so disruption of this protective autophagy system by either inhibition [72] or overactivation [73] could be therapeutic.

2.1.5. Inflammation

A degree of inflammation at early stages is protective against tumor progression, promoting coordination of responses from adaptive immune system [74] and tumor cell cytolysis [75]. Inhibition of this inflammatory state (acute phase) by factors like transforming growth factor-β (TGF-β) precludes immune responses, and thereby inducing tumorigenesis [76]. However, an elevation in the rate of inflammation (chronic phase) occurring after radio- and chemotherapy [40,77,78] and even following chemotherapy withdrawal [79] is contributed to promotion of therapy resistance. Chronic inflammation is a major inducer of CSC expansion [23] possibly causes activation of CSCs in their niche and enriches them outside the niche through inducing cancer cell/CSC dedifferentiation [46]. Thus, anti-inflammatory drugs such as aspirin can be served as an adjuvant for abrogating CSC resistance secondary to conventional therapies [80,81] (Fig. 3a).

2.1.6. From metabolic overview

Metabolic reprogramming (plasticity) is a shift in cellular bioenergetics in tumors [82] that is known as a feature of CSCs [83]. Tumor chemotherapy can be an inducer of metabolic reprogramming, and thus promoting resistance of CSCs. Resistance to chemotherapy occurs by conversion of aldehydes into weak carboxylic acids, and thereby reducing the chance of deposition for the toxic aldehydes within CSCs. This conversion is mediated by the activity of ALDHs in CSCs [30,84]. High ALDH activity is a unique feature for CSCs that distincts them from normal stem cells [1]. Among 19 members of ALDHs, ALDH1 is considered as a CSC biomarker [85] primarily linked to stemness and chemotherapy resistance [84].

CSCs would meet their metabolic demands efficiently (and further resistance to metabolic stressors) in three ways: (1) by inducing angiogenesis. CSCs (as a response to repeated radiation exposure) release factors (like insulin-like growth factor 1/IGF-1) to induce angiogenesis in the TME [86] that further allows for greater uptake of oxygen and nutrients [87]. (2) by shifting their phenotypes. CSCs exhibit diverse metabolic phenotypes across tumor types [87] or even in a same tumor [33], as for breast cancer that CSCs have epithelial- and mesenchymal-like phenotypes displaying metabolic differences. This metabolic plasticity facilitates CSC adaptation to stressors, such as nutrient burden and thus contributed to the growth of tumor [29,60]. Shifting the metabolic phenotype is possibly supported by a hypoxic TME [24], as it acts on cancer cells upon dedifferentiation into CSCs [88]. (3) by shifting the way of energy supply. CSCs would meet their metabolic demands in a versatile way. The cells can be highly glycolytic or depended on mitochondrial OXPHOS. This flexible metabolism may be due to the adaptation of the cells to a particular condition within the TME [83], and it helps them to fulfill their biosynthetic and bio-energetic demands so as to supply optimal nutrition for survival, proliferation and evasion [89]. Suppression of OXPHOS is linked to temporal necroptosis and sensitization of CSCs to chemotherapy [14,84]. However, a while after treatment with mitochondrial OXPHOS

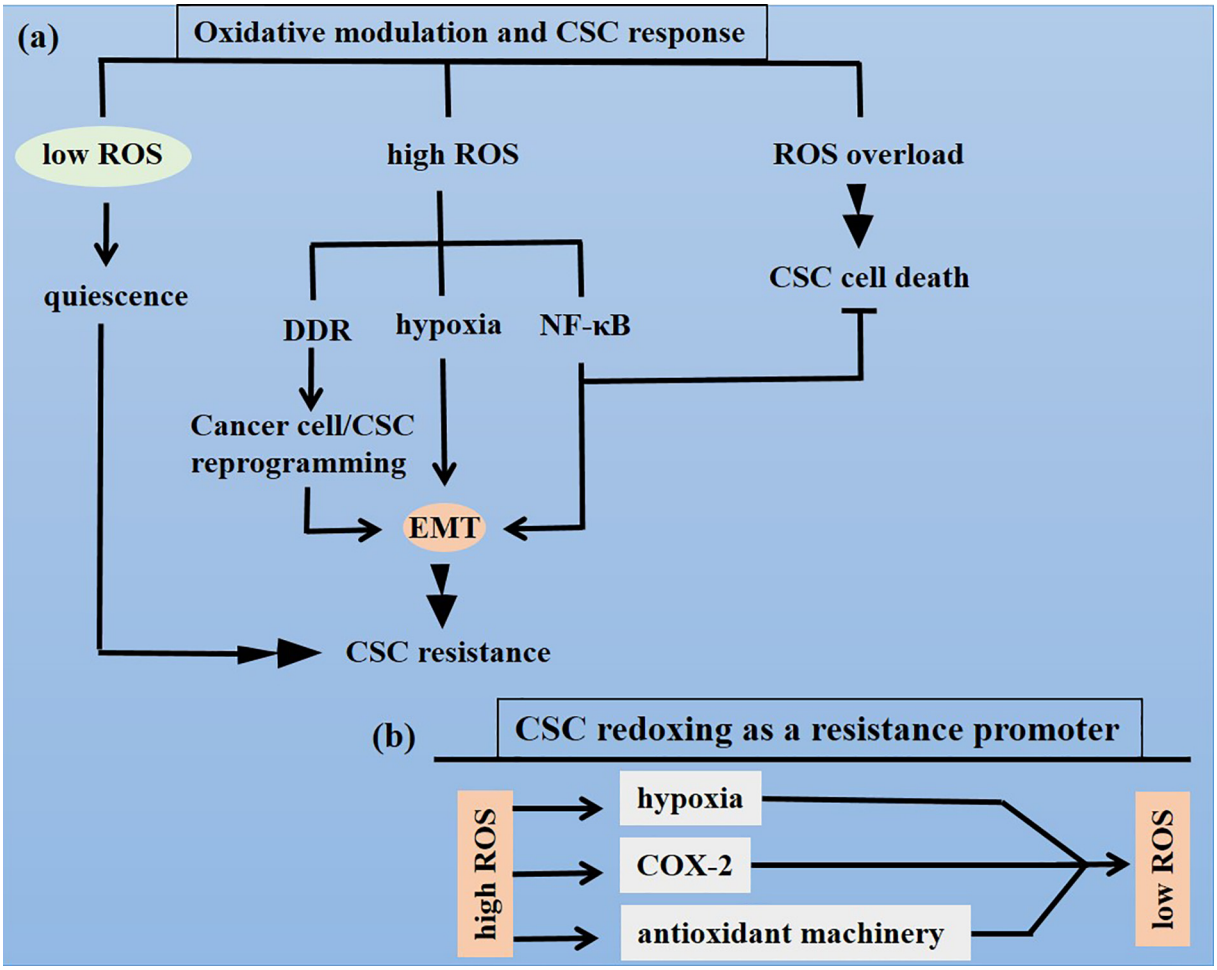

Fig. 2. Oxidative modulation in cancer stem cell (CSC) resistance and therapy. **(a)** CSCs maintain low reactive oxygen species (ROS) levels, which is for promoting CSC quiescence and therapy resistance. When encounter high ROS levels, the cells encounter hypoxia that further promotes therapy resistance. **(b)** The diagram in the lower left shows a redox balance in CSCs that acts as a promoter of therapy resistance. Disruption of this balance by ROS overloading would be an effective approach for promotion of CSC killing. DDR, DNA damage response; NF-κB, nuclear factor-κB; EMT, epithelial-mesenchymal transition; and COX-2, cyclooxygenase-2.

inhibitors like metformin, CSCs are reported to shift their metabolic demands from OXPHOS to glycolysis, and thus evolving resistance to such therapy [14,83]. Lactate is the product of this enhanced glycolysis creating an acidified milieu [87] for promotion of ECM degradation (via activation of proteases) [90] and resistance to chemotherapy [91]. The metabolic shifting capability indicates that the cells are highly adaptive to alterations in the surrounding milieu. Nicotinamide adenine dinucleotide (NAD) is a key player in cancer cell metabolism. NAD is

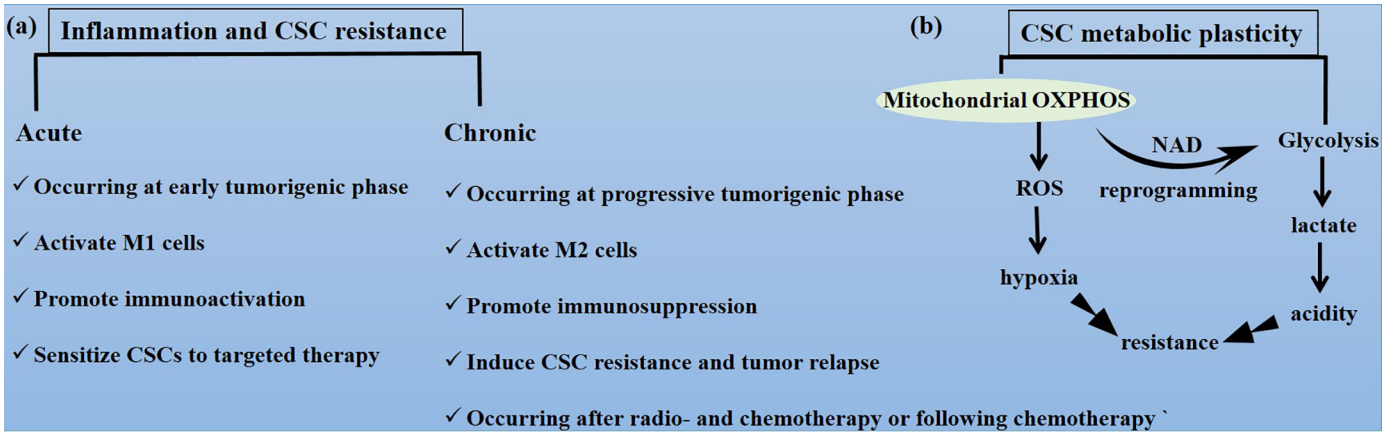

Fig. 3. Relation between inflammation metabolic plasticity with cancer stem cell (CSC) resistance. **(a)** Inflammation is divided into acute and chronic phases. The acute phase works against CSC resistance by activating immune system, while the chronic phase that could be induced by radio- and chemotherapy is a resistance promoter. **(b)** Mitochondrial oxidative phosphorylation (OXPHOS) is a primary metabolic demand in CSCs. However, upon exposure to an oxidative burden, the cells would shift the energy requirement toward glycolysis. This metabolic plasticity is thus contributed to resistance of CSCs. M1, macrophage type 1; M2, macrophage type 2; NAD, Nicotinamide adenine dinucleotide; and ROS, reactive oxygen species.

contributed to adaptation of CSCs to metabolic burden through switching their metabolism toward an energy efficient respiration [60]. NAD also renders CSC self-renewal through driving transcription of inhibitor of differentiation (ID) members [92], so maintaining CSC self-renewal is occurring in the context of a metabolic scheme (Fig. 3b).

2.1.7. CSC plasticity

CSC plasticity is a main predicament in cancer targeted therapies. CSCs are able to sequentially transition between stem- (an EMT phenotype) and non-stem (a mesenchymal-epithelial transition [MET] phenotype) [34,93], and between quiescence (drug resistance) and proliferation (drug sensitive) [36] states. This is for promotion of a heterogeneous population of cells within a tumor easily adapt with changes in the TME [94], hard to combat by conventional chemotherapies. In fact, the cellular plasticity that reconciles inherited (hierarchical) and acquired (stochastic) theories described for tumorigenesis of a cell type [95].

EMT activation in cancer cells converts them into CSCs [22]. This conversion or so called de(retro)differentiation program allows genetic reconstruction (ex, Oct4 overexpression) [96], metabolic reprogramming and therapy resistance [88,96]. CSCs keep their EMT phenotype until depositing in the distant sites of metastasis (a grow-to-go phenotype) where they change their phenotype toward attaining a MET morphology to proliferate rapidly causing tumor outgrowth (a go-to-grow phenotype) [24,54]. This rapid cellular proliferation leads to hypoxia in the nearby milieu, thereby exacerbating tumor resistance to therapy [97] (Fig. 4a). This phenotypic plasticity accounts for diverse levels of therapy resistance among CSCs, which is a potent threat [98]. Therefore, stabilization of plasticity offers a great benefit for eradication of tumors even at high stages. This is applicable by targeting EMT [47,99] using either inhibitors of EMT mediators, such as twist, slug and snail [21], or addressing central drivers of this process, including TGF-β and Notch [47]. CD24 is a marker of CSC plasticity implicated in EMT regulation and drug resistance, so it could be a potential target [98].

The pool of CSCs is in the quiescence state following radio- and chemotherapy [26,100]. The quiescence state allows maintaining their survival [100] and acquiring more resistant phenotype. When therapy is removed, they exhibit more sensitive proliferative phenotype [101]. The rate of expression for stemness markers is more pronounced in proliferative than the rate in quiescent cells [102]; this possibly helps an increase in the chance of cellular evading from therapy. The quiescent cells although have minimal metabolic activity [49], they display high ALDH activity [6] for metabolizing drugs into less toxic agents, and thus being more aggressive (invasive) upon induction [49,103]. The quiescence cells reside in avascular (hypoxic) niches [104], and the quiescent state gives CSCs sufficient time to repair damages to their genome upon exposure to radio- and chemotherapy [12,58], thus enhancing their invasiveness [12] (Fig. 4b).

Plasticity infers that not necessarily CSCs but every cells within the TME have potentials to initiate a tumor [105,106]. All cancer cells are inherently plastic and are able to function as CSCs when required [5]. This requirement is under the instruction of the TME, and that the intrinsic plasticity (rather than multipotency) is identified to be linked to tumorigenic potential of CSCs [94]. The plastic capacity infers that hierarchy is not essentially related to CSCs, as reported beforehand [107], and a desired therapeutic scheme relies on targeting plasticity within the TME (including CSC and non-CSC cells), which is an effective strategy for removing the possibility of non-CSC/CSC conversion [46,108], and so abolishing the chance of tumor relapse.

2.1.8. Drug transporters

A hallmark of CSCs (that distinct them from most other cancer cells) is that the cells robustly (and stably) express drug transporters on the cell surface, and thus exhibiting a multi-drug resistance (MDR) phenotype [15,32,109,110]. These are unidirectional cellular pumps charged by ATP, known as ATP-binding cassette (ABC) transporters. Chemotherapeutic agents are the substrates of the pumps, and the efflux of these anti-cancer agents is occurring after activation of the pumps, reducing drug accumulation within the cells [35,56,111,112], which is possibly for maintaining cellular viability [113] and stemness [112,114]. Effective targeting of ABC efflux pumps is important to make CSCs penetrable to chemotherapeutics. Examples of these drug transporters are ABC subfamily B, member 1 (ABCB1) (also known as MDR1) [32,56] and ABCG2 (also called Breast Cancer Resistance Protein/BCRP) [32].

2.1.9. Resistance receptors

Retinoic-acid-receptor-related orphan receptor gamma (RORγ) is a nuclear hormone receptor enriched highly in CSCs relative to non-stem cells. RORγ mediates interactions between CSC niche with the immune T cells for regulation of super-enhancer oncogenic network in CSCs, as well as T cell differentiation. This co-option between immune system and CSC oncogenesis is for promotion of cancer aggressiveness [26]. In addition, interaction between progranulin with its sortilin receptor expressed on cancer cells promotes their dedifferentiation into CSCs and therapy resistance [115]. Upregulation of estrogen receptors such as G-protein coupled estrogen receptor (GPER) has also been identified to induce CSC expansion and drug resistance in breast cancer [116].

2.1.10. Epigenetic regulations

2.1.10.1. MiRNAs and long non-coding RNAs (lncRNAs). MiRNAs are a class of small noncoding RNAs [117] that are bind to the target mRNAs and cause mRNA destabilization and/or translational inhibition [1]. LncRNAs are transcripts containing over 200 nucleotides that do not translate into proteins. LncRNAs may act through interactions with miRNAs and target their downstream pathways [118]. MiRNAs critically regulate CSC hallmarks, including survival [117] and drug resistance/sensitivity [119]. The effects of miRNAs and lncRNAs on CSC resistance are mainly implicated through targeting CSC stemness; these effects could be either positive [118,120–123] or negative [124–127]. These diverse functional features are tumor- and tissue-specific [1]. A same miRNA may target diverse gene/s in different tumors, and so taking different functions. A change in the level of miRNA may also influence its function. For instance, maintenance of CSC fraction is mediated in ovarian and colorectal cancers by the respective high and low levels of miR-328 [1].

3. TME is a key contributor of CSC resistance

TME has long been known as a promoter of tumor growth [24]. CSCs have symbiotic relations with the TME [128]. CSCs are exposed constitutively to multiple tumorigenic signals emanated by other TME cells. Tumor/stroma cross-talking is required for promotion of CSC resistance to chemotherapy [129]. TME promotes CSC heterogeneity and adaptation, the two resistance drivers [36]. Cancer associated fibroblasts (CAFs) are one of the key cells within the TME implicated in cancer progression. CAFs produce ECM proteins [130] to occupy a niche for CSCs [131]. A stiffened ECM hampers drug penetration to the CSC niches [131], stimulates TME hypoxia [132], and is associated with the cancer cell/CSC dedifferentiation [133], all of which are contributed to the tumor resistance and recurrence. CAFs also promote ECM degradation [134] for CSC dissemination and invasion [90]. Cellular reprogramming between cancer cells, CSCs and CAFs is one of the major mediators of therapy resistance in CSCs (Fig. 4c). Co-culture of non-lytic $CD8^+$ T cells with cancer cells is reported to induce dedifferentiation of the cells into CSC-like cells [100]. Dedifferentiation is also induced by tumor necrosis factor-α (TNF-α) [135] released by TME cells. TME is exposed constantly with conditions like hypoxia and metabolic reprogramming. Hypoxia is a promoter of cancer cell/CSC dedifferentiation (Fig. 4d). Enhanced hypoxia, as mentioned, along with glycolysis (causing increased acidity) are resistance promoters

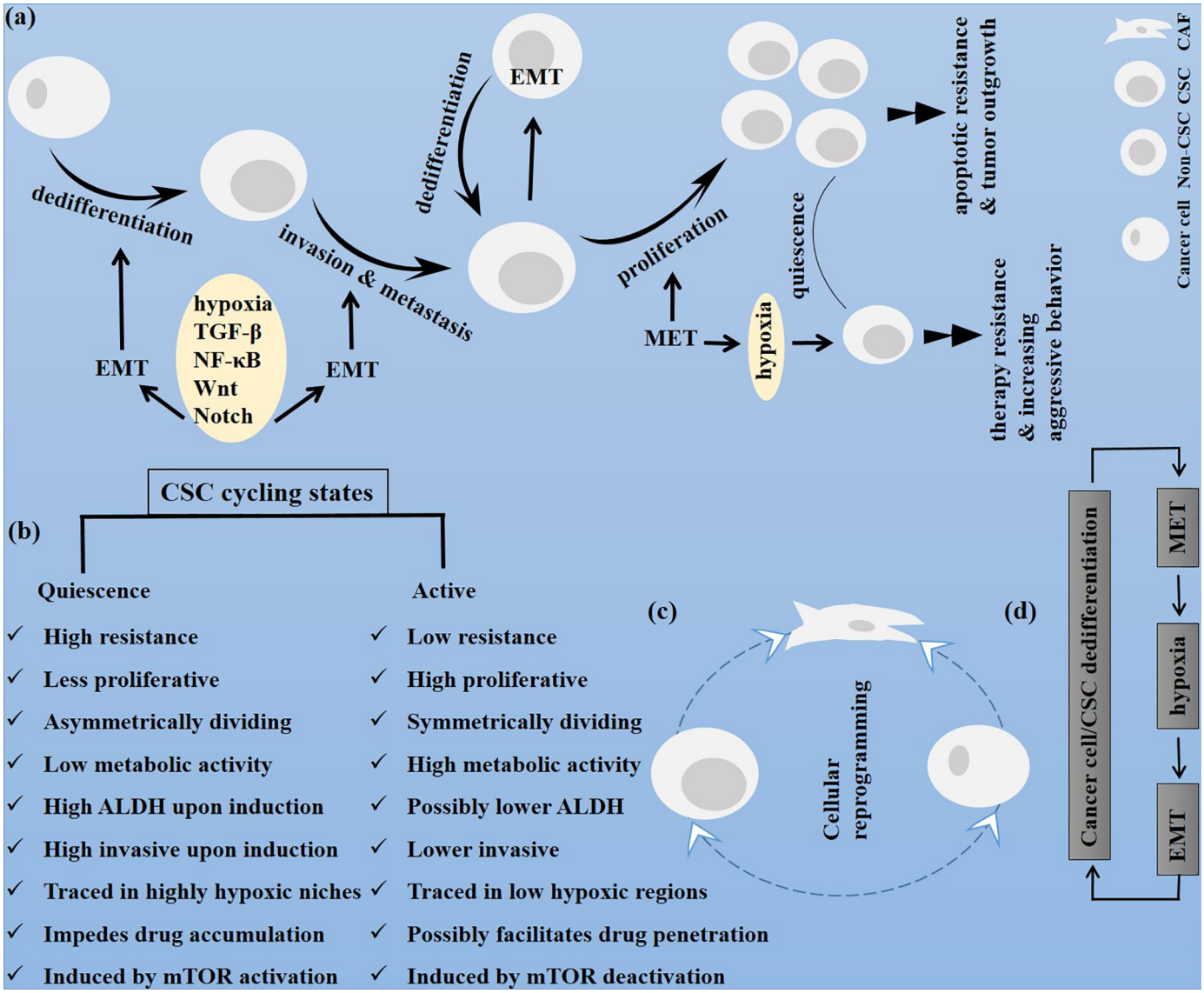

Fig. 4. Cellular plasticity as a resistant promoter. **(a)** Cancer stem cells (CSCs) are highly plastic cell types that are able to acquire diverse phenotypes (stem/non-stem) and cellular states (quiescence/proliferation) so as to evade therapy and to enhance their capacity to outgrowth the tumor vigorously. Epithelial-mesenchymal transition (EMT) induces metabolic reprogramming and dedifferentiation of cancer cells into CSCs. The cells would keep their EMT features in order to reach the distant sites of metastasis. Here, they change their phenotype by acquiring a mesenchymal-epithelial transition (MET) morphology to proliferate and expand their numbers. MET is also an inducer of CSC quiescence. CSC plasticity is controlled essentially by tumor microenvironment (TME) hypoxic condition and signaling, including transforming growth factor-β (TGF-β), nuclear factor-κB (NF-κB), Wnt and Notch. **(b)** The diagram in the lower right illustrates relation between CSC cycling states with resistance. ALDH, aldehyde dehydrogenase. **(c)** The diagram shows versatile cellular reprogramming in relation with stroma (i.e. cancer associated fibroblasts [CAFs]), and **(d)** the diagram in the lower left illustrates how EMT/MET cellular states in interaction with hypoxia promote CSC enrichment and therapy resistance.

[87,91]. TME macrophage type 2 (M2) cells are another important player in CSC resistance to therapy [136,137] by promoting both of these conditions [97], and by coordinating interplay between stemness-related signals (STAT3 and Hh) [138]. IL-4 released from CSCs to the TME tightens M2/CSC cross-talking, acting through promotion of M2 polarity [139] and protection of CSCs from apoptosis [18].

4. Signaling pathways of therapy resistance

There are a number of signaling implicated in resistance of CSCs. In the Fig. 5, a summary of the roles for these multi-tasking pathways is presented.

4.1. TGF-β

TGF-β is a major promoter of CSC stemness and resistance to radio- [140] and chemotherapy [141]. TGF-β is released by multiple cells within the TME in response to hypoxia [142]. TGF-β stimulates CSC stemness through induction of EMT [143,144] and ECM stiffness [145]. TGF-β also promotes CSC dormancy [146]. Co-activation of TGF-β and Notch signaling causes CSCs exhibiting a diversity of EMT phenotypes (phenotypic plasticity), and residing at different regions of tumor from tumor core (a place for ALDH1$^+$ hybrid E/M CSCs) to the tumor periphery or invasive edge (a site for CD44$^+$ mesenchymal CSCs) [40]. The cells reside in the core are mostly proliferative, and (despite being hypoxic) [110] they are more sensitive to nutritional and oxidative stressors than the cells in the edge [90], so a core-to-edge migration possibly gives them more chance to survive and to promote tumor

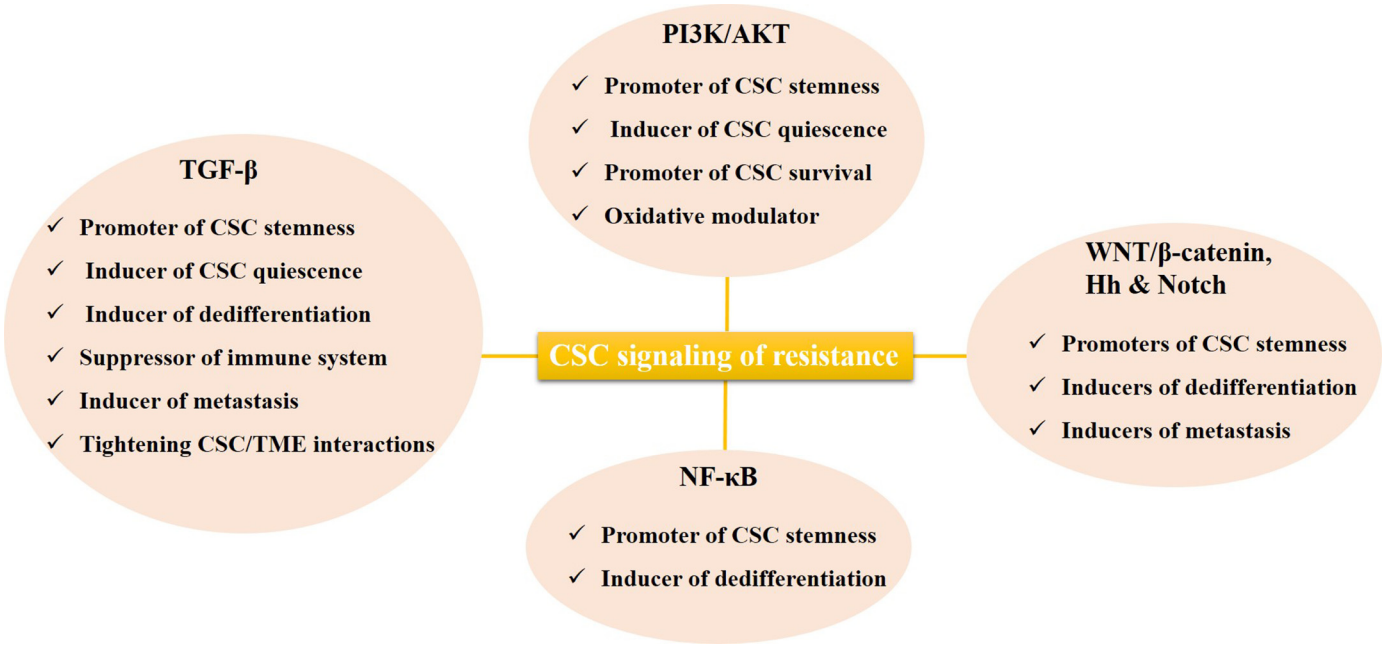

Fig. 5. The diagram displaying contribution of multi-tasking signaling pathways to resistance of cancer stem cells (CSCs). Transforming growth factor-β (TGF-β), phosphoinositide 3-kinase (PI3K)/AKT, nuclear factor-κB (NF-κB), Wnt/β-catenin.

budding (a pro-metastatic process initiated by EMT) [147].

4.2. Phosphoinositide 3-kinase (PI3K)/AKT

Dysregulation of the PI3K/AKT signaling occurs frequently in human tumors. AKT promotes survival, metastasis and chemoresistance by acting through modulation of downstream effectors, including caspases, nuclear factor-κB (NF-κB) and mammalian target of rapamycin (mTOR) family. PI3K induces constitutive activation of NF-κB to promote CSC chemoresistance [56]. The PI3K/AKT/mTOR pathway potentiates antioxidative machinery [54] and promotes quiescence [148] in CSCs. Interaction between AKT with Notch stimulated by hypoxia is for stemness maintenance and drug resistance [50]. PI3K signaling in CSCs can be targeted preferentially using pan-PI3K inhibitors, such as B591 [149].

4.3. NF-κB

NF-κB is a crucial mediator of inflammation (acute and chronic) in tumors linking inflammation with stemness [23]. NF-κB activates hypoxia related stemness signaling [150], and is associated with both early (through induction of EMT) [22,151] and late stage of tumorigenesis [22]. NF-κB reverts ROS-induced apoptotic cell death in CSCs [30], as shown in the Fig. 2a.

4.4. Signal transducer and activator of transcription3 (STAT3)

STAT3 is an immune escape mediator mainly released by cancer cells toward the TME [152]. STAT3 is important for maintaining stemness [118,153], immune escaping [136] and resistance of CSCs to radio- [137] and chemotherapy [154]. Stemness is promoted by interaction between STAT3 with NF-κB [155]. This cross-talking is tightened by TNF-α [151].

4.5. Wnt/β-catenin

Wnt/β-catenin, Hedgehog (Hh) and Notch take roles for regulation of CSC self-renewal and differentiation [156]. MRNA expression of β-catenin is increased after radiotherapy [59]. CAFs secrete Wnt into the TME [157]. Activation of Wnt/β-catenin is mediated by SOX2 expression in CSCs [20]; this activation is for promoting cancer cell/CSC dedifferentiation implicated in chemoresistance [157,158]. An EMT program is activated by Wnt in these newly formed cells acting as a promoter of tumor budding (and the resulting metastasis) and drug resistance [42,147].

4.6. Hh

Hh is another signaling related to CSC stemness [112,156] and chemoresistance [25]. To do such actions, Hh signaling regulates ABCG2 efflux pump and ALDH activity [112,159]. Cancer cells produce Hh ligand responsible for reprogramming of CAFs toward producing a supportive niche, which is for maintenance of CSC stemness [145]. Activation of Hh ligand in cancer cells stimulated by nutritional deprivation (stress) is also contributed to dedifferentiation of these cells into CSCs [9].

4.7. Notch

CSCs activate Notch for promotion of resistance to radio- [156] and chemotherapy [17,125]. Aberrant Notch activation helps acquiring EMT and self-renewal properties in CSCs [50]. P53 is placed upstream of Notch to exert protective effects against DNA damage [17]. Expression of different types of Notch in different tumors and cross-talking between this signaling with Wnt and Hh pathways add complexity for targeting these pathways [160].

5. Overcome drug resistance in CSCs

CSCs are prone dynamically to the phenotypic switching between stem and non-stem states, causing variations in their density and spatial distribution, thus complicating targeting for cancer therapy; this would possibly make different levels of resistance [8,58], and thereby variations in the response rate of CSCs to chemotherapeutics even in a same tumor [161], with a rise in the number of CSCs an increase in the dose of radio- and/or chemotherapy is required. Differentiation of CSCs from normal stem cells within tissues or organs is possibly the first step for combating therapy resistance. Disruption of normal stem cells

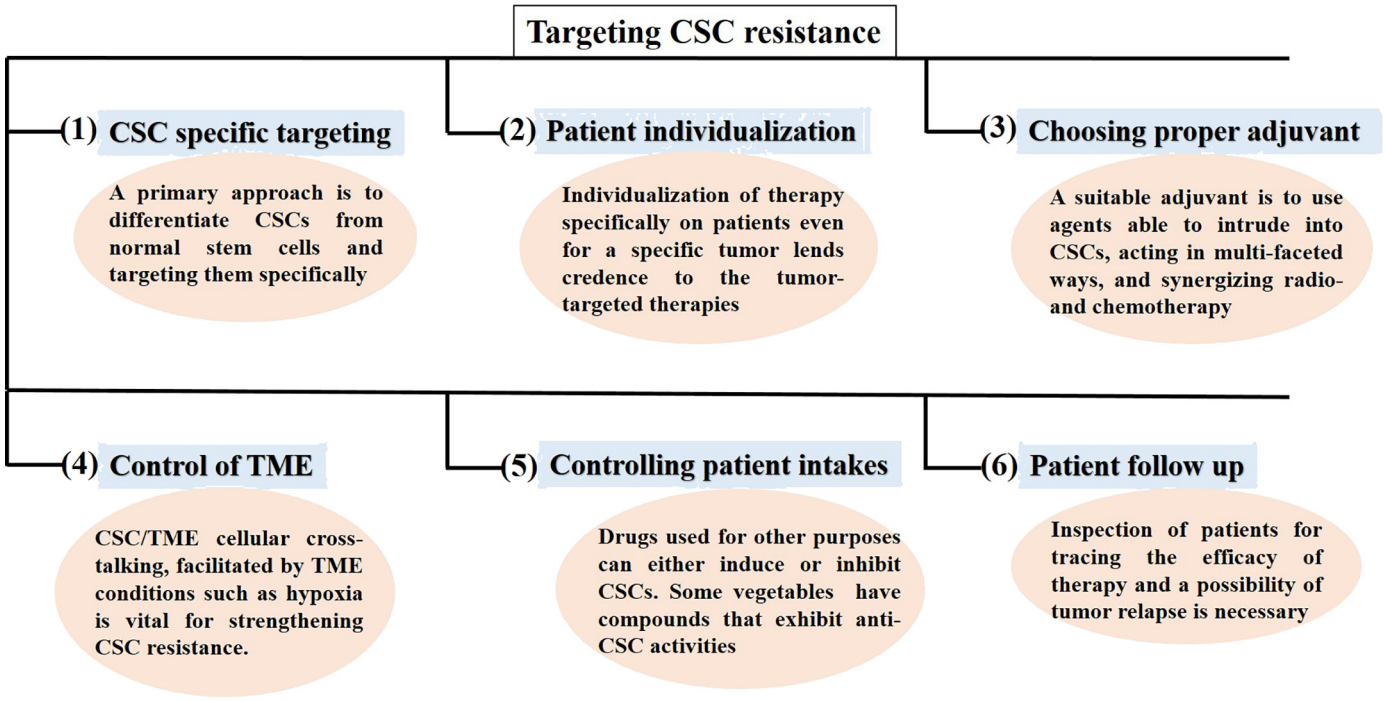

Fig. 6. The diagram shows a 6-step schedule to overcome drug resistance in cancer stem cells (CSCs). CSCs have close interactions with the nearby tumor microenvironment (TME) for promotion of therapy resistance, and due to implication of both intrinsic and extrinsic factors in promotion of CSC resistance, a multi-faceted adjuvant strategy addressing dominant cross-talking between CSCs and their nearby milieu is required. CSCs exhibit different traits among patients with the same type of tumor, so individualization of therapy, if applicable, would enhance the efficacy of therapy. Dietary and drug intake control also has its own value aiming to observe a desired response from patients to adjuvant therapy.

secondary to procedures aiming to target CSCs can cause regrowth of tumor due to the essence of normal stem cells for post-therapy tissue regeneration and homeostasis [37]. CSC profiling using nanosensors [161] or resistant embryonic stem cells [162] is promising to identify and to survey behavior of the cells within tumor tissue, being fundamental for exploring new treatment modalities and, thus, improving clinical management of tumor.

It is important to note that the behavior of CSCs is different from one patient to another, so the next step would be individualization of therapy specifically on patients, as reported for GBM [163]. When adjuvant therapy is considered, the agents chosen for such treatment modalities must be less toxic on patients [164]. CSCs have efficient drug transporter pumps restricting accumulation of drugs within the cells, so when a drug is prescribed for a patient, its penetrability must be checked beforehand. The patient may require suppressors of ABC pumps alongside, or a drug exchange.

CSCs inherit most of the resistance mechanisms from the tumorigenic milieu nearby, so a special focus in CSC-targeted therapies must be on breaking down the CSC/TME interplay; these cross connections are tightly facilitated by TME conditions, especially hypoxia; CSCs evolve a highly resistance phenotype in the hypoxic milieu, so abolishing this condition by approaches like hyperoxia could be an effective strategy for enhancing CSC sensitization [165]. Phenotypic plasticity of CSCs is determined by the TME, and it can be targeted using phenotypic stability factors [47]. It is impossible to target all signaling transduced between CSCs and the TME, thus a close inspection must be taken on recognition of major promoters of resistance and targeting them for cancer therapy. TGF-β is a key signaling for promotion of CSC resistance in a variety of tumors, so it could be a promising target. CSCs, as described, are highly responsive to any variations in the oxidant/antioxidant balance, acquiring resistance upon exposure to both low and high ROS levels (Fig. 2a). Due to the importance of low ROS levels in CSC maintenance (as described), the antioxidant delivery may not be a therapeutic choice for some cancer, instead, this approach may even cause CSC promotion [27]; this is indicative of the sensitivity of oxidative modulating approaches for cancer therapy that calls for development of modalities aiming for disruption of the redox balance in CSCs possibly by oxidative overloading as a promising strategy.

Control of patient drug intake has a critical role in CSC response to therapy. For example, morphine administered to relieve symptoms in breast cancer patients can adversely cause CSC enrichment and chemoresistance [42], while disulfiram prescribed to treat alcoholism has strong activity against CSC resistance in such tumor [164], and spironolactone (an anti-hypertensive drug) impairs DNA damage repair in CSCs [166]. In diabetic patients, high blood glucose can cause WNT activation, and thus increasing the risk of tumor [167]. Metformin prescribed for these patients reduces this risk through increasing cellular uptake of glucose. There is also a report that diabetes patients receiving metformin show more response to chemotherapy [168]. However, for tumors upon progression this therapy may not be effective due to the plastic capability of CSCs in shifting the energy demands, as discussed.

Modulation of patient dietary intake could also be effective. For instance, broccoli is enriched in sulforaphane and quercetin [169] that are known to revert apoptosis resistance in CSCs [22,169]. Curcumin found in peppers is known to hamper CSC self-renewal [22], and resveratrol found in grapes reduces CSC stemness and enhances their sensitization to radiotherapy [170]. In addition, a close inspection and control must be taken over patient's bad habits. For instance, there is a positive relation between cigarette smoking with enhanced ABCG2 pump activity [112] that demands for possibly higher doses of chemotherapy (than the dose required for non-smokers). Therefore, it is necessary to closely inspect patient intakes (drug [for other purposes] and dietary) and bad habits and modulate them in order to enhance the efficacy of targeted therapies against CSC resistance. Without a doubt, patients receiving adjuvant therapies for targeting CSC resistance will need short-term follow up, which is for tracing the efficacy of therapy. Patients may require modification of drugs (dose and type). Long-term

follow up is also essential, especially for advanced stage tumors; this is for observing a possibility of tumor relapse. The diagram in the Fig. 6 presents a summary of a 6-step schedule for targeting CSC resistance and thereby enhancing the efficacy of therapy.

6. Conclusion

Information provided in this review represents a network of the key nodes implicated in regulation of CSC resistance to therapy. CSC resistance is complex and multi-factorial. Exposure of CSCs with an array of endo- or exogenous factors that would be more intense upon tumor progression could make the cells highly compatible for regulation of tumor behavior and responding to a type of therapy. Their versatile function, plastic phenotype and diverse divisional features (a/symmetry) [46] unveil that the cells can shape the tumor for their desire, resulting in therapy failure. This would make necessitate development of neoadjuvant therapies highly qualified to penetrate into the cells and to target major drivers of tumor resistance in a multi-faceted way, having extended durability and exhibiting the least adversarial effects on surrounding normal tissues nearby. Harassment of CSC resistance mechanisms and hampering CSC re-emergence would be most effective when therapeutic approaches are designed individually for patients and the type of tumor they have [55].

Declaration of competing interest

No conflict of interest to declare.

References

[1] A.K. Srivastava, et al., Inhibition of miR-328-3p impairs cancer stem cell function and prevents metastasis in ovarian cancer, Cancer research, 2019: p. canres. (2018) 3668.
[2] K.M. Govaert, et al., Hypoxia after liver surgery imposes an aggressive cancer stem cell phenotype on residual tumor cells, Ann. Surg. 259 (4) (2014) 750–759.
[3] C. Fan, et al., Cancer-initiating cells derived from human rectal adenocarcinoma tissues carry mesenchymal phenotypes and resist drug therapies, Cell Death Dis. 4 (10) (2013) e828.
[4] T. Idowu, et al., Amphiphilic modulation of glycosylated antitumor ether lipids results in a potent triamino scaffold against epithelial cancer cell lines and BT474 cancer stem cells, J. Med. Chem. 60 (23) (2017) 9724–9738.
[5] Y. Brown, S. Hua, P.S. Tanwar, Extracellular matrix-mediated regulation of cancer stem cells and chemoresistance, Int. J. Biochem. Cell Biol. 109 (2019) 90–104.
[6] L.A. Quayle, P.D. Ottewell, I. Holen, Chemotherapy resistance and stemness in mitotically quiescent human breast cancer cells identified by fluorescent dye retention, Clinical & Experimental Metastasis 35 (8) (2018) 831–846.
[7] L. Ricci-Vitiani, et al., Identification and expansion of human colon-cancer-initiating cells, Nature 445 (7123) (2007) 111.
[8] M. Baumann, M. Krause, R. Hill, Exploring the role of cancer stem cells in radioresistance, Nat. Rev. Cancer 8 (7) (2008) 545.
[9] S. Mondal, K. Bhattacharya, C. Mandal, Nutritional stress reprograms dedifferentiation in glioblastoma multiforme driven by PTEN/Wnt/Hedgehog axis: a stochastic model of cancer stem cells, Cell death discovery 4 (1) (2018) 110.
[10] C. Yan, et al., Enhanced autophagy in colorectal cancer stem cells does not contribute to radio-resistance, Oncotarget 7 (29) (2016) 45112.
[11] F. Karandish, et al., Peptide-targeted, stimuli-responsive polymersomes for delivering a cancer stemness inhibitor to cancer stem cell microtumors, Colloids Surf. B: Biointerfaces 163 (2018) 225–235.
[12] R.J. Atkins, et al., Cell quiescence correlates with enhanced glioblastoma cell invasion and cytotoxic resistance, Exp. Cell Res. 374 (2) (2019) 353–364.
[13] P. Wang, et al., Cancer stem-like cells can be induced through dedifferentiation under hypoxic conditions in glioma, hepatoma and lung cancer, Cell death discovery 3 (2017) 16105.
[14] P. Sancho, et al., MYC/PGC-1α balance determines the metabolic phenotype and plasticity of pancreatic cancer stem cells, Cell Metab. 22 (4) (2015) 590–605.
[15] D. Wang, J.T.M. Plukker, R. Coppes, Cancer stem cells with increased metastatic potential as a therapeutic target for esophageal cancer, Seminars in Cancer Biology, Elsevier, 2017.
[16] E. Yokoi, et al., Lurbinectedin (PM01183), a selective inhibitor of active transcription, effectively eliminates both cancer cells and cancer stem cells in preclinical models of uterine cervical cancer, Investig. New Drugs (2018) 1–10.
[17] L. Yu, et al., Cisplatin selects for stem-like cells in osteosarcoma by activating Notch signaling, Oncotarget 7 (22) (2016) 33055.
[18] M. Todaro, et al., Colon cancer stem cells dictate tumor growth and resist cell death by production of interleukin-4, Cell Stem Cell 1 (4) (2007) 389–402.
[19] A.K. Iyer, et al., Role of integrated cancer nanomedicine in overcoming drug resistance, Adv. Drug Deliv. Rev. 65 (13–14) (2013) 1784–1802.
[20] T. Andey, et al., Cationic lipoplexes for treatment of cancer stem cell-derived murine lung tumors Transforming growth factor-β (TGF-β), Nanomedicine 18 (2019) 31–43.
[21] M.-Y. Chou, et al., Sox2 expression involvement in the oncogenicity and radiochemoresistance of oral cancer stem cells, Oral Oncol. 51 (1) (2015) 31–39.
[22] W. Zhou, et al., Dietary polyphenol quercetin targets pancreatic cancer stem cells, Int. J. Oncol. 37 (3) (2010) 551–561.
[23] D.-W. Yeh, et al., Interplay between inflammation and stemness in cancer cells: the role of toll-like receptor signaling, J Immunol Res 2016 (2016).
[24] A. Dhawan, et al., Mathematical modelling of phenotypic plasticity and conversion to a stem-cell state under hypoxia, Sci. Rep. 6 (2016) 18074.
[25] X. Chen, et al., Epithelial mesenchymal transition and hedgehog signaling activation are associated with chemoresistance and invasion of hepatoma subpopulations, J. Hepatol. 55 (4) (2011) 838–845.
[26] N.K. Lytle, et al., A multiscale map of the stem cell state in pancreatic adenocarcinoma, Cell 177 (3) (2019) 572–586 (e22).
[27] S. Ma, et al., CD133+ HCC cancer stem cells confer chemoresistance by preferential expression of the Akt/PKB survival pathway, Oncogene 27 (12) (2008) 1749.
[28] L. Wang, et al., Enrichment of prostate cancer stem-like cells from human prostate cancer cell lines by culture in serum-free medium and chemoradiotherapy, Int. J. Biol. Sci. 9 (5) (2013) 472.
[29] M. Luo, et al., Targeting breast cancer stem cell state equilibrium through modulation of redox signaling, Cell Metab. 28 (1) (2018) 69–86 (e6).
[30] N. Yip, et al., Disulfiram modulated ROS–MAPK and NFκB pathways and targeted breast cancer cells with cancer stem cell-like properties, Br. J. Cancer 104 (10) (2011) 1564.
[31] M.T. Mueller, et al., Combined targeted treatment to eliminate tumorigenic cancer stem cells in human pancreatic cancer, Gastroenterology 137 (3) (2009) 1102–1113.
[32] A. Eramo, et al., Chemotherapy resistance of glioblastoma stem cells, Cell Death Differ. 13 (7) (2006) 1238.
[33] K.-m. Lee, et al., MYC and MCL1 cooperatively promote chemotherapy-resistant breast cancer stem cells via regulation of mitochondrial oxidative phosphorylation, Cell Metab. 26 (4) (2017) 633–647 (e7).
[34] A. Fesler, et al., Overcoming chemoresistance in cancer stem cells with the help of microRNAs in colorectal cancer, Future Medicine 9 (6) (2017) 793–796.
[35] H. Dianat-Moghadam, et al., Cancer stem cells-emanated therapy resistance: implications for liposomal drug delivery systems, J. Control. Release 288 (2018) 62–83.
[36] B.C. Prager, et al., Cancer stem cells: the architects of the tumor ecosystem, Cell Stem Cell 24 (1) (2019) 41–53.
[37] S. Dingwall, et al., Neoplastic human embryonic stem cells as a model of radiation resistance of human cancer stem cells, Oncotarget 6 (26) (2015) 22258.
[38] M. Reina-Campos, et al., Metabolic reprogramming of the tumor microenvironment by p62 and its partners, Biochimica et Biophysica Acta (BBA)-Reviews on Cancer 1870 (1) (2018) 88–95.
[39] D. Buenrostro, et al., Early TGF-β inhibition in mice reduces the incidence of breast cancer induced bone disease in a myeloid dependent manner, Bone 113 (2018) 77–88.
[40] F. Bocci, et al., Toward understanding cancer stem cell heterogeneity in the tumor microenvironment, Proc. Natl. Acad. Sci. 116 (1) (2019) 148–157.
[41] K.J. Lenos, et al., Stem cell functionality is microenvironmentally defined during tumour expansion and therapy response in colon cancer, Nat. Cell Biol. 20 (10) (2018) 1193.
[42] D.-G. Niu, et al., Morphine promotes cancer stem cell properties, contributing to chemoresistance in breast cancer, Oncotarget 6 (6) (2015) 3963.
[43] M.-J. Wu, et al., Epithelial-mesenchymal transition directs stem cell polarity via regulation of mitofusin, Cell Metab. 29 (4) (2019) 993–1002 (e6).
[44] J. Caramel, M. Ligier, A. Puisieux, Pleiotropic roles for ZEB1 in cancer, Cancer Res. 78 (1) (2018) 30–35.
[45] Z. Wang, et al., Pancreatic cancer-initiating cell exosome message transfer into noncancer-initiating cells: the importance of CD44v6 in reprogramming, J. Exp. Clin. Cancer Res. 38 (1) (2019) 132.
[46] M. Najafi, K. Mortezaee, R. Ahadi, Cancer stem cell (a) symmetry & plasticity: tumorigenesis and therapy relevance, Life Sci. 231 (2019) 116520.
[47] P. Yaswen, Reinforcing targeted therapeutics with phenotypic stability factors, Cell Cycle 13 (24) (2014) 3818–3822.
[48] D. Filipponi, et al., DNA damage signaling-induced cancer cell reprogramming as a driver of tumor relapse, Mol. Cell 74 (4) (2019) 651–663.
[49] I.-C. Lee, Y.-C. Wu, W.-S. Hung, Hyaluronic acid-based multilayer films regulate hypoxic multicellular aggregation of pancreatic cancer cells with distinct cancer stem-cell-like properties, ACS Appl. Mater. Interfaces 10 (45) (2018) 38769–38779.
[50] Z. Zhang, et al., Hypoxia potentiates gemcitabine-induced stemness in pancreatic cancer cells through AKT/Notch1 signaling, J. Exp. Clin. Cancer Res. 37 (1) (2018) 291.
[51] S. Cannito, et al., Redox mechanisms switch on hypoxia-dependent epithelial–mesenchymal transition in cancer cells, Carcinogenesis 29 (12) (2008) 2267–2278.
[52] J.P. Joseph, et al., Hypoxia induced EMT: a review on the mechanism of tumor progression and metastasis in OSCC, Oral Oncol. 80 (2018) 23–32.
[53] P. Dauer, et al., ER stress sensor, glucose regulatory protein 78 (GRP78) regulates redox status in pancreatic cancer thereby maintaining “stemness”, Cell Death Dis. 10 (2) (2019) 132.

[54] F. Eckert, et al., Rationale for combining radiotherapy and immune checkpoint inhibition for patients with hypoxic tumors, Front. Immunol. 10 (2019).
[55] M. Diehn, et al., Association of reactive oxygen species levels and radioresistance in cancer stem cells, nature 458 (7239) (2009) 780.
[56] Y. Hu, et al., Effects of PI3K inhibitor NVP-BKM120 on overcoming drug resistance and eliminating cancer stem cells in human breast cancer cells, Cell Death Dis. 6 (12) (2015) e2020.
[57] Y. Wang, et al., Lipolytic inhibitor G0S2 modulates glioma stem-like cell radiation response, J. Exp. Clin. Cancer Res. 38 (1) (2019) 147.
[58] F.P. D'Andrea, et al., Cancer stem cell overexpression of nicotinamide N-methyltransferase enhances cellular radiation resistance, Radiother. Oncol. 99 (3) (2011) 373–378.
[59] K. Anuja, et al., Radiation-induced DNA damage response and resistance in colorectal cancer stem-like cells, Int. J. Radiat. Biol. (2019) 1–13.
[60] H.-K. Park, et al., Interplay between TRAP1 and sirtuin-3 modulates mitochondrial respiration and oxidative stress to maintain stemness of glioma stem cells, Cancer Res. 79 (7) (2019) 1369–1382.
[61] J.E. Visvader, G.J. Lindeman, Cancer stem cells in solid tumours: accumulating evidence and unresolved questions, Nat. Rev. Cancer 8 (10) (2008) 755.
[62] H. Lu, et al., Chemotherapy triggers HIF-1–dependent glutathione synthesis and copper chelation that induces the breast cancer stem cell phenotype, Proc. Natl. Acad. Sci. 112 (33) (2015) E4600–E4609.
[63] W.R. Taylor, et al., Small-molecule ferroptotic agents with potential to selectively target cancer stem cells, Sci. Rep. 9 (1) (2019) 5926.
[64] H. Huang, et al., Inhibition of PGE 2/EP4 receptor signaling enhances oxaliplatin efficacy in resistant colon cancer cells through modulation of oxidative stress, Sci. Rep. 9 (1) (2019) 4954.
[65] E.-H. Kim, et al., 15-deoxy-Δ 12, 14-prostaglandin J 2 induces COX-2 expression through Akt-driven AP-1 activation in human breast cancer cells: a potential role of ROS, Carcinogenesis 29 (4) (2008) 688–695.
[66] D. Wang, et al., Prostaglandin E2 promotes colorectal cancer stem cell expansion and metastasis in mice, Gastroenterology 149 (7) (2015) 1884–1895 (e4).
[67] A.P. Kipp, et al., Time-and cell-resolved dynamics of redox-sensitive Nrf2, HIF and NF-κB activities in 3D spheroids enriched for cancer stem cells, Redox Biol. 12 (2017) 403–409.
[68] Y. You, et al., BRCA1 affects the resistance and stemness of SKOV3-derived ovarian cancer stem cells by regulating autophagy, Cancer Medicine 8 (2) (2019) 656–668.
[69] S. Talukdar, et al., MDA-9/Syntenin regulates protective autophagy in anoikis-resistant glioma stem cells, Proc. Natl. Acad. Sci. 115 (22) (2018) 5768–5773.
[70] R. Ojha, S. Singh, S. Bhattacharyya, JAK-mediated autophagy regulates stemness and cell survival in cisplatin resistant bladder cancer cells, Biochimica et Biophysica Acta (BBA)-General Subjects 1860 (11) (2016) 2484–2497.
[71] S.M. Frisch, R.A. Screaton, Anoikis mechanisms, Curr. Opin. Cell Biol. 13 (5) (2001) 555–562.
[72] P. Baquero, et al., Targeting quiescent leukemic stem cells using second generation autophagy inhibitors, Leukemia 33 (4) (2019) 981.
[73] S. Talukdar, et al., Regulation of protective autophagy in anoikis-resistant glioma stem cells by SDCBP/MDA-9/Syntenin, Autophagy 14 (10) (2018) 1845–1846.
[74] E. Ubil, et al., Tumor-secreted Pros1 inhibits macrophage M1 polarization to reduce antitumor immune response, J. Clin. Invest. (6) (2018) 128.
[75] S.L. Topalian, et al., Mechanism-driven biomarkers to guide immune checkpoint blockade in cancer therapy, Nat. Rev. Cancer 16 (5) (2016) 275.
[76] P. Zhao, et al., Dual-targeting biomimetic delivery for anti-glioma activity via remodeling the tumor microenvironment and directing macrophage-mediated immunotherapy, Chem. Sci. 9 (10) (2018) 2674–2689.
[77] V. Rausch, et al., Synergistic activity of sorafenib and sulforaphane abolishes pancreatic cancer stem cell characteristics, Cancer Res. 70 (12) (2010) 5004–5013.
[78] B. Thakur, P. Ray, Cisplatin triggers cancer stem cell enrichment in platinum-resistant cells through NF-κB-TNFα-PIK3CA loop, J. Exp. Clin. Cancer Res. 36 (1) (2017) 164.
[79] D. Jia, et al., An autocrine inflammatory forward-feedback loop after chemotherapy withdrawal facilitates the repopulation of drug-resistant breast cancer cells, Cell Death Dis. 8 (7) (2017) e2932.
[80] Z. Chen, et al., Aspirin cooperates with p300 to activate the acetylation of H3K9 and promote FasL-mediated apoptosis of cancer stem-like cells in colorectal cancer, Theranostics 8 (16) (2018) 4447.
[81] S. Saha, et al., Aspirin suppresses the acquisition of chemoresistance in breast cancer by disrupting an NFκB–IL6 signaling axis responsible for the generation of cancer stem cells, Cancer Res. 76 (7) (2016) 2000–2012.
[82] G.J. Yoshida, Metabolic reprogramming: the emerging concept and associated therapeutic strategies, J. Exp. Clin. Cancer Res. 34 (1) (2015) 111.
[83] A. Banerjee, et al., Stem cell-like breast cancer cells with acquired resistance to metformin are sensitive to inhibitors of NADH-dependent CtBP dimerization, Carcinogenesis 40 (7) (2019) 871–882.
[84] I. Chefetz, et al., A pan-ALDH1A inhibitor induces necroptosis in ovarian Cancer stem-like cells, Cell Rep. 26 (11) (2019) 3061–3075 (e6).
[85] G. Mazor, et al., The lncRNA TP73-AS1 is linked to aggressiveness in glioblastoma and promotes temozolomide resistance in glioblastoma cancer stem cells, Cell Death Dis. 10 (3) (2019) 246.
[86] N. Colwell, et al., Hypoxia in the glioblastoma microenvironment: shaping the phenotype of cancer stem-like cells, Neuro-oncology 19 (7) (2017) 887–896.
[87] C. Chen, et al., Targeting LIN28B reprograms tumor glucose metabolism and acidic microenvironment to suppress cancer stemness and metastasis, Oncogene (2019) 1.
[88] K. Fekir, et al., Retrodifferentiation of human tumor hepatocytes to stem cells leads to metabolic reprogramming and chemoresistance, Cancer Res. 79 (8) (2019) 1869–1883 (p. canres. 2110.2018).
[89] J.A. Kruger, et al., Characterization of stem cell–like cancer cells in immune-competent mice, Blood 108 (12) (2006) 3906–3912.
[90] C. Melzer, et al., Cancer stem cell niche models and contribution by mesenchymal stroma/stem cells, Mol. Cancer 16 (1) (2017) 28.
[91] H. Zhao, et al., Magnesium-stabilized multifunctional DNA nanoparticles for tumor-targeted and pH-responsive drug delivery, ACS Appl. Mater. Interfaces 10 (18) (2018) 15418–15427.
[92] A.D. Gujar, et al., An NAD + -dependent transcriptional program governs self-renewal and radiation resistance in glioblastoma, Proc. Natl. Acad. Sci. 113 (51) (2016) E8247–E8256.
[93] N.A. Fonseca, et al., The cancer stem cell phenotype as a determinant factor of the heterotypic nature of breast tumors, Crit. Rev. Oncol. Hematol. 113 (2017) 111–121.
[94] A. Dirkse, et al., Stem cell-associated heterogeneity in glioblastoma results from intrinsic tumor plasticity shaped by the microenvironment, Nat. Commun. 10 (1) (2019) 1787.
[95] V. Plaks, N. Kong, Z. Werb, The cancer stem cell niche: how essential is the niche in regulating stemness of tumor cells? Cell Stem Cell 16 (3) (2015) 225–238.
[96] S.M. Kumar, et al., Acquired cancer stem cell phenotypes through Oct4-mediated dedifferentiation, Oncogene 31 (47) (2012) 4898.
[97] H. Jeong, et al., Tumor-associated macrophages enhance tumor hypoxia and aerobic glycolysis, Cancer Res. 79 (4) (2019) 795–806.
[98] A. Biddle, et al., Phenotypic plasticity determines cancer stem cell therapeutic resistance in oral squamous cell carcinoma, EBioMedicine 4 (2016) 138–145.
[99] D. Nantajit, D. Lin, J.J. Li, The network of epithelial–mesenchymal transition: potential new targets for tumor resistance, J. Cancer Res. Clin. Oncol. 141 (10) (2015) 1697–1713.
[100] R.G. Stein, et al., Cognate non-lytic interactions between CD8 + T cells and breast cancer cells induce cancer stem cell-like properties, Cancer Res. 79 (7) (2019) 1507–1519 (p. canres. 0387.2018).
[101] X. Liu, et al., KDM5B promotes drug resistance by regulating melanoma-propagating cell subpopulations, Mol. Cancer Ther. 18 (3) (2019) 706–717.
[102] R. Tejero, et al., Gene signatures of quiescent glioblastoma cells reveal mesenchymal shift and interactions with niche microenvironment, EBioMedicine 42 (2019) 252–269.
[103] B.J. Wilson, et al., ABCB5 maintains melanoma-initiating cells through a proinflammatory cytokine signaling circuit, Cancer Res. 74 (15) (2014) 4196–4207.
[104] X. Zhang, et al., Targeting mitochondrial function to treat quiescent tumor cells in solid tumors, Int. J. Mol. Sci. 16 (11) (2015) 27313–27326.
[105] I.S. Kim, et al., Microenvironment-derived factors driving metastatic plasticity in melanoma, Nat. Commun. 8 (2017) 14343.
[106] P.B. Gupta, et al., Phenotypic plasticity: driver of cancer initiation, progression, and therapy resistance, Cell Stem Cell 24 (1) (2018) 65–78.
[107] C.A. O'Brien, et al., A human colon cancer cell capable of initiating tumour growth in immunodeficient mice, Nature 445 (7123) (2007) 106.
[108] R. Ishida, et al., The tissue-reconstructing ability of colon CSCs is enhanced by FK506 and suppressed by GSK3 inhibition, Mol. Cancer Res. 15 (10) (2017) 1455–1466.
[109] M. Boesch, et al., Drug transporter-mediated protection of cancer stem cells from ionophore antibiotics, Stem Cells Transl. Med. 4 (9) (2015) 1028–1032.
[110] M. Zhao, et al., Hypoxia-induced cell stemness leads to drug resistance and poor prognosis in lung adenocarcinoma, Lung Cancer 87 (2) (2015) 98–106.
[111] S.Y. Koh, et al., Baicalein suppresses stem cell-like characteristics in radio-and chemoresistant MDA-MB-231 human breast cancer cells through up-regulation of IFIT2, Nutrients 11 (3) (2019) 624.
[112] M. Zhang, et al., Mithramycin represses basal and cigarette smoke–induced expression of ABCG2 and inhibits stem cell signaling in lung and esophageal cancer cells, Cancer Res. 72 (16) (2012) 4178–4192.
[113] X. Ling, et al., An ABCG2 non-substrate anticancer agent FL118 targets drug-resistant cancer stem-like cells and overcomes treatment resistance of human pancreatic cancer, J. Exp. Clin. Cancer Res. 37 (1) (2018) 240.
[114] L.Y. Bourguignon, et al., Hyaluronan-CD44 interaction activates stem cell marker Nanog, Stat-3-mediated MDR1 gene expression, and ankyrin-regulated multidrug efflux in breast and ovarian tumor cells, J. Biol. Chem. 283 (25) (2008) 17635–17651.
[115] S. Rhost, et al., Sortilin inhibition limits secretion-induced progranulin-dependent breast cancer progression and cancer stem cell expansion, Breast Cancer Res. 20 (1) (2018) 137.
[116] Y. Li, et al., Zinc finger protein 32 promotes breast cancer stem cell-like properties through directly promoting GPER transcription, Cell Death Dis. 9 (12) (2018) 1162.
[117] Y. Zhu, et al., Suppression of miR-21-3p enhances TRAIL-mediated apoptosis in liver cancer stem cells by suppressing the PI3K/Akt/Bad cascade via regulating PTEN, Cancer Manag. Res. 11 (2019) 955.
[118] S. Ouyang, et al., LncRNA BCAR4, targeting to miR-665/STAT3 signaling, maintains cancer stem cells stemness and promotes tumorigenicity in colorectal cancer, Cancer Cell Int. 19 (1) (2019) 72.
[119] R.H. Mohamed, et al., Co-regulatory network of oncosuppressor miRNAs and transcription factors for pathology of Human Hepatic Cancer stem Cells (HCsC), Sci. Rep. 9 (1) (2019) 5564.
[120] J. Wu, et al., The long non-coding RNA LncHDAC2 drives the self-renewal of liver cancer stem cells via activation of hedgehog signaling, J. Hepatol. 70 (5) (2019) 918–929.
[121] P. Zhu, et al., Lnc-β-Catm elicits EZH2-dependent β-catenin stabilization and

sustains liver CSC self-renewal, Nat. Struct. Mol. Biol. 23 (7) (2016) 631.
[122] L.Y. Bourguignon, et al., Hyaluronan-CD44v3 interaction with Oct4-Sox2-Nanog promotes miR-302 expression leading to self-renewal, clonal formation, and cisplatin resistance in cancer stem cells from head and neck squamous cell carcinoma, J. Biol. Chem. 287 (39) (2012) 32800–32824.
[123] V.Y. Shin, et al., Long non-coding RNA NEAT1 confers oncogenic role in triple-negative breast cancer through modulating chemoresistance and cancer stemness, Cell Death Dis. 10 (4) (2019) 270.
[124] Y.-S. Weng, et al., MCT-1/miR-34a/IL-6/IL-6R signaling axis promotes EMT progression, cancer stemness and M2 macrophage polarization in triple-negative breast cancer, Mol. Cancer 18 (1) (2019) 42.
[125] X. Wang, et al., miR-181b/Notch2 overcome chemoresistance by regulating cancer stem cell-like properties in NSCLC, Stem Cell Res Ther 9 (1) (2018) 327.
[126] Y. Jia, et al., Tumorigenicity of cancer stem-like cells derived from hepatocarcinoma is regulated by microRNA-145, Oncol. Rep. 27 (6) (2012) 1865–1872.
[127] R.-Z. Ran, et al., miR-194 inhibits liver cancer stem cell expansion by regulating RAC1 pathway, Exp. Cell Res. 378 (1) (2019) 66–75.
[128] D.L. Schonberg, et al., Brain tumor stem cells: molecular characteristics and their impact on therapy, Mol. Asp. Med. 39 (2014) 82–101.
[129] N. Li, et al., An FBXW7-ZEB2 axis links EMT and tumour microenvironment to promote colorectal cancer stem cells and chemoresistance, Oncogenesis 8 (3) (2019) 13.
[130] K. Alamoud, M. Kukuruzinska, Emerging insights into Wnt/β-catenin signaling in head and neck cancer, J. Dent. Res. 97 (6) (2018) 665–673.
[131] Q. Yu, et al., Fibronectin promotes the malignancy of glioma stem-like cells via modulation of cell adhesion, differentiation, proliferation and chemoresistance, Front. Mol. Neurosci. 11 (2018) 130.
[132] D.E. Biancur, A.C. Kimmelman, The plasticity of pancreatic cancer metabolism in tumor progression and therapeutic resistance, Biochimica et Biophysica Acta (BBA)-Reviews on Cancer 1870 (1) (2018) 67–75.
[133] V. Gkretsi, T. Stylianopoulos, Cell adhesion and matrix stiffness: coordinating Cancer cell invasion and metastasis, Front. Oncol. 8 (2018) 145.
[134] D. Bourboulia, W.G. Stetler-Stevenson, Matrix metalloproteinases (MMPs) and tissue inhibitors of metalloproteinases (TIMPs): positive and negative regulators in tumor cell adhesion, Seminars in Cancer Biology, Elsevier, 2010.
[135] J. Landsberg, et al., Melanomas resist T-cell therapy through inflammation-induced reversible dedifferentiation, Nature 490 (7420) (2012) 412.
[136] D. Zhang, D.G. Tang, K. Rycaj, Cancer stem cells: regulation programs, immunological properties and immunotherapy, Seminars in Cancer Biology, Elsevier, 2018.
[137] Y. Shi, et al., Ibrutinib inactivates BMX-STAT3 in glioma stem cells to impair malignant growth and radioresistance, Sci. Transl. Med. 10 (443) (2018) eaah6816.
[138] M. Jinushi, et al., Tumor-associated macrophages regulate tumorigenicity and anticancer drug responses of cancer stem/initiating cells, Proc. Natl. Acad. Sci. 108 (30) (2011) 12425–12430.
[139] H.T. Tzeng, et al., Rab37 in lung cancer mediates exocytosis of soluble ST2 and thus skews macrophages toward tumor-suppressing phenotype, Int. J. Cancer 143 (7) (2018) 1753–1763.
[140] P. Yadav, B.S. Shankar, Radio resistance in breast cancer cells is mediated through TGF-β signalling, hybrid epithelial-mesenchymal phenotype and cancer stem cells, Biomed. Pharmacother. 111 (2019) 119–130.
[141] J. Zhuang, et al., TGFβ1 promotes gemcitabine resistance through regulating the lncRNA-LET/NF90/miR-145 signaling axis in bladder cancer, Theranostics 7 (12) (2017) 3053.
[142] Y.-A. Tang, et al., Hypoxic tumor microenvironment activates GLI2 via HIF-1α and TGF-β2 to promote chemoresistance in colorectal cancer, Proc. Natl. Acad. Sci. 115 (26) (2018) E5990–E5999.
[143] Y. Katsuno, et al., Chronic TGF-β exposure drives stabilized EMT, tumor stemness, and cancer drug resistance with vulnerability to bitopic mTOR inhibition, Sci. Signal. 12 (570) (2019) eaau8544.
[144] K. Chikamatsu, et al., Alteration of cancer stem cell-like phenotype by histone deacetylase inhibitors in squamous cell carcinoma of the head and neck, Cancer Sci. 104 (11) (2013) 1468–1475.
[145] A.S. Cazet, et al., Targeting stromal remodeling and cancer stem cell plasticity overcomes chemoresistance in triple negative breast cancer, Nat. Commun. (9) (2018).
[146] J.A. Brown, et al., TGF-β-induced quiescence mediates chemoresistance of tumor-propagating cells in squamous cell carcinoma, Cell Stem Cell 21 (5) (2017) 650–664 (e8).
[147] H. Li, et al., The tumor microenvironment: an irreplaceable element of tumor budding and epithelial-mesenchymal transition-mediated cancer metastasis, Cell Adhes. Migr. 10 (4) (2016) 1–13.
[148] M. Marhold, et al., HIF1α regulates mTOR signaling and viability of prostate cancer stem cells, Mol. Cancer Res. 13 (3) (2015) 556–564.
[149] H. Zhou, et al., B591, a novel specific pan-PI3K inhibitor, preferentially targets cancer stem cells, Oncogene (2019) 1.
[150] J. Qin, et al., Hypoxia-inducible factor 1 alpha promotes cancer stem cells-like properties in human ovarian cancer cells by upregulating SIRT1 expression, Sci. Rep. 7 (1) (2017) 10592.
[151] C.C. da Hora, et al., Sustained NF-κB-STAT3 signaling promotes resistance to Smac mimetics in glioma stem-like cells but creates a vulnerability to EZH2 inhibition, Cell Death Discovery 5 (1) (2019) 72.
[152] S.D. Alipoor, et al., The potential biomarkers and immunological effects of tumor-derived exosomes in lung cancer, Front. Immunol. 9 (2018).
[153] K. Balamurugan, et al., C/EBPδ links IL-6 and HIF-1 signaling to promote breast cancer stem cell-associated phenotypes, Oncogene 38 (20) (2019) 3765.
[154] W. Li, et al., 2-Ethoxystypandrone, a novel small-molecule STAT3 signaling inhibitor from Polygonum cuspidatum, inhibits cell growth and induces apoptosis of HCC cells and HCC Cancer stem cells, BMC Complement. Altern. Med. 19 (1) (2019) 38.
[155] B.A. Tannous, C.E. Badr, A TNF-NF-κB-STAT3 loop triggers resistance of glioma-stem-like cells to Smac mimetics while sensitizing to EZH2 inhibitors, Cell Death Dis. 10 (4) (2019) 268.
[156] Z.-J. Hou, et al., Flubendazole, FDA-approved anthelmintic, targets breast cancer stem-like cells, Oncotarget 6 (8) (2015) 6326.
[157] Y.-B. Hu, et al., Exosomal Wnt-induced dedifferentiation of colorectal cancer cells contributes to chemotherapy resistance, Oncogene 38 (11) (2019) 1951.
[158] P.-C. Hou, et al., Hypoxia-induced downregulation of DUSP-2 phosphatase drives colon cancer stemness, Cancer Res. 77 (16) (2017) 4305–4316.
[159] V. Justilien, A.P. Fields, Molecular pathways: novel approaches for improved therapeutic targeting of hedgehog signaling in cancer stem cells, Clin. Cancer Res. 21 (3) (2015) 505–513.
[160] N. Takebe, et al., Targeting notch, hedgehog, and Wnt pathways in cancer stem cells: clinical update, Nat. Rev. Clin. Oncol. 12 (8) (2015) 445.
[161] Y. Geng, et al., Rapid phenotyping of cancer stem cells using multichannel nanosensor arrays, Nanomedicine 14 (6) (2018) 1931–1939.
[162] S.A. Mousa, et al., Stress resistant human embryonic stem cells as a potential source for the identification of novel cancer stem cell markers, Cancer Lett. 289 (2) (2010) 208–216.
[163] S.D. Fouse, et al., Response of primary glioblastoma cells to therapy is patient specific and independent of cancer stem cell phenotype, Neuro-oncology 16 (3) (2013) 361–371.
[164] Z. Yang, et al., Disulfiram modulates ROS accumulation and overcomes synergistically cisplatin resistance in breast cancer cell lines, Biomed. Pharmacother. 113 (2019) 108727.
[165] P. Wang, et al., HIF1α regulates glioma chemosensitivity through the transformation between differentiation and dedifferentiation in various oxygen levels, Sci. Rep. 7 (1) (2017) 7965.
[166] A. Gold, et al., Spironolactone inhibits the growth of cancer stem cells by impairing DNA damage response, Oncogene (2019) 1.
[167] U. Kahlert, et al., Targeting cancer stem-like cells in glioblastoma and colorectal cancer through metabolic pathways, Int. J. Cancer 140 (1) (2017) 10–22.
[168] Y. Feng, et al., Metformin reverses stem cell-like HepG2 sphere formation and resistance to sorafenib by attenuating epithelial-mesenchymal transformation, Mol. Med. Rep. 18 (4) (2018) 3866–3872.
[169] M. Appari, et al., Sulforaphane, quercetin and catechins complement each other in elimination of advanced pancreatic cancer by miR-let-7 induction and K-ras inhibition, Int. J. Oncol. 45 (4) (2014) 1391–1400.
[170] L. Wang, et al., Resveratrol, a potential radiation sensitizer for glioma stem cells both in vitro and in vivo, J. Pharmacol. Sci. 129 (4) (2015) 216–225.