antioxidants

MDPI

Review

NRF2 and Aquaglyceroporins: Orchestrating Metabolic and Redox Homeostasis in Cancer Resistance

Monika Mlinarić [1,†], Anita Bijelić [2,†], Ana Josipa Jerončić [1], Juraj Kolak [3], Kristina Kanceljak [3], Martina Matec [3], Anita Zenko Sever [3], Hrvoje Silovski [3,4], Tajana Silovski [3,5,*] and Ana Čipak Gašparović [1,*]

[1] Division of Molecular Medicine, Ruđer Bošković Institute, 10000 Zagreb, Croatia; mmlinar@irb.hr (M.M.); anjeronci@irb.hr (A.J.J.)
[2] Faculty of Dental Medicine and Health, Josip Juraj Strossmayer University of Osijek, 31000 Osijek, Croatia; anitabijelic0511@gmail.com
[3] University Hospital Centre Zagreb, 10000 Zagreb, Croatia; jkolak@kbc-zagreb.hr (J.K.); kkancelj@kbc-zagreb.hr (K.K.); mmatec@kbc-zagreb.hr (M.M.); anita.zenko.sever@kbc-zagreb.hr (A.Z.S.); hsilovsk@kbc-zagreb.hr (H.S.)
[4] Department of Surgery, School of Medicine, University of Zagreb, 10000 Zagreb, Croatia
[5] Department of Pathophysiology, School of Medicine, University of Zagreb, 10000 Zagreb, Croatia
* Correspondence: tsilovsk@kbc-zagreb.hr (T.S.); acipak@irb.hr (A.Č.G.); Tel.: +385-12388581 (T.S.); +385-14571212 (A.Č.G.)
† These authors contributed equally to this work.

Abstract

Cancer remains a primary global health challenge, characterized by high metabolic plasticity and therapy resistance. A critical factor in tumor survival is the fine-tuning of redox homeostasis, governed by the NRF2 signaling pathway. This redox balance is inevitably linked to the cellular metabolic network, where NRF2 drives the pentose phosphate pathway, NADPH production, and lipid metabolism to fuel rapid proliferation and drug detoxification. Recent evidence highlights a functional link between NRF2 and specific aquaporin (AQP) isoforms, peroxiporins and aquaglyceroporins, which facilitate the transmembrane flux of hydrogen peroxide and glycerol. By regulating the movement of these molecules, AQPs can actively contribute to both metabolic reprogramming and redox signaling. This review explores the mechanisms of NRF2 and AQP co-regulation in breast, colon, and liver cancers, illustrating their combined role in driving tumor progression and malignancy. Furthermore, we evaluate the clinical potential of targeting these pathways and discuss the utility of NRF2/AQP co-expression as a predictive biomarker for therapy outcomes. Identifying these molecular signatures offers a promising avenue for patient stratification and the development of novel therapeutic strategies to overcome tumor resistance.

Keywords: NRF2; aquaporins; aquaglyceroporins; cancer; therapy resistance

Check for updates

Academic Editor: Sander Bekeschus

Received: 12 October 2026
Revised: 29 October 2026
Accepted: 2 November 2026
Published: 6 November 2026

Copyright: © 2026 by the authors. Licensee MDPI, Basel, Switzerland. This article is an open access article distributed under the terms and conditions of the Creative Commons Attribution (CC BY) license.

1. Introduction

Reactive oxygen species (ROS) have a paradoxical role in cancer biology, promoting malignant transformation and tumor progression at moderate levels while underlying the cytotoxic mechanism of action of many anticancer therapies at higher levels. Consequently, oxidative stress is linked to cancer at every stage of development and progression, acting simultaneously as a causative factor, a consequence of malignant transformation, and a mechanism exploited by chemo-, radio-, immune-, and biotherapy to induce cancer cell death [1]. As tumors evolve, their genetic instability generates resistant clones capable of tolerating and adapting to therapy-induced oxidative stress. This adaptation is frequently

accompanied by elevated basal ROS levels and an upregulated antioxidant defense system mediated by NRF2 (nuclear factor erythroid 2-related factor 2), the major transcription factor regulating the cellular antioxidant response [2]. In addition to its canonical antioxidant role, NRF2 also reprograms cancer cell metabolism, shaping glucose, lipid, and amino acid handling in ways that support tumor survival under stress (reviewed in [3]). Emerging evidence suggests that this metabolic dimension of NRF2 signaling could intersect with aquaglyceroporins, a subfamily of membrane channels that facilitate the transport of water, glycerol, and other small solutes such as hydrogen peroxide (H_2O_2), thereby contributing to cellular metabolic flexibility. Here, we focus on the interplay between NRF2 and aquaglyceroporins, proposing a pathway through which this crosstalk may reshape cancer cell redox and metabolic homeostasis and drive resistance to therapy.

2. Redox-Metabolic Regulation in Cancer

Cancer cells reprogram their metabolism to meet their increased metabolic demands and support rapid proliferation. This includes increased glucose uptake and glycolytic flux, altered mitochondrial respiration, enhanced glutamine utilization, and activation of NADPH-generating pathways [4]. Together, these adaptations provide energy, biosynthetic precursors, and reducing power, but also increase metabolic activity and alter mitochondrial function, resulting in elevated basal ROS levels compared with normal cells [5]. This increase in ROS, in turn, modulates cellular signaling pathways, further promoting cell proliferation and survival while enhancing stress resistance, ultimately leading to therapy resistance. Given their high reactivity, excess ROS interact with DNA, lipids, and proteins, causing DNA single- and double-strand breaks that can activate proto-oncogenes or inhibit tumor suppressor genes, thereby driving carcinogenesis [6].

Cancer development is a multistep process driven by genetic alterations that disrupt the normal regulation of cell proliferation and differentiation. Rapid and uncontrolled proliferation generates a heterogeneous tumor mass composed of cells with various genetic and adaptive properties. As tumors expand, their growth outpaces the development of functional blood vessels, limiting oxygen and nutrient delivery.

Although tumors initiate angiogenesis to compensate for this hypoxic demand, the resulting vasculature is often structurally abnormal and functionally inadequate, leading to chronic hypoxia. This is further exacerbated by the high metabolic activity of cancer cells, which impairs oxygen delivery and perfusion. Under these conditions, cancer cells often shift their metabolism toward glycolysis. However, increased glycolysis is not restricted to hypoxic regions, as many cancer cells convert glucose to lactate even when oxygen is available, a phenomenon known as aerobic glycolysis or the Warburg effect. Hypoxia can further alter tumor metabolism by reducing mitochondrial oxidative phosphorylation, promoting electron leakage from the electron transport chain, and activating ROS-generating enzymes. Thus, oxygen availability, mitochondrial electron transport, and ROS-generating enzymes make distinct but interconnected contributions to cancer-associated redox imbalance. Under hypoxic conditions, mitochondrial dysfunction can enhance superoxide generation, whereas aerobic glycolysis may support biosynthesis and redox adaptation independently of oxygen limitation. Elevated ROS, in turn, promote tumor cell adaptation, aggressiveness, metastasis, and resistance to therapy [7].

ROS play a dual role in both normal and cancer cells: moderate levels facilitate proliferation by activating pro-survival signaling pathways, while excess ROS induce DNA damage, senescence, and cell death [6,8,9].

To balance this, cancer cells upregulate their antioxidant systems, a process termed "redox reprogramming", enabling resistance to chemotherapy, radiotherapy, targeted therapy, and immunotherapy [10,11]. This is primarily mediated by transcription factor NRF2,

which drives the transcription of genes involved in ROS detoxification, glutathione and thioredoxin pathways, and NADPH production [12]. However, NRF2 acts as a double-edged sword: while its cytoprotective response protects normal cells, constitutive NRF2 activation in cancer allows tumor cells to tolerate elevated oxidative stress, promoting their survival and progression [6,8,9,13]. This adapted antioxidant machinery allows tumors to harness ROS for proliferative signaling while neutralizing ROS-mediated cytotoxicity from anticancer therapies [14].

Beyond redox reprogramming, cancer cells employ additional mechanisms to avoid therapy-mediated toxicity, including enhanced DNA repair, apoptosis inhibition, altered drug metabolism, and increased drug efflux [15]. One of the mechanisms is the overexpression of ABC (ATP-binding cassette) transporters, including P-gp/ABCB1 (P-glycoprotein), MRPs/ABCCs (multidrug resistance-associated protein), and BCRP/ABCG2 (breast cancer resistance protein), which actively export anticancer drugs from the cell, lowering intracellular drug concentrations and reducing cytotoxicity [16]. Interestingly, their expression and activity are also influenced by oxidative stress and redox-regulating pathways, linking ROS signaling to multidrug resistance [11].

Increased ROS levels have been shown to induce MDR1 expression in Caco-2 colon cancer cells exposed to H_2O_2 [17]. ROS activate NRF2, which can promote the expression and activity of P-gp and consequently reduce intracellular drug accumulation [18]. Similarly, oxidative stress can activate FOXO transcription factors, which have also been implicated in the regulation of drug efflux transporters mediating chemotherapy resistance [19]. Other redox-responsive pathways, such as APE1/Ref-1 signaling, can further promote the expression of MDR, MRP, and BCRP transporters. Thus, beyond causing significant cellular damage, ROS can also trigger adaptive responses, including drug efflux, that allow cancer cells to maintain redox homeostasis and reduce their sensitivity to anticancer therapy.

ROS may also induce hypermethylation in various cancers. For example, prolonged ROS stress in hepatocellular carcinoma cells down-regulates E-cadherin expression [20], an adhesion molecule whose loss may contribute to the early stages of the metastatic process. Furthermore, ROS activates the PI3K/Akt/GSK3 (phosphoinositide 3-kinase/protein kinase B/glycogen synthase kinase-3) signaling pathway, affecting several biological processes, some of which are mediated by the transcription factor Snail. Snail binds to E-boxes within the E-cadherin promoter and promotes the methylation of its CpG sites through the recruitment of HDAC1 (histone deacetylase 1) and DNMT1 (DNA methyltransferase 1) [20]. In human colorectal cancer cells, H_2O_2 down-regulated the tumor suppressor RUNX3 through DNMT1/HDAC1-mediated promoter hypermethylation and inhibition of RUNX3 nuclear localization [21]. These findings demonstrate a link between oxidative stress-driven epigenetic silencing of RUNX3 and enhanced cell proliferation and colorectal cancer progression.

As mentioned, ROS can modulate PI3K/Akt and MAPK (mitogen-activated protein kinase) pathways, driving not only tumor cell proliferation but also survival, chemoresistance, and protection from cell death [22]. The MAPK pathway regulates cell proliferation, differentiation, survival, and cell death through several alternative branches, including ERK1/2 (extracellular signal-regulated kinases 1/2), JNK (c-Jun N-terminal kinases), and p38 MAPK. ROS can activate MAPK signaling through multiple mechanisms: ligand-independent activation of specific tyrosine kinase receptors stimulates the ERK pathway [23], while ROS-mediated activation of PLC-γ (phospholipase C-γ) increases intracellular Ca^{2+} levels, activating PKC (protein kinase C) and further promoting ERK pathway activation [24]. ROS can also activate JNK and p38 by modifying redox-sensitive regulatory proteins, particularly through ASK1 activation, which drives downstream JNK/p38 signaling and contributes to cellular stress responses and tumor survival [23].

The PI3K/Akt pathway similarly regulates proliferation, survival, apoptosis, autophagy, and drug resistance, with downstream targets including GSK3, FOXO, BAD, mTOR, and p53 [25]. ROS enhance this pathway by oxidizing reactive cysteine residues in PTEN, a key negative regulator of PI3K/Akt signaling, thereby reducing PTEN activity and increasing PI3K/Akt signaling [26].

3. NRF2

NRF2 belongs to the cap'n'collar family of basic leucine zipper proteins. NRF2 is continuously synthesized but rapidly degraded through the action of KEAP1 (Kelch-like ECH-associated protein 1). Under physiological conditions, KEAP1 functions as a negative regulator of NRF2 by serving as a substrate adaptor for the CUL3–Rbx1-based E3 ubiquitin ligase complex, thereby targeting NRF2 for ubiquitination and proteasomal degradation [27–29]. This tight posttranslational control maintains low basal NRF2 levels and ensures that its transcriptional program is engaged only in response to cellular stress (Figure 1) [30,31].

Under basal conditions, canonical regulation of NRF2 depends on the interaction between KEAP1 and two motifs located within the Neh2 domain of NRF2, the high-affinity ETGE motif and the lower-affinity DLG motif. The cooperative binding of both Neh2 motifs to the KEAP1 homodimer underlies the "hinge-and-latch" mechanism, whereby the geometry of this interaction places NRF2 in a configuration that facilitates its ubiquitination and subsequent proteasomal degradation [27,32,33]. In this way, KEAP1 maintains low intracellular NRF2 levels and prevents persistent activation of the antioxidant response pathway [33].

KEAP1 contains multiple redox-sensitive cysteine residues, of which Cys151, Cys273, and Cys288 are particularly important as electrophile and oxidant sensors. Oxidative or electrophilic modification of reactive cysteines disrupts DLG-KEAP1 binding, consequently disabling NRF2 ubiquitination and degradation [9,34]. As a result, newly synthesized NRF2 escapes degradation, accumulates, and translocates into the nucleus [35,36]. In the nucleus, NRF2 heterodimerizes with small Maf proteins and binds to antioxidant response elements (AREs) within the promoters of target genes [3,37].

In the absence of stress, BACH1 (BTB and CNC homology 1) competes with NRF2 for binding to ARE sequences by forming heterodimers with small Maf proteins, thereby repressing transcription of NRF2 target genes. Oxidative stress promotes BACH1 dissociation from ARE sites and subsequent degradation, allowing NRF2 to occupy AREs and activate cytoprotective gene expression [38].

Beyond this canonical, redox-regulated control, NRF2 stability is also governed by a KEAP1-independent pathway. GSK-3β (glycogen synthase kinase-3β) phosphorylates NRF2, which promotes its recognition by the β-TrCP (β-transducin repeat-containing protein) and subsequent ubiquitination by the SCF^β-TrCP/CUL1 E3 ubiquitin ligase complex. This alternative degradation pathway provides an additional level of NRF2 regulation and links its activity to broader growth factor signaling, such as the PI3K/AKT [39].

Once NRF2 is activated and translocated to the nucleus, it initiates the transcription of more than 200 genes [40]. These target genes fall broadly into three functional groups. The first comprises antioxidant and cytoprotective enzymes that neutralize ROS and electrophiles directly, such as NQO1, heme oxygenase-1 (HMOX1), glutamate–cysteine ligase catalytic and modifier subunits (GCLC and GCLM), and other components of the glutathione and thioredoxin antioxidant systems [41]. The second comprises phase I and phase II drug-metabolizing (biotransformation) enzymes responsible for xenobiotic detoxification, including cytochrome P450 enzymes (phase I) and glutathione S-transferases, UDP-glucuronosyltransferases, and aldo-keto reductases (phase II) [41]. The third com-

prises phase III transporters, which mediate the cellular efflux of xenobiotics and their glutathione, glucuronide, or sulfate conjugates, including members of the ATP-binding cassette (ABC) transporter family such as the multidrug resistance-associated proteins (MRPs/ABCCs) and P-glycoprotein (ABCB1) [36,42]. Some of these target genes are discussed in more detail below.

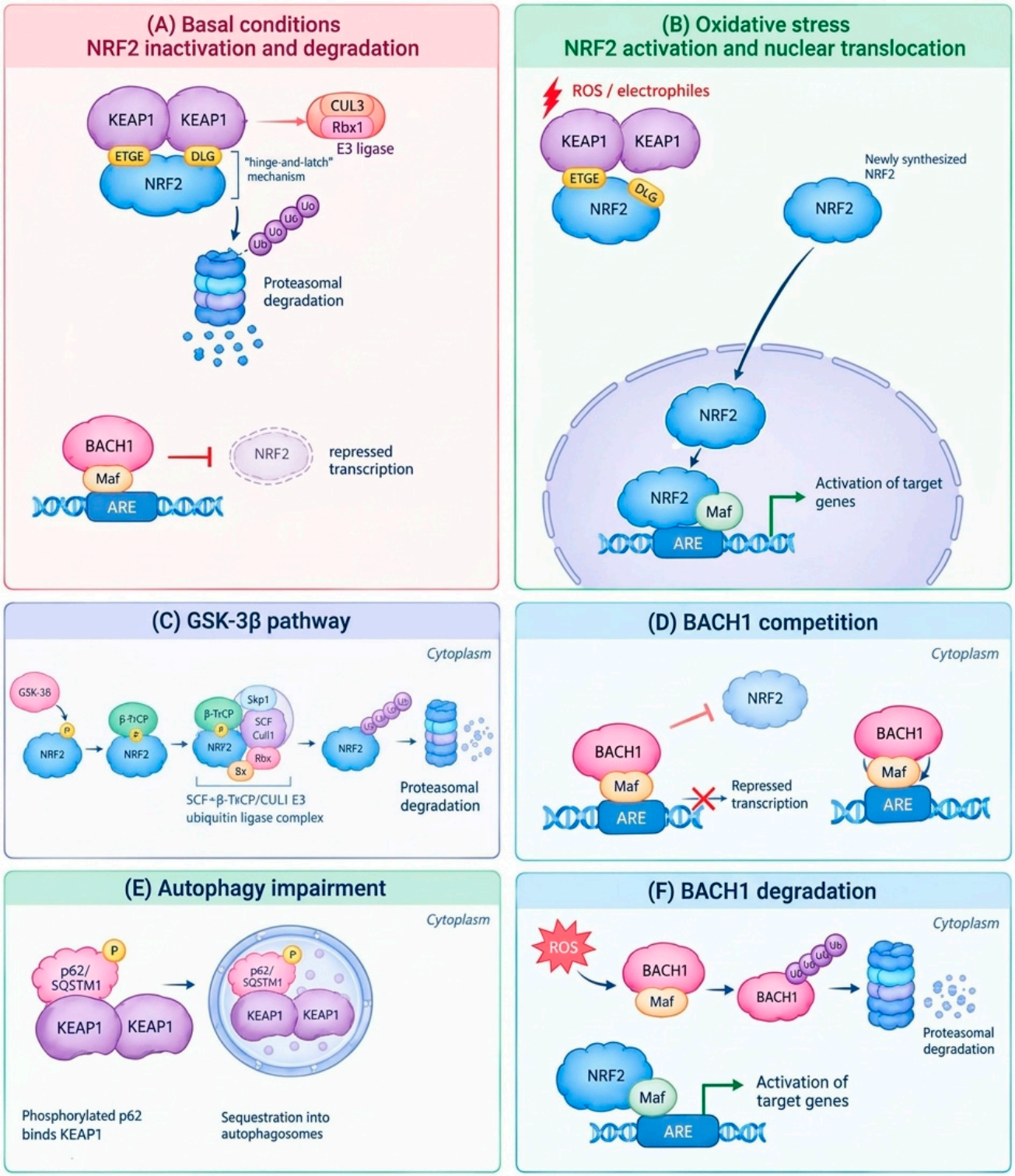


Figure 1. ROS-dependent regulation of NRF2 activation. (**A**) Under basal conditions, NRF2 is continuously targeted for degradation by the KEAP1–CUL3 E3 ubiquitin ligase complex. (**B**) Increased ROS levels modify KEAP1, leading to NRF2 stabilization, accumulation, and translocation into the

nucleus, where it activates the expression of antioxidant and cytoprotective genes. (**C**) In addition to KEAP1, GSK3β can inactivate NRF2 by phosphorylating it, thus making it a target for degradation by the SCF^β-TrCP/CUL1 E3 ubiquitin ligase complex. (**D**) In the absence of stress, BACH1 competes with NRF2 for binding to ARE sequences by forming heterodimers with small Maf proteins, thereby repressing transcription of NRF2 target genes. (**E**) Impaired autophagy causes accumulation of phosphorylated p62/SQSTM1, which binds to KEAP1, sequestering it into autophagosomes and disrupting the KEAP1-NRF2 complex/p62, amplifying pathway activation. (**F**) In the presence of oxidative stress, BACH1 is ubiquitinized and targeted for proteasomal degradation, allowing NRF2 to bind to ARE regions.

3.1. Anti-Tumor Effects and Contribution to Resistance

Given the multiple layers of control that normally keep NRF2 activity in check, it is notable that this regulation is frequently subverted in cancer. Several molecular mechanisms underlie this constitutive activation, including somatic alterations in NFE2L2 (encoding NRF2) or KEAP1. Gain-of-function mutations in NFE2L2 often cluster in the ETGE and DLG motifs, weakening the interaction with KEAP1 and impairing NRF2 degradation [43]. Conversely, loss-of-function mutations in KEAP1 abolish its E3 adaptor function, likewise resulting in persistent NRF2 stabilization [31,35,44]. These alterations are especially relevant in non-small cell lung cancer, hepatocellular carcinoma, and head and neck squamous cell carcinoma, where KEAP1 mutations are observed in a substantial proportion of tumors [28,30].

Impaired autophagy represents an additional mechanism driving persistent NRF2 activation through the accumulation of SQSTM1/p62 (sequestosome 1), an autophagy receptor that accumulates in settings of impaired autophagic flux, a common feature of many tumors [45]. When phosphorylated, p62 directly binds KEAP1 through a KEAP1-interacting region, sequestering it into autophagosomes and disrupting the KEAP1-NRF2 complex [32,45]. This interaction competitively displaces NRF2 from KEAP1, thereby impairing KEAP1-mediated ubiquitination and degradation of NRF2, allowing NRF2 to accumulate and constitutively activate the transcription of antioxidant and cytoprotective genes [22]. This p62-dependent, non-canonical activation of NRF2 can create a feed-forward loop, as NRF2 itself transcriptionally upregulates SQSTM1/p62, amplifying pathway activation [22].

Additional alterations affecting components of the NRF2 degradation machinery further contribute to persistent pathway activation. CUL3 (Cullin 3) encodes the scaffold protein of the Cullin-3 E3 ubiquitin ligase complex, which partners with KEAP1 to mediate NRF2 ubiquitination; alterations in CUL3 impair this ubiquitination process, resulting in constitutive NRF2 stabilization [46]. Similarly, alterations in chromatin remodeling factors, particularly SMARCA4 (BRG1), have been implicated in dysregulation of NRF2 signaling, affecting transcriptional regulation and contributing to persistent activation of NRF2-dependent gene expression programs [47]. Further, additional NRF2-modulating proteins have been identified: PALB2 competes with NRF2 for KEAP1 binding [48], while BRCA1 directly interacts with NRF2 itself to promote its stability and activation, thereby providing yet another layer through which NRF2 degradation can be subverted in cancer [49].

Beyond genetic alterations, epigenetic mechanisms also contribute to persistent NRF2 activation. Hypermethylation of the KEAP1 promoter reduces KEAP1 expression, thereby impairing NRF2 ubiquitination and degradation, allowing NRF2 to accumulate and constitutively activate the transcription of antioxidant and cytoprotective genes in several human malignancies, including colorectal, lung, gastric, breast and prostate cancer, clear renal cell carcinoma, and malignant gliomas [50–55]. Similarly, post-transcriptional regulation by microRNAs influences NRF2 signaling. For example, in breast cancer cells, miR-200a directly targets KEAP1 mRNA, reduces KEAP1 protein expression, and thereby promotes

NRF2 activation [56]. Similarly, miR-141-3p suppresses the KEAP1-NRF2 axis and contributes to paclitaxel resistance by attenuating ferroptosis [57]. Together, dysregulation of NRF2-regulating microRNAs may contribute to aberrant pathway activation in cancer.

In parallel with these genetic and epigenetic mechanisms, NRF2 can also be activated through atypical and KEAP1-independent routes. Oncogenic signaling through KRAS, BRAF, and MYC can transcriptionally upregulate NFE2L2, thereby increasing the amount of NRF2 protein available to escape degradation [3,58]. Similarly, activation of the PI3K/AKT pathway suppresses GSK-3β, which, as described above, normally promotes NRF2 degradation through the β-TrCP pathway, resulting in further stabilization of NRF2 [31,33].

In addition to its redox and anabolic functions, NRF2 is a major regulator of drug detoxification and therapy resistance, and these roles overlap substantially with the phase I/II and cytoprotective genes described above [36,37,42]. By coordinately inducing these detoxifying and conjugating enzymes, NRF2 enhances the ability of cancer cells to metabolize and neutralize cytotoxic agents. It further promotes drug efflux by upregulating ABC transporters such as ABCG2, ABCC1, and ABCC2, which actively export chemotherapeutics including platinum compounds, anthracyclines, and taxanes [36,42]. Together with NRF2-driven increases in glutathione levels, which support conjugation and inactivation of electrophilic anticancer agents [37], this network underlies much of NRF2's contribution to multidrug resistance in cancer.

This transcriptional program carries profound clinical significance. Constitutive or high nuclear NRF2 expression is associated with resistance to cisplatin in lung and ovarian cancers, sorafenib in hepatocellular carcinoma, imatinib in chronic myeloid leukemia, and bortezomib in multiple myeloma [32,36,44], and high NRF2 activity consistently correlates with poorer patient outcomes and shorter overall survival across several malignancies [28,30,31]. Emerging evidence also suggests that NRF2 influences responses to targeted therapies and immunotherapies by modulating antigen presentation, shaping an immunosuppressive tumor microenvironment, and interacting with the endoplasmic reticulum stress response and the unfolded protein response [59,60]. Thus, NRF2 contributes not only to intrinsic cytoprotection but also to broader adaptive programs that can limit the effectiveness of therapy.

Critically, however, the effects of NRF2 in cancer depend strongly on biological context, including tumor stage and cellular background. In normal tissues and early carcinogenesis, NRF2 acts predominantly as a tumor suppressor, limiting oxidative DNA damage, reducing mutagenesis, enhancing carcinogen detoxification, and restraining inflammatory signaling [30,58]. NRF2-deficient models confirm that intact NRF2 signaling protects against malignant initiation [12,35]. Once malignant transformation occurs, however, sustained NRF2 activation is hijacked by tumor cells to support survival, metabolic plasticity, and resistance to treatment [35]. This dual role, referred to as the "NRF2 paradox" [35], presents a major therapeutic challenge: the very pathway that shields healthy tissues from carcinogenic injury also equips established tumors with the tools needed for persistence, progression, and multidrug resistance [3,35,59].

NRF2 is therefore far more than an antioxidant guardian. In advanced tumors, it becomes a liability, protecting cells from chemotherapy, rewiring metabolism for survival, and locking in drug resistance. NRF2 activation reshapes tumor biology across several interconnected processes, spanning redox defense, anabolic metabolism, and drug detoxification. At the same time, its protective role in normal tissues nonetheless underscores the need for therapeutic strategies tailored to disease context and tumor genotype [2,3,37,58].

3.2. NRF2-Mediated Metabolic Reprogramming

Metabolic abnormalities within tumors provide an additional layer of NRF2 regulation. Oncometabolites such as fumarate, which accumulates in fumarate hydratase-deficient tumors, and succinate, which accumulates in SDH-mutant tumors, can modify KEAP1 cysteine residues and disrupt its regulatory capacity [3,61,62]. Pathways involved in nutrient sensing and metabolic status provide additional links between cellular energy state and NRF2 activity. AMPK can directly phosphorylate NRF2 at three serine residues, thereby enhancing expression of its target genes [63] and can also influence factors involved in NRF2 stabilization, integrating redox defense with energy availability and biosynthetic demand. Furthermore, AMPK also acts as a key negative regulator of mTOR: activated by cell stress signals such as ROS, AMPK inhibits mTOR signaling. thereby restraining mTOR-driven glycolysis and tumor progression [64]. Together, these findings indicate that NRF2 activation in cancer cannot be explained solely by KEAP1 or NFE2L2 mutations, as tumor metabolism itself can also modulate pathway activity [3,60].

Once activated, NRF2 drives a coordinated metabolic program that supports tumor growth and stress adaptation, a phenomenon often termed "NRF2 addiction" [65,66]. This reprogramming affects glutathione biosynthesis, NADPH generation, and anabolic carbon flux [3,66]. Among the established direct NRF2 targets, GCLC and GCLM encode the catalytic and modifier subunits of glutamate–cysteine ligase, the rate-limiting enzyme complex in de novo glutathione biosynthesis [67]. NRF2 also induces glutathione reductase (GSR), which reduces oxidized glutathione (GSSG) back to reduced glutathione (GSH), thereby supporting glutathione recycling and the maintenance of a reduced intracellular environment [68]. In addition, NRF2 regulates glutathione S-transferases (GSTs), which catalyze the conjugation of GSH with electrophiles and cytotoxic compounds, thereby contributing to detoxification and, in cancer cells, to the inactivation of certain anticancer agents [69]. By increasing intracellular GSH pools, tumor cells gain an enhanced capacity to neutralize ROS, preserve a reduced intracellular environment, and protect DNA, lipids, and proteins from oxidative damage [3,59,70,71]. Furthermore, NRF2 also promotes anabolic metabolism by redirecting carbon flux through the pentose phosphate pathway (PPP) and associated biosynthetic routes. Established direct NRF2 target genes include glucose-6-phosphate dehydrogenase (G6PD) and phosphogluconate dehydrogenase (PGD), which catalyze the oxidative branch of the pentose phosphate pathway (PPP) and generate NADPH, as well as transketolase (TKT) and transaldolase 1 (TALDO1), which participate in the non-oxidative branch of the pathway [71]. Through these genes, NRF2 diverts glucose into the PPP to generate NADPH for antioxidant defense and reductive biosynthesis, and ribose-5-phosphate for nucleotide synthesis. In cancer models, NRF2 activation has been shown to redirect glucose and glutamine into anabolic pathways, supporting nucleotide and lipid synthesis, and the maintenance of redox homeostasis [71]. Concurrently, NRF2 influences glutamine utilization and enhances the transcription of genes involved in serine biosynthesis and one-carbon metabolism [3,33]. This redistribution of carbon into biosynthetic pathways fulfills the biosynthetic demands of proliferating tumor cells while enabling them to survive hostile microenvironmental conditions, such as hypoxia, nutrient deprivation, and elevated oxidative stress [3,28,31,35,59,60].

4. Aquaporins

Aquaporins (AQPs) are a conserved family of small integral membrane proteins that facilitate the passive, concentration gradient-dependent transport of water and other small solutes across membranes [72,73]. The first aquaporin, AQP1, was discovered in 1992 by Peter Agre and colleagues, and this contribution was recognized with the 2003 Nobel Prize in Chemistry [74]. Since then, 13 human isoforms (AQP0-AQP12) have been identified,

each with a distinct tissue distribution, substrate selectivity, and physiological roles [75]. Structurally, each aquaporin monomer consists of six transmembrane alpha-helical domains connected by five interhelical loops (A–E), with both termini in the cytoplasm [75] (Figure 2). Two highly conserved NPA (asparagine–proline–alanine) motifs, located in loops B and E, meet at the center of the pore to form the characteristic hourglass constriction responsible for water selectivity and proton exclusion, while an aromatic/arginine (ar/R) selectivity filter narrows the pore to approximately 3 Å, creating an electrostatic barrier against ions [75,76]. In the membrane, aquaporin monomers form homotetramers, with each monomer functioning as an independent channel [72]. Based on substrate selectivity, aquaporins are classified into three groups: classical aquaporins (AQP0, AQP1, AQP2, AQP4, AQP5, AQP6, AQP8), which are primarily selective for water; aquaglyceroporins (AQP3, AQP7, AQP9, AQP10), which also transport glycerol, urea, and other small neutral solutes; and superaquaporins (AQP11, AQP12), which are structurally different and localized intracellularly [75,77]. Their tissue-specific expression supports a broad range of physiological functions, including renal water reabsorption, cerebrospinal fluid secretion, brain osmoregulation, exocrine gland secretion, skin hydration, and sperm maturation, and their dysregulation is associated with pathological conditions ranging from nephrogenic diabetes insipidus and neuromyelitis optica to cancer [72].

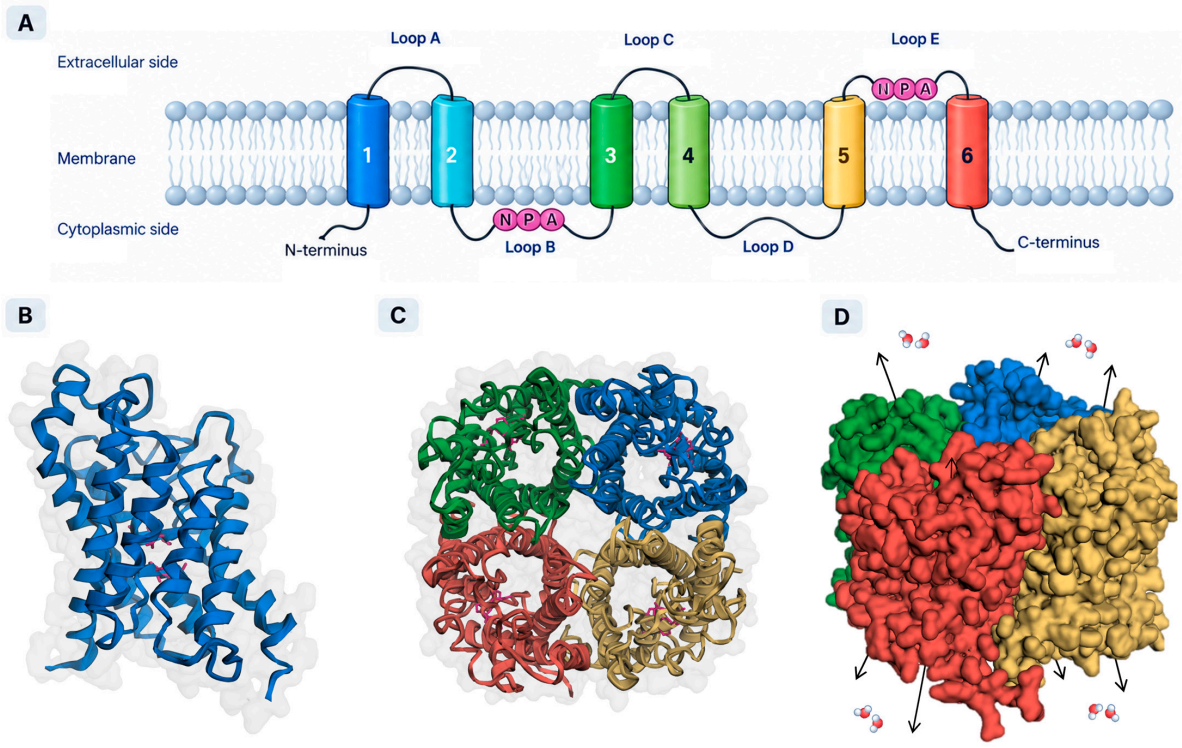


Figure 2. Aquaporin structure. Panel (**A**) each aquaporin monomer consists of six transmembrane alpha-helical domains connected by five interhelical loops (A–E); panel (**B**) ribbon representation of a single aquaporin monomer, with the NPA motifs highlighted in magenta; panel (**C**) top view of the aquaporin homotetramer, with each monomer shown in a different color. Each monomer forms its own channel and functions independently. (**D**) Surface representation of the tetramer. The arrows and water molecules illustrate the passage of water through the individual monomeric channels.

Beyond their role in water transport, several aquaporins function as peroxiporins, channels that facilitate the transport of hydrogen peroxide (H_2O_2) across the membrane. As AQP-mediated transport is passive and concentration gradient-dependent, solutes can move in either direction; for H_2O_2, this means that AQPs may mediate either influx or efflux

depending on the local redox environment. Because H_2O_2 is an important second messenger in redox signaling, which becomes damaging at higher concentrations, its production and intracellular localization are tightly controlled. Aquaporins have not only been shown to mediate H_2O_2 transport, but their expression profiles also influence cellular susceptibility to extracellular H_2O_2 signaling [78]. AQP8 was the first experimentally confirmed peroxiporin, and since then AQP1, AQP3, AQP5, AQP9, and AQP11 have been shown to be able to transport H_2O_2. As a model for understanding selective H_2O_2 transport by aquaporins, AQP8 has been shown to function as more than a passive, constitutively open channel. Instead, AQP8 undergoes reversible gating during cellular stress, enabling the modulation of the strength and duration of H_2O_2 signaling while preventing its uncontrolled accumulation [79]. This gating is feedback-regulated as persulfidation of the conserved cysteine residue, driven by H_2S generated locally through cystathionine β-synthase activity, closes the channel and terminates H_2O_2 transport [80]. Through this mechanism, AQP8 senses H_2O_2 flux and uses a second transmitter, H_2S, to switch itself off. Besides AQP8, AQP3 has also been shown to facilitate the uptake of both extracellular and NADPH oxidase (Nox)-derived H_2O_2. By increasing cellular permeability to H_2O_2, AQP3 modulates intracellular redox signaling and amplifies downstream signaling pathways that rely on ROS as second messengers [81]. Similarly to AQP8, AQP3 activity is regulated to ensure balanced H_2O_2 transport and prevent excessive intracellular ROS accumulation. One level of regulation is provided by pH-dependent gating, where AQP3 remains active at physiological pH but becomes inhibited under acidic conditions [82]. In addition, AQP3 exhibits H_2O_2-dependent autoregulation, where high H_2O_2 concentrations induce channel closure and reduce membrane permeability [82]. Together, these mechanisms limit excessive ROS influx, thereby protecting cells from oxidative damage while maintaining redox homeostasis. A similar regulatory mechanism has been described for AQP5, whose activity is controlled by extracellular pH in a phosphorylation-dependent manner, with higher channel activity at physiological pH [83]. Importantly, aquaporin regulation extends beyond channel gating and includes changes in gene expression and phosphorylation-dependent membrane trafficking, including translocation to and from the plasma membrane via endocytosis, recycling, and exocytosis, as well as through changes in protein stability and turnover, thereby enabling rapid and tissue-specific control of membrane permeability [84,85]. Furthermore, aquaporin activity is influenced by the upstream regulation of kinases and phosphatases, adding an additional layer of complexity [85]. Collectively, these findings show that peroxiporins are active modulators of redox signaling rather than passive channels for H_2O_2 diffusion. By controlling not just whether, but how much, where, and for how long H_2O_2 crosses the membrane, they directly shape cellular redox homeostasis and influence downstream signaling, adaptation, and stress responses. The relevance of peroxiporins extends beyond physiological redox signaling, as altered aquaporin expression is frequently observed in cancer. A hallmark of cancer cells is the presence of elevated ROS levels resulting from increased metabolic activity, mitochondrial dysfunction, oncogenic signaling, and enhanced ROS production. While excessive ROS levels can be cytotoxic, moderate increases in ROS act as signaling molecules that promote proliferation, survival, migration, angiogenesis, and therapy resistance [86]. Given their ability to regulate the scale, localization, and duration of H_2O_2 signaling, peroxiporins influence these cancer-related processes. Consequently, their dysregulated expression may provide a selective advantage to cancer cells by facilitating redox-dependent signaling while preventing the accumulation of toxic ROS levels.

4.1. Aquaglyceroporins and Cellular Metabolism

Another important role of aquaporins is the transport of glycerol, which makes aquaglyceroporins important regulators of cellular metabolism. Among the 13 human

aquaporins, AQP3, AQP7, AQP9, and AQP10 are classified as aquaglyceroporins and, depending on the isoform, may also facilitate H_2O_2 transport. In this group, AQP3 and AQP9 have been shown to function as both aquaglyceroporins and peroxiporins, thereby linking metabolic regulation with cellular redox homeostasis. Aquaglyceroporins display distinct tissue-specific expression patterns that reflect their physiological functions. The most extensively studied metabolic roles are found in adipose tissue and liver, where coordinated glycerol transport regulates energy homeostasis. AQP3 and AQP7 facilitate glycerol flux from adipocytes during lipolysis, and the released glycerol is then taken up by hepatocytes via AQP9 to serve as a substrate for gluconeogenesis and triglyceride synthesis [87,88]. Once internalized, glycerol is phosphorylated by glycerol kinase to glycerol-3-phosphate, an important metabolic intermediate that can enter glycolysis or gluconeogenesis and contributes to lipid biosynthesis, ATP production, and cellular energy homeostasis. Through the regulation of glycerol transport, aquaglyceroporins influence the balance between energy storage and consumption and participate in the metabolic adaptation of cells and tissues. Besides adipose tissue and liver, different aquaglyceroporins modulate glycerol transport in the kidney, skeletal muscle, heart, pancreas, skin, brain, reproductive system, and placenta [89]. Because the regulation of glycerol flux plays a crucial role in maintaining energy homeostasis, dysregulation of aquaglyceroporins has been associated with a range of metabolic disorders, including obesity, insulin resistance, non-alcoholic fatty liver disease, and cardiac hypertrophy [90]. Furthermore, increasing evidence indicates that dysregulated aquaglyceroporin expression also contributes to cancer, where enhanced glycerol uptake supports the metabolic reprogramming required for tumor growth, survival, and resistance to metabolic stress [91–93].

4.2. Aquaglyceroporins in Cancer

Through their ability to transport both H_2O_2 and glycerol, aquaporins influence redox signaling and cellular metabolism, two processes that are widely reprogrammed in cancer. Cancer cells must continuously adapt to elevated ROS levels and increased metabolic demands to sustain proliferation, survival, invasion, and therapy resistance. As shown in Table 1, dysregulated expression of aquaporins has been documented in numerous cancer types and was associated with cancer progression and poor clinical outcomes across a range of malignancies. Among aquaporins, AQP3 is one of the most extensively studied and consistently upregulated isoforms. Although expressed in normal tissues, AQP3 levels are frequently elevated in cancer compared with the corresponding healthy tissue [94]. Its frequent overexpression may be related to its dual role as both an aquaglyceroporin and a peroxiporin, enabling it to regulate cellular metabolism and redox signaling simultaneously, thereby promoting multiple hallmarks of cancer progression. AQP-mediated H_2O_2 transport can amplify or diminish downstream signaling cascades that rely on ROS as second messengers. AQP3-mediated H_2O_2 uptake facilitates Akt activation, while AQP3 silencing attenuates PI3K/Akt signaling in breast cancer cells [95]. Through activation or inhibition of cellular signaling pathways, AQP3 regulates several processes associated with cancer progression, including matrix metalloproteinase expression, epithelial–mesenchymal transition (EMT), proliferation, migration, invasion, and chemoresistance. Although AQP-mediated H_2O_2 transport can occur in either direction, the cancer models discussed here mainly demonstrate AQP-mediated influx of extracellular, often NOX-derived H_2O_2. Mechanistically, AQP3-mediated H_2O_2 uptake has been shown to oxidize and inactivate PTEN, resulting in sustained Akt signaling and activation of downstream mTOR-dependent pathways [96]. In lung adenocarcinoma, this mechanism suppresses autophagy and promotes cancer cell proliferation [96], whereas in breast cancer, AQP3-dependent transport of Nox2-derived H_2O_2 enhances Akt signaling and promotes migration and metastasis [97]. Simi-

larly, in cervical cancer, AQP3 promotes invasion and metastatic dissemination through a Nox4/H_2O_2/Syk/PI3K/Akt signaling axis [81], while in gastric cancer, AQP3 contributes to cisplatin resistance through the regulation of autophagy [98]. This was further supported by studies showing that AQP3 silencing decreases glycerol uptake, impairs lipogenesis, inhibits autophagy, and promotes apoptosis [91]. Moreover, AQP3 inhibition reduces ATP and lipid production and suppresses PI3K/Akt signaling in gastric cancer [92], highlighting the importance of AQP3-mediated glycerol metabolism in sustaining tumor growth and survival. In EBV-associated gastric cancer, AQP3 contributes to cancer cell proliferation, migration, and autophagy by activating ERK-dependent 4E-BP1 signaling [99]. Importantly, not only AQP3 expression and activity, but also its subcellular localization appear to influence tumorigenesis. In breast and prostate cancer, altered localization of aquaporins from the plasma membrane to the cytoplasm has been observed during disease progression [100,101], suggesting that changes in channel trafficking and cellular distribution may contribute to cancer development and aggressiveness. Together, these studies demonstrate a predominantly pro-tumorigenic role for AQP3 and support its potential value as both a prognostic biomarker and therapeutic target. While the oncogenic role of AQP3 has been extensively characterized, less is known about the contribution of other aquaglyceroporins. Emerging evidence indicates that AQP7 also contributes to cancer progression through the regulation of cellular metabolism. AQP7 expression varies among cancer types, suggesting a context-dependent role and potential prognostic value [94]. In breast cancer, elevated AQP7 expression has been associated with poor overall survival [93]. Consistent with these observations, AQP7 depletion in a mouse model reduced primary tumor growth and lung metastasis while altering multiple metabolic pathways [93], highlighting its role in metabolic adaptation during cancer progression. In contrast, AQP10 mRNA expression has been reported to be upregulated in breast cancer, and higher AQP10 levels were associated with favorable relapse-free survival [102]. However, the role of AQP10 in cancer remains poorly understood [94]. AQP9 displays different expression patterns in cancer. In most malignancies, increased AQP9 expression is associated with cancer progression and poorer patient survival, whereas in hepatocellular carcinoma (HCC) it is frequently downregulated [94]. This observation is particularly noteworthy because AQP9 is the predominant aquaglyceroporin in normal hepatocytes, but its reduced expression has been associated with enhanced proliferation, EMT, and poor prognosis in HCC [103–105]. In contrast, AQP3 expression is frequently increased in HCC and has been linked to enhanced proliferation, migration, invasion, and metastatic potential [105–107]. Because both proteins regulate glycerol metabolism and redox homeostasis, their opposing expression patterns may contribute to the metabolic reprogramming and stress adaptation characteristic of HCC. Collectively, these findings indicate that aquaglyceroporins have distinct, tissue-dependent functions in cancer. While AQP3 generally promotes cancer progression through the coordinated regulation of redox signaling and metabolic adaptation, the roles of AQP7, AQP9, and AQP10 appear to be more context-dependent and influenced by tissue-specific metabolic requirements. Moreover, through their ability to regulate ROS signaling and metabolic substrate availability, aquaglyceroporins may facilitate adaptation to the oxidative and metabolic stresses imposed by the tumor microenvironment and anticancer therapies. This adaptive capacity is particularly relevant in the context of NRF2-dependent antioxidant responses, which are increasingly recognized as major determinants of cancer progression and therapy resistance.

Table 1. Aquaporins associated with cancer.

	Associated Cancer	Reference
AQP1	brain, lung, colorectal, breast, ovarian, and endometrial cancer, multiple myeloma	[108–120]
AQP2	lung and endometrial cancer	[121,122]
AQP3	pituitary, salivary gland, lung, thymic, gastric, esophageal, pancreatic, colorectal, breast, ovarian, prostate, testicular, urothelial, and skin cancer	[91,92,96,106,107,111,116,117,123–129]
AQP4	brain, thyroid, and colorectal cancer	[130–134]
AQP5	lung, gastric, pancreatic, colorectal, breast, ovarian, and cervical cancer	[111,116,117,119,128,135–145]
AQP6	ovarian cancer	[146]
AQP7	breast cancer	[93]
AQP8	leukemia, ovarian cancer	[146–148]
AQP9	liver, lung, and prostate cancer	[103–105,121,142,143,149]
AQP10	breast cancer	[102]

5. Organ-Specific Mechanisms of Cancer Resistance

The development of resistance depends on the ability of tumor cells to adapt to persistent oxidative, metabolic, and therapeutic stress. As discussed above, NRF2 can coordinate much of this adaptive capacity by regulating antioxidant defense, detoxification, and metabolic reprogramming [27,58,59]. AQPs can contribute to these stress axes through distinct mechanisms: peroxiporins can control membrane flux of H_2O_2, thereby shaping the intensity and duration of ROS signaling, while some isoforms control glycerol flux, further affecting energy metabolism and lipid biosynthesis [79,87,88,150]. Because AQPs can directly influence intracellular levels of ROS and availability of metabolic substrates, they intersect with NRF2-driven antioxidant and metabolic programs, positioning the NRF2–AQP axis as a coordinated system for tumor stress adaptation. It is important to note that no study has yet directly demonstrated NRF2-AQP regulation or a causal NRF2-AQP axis in treatment-resistant tumors. Available studies have largely examined NRF2 or AQPs separately, and direct evidence linking AQP dysregulation to NRF2-driven therapy resistance remains limited. Nevertheless, because both NRF2 and AQPs regulate oxidative stress and H_2O_2-dependent signaling, they are discussed in parallel as potentially interacting components of tumor stress adaptation. Rapid proliferation and treatment-related stress often increase ROS levels, and NRF2 helps cells cope with this pressure by upregulating antioxidant defenses and reducing oxidative cell death [28,30,35], while peroxiporin-mediated H_2O_2 transport further modulates the scale and localization of this redox signaling [79,150]. NRF2 also supports therapy resistance by reprogramming metabolic pathways, diverting glucose and glutamine into anabolic pathways that fuel nucleotide synthesis, NADPH generation, glutathione maintenance, and biomass production [3,31–33,37,42,45,71,151], which runs parallel to the metabolic role of aquaglyceroporins in supplying glycerol-derived substrates for lipogenesis and energy production [87,88,91,92]. This intertwining of redox and metabolic adaptation, mediated by NRF2 and AQPs, may be particularly relevant in tissues such as breast, colon, and liver, where both pathways have been independently linked to therapy resistance. The sections below examine how NRF2- and AQP-driven adaptation contribute to resistance in each of these cancers.

5.1. Breast Cancer

Breast cancer is the most frequently diagnosed malignancy among women worldwide. The disease is a heterogeneous group of malignancies that arise in the mammary glands, with carcinomas making up the majority of cancers. Sarcomas and angiosarcomas are rarely seen. Incidence is highest in industrialized countries, largely owing to lifestyle-related risk factors such as poor diet, smoking, chronic stress, and low physical activity [152,153].

Based on criteria of pathological features and invasiveness, breast cancer can be divided into three major groups: non-invasive (or in situ), invasive, and metastatic breast cancer. Invasive ductal carcinoma is the most common form of invasive breast cancer and is characterized by the invasion of malignant ductal cells into the surrounding breast stroma. Invasive lobular carcinoma is the second most common invasive subtype, originating from the mammary lobules and typically exhibiting an infiltrative growth pattern of single tumor cells or cells arranged in linear formations. Metastatic breast cancer (mBC) represents advanced disease in which tumor cells disseminate beyond the primary breast tumor to distant sites, most commonly the bones, liver, lungs, and brain [153,154].

Although early-stage disease is often curable, the transition to the metastatic state marks a fundamental shift in clinical outcome. Despite numerous therapeutic options, including chemotherapy, immunotherapy, endocrine therapy, targeted therapy, and radiotherapy, mBC inevitably develops resistance and progresses. This resistance arises from a complex interplay of mechanisms, including the accumulation of genetic mutations, epigenetic modifications, and alterations in the tumor microenvironment [154,155].

Among the diverse resistance mechanisms in mBC, NRF2 and aquaglyceroporins are increasingly implicated, both regulating transmembrane transport, antioxidant status, metabolism, and H_2O_2 levels to activate survival pathways [59,77,156,157]. This is particularly relevant in breast cancer, where aggressive tumor behavior is strongly linked to altered membrane transport, redox signaling, and metabolic flexibility [27,58–60]. No study has yet directly demonstrated a mechanistic link between NRF2 and aquaglyceroporins in breast cancer. The available data support functional overlap rather than established causality: NRF2 enables tumor cell survival by activating antioxidant programs, detoxification systems, and anabolic metabolism, while aquaglyceroporins may facilitate the transmembrane movement of substrates and signaling molecules required for these same adaptive processes. Together, these systems may help breast cancer cells maintain intracellular homeostasis during oxidative stress, remain metabolically active under shifting nutrient availability, and better tolerate treatment-related pressure, contributing to a resistant phenotype marked by enhanced stress tolerance, survival, and potentially increased invasive capacity [3,12,31,45]. We next examine how these two systems intersect across the major therapeutic modalities used in mBC—endocrine therapy, chemotherapy, targeted therapy, immunotherapy, and radiotherapy—highlighting the resistance mechanisms involved and the proposed contribution of NRF2 and aquaglyceroporins.

Endocrine therapy. In hormone receptor-positive (HR+) mBC, resistance to endocrine therapy evolves through several well-defined routes. One involves mutations in the ESR1 gene under aromatase inhibitor pressure, producing structural changes in the estrogen receptor that render it constitutively active independent of estrogen [158,159]. Another involves activation of alternative growth factor pathways, such as PI3K/AKT/mTOR hyperactivation driven by PIK3CA mutations or loss of the tumor suppressor PTEN [160]. HR+ BC cells can also acquire resistance by shifting reliance from estrogen receptor signaling to alternative pathways, including MAPK or HER2 signaling cascades [161,162]. Among these routes, the interplay between estrogen signaling, AQP3, and NRF2 has not been fully characterized, but existing data suggest it merits attention.

Estrogen signaling directly upregulates AQP3 through estrogen response elements (EREs) in its promoter [163]. Even after the onset of endocrine resistance, AQP3 continues to support cell motility and invasion in resistant breast cancer cells, pointing to a broader role beyond its initial estrogen-driven induction [164]. AQP3 may mediate this resistance by facilitating transmembrane H_2O_2 influx [150]: intracellular H_2O_2 accumulation inactivates phosphatases such as PTEN via oxidation, cross-activating HER2 and PI3K/AKT signaling, while imported H_2O_2 can also oxidize key cysteine residues on KEAP1, triggering NRF2 nuclear translocation [165]. Once in the nucleus, NRF2 coordinates metabolic reprogramming and upregulates cytoprotective and detoxification pathways, enabling HR+ breast cancer cells to resist endocrine therapy and its associated therapeutic stress [166].

Chemotherapy. In mBC, chemotherapy is used alone or in combination with targeted therapy or immunotherapy, particularly in more aggressive subtypes such as HER2+ or triple-negative breast cancer (TNBC); in HR+ disease, it is typically introduced after endocrine resistance develops. Because chemotherapy preferentially targets rapidly dividing cells, cancer cells employ several metabolic and mechanical strategies to evade its effects. These include overexpression of ABC transporter proteins that actively efflux cytotoxic agents such as taxanes and anthracyclines before they can cause intracellular damage [167–169], the presence of cancer stem cells, a slow-cycling or quiescent subpopulation with reduced susceptibility to cell-cycle-dependent agents and frequently enhanced drug-efflux capacity [170], and, against DNA-damaging agents such as platinum salts, upregulation of DNA damage repair pathways, including mismatch repair machinery and PARP enzymes, allowing faster repair of genomic damage than chemotherapy can inflict [171,172].

Chemotherapy-induced ROS, such as H_2O_2, causes DNA damage and can increase intracellular NRF2 levels, which in turn induce ABC efflux pump expression to export paclitaxel and doxorubicin, and stimulate glutathione synthesis to neutralize ROS [69,173]. In parallel, overexpression of AQP3 and AQP7 enhances glycerol uptake, driving lipid synthesis and oxidative phosphorylation; the resulting ATP production supplies the energy needed for efflux pump activity, antioxidant protection, and cytoprotective autophagy [93,94,174].

Targeted therapy. Targeted therapies such as anti-HER2 agents, CDK4/6 inhibitors, and PI3KCA inhibitors precisely block specific oncogenic drivers, but tumors eventually evolve mechanisms to bypass this suppression. In HER2+ tumors, one such mechanism is truncation of the HER2 receptor (p95HER2), which loses its extracellular domain—preventing trastuzumab binding—while retaining an intact, active intracellular kinase domain [175]. Another is compensatory upregulation of alternative receptor tyrosine kinases, such as HER3 or MET, which share downstream effector pathways and effectively reroute signaling [175,176].

Resistance to CDK4/6 inhibitors combined with endocrine therapy can be mediated by loss-of-function mutations in the tumor suppressor RB1 [177], or by amplification/gain-of-function of CCNE1 (encoding cyclin E) [178] or CDK2/CDK6 [179], which drive cell division independent of CDK4/6 blockade. Beyond these genetic alterations, metabolic adaptation offers a complementary route to resistance: in addition to the role of NRF2 in metabolic reprogramming that enables tumor cell escape from other targeted therapies, a similar NRF2-driven acceleration of the PPP may support S-phase progression and nucleotide synthesis, potentially contributing to escape from CDK4/6 blockade in HR+ breast cancer, though this specific mechanism remains to be directly demonstrated [77].

Immunotherapy. Cancer cells evade immune-mediated killing through at least three complementary strategies: creating an immunologically silent microenvironment, recruiting immunosuppressive cell populations, and upregulating alternative checkpoint pathways. Immune checkpoint inhibitors expose cancer cells to T-cell-mediated immunity, activating

cytotoxic T-cells that generate ROS to destroy tumor cells. To evade this, cancer cells can create an "immune desert" by downregulating MHC class I molecules or mutating beta-2-microglobulin, rendering them invisible to T-cells [180].

High NRF2 expression lowers overall intracellular ROS levels within the tumor, reducing the oxidative stress signals that normally mobilize immune cells and thereby fostering an "immune cold" environment [181]. Cancer cells also recruit immunosuppressive populations such as myeloid-derived suppressor cells, regulatory T-cells, and M2-polarized macrophages, which actively inhibit cytotoxic T-cells [182]. When checkpoint inhibitors block the PD-1/PD-L1 axis, cancer cells can further upregulate alternative checkpoints such as LAG-3 or TIM-3 to suppress immune activity again [183,184]. AQP3 and AQP5 may similarly contribute to immune evasion by exporting H_2O_2 from the cancer cell cytoplasm, since cytotoxic T-cells rely on ROS to kill tumor cells; this hypothesis is of particular interest in triple-negative breast cancer, where both transporters serve as prognostic markers [100], though direct evidence for this immune-shielding role is still lacking.

Radiotherapy. Radiotherapy induces double-strand DNA breaks and ROS formation, ultimately driving cancer cell death, but cancers can acquire features that promote radioresistance. Advanced, disseminated tumors often have a hypoxic core, limiting the oxygen needed to generate ROS from radiation. In hypoxic cancer cells, NRF2 stabilizes HIF-1α, shifting cellular metabolism toward oxygen independence and promoting radioresistance; these cells also produce free-radical scavengers such as glutathione to neutralize ROS before structural damage occurs [69,185]. Radiotherapy can also trigger epithelial-to-mesenchymal transition, conferring breast cancer cells with increased motility, a stem-cell-like phenotype, and resistance to radiation-induced cell death [186,187].

Across endocrine, cytotoxic, targeted, immune, and radiation-based therapies, NRF2 consistently emerges as a central driver of resistance through coordinated redox control, metabolic adaptation, and cytoprotective gene expression. Direct mechanistic evidence linking aquaglyceroporins to NRF2-driven resistance remains limited, but the available data support a functional convergence between NRF2-mediated stress adaptation and aquaglyceroporin-facilitated transport across all these treatment contexts, with both mechanisms jointly contributing to the maintenance of metabolic and redox homeostasis in breast cancer cells under therapeutic pressure [31,37,59].

5.2. Colorectal Cancer

Colorectal cancer (CRC) is one of the most frequent cancers worldwide, along with prostate, lung, and breast cancer. With over 1.9 million new cases and 903,000 deaths estimated in 2020, incidence is predicted to increase to 3.2 million new cases by 2040 [188]. Most CRC cases are diagnosed in adults over 50 years old; however, over the past decade, incidence has risen among young adults, and CRC is now the third leading cause of cancer death in this age group [189–191].

CRC development begins with the accumulation of genetic and epigenetic alterations in normal colon epithelial cells, leading to abnormal proliferation and the formation of precancerous lesions such as adenomatous polyps, which progressively increase in size, accumulate additional molecular alterations, and can subsequently progress to carcinoma [190,192]. In some cases, these alterations arise in colonic stem cells located at the base of the crypts of Lieberkühn, which continuously generate differentiated epithelial cells and drive the rapid renewal of the intestinal epithelium, occurring approximately every 2–7 days, with mutations affecting oncogenes, tumor suppressors, and pathways regulating proliferation and differentiation promoted by genetic and environmental factors, including carcinogens and infectious agents [190,192]. Although these molecular alterations are central to CRC development, chronic inflammation and dysregulation of the host immune response also

contribute to tumor initiation and progression, with inflammatory conditions of the colon associated with increased CRC risk [193]. Lifestyle and dietary factors further influence CRC development: high consumption of processed and red meat, alcohol intake, and smoking, together with low dietary fiber consumption, have all been associated with increased risk [194].

CRC is a genetically and epigenetically heterogeneous disease. Transformation of normal colonic cells involves mutations in oncogenes and tumor suppressor genes, together with epigenetic alterations such as DNA methylation, histone modifications, and changes in microRNA expression, giving rise to distinct molecular features including chromosomal instability (CIN), microsatellite instability (MSI), mismatch repair (MMR) deficiency, and the CpG island methylator phenotype (CIMP). To account for this heterogeneity, the Colorectal Cancer Subtyping Consortium integrated six previously established gene-expression-based classification systems in 2015; analysis of 4151 patients across 18 datasets led to the Consensus Molecular Subtypes (CMS), which classify CRC into four subtypes. CMS1 (MSI-immune) is characterized by microsatellite instability, frequent MMR deficiency, hypermutation, and a strong immune response. CMS2 (canonical) exhibits epithelial differentiation and activation of canonical signaling pathways, particularly WNT and MYC, and is frequently associated with chromosomal instability [195,196]. CMS3 (metabolic) is characterized by metabolic dysregulation and a higher frequency of KRAS mutations. CMS4 (mesenchymal) displays prominent TGF-β signaling, epithelial–mesenchymal transition, stromal infiltration, and angiogenesis, and is associated with a more aggressive phenotype and poorer prognosis [195,196].

Once diagnosed, CRC is treated with surgery, chemotherapy, or radiotherapy depending on stage; for lower-stage, localized tumors, surgery is the best available option [192]. Despite these options, CRC mortality remains high, as many patients are diagnosed at advanced stages—only 39% are diagnosed early, while the tumor is still localized. The 5-year survival rate is approximately 90% for patients diagnosed at an early stage but declines to between 71% and 14% for those diagnosed at more regional or distant stages; these high mortality rates are largely attributable to late-stage diagnosis and metastasis [197].

5-fluorouracil (5-FU) is a synthetic fluorinated pyrimidine analog and the most widely used chemotherapeutic agent for CRC. Its anticancer effects are exerted by its active metabolites, which disrupt DNA and RNA synthesis [198]. In addition, 5-FU kills cancer cells via excessive ROS generation, overwhelming the already elevated basal ROS levels in cancer cells and triggering oxidative stress-mediated apoptosis [199].

Kang et al. (2014) investigated NRF2 expression in the 5-FU-resistant CRC cell line SNUC5 [200], finding that SNUC5/5-FUR cells expressed higher NRF2 than non-resistant SNUC5 cells. Through epigenetic modifications, NRF2 overexpression allowed cells to overcome high ROS levels and evade oxidative stress; silencing NRF2 (siNRF2) reversed this effect, resensitizing cells to 5-FU, reducing their proliferation rate, and increasing cytotoxicity. In the HT29 cell line, NRF2 is additionally activated by 5-FU itself, possibly due to elevated ROS levels [201]. In another study, oncogene-induced NRF2 (via K-Ras, B-Raf, or Myc) transcription promoted ROS detoxification and carcinogenesis [202].

Radiotherapy is another widely used therapy for CRC patients, particularly those with more advanced disease. It harms cancer cells both directly, by damaging biomolecules such as proteins, lipids, and DNA, and indirectly, through the production of radiation-derived ROS [203]. Due to cancer cell heterogeneity, subpopulations within advanced-stage tumors can similarly adapt to radiation-induced oxidative stress via NRF2-driven antioxidant and metabolic reprogramming, extending the resistance phenotype described above for chemotherapy to radiotherapy as well [204,205].

In addition to their metabolic roles discussed above, several peroxiporin isoforms are also dysregulated in CRC and support tumor progression through distinct mechanisms: AQP1 contributes to tumor angiogenesis and endothelial cell migration, AQP3 promotes cancer cell migration and is linked to metastasis via EGF-driven PI3K/Akt signaling, and AQP5 drives proliferation and EMT through Ras and Smad2/3 signaling, respectively [73]. In HT20 colorectal cancer cells, AQP1-mediated plasma membrane water permeability was shown to promote cell migration, and AQP1 expression increased extravasation of these cells in the lungs of nude mice following tail-vein injection, implicating AQP1 in colon cancer invasion and metastasis [206]. AQP1 blockade using inhibitor AqB013 in HT29 and HCT-116 cell lines reduced migration in HT29 cells, which have high baseline AQP1 expression, but had no effect on HCT-116 cells, which express lower levels of AQP1 [207]. AqB013 also achieved near-complete inhibition of endothelial tube formation in an angiogenesis assay [207]. Further, AQP5 was detected in about 62% of human CRC tissue samples, with no staining in surrounding normal mucosa, and was highly expressed across the CRC cell lines RKO, HCT116, DLD1, and SW480 relative to the NSCLC line H838, with AQP5 promoting proliferation via ERK phosphorylation through the Ras signaling pathway [141].

5.3. Liver

Liver cancer is one of the most frequent cancers globally, with an estimated 529,202 incident cases and 483,875 deaths caused by this disease. In the span of eleven years, from 2010 to 2021, the global numbers for incidence and death from liver cancer rose by 26% and 25%, respectively [208]. In contrast to cancers such as breast, lung, and colorectal cancer, liver cancer commonly develops in chronically damaged liver tissue as a consequence of multiple interacting risk factors. Thus, lifestyle and environmental factors play an important role in liver cancer development. The development of liver cancer is associated with multiple risk factors, including alcohol-related cirrhosis, smoking, obesity, diabetes, iron overload, dietary exposures, hepatitis B and C virus infections, and fatty liver disease [209]. HCC accounts for almost 90% of liver cases diagnosed globally, while cholangiocarcinoma (CCA) is the second most prevalent primary liver malignancy after HCC [210].

The prognosis for liver cancer is poor, with only a limited proportion of patients being eligible for surgical removal, which is primarily applicable to patients diagnosed at an early stage [211]. For those patients diagnosed with more advanced-stage liver cancer, treatment options include transarterial chemoembolization (TACE), radiotherapy, or oral dosing with sorafenib, a kinase inhibitor and the established option for late-stage cases. In a multicenter, phase 3, double-blind, placebo-controlled trial, Llovet et al. evaluated the efficacy and safety of sorafenib in 602 patients with advanced hepatocellular carcinoma who had not previously received systemic treatment, reporting a nearly three-month improvement in median overall survival and time to radiologic progression compared with placebo [212].

Although sorafenib improved overall survival in advanced hepatocellular carcinoma, objective responses were observed in only a small proportion of patients, while the median time to radiologic progression was 5.5 months. These findings highlight the limited durability of the therapeutic response and the development of treatment resistance in a substantial proportion of patients [211].

One factor potentially contributing to therapy resistance in liver cancer is the highly oxidative environment of the liver. The liver is one of the most mitochondria-rich organs, reflecting its high metabolic demands and central role in glucose, lipid, and protein metabolism. Mitochondrial oxidative metabolism, including fatty acid β-oxidation, is an important source of ROS, which are generated in part through electron leakage from the mitochondrial electron transport chain [213]. At the same time, chronic inflammation associated with liver disease can further increase ROS production through the activation of

neutrophils and Kupffer cells. Consequently, hepatocytes can be exposed to persistently elevated levels of ROS, including H_2O_2, which can cross cellular membranes and modulate redox signaling [214].

Increasing evidence suggests that adaptation to oxidative stress contributes to sorafenib resistance in HCC [215–218]. In an integrative transcriptomic study combined with functional pharmacological interventions, Michilli et al. demonstrated that adaptation to oxidative stress and activation of antioxidant defense mechanisms contribute to the maintenance of tumor cell survival during prolonged sorafenib exposure [215]. Their findings suggest that NRF2-driven transcriptional reprogramming is a key adaptive mechanism underlying sorafenib resistance by enhancing antioxidant defenses, reducing oxidative stress, and promoting metabolic adaptation. Constitutive NRF2 activation has also been associated with resistance to multiple chemotherapeutic agents and radiotherapy, suggesting that NRF2-mediated antioxidant responses may contribute to therapeutic resistance across different treatment modalities in HCC and other malignancies [215].

Ma-on et al. analyzed paraffin-embedded HCC tissues and found that elevated ROS levels correlated with increased NRF2 activation and higher levels of 8-hydroxydeoxyguanosine (8-OHdG), a marker of oxidative DNA damage [219]. These findings were confirmed in vitro: H_2O_2-induced oxidative stress in HepG2 cells similarly increased NRF2 activation and 8-OHdG levels, and also enhanced cell migration and invasion in wound-healing assays, supporting a role for oxidative stress in promoting HCC progression [219].

NRF2-mediated adaptation may further promote tumor cell survival by regulating anti-apoptotic proteins such as Bcl-xL [220]. NRF2 activation by tBHQ (tert-butylhydroquinone) increased Bcl-xL expression in Hepa-1 and HepG2 cells, and an antioxidant response element (ARE) was identified at nucleotides −608 to −600 in the Bcl-xL promoter that bound NRF2 and drove its expression. NRF2 inhibition and overexpression correspondingly decreased and increased Bcl-xL levels, confirming a direct regulatory relationship. Furthermore, inhibiting NRF2 or Bcl-xL increased susceptibility to etoposide-induced cell death, demonstrating that NRF2 promotes cell survival and apoptosis resistance through Bcl-xL upregulation [220].

In addition, the NRF2 signaling pathway acts as a key negative regulator of ferroptosis in HCC cells [221]. Ferroptosis is an iron-dependent form of regulated cell death driven by lipid ROS accumulation, and can be pharmacologically inhibited by iron chelators and lipid peroxidation inhibitors. p62 promotes Keap1 inactivation, preventing NRF2 degradation and facilitating its nuclear accumulation, where it induces cytoprotective genes involved in redox and iron homeostasis, including NQO1, HO-1, and FTH1. NQO1 and HO-1 support the antioxidant response, while FTH1 promotes iron storage, together limiting oxidative damage and lipid peroxidation. Accordingly, NRF2 suppression increased ferroptosis sensitivity, whereas inhibition of NQO1, HO-1, or FTH1 enhanced ferroptosis in HCC cells [221].

Beyond NRF2, aquaporins provide an additional level of H_2O_2 regulation by facilitating its transmembrane transport, potentially influencing intracellular redox homeostasis. Persistent oxidative stress can therefore promote cellular adaptations that enhance tumor cell survival under unfavorable conditions and may contribute to reduced sensitivity to anticancer therapies. Numerous studies have reported that AQPs play key roles in tumor biology, facilitating cancer cell migration, proliferation, and angiogenesis [73].

In liver cancer, AQP1, AQP3, and AQP5 have been associated with tumor progression and poor prognosis, highlighting their potential as therapeutic targets and biomarkers, while AQP9 has been implicated in modulation of the tumor microenvironment [106,142,145]. Among these, AQP3 and AQP5 have been particularly investigated in HCC.

A study of 130 HCC tissues and adjacent nonneoplastic tissues found that AQP3 and AQP5 were overexpressed in HCC tissue, with AQP3 localized mainly to the cytoplasm and cell membrane [143]. Coexpression of AQP3 and AQP5 was associated with tumor stage, grade, and metastasis, suggesting their potential as diagnostic and prognostic markers in HCC [143].

AQP3 has also been implicated in the malignant progression of HCC; its overexpression promotes HCC cell proliferation and migration, whereas AQP3 silencing suppresses these phenotypes [144]. This regulation appears to involve miR-124, which reduces AQP3 expression and thereby inhibits HCC cell proliferation, while circular RNA HIPK3 (circHIPK3) acts as a miR-124 sponge to enhance AQP3 expression. Consistent with these in vitro findings, circHIPK3 knockdown suppressed xenograft tumor growth in vivo, suggesting that the circHIPK3–miR-124–AQP3 axis contributes to HCC progression and may represent a potential therapeutic target [144]. Further, AQP3 expression correlated with the liver cancer stem cell (LCSC) marker CD133 [106]. In HCCLM3 and HUH7 cells, AQP3 increased the CD133+ cell proportion and CD133 expression while promoting stem-cell-like properties, effects that were abolished by pharmacological STAT3 inhibition, indicating that AQP3 regulates cancer stemness through the STAT3/CD133 axis [106]. AQP5 has also been implicated in HCC progression and metastasis, promoting cell migration, invasion, and metastasis by activating NF-κB signaling, which in turn drives epithelial–mesenchymal transition [145].

In contrast, AQP7, AQP8 and AQP9 are usually downregulated in HCC [142,222]. An in vivo HCC study showed opposing roles of AQP3 and AQP9 in tumor development [142]. AQP3 was overexpressed in HCC tissues, particularly in poorly differentiated tumors, whereas AQP9 expression was reduced compared with nonneoplastic liver tissue. Pharmacological reduction in AQP3 with Auphen suppressed xenograft tumor growth and increased tumor cell apoptosis. Conversely, increasing AQP9 expression using dibutyryl cAMP also inhibited tumor growth [142].

Reduced AQP8 and AQP9 expression has also been reported in HCC compared with normal liver tissue, using H4IIE rat HCC cells in vitro and also in vivo [222]. Under hypotonic challenge, normal hepatocytes rapidly swelled in an AQP-dependent manner (abolished by the inhibitor $HgCl_2$), whereas H4IIE cells failed to swell significantly. The study proposed that this reduced AQP-dependent water movement may impair the apoptotic volume decrease, an early apoptotic event, contributing to HCC resistance to apoptosis, consistent with the lower caspase-3 activity observed in HCC cells. Similar AQP8 and AQP9 downregulation was observed in a progressive neonatal DEN mouse model of HCC [222].

Taken together, these findings indicate that dysregulated AQP expression is closely associated with HCC progression and tumor cell survival. AQP3 and AQP5 are generally upregulated and promote proliferation, migration, invasion, metastasis, and cancer stemness, whereas AQP7–9 tend to be downregulated and may have distinct roles in tumor development [106,142–144,222]. Importantly, reduced AQP8 and AQP9 expression in HCC has also been linked to impaired apoptotic responsiveness, suggesting that AQP-dependent mechanisms may contribute to tumor cell survival [222]. However, despite these findings, direct evidence linking AQP dysregulation to resistance to anticancer therapy in HCC remains limited, highlighting the need for further investigation.

6. Clinical Potential in Therapeutic Targeting

The clinical translation of molecular oncology reflects a shift from mapping the basic biology of cancer cells toward designing integrated therapeutic interventions. Advanced malignancies have traditionally been treated by blocking isolated pathways, but this approach is increasingly giving way to strategies that target cooperative defense networks within the tumor microenvironment. As described, NRF2 and the aquaglyceroporin AQP3

can form a coordinated cytoprotective axis: NRF2 clears oxidative threats transcriptionally, while AQP3-mediated H_2O_2 transport feeds downstream pro-survival and anabolic signaling [28,32,150,223]. Jointly targeting both systems is therefore a molecularly rational strategy worth exploring for aggressive, multidrug-resistant tumors.

As discussed, constitutive NRF2 activation is a central clinical obstacle, allowing cancer cells to rapidly clear the ROS generated by radiotherapy and chemotherapy and cementing multidrug resistance [36,151]. NRF2-targeted drug development splits into two strategies—chemoprevention and direct therapeutic inhibition [224]. While NRF2 activators aim to shield healthy tissues from oxidative damage and prevent early carcinogenesis, the goal of chemical inhibitors targeting the NRF2-ARE signaling pathway is exactly the opposite: to tear down this antioxidant shield inside the existing tumor mass [224,225]. By deploying small-molecule NRF2 inhibitors, clinicians can stop this transcription factor from binding to its genomic target sequences. This intervention rapidly depletes intracellular glutathione pools, pushing highly stressed malignant cells past their survival threshold and into apoptosis [225,226]. The most valuable clinical benefit of this approach is its ability to resensitize refractory tumor lines to standard chemotherapeutic drugs, helping to bypass acquired resistance mechanisms [225,226].

In parallel with managing the cell's redox balance, targeting specific aquaporin channels offers an independent clinical advantage because it addresses the physical and metabolic drivers of tumor progression. Beyond its peroxiporin activity, AQP3 promotes actin-cytoskeleton reorganization and EMT-associated migration and invasion [163], while AQP3 and AQP5 together shape cell stiffness, membrane biomechanics, and cell–cell adhesion [227]. This cytoskeletal crosstalk, layered onto AQPs' canonical role in cell volume regulation, gives malignant cells the structural flexibility and mobility needed to invade surrounding tissue, properties that can extend to metastatic dissemination, though this link remains to be directly demonstrated in vivo. AQP3-mediated H_2O_2 influx further reinforces this phenotype by acting as a second messenger for downstream proliferative signaling [150]. Clinically, this dual physical and metabolic role is already evident: in ovarian cancer models, aquaporin expression directly affects how well platinum-based drugs like cisplatin can halt cell proliferation [228]. Pharmacological blockade of these channels accordingly delivers immediate anti-invasive benefit, stripping tumor cells of their migration signals and their ability to remodel shape during neovascularization and tissue matrix penetration [150].

Although NRF2 and AQP3 may not be directly coupled at the molecular level, we hypothesize that targeting either system in isolation could trigger a compensatory response from the other: for instance, pharmacological AQP inhibition might reduce intracellular H_2O_2 influx and relieve oxidative pressure on KEAP1, potentially blunting NRF2 activation, while conversely, residual or compensatory NRF2 activity could offset the loss of AQP-mediated redox signaling by other means [150]. This is the rationale for a dual-axis strategy. Combining NRF2 inhibition with pharmacological AQP3 blockade could disrupt AQP3-driven cytoskeletal remodeling and biomechanical adaptation, as shown in vitro [163,227], while simultaneously removing the cell's capacity to clear oxidative damage [225,226], increasing overall cell stress and potentially reducing resistance. This remains a proposed combination strategy rather than one with established clinical validation.

Translating NRF2 and AQP inhibitors into clinical use faces substantial pharmacological hurdles, echoing the historical struggle to develop selective, non-toxic metal-based anticancer drugs [229]. Early AQP inhibitors, mercury, silver, and gold compounds, showed potency but poor selectivity and significant systemic toxicity [230,231]. However, their lack of selectivity and severe systemic toxicity mirrored the early clinical setbacks seen with ruthenium and platinum metal complexes [229–231]. NRF2 pathway inhibition faces

a parallel challenge: because basal NRF2 activity is essential for normal tissue function, non-selective inhibitors risk damaging healthy organs [224]. Clinical development has accordingly shifted toward non-electrophilic small molecules and natural products aimed at reducing NRF2 or AQP expression with a more tolerable toxicity profile [224,225,230]. Successfully introducing these targeted agents into clinical practice depends entirely on a clear screening framework and biomarker-driven patient stratification. Identifying high-risk patients begins by looking at initial tumor biopsies using immunohistochemistry or transcriptomic profiling.

Biomarker-guided patient stratification is a logical next step, but there is still not enough evidence. KEAP1/NFE2L2 mutations, including when detected in blood-based sequencing, are associated with worse outcomes in NSCLC [232] and with radioresistance [233]. AQP1, AQP3, and AQP5 expression has been linked to nodal and distant metastasis in colorectal cancer, but associations with survival are inconsistent across studies and tumor types [116,234]. Whether tumors combining NRF2 pathway activation with high AQP3/AQP5 expression form a clinically distinct, poorer-prognosis subgroup has not been tested and would require validation in patient cohorts.

Synchronized targeting of the NRF2 antioxidant pathway and peroxiporins therefore represents a promising direction in precision oncology, although realizing it will require overcoming selectivity and toxicity challenges and validating biomarkers for patient stratification (Summarized in Figure 3).

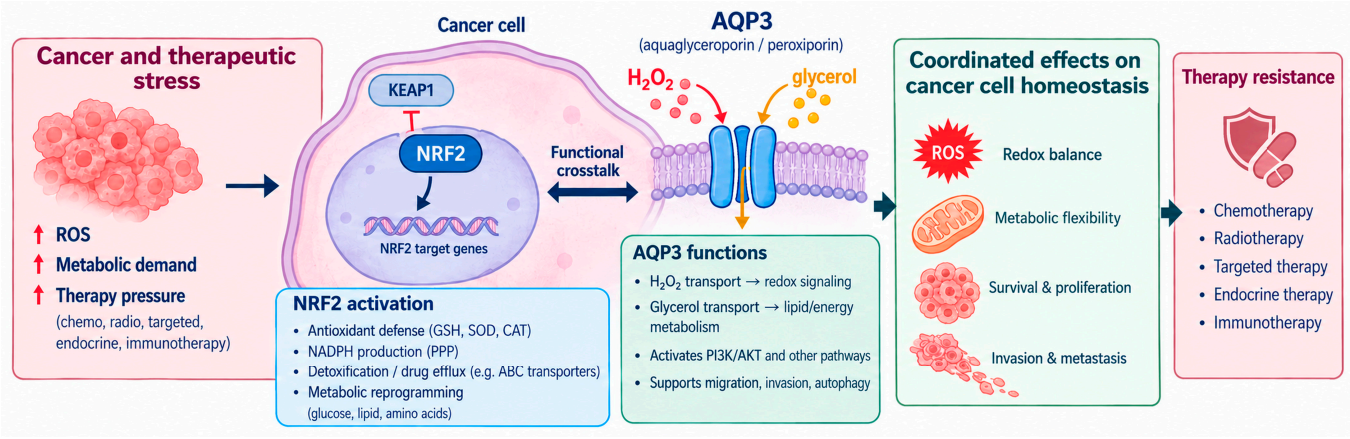


Figure 3. Coordinated NRF2 and AQP3 activities in cancer cell redox and metabolic adaptation.

Conceptual overview of the proposed interplay between NRF2 signaling and AQP3-mediated transport in cancer cells exposed to oxidative, metabolic, and therapeutic stress. Increased ROS, metabolic demand, and therapy pressure can promote NRF2 activation through disruption of KEAP1-mediated regulation. NRF2 activation induces antioxidant defense, NADPH production, detoxification and drug-efflux mechanisms, and metabolic reprogramming. In parallel, AQP3, acting as an aquaglyceroporin and peroxiporin, facilitates the transport of H_2O_2 and glycerol, thereby contributing to redox signaling and lipid/energy metabolism and influencing signaling pathways and cellular processes associated with cancer cell adaptation. NRF2 and AQP3 activities may therefore converge on redox homeostasis, metabolic flexibility, survival and proliferation, and invasion and metastasis, contributing to therapy resistance. The figure represents a conceptual synthesis of the evidence discussed in this review; a direct molecular interaction between NRF2 and AQP3 has not been established, and the proposed functional relationship requires further experimental validation. The figure was developed with graphical assistance from OpenAI's ChatGPT and subsequently reviewed, modified, and scientifically validated by the authors.

7. Immunohistochemistry-Based NRF2/AQP Co-Expression

7.1. Immunohistochemistry as the Basis of Clinical Potential

A biomarker may be biologically relevant but offers little clinical value if its expression cannot be measured reproducibly using routine specimens. Immunohistochemistry (IHC) preserves tissue architecture and allows the staining signal to be assigned to defined cell populations and intracellular compartments in formalin-fixed, paraffin-embedded (FFPE) tissue [235,236]. Localization affects the interpretation of both NRF2 and aquaporins (AQPs). In the studies reviewed here, NRF2 was assessed separately in the nuclear and cytoplasmic compartments, whereas AQP evaluation also included membranous staining [163,237,238]. Tissue microarrays (TMAs) are practical for studying large, clinically annotated tumor cohorts under comparable staining conditions, while requiring relatively little archival tissue and reagents [239]. The main drawback is limited sampling: a small number of cores may miss variation in expression across the tumor, so findings that depend on spatial distribution may require confirmation in additional cores or whole sections [240].

Associations with clinicopathological characteristics or survival are difficult to interpret unless the performance of the underlying assay is established. Current guidance recommends analytical validation or verification before introducing an IHC assay into clinical practice [241]. The REMARK recommendations also require sufficient methodological detail to evaluate staining procedures, scoring, quality control, and reproducibility in prognostic biomarker studies [242]. NRF2 provides a clear example of why this matters: commercially available antibodies have differed in their ability to distinguish specific from nonspecific staining [237].

Digital image analysis may improve scoring consistency and preserve continuous expression measurements, though its reliability still depends on assay quality and pathologist oversight [235,243]. Multiplex antibody-based imaging makes it possible to assess several proteins in the same section while retaining spatial information, but both individual antibodies and assembled panels must be appropriately qualified [230]. Applied to NRF2 and AQPs, this approach could allow candidate expression profiles to be examined within the same tissue context [235,236].

7.2. Immunohistochemical Quantification of NRF2 and Aquaporins

Reliable IHC assessment of NRF2 requires a clear definition of the analyte and cellular compartment being evaluated. Reports should specify whether the assay detects NRF2 or phosphorylated NRF2, and whether scoring is based on nuclear or cytoplasmic staining, since these are distinct readouts that should be reported separately [237,238,244]. In a cohort of 203 TNBC patients, nuclear NRF2 and cytoplasmic KEAP1 were evaluated using H-scores, weighting the percentage of tumor cells at each staining intensity to yield a continuous score from 0 to 300 [244]. Because no universally accepted H-score cutoffs are available, the cohort medians, 200 for NRF2 and 100 for KEAP1, were used to define low and high expression [244].

The choice of antibody can alter NRF2 classification. Torrente et al. tested two commercially available antibodies in FFPE cell pellets from isogenic NRF2 gain-of-function and NRF2-knockout DLD1 colorectal cancer cells [237]. One antibody failed to distinguish the two controls; the other produced strong nuclear staining in gain-of-function cells and no staining in knockout cells, and was selected for the clinically annotated CRC TMA analysis [237]. A separate HCC study used different scoring approaches for two NRF2 readouts: cytoplasmic NRF2 was graded 0–3+, while phosphorylated NRF2 (pNRF2) was recorded as the percentage of positive tumor cell nuclei, with ≥10% classified as high [238]. Antibody, analyte, cellular compartment, and scoring method should therefore be specified whenever NRF2 IHC findings are reported [237,238,244].

Published AQP studies are similarly difficult to compare, as they used different scoring systems. In 56 ER-positive invasive ductal BC patients, membranous/cytoplasmic AQP3 staining was scored 0–12 using an immunoreactivity score derived from intensity and proportion of positive cells [163]. A separate 591-specimen TMA graded AQP5 intensity 0–3 combined with a binary proportion threshold, with a product score ≥ 1 considered positive [245]. Because these scoring rules differ, the resulting expression categories are not directly interchangeable [163,245]. In CRC, QuPath was used to calculate Allred scores (0–8) alongside H-scores for AQP1, AQP3, and AQP5, with thresholds derived from time-dependent ROC curves using 5-year progression-free survival as the outcome [234].

Because these cutoffs were derived from outcome data within the same cohort, the derivation method should be reported transparently and the thresholds validated in an independent cohort. Continuous measurements should also be retained alongside categorical groups to limit information loss from dichotomization [242].

7.3. Co-Expression Panels for Breast, Colorectal and Hepatocellular Cancer

The tissue studies reviewed here follow two broad approaches: some evaluated several proteins in parallel but analyzed each marker separately, while others combined marker-specific expression categories into a single immunophenotype variable [100,143,244]. The CRC studies discussed below did not use a combined classifier [116,234,237].

Combined classifications were examined in two retrospective TNBC cohorts. In 203 surgically treated patients, NRF2 and KEAP1 expression was divided into four immunophenotypic groups; the NRF2-high/KEAP1-low phenotype was associated with poorer OS and DFS, while NRF2-low/KEAP1-high tumors had more favorable outcomes, and the four-group classification remained independently associated with both endpoints [244]. A separate 106-patient cohort assessed NRF2 alone (not combined with AQPs) showed that nuclear NRF2 was positive in 44% of cases, with significantly higher mean NRF2-positive nuclei in carcinoma (43.21%) versus fibroadenoma (7.12%), and grade 3 tumors showing higher expression than grade 1 [246]. A second TNBC cohort of 96 patients found high AQP3 and AQP5 each associated with tumor size, nodal status, and local recurrence or distant metastasis; patients with high expression of both aquaporins had poorer 5-year DFS and OS, and the combined AQP3/AQP5 pattern remained prognostically significant in multivariate analysis [100].

In CRC, markers were assessed separately rather than combined into a predefined score. Among 486 surgically resected colon cancers, AQP1, AQP3, and AQP5 were independently associated with lymph node metastasis in multivariate logistic analysis, but none independently predicted OS or DFS [116]. In a later, digitally quantified TMA series of 112 CRC cases, only AQP1 retained an independent association with 5-year OS [234]. High nuclear NRF2 expression was associated with shorter cancer-specific survival in a separate CRC cohort [234]. An additional 83-patient CRC tissue-array study assessed NRF2-pathway molecules alone, without AQPs, identifying KEAP1 and Bach1 as the strongest predictors of NRF2-related oxidative activity in tumor and normal tissue, respectively [247].

In HCC, pNRF2, cytoplasmic NRF2, and KEAP1 were measured in the same tissue cohort but analyzed as separate biomarkers rather than a predefined combined classifier [238]. A retrospective 130-patient cohort evaluated AQP3 and AQP5 expression: combined high expression was associated with serum alpha-fetoprotein, tumor stage, and histological grade, and patients with high expression of both proteins had poorer 5-year DFS and OS, with the combined pattern remaining independently associated with both endpoints [143].

One possible future step would be the development of a combined NRF2-AQP marker panel assessed by IHC. To date, no such panel has been established or clinically validated;

its development would require clear scoring rules and testing in an independent patient cohort before its prognostic value could be determined.

8. Conclusions

Cancer therapy resistance encompasses a wide range of cellular processes. Some are recognized as primary drivers of resistance, while others remain less visible, yet still shape the antioxidant and metabolic adaptations that allow tumor cells to persist under treatment. NRF2 and aquaporins are two such proteins, each regulating antioxidant status and cell metabolism in its own distinct manner, thereby shaping the response to therapy. In breast, colorectal, and liver cancer, the evidence we reviewed shows that NRF2 supports glutathione-based antioxidant defense, NADPH production, and anabolic metabolism, whereas aquaglyceroporins/peroxiporins control the transport of H_2O_2 and glycerol, and thereby influence redox-sensitive signaling and metabolic adaptation. AQP expression is dysregulated in an isoform- and tumor-type-specific manner, with both upregulation and downregulation reported, and its prognostic associations differ between tumor types. Importantly, no direct molecular interaction or causal NRF2-AQP axis has been demonstrated in treatment-resistant tumors. The available experimental evidence supports AQP-mediated H_2O_2 transport, which could plausibly contribute to KEAP1 oxidation and NRF2 activation.

To date, however, no study has evaluated NRF2 and AQP expression as a combined biomarker panel within the same tumor specimens, and the underlying IHC literature remains heterogeneous: antibody specificity, scoring systems, and staining compartments vary substantially between studies, limiting direct comparison and complicating any near-term move toward a standardized combined assay. This reflects a broader pattern in cancer biology: the more we learn, the more complex the underlying interactions prove to be. Based on this evidence, we see three priorities for future work. First, the relationship between NRF2 and AQPs should be tested directly. Second, NRF2 and the relevant AQP isoforms should be quantified together in the same specimens, using validated antibodies, standardized and digitally supported scoring, and independent cohorts. Third, if a functional link is confirmed, dual NRF2/AQP targeting should be tested in therapy-resistant models. Only then can the combined NRF2/AQP assessment be considered as a potential biomarker panel or therapeutic strategy for addressing cancer therapy resistance.

Author Contributions: Conceptualization, M.M. (Monika Mlinarić) and A.Č.G.; writing—original draft preparation, M.M. (Monika Mlinarić), A.B., A.J.J., J.K., K.K., M.M. (Martina Matec), A.Z.S., H.S., T.S. and A.Č.G.; writing—review and editing, M.M. (Monika Mlinarić), A.J.J., T.S. and A.Č.G.; visualization, M.M. (Monika Mlinarić); supervision, H.S., T.S. and A.Č.G.; funding acquisition, T.S. and A.Č.G. All authors have read and agreed to the published version of the manuscript.

Funding: This research was funded by HRZZ, grant number IP-2025-02-8980, and by the Croatian Society for Medical Oncology/Croatian Medical Association.

Institutional Review Board Statement: Not applicable.

Informed Consent Statement: Not applicable.

Data Availability Statement: No new data were created or analyzed in this study. Data sharing is not applicable to this article.

Acknowledgments: During the preparation of this manuscript, the authors used OpenAI's ChatGPT-5.6 Luna (image-generation functionality) to assist with the development and refinement of the conceptual graphical illustration presented in Figures 1–3. The tool was used to generate an initial visual representation based on the scientific concepts and information presented in this review and to assist with the visual organization and graphical refinement of the figure. The authors reviewed and edited the output for scientific accuracy, terminology, graphical clarity, and consistency with the manuscript and take full responsibility for the content of this publication.

Conflicts of Interest: The authors declare no conflicts of interest.

Abbreviations

The following abbreviations are used in this manuscript:

4E-BP1	eukaryotic translation initiation factor 4E-binding protein 1
5-FU	5-fluorouracil
8-OHdG	8-hydroxydeoxyguanosine
ABC	ATP-binding cassette
ABCB1	ATP-binding cassette subfamily B member 1 (P-glycoprotein)
ABCC1/ABCC2	ATP-binding cassette subfamily C member 1/2
ABCG2	ATP-binding cassette subfamily G member 2 (breast cancer resistance protein)
Akt	protein kinase B
AMPK	AMP-activated protein kinase
APE1/Ref-1	apurinic/apyrimidinic endonuclease 1/redox effector factor-1
AQP	aquaporin
ar/R	aromatic/arginine (selectivity filter)
ARE	antioxidant response element
ASK1	apoptosis signal-regulating kinase 1
AVD	apoptotic volume decrease
BACH1	BTB and CNC homology 1
BC	breast cancer
BCRP	breast cancer resistance protein
Bcl-xL	B-cell lymphoma-extra large
BRAF	B-Raf proto-oncogene, serine/threonine kinase
BRCA1	breast cancer gene 1
BRG1	Brahma-related gene 1
β-TrCP	β-transducin repeat-containing protein
cAMP	cyclic adenosine monophosphate
CCA	cholangiocarcinoma
CCNE1	cyclin E1
CD133	prominin-1
CDK2/CDK4/6	cyclin-dependent kinase 2/4/6
CIMP	CpG island methylator phenotype
CIN	chromosomal instability
circHIPK3	circular RNA HIPK3
CMS	Consensus Molecular Subtypes
CRC	colorectal cancer
CUL1/CUL3	Cullin 1/Cullin 3
DEN	diethylnitrosamine
DFS	disease-free survival
DNMT1	DNA methyltransferase 1
EGF	epidermal growth factor
EMT	epithelial–mesenchymal transition
ER	estrogen receptor
ERE(s)	estrogen response element(s)
ERK1/2	extracellular signal-regulated kinases 1/2
ESR1	estrogen receptor 1 (gene)
FFPE	formalin-fixed, paraffin-embedded
FOXO	forkhead box O
FTH1	ferritin heavy chain 1
G6PD	glucose-6-phosphate dehydrogenase
GCLC/GCLM	glutamate–cysteine ligase catalytic/modifier subunit
GSH	glutathione
GSK-3β (GSK3)	glycogen synthase kinase-3β

GSR	glutathione reductase
GST	glutathione S-transferase
H_2O_2	hydrogen peroxide
H_2S	hydrogen sulfide
HCC	hepatocellular carcinoma
HDAC1	histone deacetylase 1
HER2/HER3	human epidermal growth factor receptor 2/3
$HgCl_2$	mercury chloride
HIF-1α	hypoxia-inducible factor 1-alpha
HMOX1/HO-1	heme oxygenase-1
HR+	hormone receptor-positive
IHC	immunohistochemistry
JNK	c-Jun N-terminal kinase
KEAP1	Kelch-like ECH-associated protein 1
KRAS	Kirsten rat sarcoma viral oncogene homolog
LAG-3	lymphocyte-activation gene 3
LCSC	liver cancer stem cell
MAPK	mitogen-activated protein kinase
mBC	metastatic breast cancer
MDR1	multidrug resistance protein 1
MET	MET proto-oncogene (receptor tyrosine kinase)
MHC	major histocompatibility complex
miR-124/miR-200a	microRNA-124/microRNA-200a
MMR	mismatch repair
MRP(s)	multidrug resistance-associated protein(s)
MSI	microsatellite instability
mTOR	mechanistic target of rapamycin
MYC/Myc	MYC proto-oncogene
NADPH	nicotinamide adenine dinucleotide phosphate (reduced)
NF-κB	nuclear factor kappa-light-chain-enhancer of activated B cells
NFE2L2	nuclear factor erythroid 2-like 2 (gene encoding NRF2)
NPA	asparagine–proline–alanine (motif)
NQO1	NAD(P)H:quinone oxidoreductase 1
NRF2	nuclear factor erythroid 2-related factor 2
NSCLC	non-small cell lung cancer
Nox/Nox2/Nox4	NADPH oxidase/isoform 2/isoform 4
OS	overall survival
p62	sequestosome 1
p95HER2	truncated HER2 receptor
PALB2	partner and localizer of BRCA2
PARP	poly(ADP-ribose) polymerase
PD-1/PD-L1	programmed cell death protein 1/programmed death-ligand 1
PGD	6-phosphogluconate dehydrogenase
PI3K	phosphoinositide 3-kinase
PIK3CA	phosphatidylinositol-4,5-bisphosphate 3-kinase catalytic subunit alpha
PKC	protein kinase C
PLC-γ	phospholipase C-gamma
pNRF2	phosphorylated NRF2
PPP	pentose phosphate pathway
P-gp	P-glycoprotein
PTEN	phosphatase and tensin homolog
Rbx1	RING-box protein 1
RB1	retinoblastoma 1
REMARK	Reporting Recommendations for Tumor Marker Prognostic Studies
ROC	receiver operating characteristic

ROS	reactive oxygen species
RUNX3	Runt-related transcription factor 3
SCF	Skp1–Cullin1–F-box protein complex
SDH	succinate dehydrogenase
siNRF2	small interfering RNA targeting NRF2
SMARCA4	SWI/SNF-related, matrix-associated, actin-dependent regulator of chromatin, subfamily A, member 4
Smad2/3	mothers against decapentaplegic homolog 2/3
SQSTM1	sequestosome 1
STAT3	signal transducer and activator of transcription 3
Syk	spleen tyrosine kinase
TACE	transarterial chemoembolization
TALDO1	transaldolase 1
tBHQ	tert-butylhydroquinone
TGF-β	transforming growth factor beta
TIM-3	T-cell immunoglobulin and mucin-domain containing-3
TKT	transketolase
TMA(s)	tissue microarray(s)
TNBC	triple-negative breast cancer
WNT	wingless-related integration site (signaling pathway)

References

1. Van Loenhout, J.; Peeters, M.; Bogaerts, A.; Smits, E.; Deben, C. Oxidative Stress-Inducing Anticancer Therapies: Taking a Closer Look at Their Immunomodulating Effects. *Antioxidants* **2020**, *9*, 1188. [CrossRef] [PubMed]
2. Barrera, G.; Cucci, M.A.; Grattarola, M.; Dianzani, C.; Muzio, G.; Pizzimenti, S. Control of Oxidative Stress in Cancer Chemoresistance: Spotlight on Nrf2 Role. *Antioxidants* **2021**, *10*, 510. [CrossRef] [PubMed]
3. DeBlasi, J.M.; DeNicola, G.M. Dissecting the Crosstalk between NRF2 Signaling and Metabolic Processes in Cancer. *Cancers* **2020**, *12*, 3023. [CrossRef] [PubMed]
4. Pavlova, N.N.; Thompson, C.B. The Emerging Hallmarks of Cancer Metabolism. *Cell Metab.* **2016**, *23*, 27–47. [CrossRef] [PubMed]
5. Galadari, S.; Rahman, A.; Pallichankandy, S.; Thayyullathil, F. Reactive Oxygen Species and Cancer Paradox: To Promote or to Suppress? *Free Radic. Biol. Med.* **2017**, *104*, 144–164. [CrossRef] [PubMed]
6. Srinivas, U.S.; Tan, B.W.Q.; Vellayappan, B.A.; Jeyasekharan, A.D. ROS and the DNA Damage Response in Cancer. *Redox Biol.* **2019**, *25*, 101084. [CrossRef] [PubMed]
7. Mendoza, E.N.; Ciriolo, M.R.; Ciccarone, F. Hypoxia-Induced Reactive Oxygen Species: Their Role in Cancer Resistance and Emerging Therapies to Overcome It. *Antioxidants* **2025**, *14*, 94. [CrossRef] [PubMed]
8. Glorieux, C.; Enríquez, C.; González, C.; Aguirre-Martínez, G.; Buc Calderon, P. The Multifaceted Roles of NRF2 in Cancer: Friend or Foe? *Antioxidants* **2024**, *13*, 70. [CrossRef] [PubMed]
9. Panda, H.; Wen, H.; Suzuki, M.; Yamamoto, M. Multifaceted Roles of the KEAP1–NRF2 System in Cancer and Inflammatory Disease Milieu. *Antioxidants* **2022**, *11*, 538. [CrossRef] [PubMed]
10. Bhardwaj, V.; He, J. Reactive Oxygen Species, Metabolic Plasticity, and Drug Resistance in Cancer. *Int. J. Mol. Sci.* **2020**, *21*, 3412. [CrossRef] [PubMed]
11. Li, Y.; Zhang, X.; Wang, Z.; Li, B.; Zhu, H. Modulation of Redox Homeostasis: A Strategy to Overcome Cancer Drug Resistance. *Front. Pharmacol.* **2023**, *14*, 1156538. [CrossRef] [PubMed]
12. He, F.; Ru, X.; Wen, T. NRF2, a Transcription Factor for Stress Response and Beyond. *Int. J. Mol. Sci.* **2020**, *21*, 4777. [CrossRef] [PubMed]
13. Gañán-Gómez, I.; Wei, Y.; Yang, H.; Boyano-Adánez, M.C.; García-Manero, G. Oncogenic Functions of the Transcription Factor Nrf2. *Free Radic. Biol. Med.* **2013**, *65*, 750–764. [CrossRef] [PubMed]
14. Gu, X.; Mu, C.; Zheng, R.; Zhang, Z.; Zhang, Q.; Liang, T. The Cancer Antioxidant Regulation System in Therapeutic Resistance. *Antioxidants* **2024**, *13*, 778. [CrossRef] [PubMed]
15. Cheng, X.; Xu, M.; Wei, J.; Cao, C. Cancer Drug Response and Resistance: Molecular Mechanisms and Combating Strategies. *Signal Transduct. Target. Ther.* **2026**, *11*, 307. [CrossRef] [PubMed]
16. Xiao, H.; Zheng, Y.; Ma, L.; Sun, Q. Clinically-Relevant ABC Transporter for Anti-Cancer Drug Resistance. *Front. Pharmacol.* **2021**, *12*, 648407. [CrossRef] [PubMed]

17. Abalenikhina, Y.V.; Shchulkin, A.V.; Mylnikov, P.Y.; Rokunov, E.D.; Yakusheva, E.N. Mechanisms of P-Glycoprotein Regulation Under Exogenous and Endogenous Oxidative Stress In Vitro. *Acta Naturae* **2022**, *14*, 69–78. [CrossRef] [PubMed]
18. Shchulkin, A.V.; Abalenikhina, Y.V.; Erokhina, P.D.; Chernykh, I.V.; Yakusheva, E.N. The Role of P-Glycoprotein in Decreasing Cell Membranes Permeability during Oxidative Stress. *Biochemistry* **2021**, *86*, 197–206. [CrossRef] [PubMed]
19. Beretta, G.L.; Corno, C.; Zaffaroni, N.; Perego, P. Role of FoxO Proteins in Cellular Response to Antitumor Agents. *Cancers* **2019**, *11*, 90. [CrossRef] [PubMed]
20. Lim, S.-O.; Gu, J.-M.; Cho, J.W.; Park, Y.N.; Park, C.K.; Cho, J.W.; Park, Y.M.; Jung, G. Epigenetic Changes Induced by Reactive Oxygen Species in Hepatocellular Carcinoma: Methylation of the E-Cadherin Promoter. *Gastroenterology* **2008**, *135*, 2128–2140. [CrossRef] [PubMed]
21. Damiano, O.M.; Stevens, A.J.; Kenwright, D.N.; Seddon, A.R. Chronic Inflammation to Cancer: The Impact of Oxidative Stress on DNA Methylation. *Front. Biosci. Landmark Ed.* **2025**, *30*, 26142. [CrossRef] [PubMed]
22. Aggarwal, V.; Tuli, H.S.; Varol, A.; Thakral, F.; Yerer, M.B.; Sak, K.; Varol, M.; Jain, A.; Khan, M.A.; Sethi, G. Role of Reactive Oxygen Species in Cancer Progression: Molecular Mechanisms and Recent Advancements. *Biomolecules* **2019**, *9*, 735. [CrossRef] [PubMed]
23. O'Neill, E.J.; Kornel, A.; Irwin, E.C.; Tsiani, E. ASK1 in Cancer Cell Death: Insights from In Vitro and In Vivo Studies. *Cells* **2026**, *15*, 1282. [CrossRef] [PubMed]
24. Perillo, B.; Di Donato, M.; Pezone, A.; Di Zazzo, E.; Giovannelli, P.; Galasso, G.; Castoria, G.; Migliaccio, A. ROS in Cancer Therapy: The Bright Side of the Moon. *Exp. Mol. Med.* **2020**, *52*, 192–203. [CrossRef] [PubMed]
25. Glaviano, A.; Foo, A.S.C.; Lam, H.Y.; Yap, K.C.H.; Jacot, W.; Jones, R.H.; Eng, H.; Nair, M.G.; Makvandi, P.; Geoerger, B.; et al. PI3K/AKT/mTOR Signaling Transduction Pathway and Targeted Therapies in Cancer. *Mol. Cancer* **2023**, *22*, 138. [CrossRef] [PubMed]
26. Trinh, V.H.; Nguyen Huu, T.; Sah, D.K.; Choi, J.M.; Yoon, H.J.; Park, S.C.; Jung, Y.S.; Lee, S.-R. Redox Regulation of PTEN by Reactive Oxygen Species: Its Role in Physiological Processes. *Antioxidants* **2024**, *13*, 199. [CrossRef] [PubMed]
27. Hayes, J.D.; Dinkova-Kostova, A.T. The Nrf2 Regulatory Network Provides an Interface between Redox and Intermediary Metabolism. *Trends Biochem. Sci.* **2014**, *39*, 199–218. [CrossRef] [PubMed]
28. Kang, J.-S.; Nam, L.B.; Yoo, O.-K.; Keum, Y.-S. Molecular Mechanisms and Systemic Targeting of NRF2 Dysregulation in Cancer. *Biochem. Pharmacol.* **2020**, *177*, 114002. [CrossRef] [PubMed]
29. Luchkova, A.; Mata, A.; Cadenas, S. Nrf2 as a Regulator of Energy Metabolism and Mitochondrial Function. *FEBS Lett.* **2024**, *598*, 2092–2105. [CrossRef] [PubMed]
30. Jung, B.-J.; Yoo, H.-S.; Shin, S.; Park, Y.-J.; Jeon, S.-M. Dysregulation of NRF2 in Cancer: From Molecular Mechanisms to Therapeutic Opportunities. *Biomol. Ther.* **2018**, *26*, 57–68. [CrossRef] [PubMed]
31. Panieri, E.; Telkoparan-Akillilar, P.; Suzen, S.; Saso, L. The NRF2/KEAP1 Axis in the Regulation of Tumor Metabolism: Mechanisms and Therapeutic Perspectives. *Biomolecules* **2020**, *10*, 791. [CrossRef] [PubMed]
32. Paramasivan, P.; Kankia, I.H.; Langdon, S.P.; Deeni, Y.Y. Emerging Role of Nuclear Factor Erythroid 2-Related Factor 2 in the Mechanism of Action and Resistance to Anticancer Therapies. *Cancer Drug Resist.* **2019**, *2*, 490–515. [CrossRef] [PubMed]
33. Lee, S.B.; Sellers, B.N.; DeNicola, G.M. The Regulation of NRF2 by Nutrient-Responsive Signaling and Its Role in Anabolic Cancer Metabolism. *Antioxid. Redox Signal.* **2018**, *29*, 1774–1791. [CrossRef] [PubMed]
34. Yamamoto, M.; Kensler, T.W.; Motohashi, H. The KEAP1-NRF2 System: A Thiol-Based Sensor-Effector Apparatus for Maintaining Redox Homeostasis. *Physiol. Rev.* **2018**, *98*, 1169–1203. [CrossRef] [PubMed]
35. Menegon, S.; Columbano, A.; Giordano, S. The Dual Roles of NRF2 in Cancer. *Trends Mol. Med.* **2016**, *22*, 578–593. [CrossRef] [PubMed]
36. Hallis, S.P.; Go, B.J.; Yoo, J.M.; Cho, G.H.; Kwak, M.-K. Toward a Better Understanding of NRF2/NFE2L2 and BCRP/ABCG2 in Therapy Resistance in Cancer. *Drug Targets Ther.* **2023**, *2*, 111–123. [CrossRef]
37. Verza, F.A.; Da Silva, G.C.; Nishimura, F.G. The Impact of Oxidative Stress and the NRF2-KEAP1-ARE Signaling Pathway on Anticancer Drug Resistance. *Oncol. Res.* **2025**, *33*, 1819–1834. [CrossRef] [PubMed]
38. Davies, K.J.A.; Forman, H.J. Does Bach1 & C-Myc Dependent Redox Dysregulation of Nrf2 & Adaptive Homeostasis Decrease Cancer Risk in Ageing? *Free Radic. Biol. Med.* **2019**, *134*, 708–714. [CrossRef] [PubMed]
39. Patibandla, C.; van Aalten, L.; Dinkova-Kostova, A.T.; Honda, T.; Cuadrado, A.; Fernández-Ginés, R.; McNeilly, A.D.; Hayes, J.D.; Cantley, J.; Sutherland, C. Inhibition of Glycogen Synthase Kinase-3 Enhances NRF2 Protein Stability, Nuclear Localisation and Target Gene Transcription in Pancreatic Beta Cells. *Redox Biol.* **2024**, *71*, 103117. [CrossRef] [PubMed]
40. Malhotra, D.; Portales-Casamar, E.; Singh, A.; Srivastava, S.; Arenillas, D.; Happel, C.; Shyr, C.; Wakabayashi, N.; Kensler, T.W.; Wasserman, W.W.; et al. Global Mapping of Binding Sites for Nrf2 Identifies Novel Targets in Cell Survival Response through ChIP-Seq Profiling and Network Analysis. *Nucleic Acids Res.* **2010**, *38*, 5718–5734. [CrossRef] [PubMed]

41. Morgenstern, C.; Lastres-Becker, I.; Demirdöğen, B.C.; Costa, V.M.; Daiber, A.; Foresti, R.; Motterlini, R.; Kalyoncu, S.; Arioz, B.I.; Genc, S.; et al. Biomarkers of NRF2 Signalling: Current Status and Future Challenges. *Redox Biol*. **2024**, *72*, 103134. [CrossRef] [PubMed]
42. Bai, X.; Chen, Y.; Hou, X.; Huang, M.; Jin, J. Emerging role of NRF2 in chemoresistance by regulating drug-metabolizing enzymes and efflux transporters. *Drug Metab. Rev.* **2016**, *48*, 541–567. [CrossRef] [PubMed]
43. Shibata, T.; Kokubu, A.; Saito, S. NRF2 Mutation Confers Malignant Potential and Resistance to Chemotherapy in Human Lung Cancer. *Cancer Res*. **2011**, *71*, 831–840. [CrossRef] [PubMed]
44. Hasan, S.K.; Jayakumar, S.; Espina Barroso, E.; Jha, A.; Catalano, G.; Sandur, S.K. Molecular Targets of Oxidative Stress: Focus on Nuclear Factor Erythroid 2–Related Factor 2 Function in Leukemia and Other Cancers. *Cells* **2025**, *14*, 713. [CrossRef] [PubMed]
45. Smolková, K.; Mikó, E.; Kovács, T.; Leguina-Ruzzi, A.; Sipos, A.; Bai, P. Nuclear Factor Erythroid 2-Related Factor 2 in Regulating Cancer Metabolism. *Antioxid. Redox Signal*. **2020**, *33*, 966. [CrossRef] [PubMed]
46. Kerins, M.J.; Ooi, A. A Catalogue of Somatic NRF2 Gain-of-Function Mutations in Cancer. *Sci. Rep*. **2018**, *8*, 12846. [CrossRef] [PubMed]
47. Song, S.; Nguyen, V.; Schrank, T.; Mulvaney, K.; Walter, V.; Wei, D.; Orvis, T.; Desai, N.; Zhang, J.; Hayes, D.N.; et al. Loss of SWI/SNF Chromatin Remodeling Alters NRF2 Signaling in Non-Small Cell Lung Carcinoma. *Mol. Cancer Res*. **2020**, *18*, 1777–1788. [CrossRef] [PubMed]
48. Oskomić, M.; Tomić, A.; Barbarić, L.; Matić, A.; Kindl, D.C.; Matovina, M. KEAP1-NRF2 Interaction in Cancer: Competitive Interactors and Their Role in Carcinogenesis. *Cancers* **2025**, *17*, 447. [CrossRef] [PubMed]
49. Gorrini, C.; Baniasadi, P.S.; Harris, I.S.; Silvester, J.; Inoue, S.; Snow, B.; Joshi, P.A.; Wakeham, A.; Molyneux, S.D.; Martin, B.; et al. BRCA1 Interacts with Nrf2 to Regulate Antioxidant Signaling and Cell Survival. *J. Exp. Med*. **2013**, *210*, 1529–1544. [CrossRef] [PubMed]
50. Hanada, N.; Takahata, T.; Zhou, Q.; Ye, X.; Sun, R.; Itoh, J.; Ishiguro, A.; Kijima, H.; Mimura, J.; Itoh, K.; et al. Methylation of the KEAP1 Gene Promoter Region in Human Colorectal Cancer. *BMC Cancer* **2012**, *12*, 66. [CrossRef] [PubMed]
51. Fabrizio, F.P.; Sparaneo, A.; Centra, F.; Trombetta, D.; Storlazzi, C.T.; Graziano, P.; Maiello, E.; Fazio, V.M.; Muscarella, L.A. Methylation Density Pattern of KEAP1 Gene in Lung Cancer Cell Lines Detected by Quantitative Methylation Specific PCR and Pyrosequencing. *Int. J. Mol. Sci*. **2019**, *20*, 2697. [CrossRef] [PubMed]
52. Barbano, R.; Muscarella, L.A.; Pasculli, B.; Valori, V.M.; Fontana, A.; Coco, M.; la Torre, A.; Balsamo, T.; Poeta, M.L.; Marangi, G.F.; et al. Aberrant Keap1 Methylation in Breast Cancer and Association with Clinicopathological Features. *Epigenetics* **2013**, *8*, 105–112. [CrossRef] [PubMed]
53. Zhang, P.; Singh, A.; Yegnasubramanian, S.; Esopi, D.; Kombairaju, P.; Bodas, M.; Wu, H.; Bova, G.S.; Biswal, S. Loss of Keap1 Function in Prostate Cancer Cells Causes Chemo- and Radio-Resistance and Promotes Tumor Growth. *Mol. Cancer Ther.* **2010**, *9*, 336. [CrossRef] [PubMed]
54. Muscarella, L.A.; Barbano, R.; D'Angelo, V.; Copetti, M.; Coco, M.; Balsamo, T.; la Torre, A.; Notarangelo, A.; Troiano, M.; Parisi, S.; et al. Regulation of KEAP1 Expression by Promoter Methylation in Malignant Gliomas and Association with Patient's Outcome. *Epigenetics* **2011**, *6*, 317–325. [CrossRef] [PubMed]
55. Fabrizio, F.P.; Costantini, M.; Copetti, M.; la Torre, A.; Sparaneo, A.; Fontana, A.; Poeta, L.; Gallucci, M.; Sentinelli, S.; Graziano, P.; et al. Keap1/Nrf2 Pathway in Kidney Cancer: Frequent Methylation of KEAP1 Gene Promoter in Clear Renal Cell Carcinoma. *Oncotarget* **2017**, *8*, 11187–11198. [CrossRef] [PubMed]
56. Panahizadeh, R.; Vatankhah, M.A.; Safari, A.; Danesh, H.; Nazmi, N.; Gholizadeh, P.; Soozangar, N.; Jeddi, F. The Interplay between microRNAs and Nrf2 Signaling in Human Cancers. *Cancer Cell Int*. **2024**, *24*, 234. [CrossRef] [PubMed]
57. Duan, W.-L.; Wang, X.-J.; Guo, A.; Gu, L.-H.; Sheng, Z.-M.; Luo, H.; Yang, L.-X.; Wang, W.-H.; Zhang, B.-G. miR-141-3p Promotes Paclitaxel Resistance by Attenuating Ferroptosis via the Keap1-Nrf2 Signaling Pathway in Breast Cancer. *J. Cancer* **2024**, *15*, 5622–5635. [CrossRef] [PubMed]
58. He, F.; Antonucci, L.; Karin, M. NRF2 as a Regulator of Cell Metabolism and Inflammation in Cancer. *Carcinogenesis* **2020**, *41*, 405–416. [CrossRef] [PubMed]
59. Rojo de la Vega, M.; Chapman, E.; Zhang, D.D. NRF2 and the Hallmarks of Cancer. *Cancer Cell* **2018**, *34*, 21–43. [CrossRef] [PubMed]
60. Joof, A.N.; Bajinka, O.; Zhou, Y.; Ren, F.; Tan, Y. The Central Role of NRF2 in Cancer Metabolism and Redox Signaling: Novel Insights into Crosstalk with ER Stress and Therapeutic Modulation. *Cell Biochem. Biophys*. **2025**, *83*, 4145–4161. [CrossRef] [PubMed]
61. Adam, J.; Hatipoglu, E.; O'Flaherty, L.; Ternette, N.; Sahgal, N.; Lockstone, H.; Baban, D.; Nye, E.; Stamp, G.W.; Wolhuter, K.; et al. Renal Cyst Formation in Fh1-Deficient Mice Is Independent of the Hif/Phd Pathway: Roles for Fumarate in KEAP1 Succination and Nrf2 Signaling. *Cancer Cell* **2011**, *20*, 524–537. [CrossRef] [PubMed]
62. Cheng, P.; Xia, R.; Wang, X. Ferroptosis: A Promising Target for Fumarate Hydratase-Deficient Tumor Therapeutics Literature Review. *Transl. Cancer Res*. **2024**, *13*, 3126–3141. [CrossRef] [PubMed]

63. Matzinger, M.; Fischhuber, K.; Pölöske, D.; Mechtler, K.; Heiss, E.H. AMPK Leads to Phosphorylation of the Transcription Factor Nrf2, Tuning Transactivation of Selected Target Genes. *Redox Biol*. **2019**, *29*, 101393. [CrossRef] [PubMed]
64. Tang, Z.; Shi, Y.; Jiang, X.; Jiang, S.; Maihemuti, N.; Zhang, J.; Tang, B.; Xu, Z. ZIP7 Drives Glycolytic Reprogramming and Lactate-Mediated Immune Remodeling in Lung Adenocarcinoma Through GSK3β-NRF2 Signaling. *Biomedicines* **2026**, *14*, 1262. [CrossRef] [PubMed]
65. Kitamura, H.; Motohashi, H. NRF2 Addiction in Cancer Cells. *Cancer Sci*. **2018**, *109*, 900–911. [CrossRef] [PubMed]
66. Okazaki, K.; Papagiannakopoulos, T.; Motohashi, H. Metabolic Features of Cancer Cells in NRF2 Addiction Status. *Biophys*. *Rev*. **2020**, *12*, 435–441. [CrossRef] [PubMed]
67. Wild, A.C.; Moinova, H.R.; Mulcahy, R.T. Regulation of Gamma-Glutamylcysteine Synthetase Subunit Gene Expression by the Transcription Factor Nrf2. *J*. *Biol*. *Chem*. **1999**, *274*, 33627–33636. [CrossRef] [PubMed]
68. Harvey, C.J.; Thimmulappa, R.K.; Singh, A.; Blake, D.J.; Ling, G.; Wakabayashi, N.; Fujii, J.; Myers, A.; Biswal, S. Nrf2-Regulated Glutathione Recycling Independent of Biosynthesis Is Critical for Cell Survival during Oxidative Stress. *Free Radic*. *Biol*. *Med*. **2009**, *46*, 443–453. [CrossRef] [PubMed]
69. Itoh, K.; Chiba, T.; Takahashi, S.; Ishii, T.; Igarashi, K.; Katoh, Y.; Oyake, T.; Hayashi, N.; Satoh, K.; Hatayama, I.; et al. An Nrf2/Small Maf Heterodimer Mediates the Induction of Phase II Detoxifying Enzyme Genes through Antioxidant Response Elements. *Biochem*. *Biophys*. *Res*. *Commun*. **1997**, *236*, 313–322. [CrossRef] [PubMed]
70. Purohit, V.; Simeone, D.M.; Lyssiotis, C.A. Metabolic Regulation of Redox Balance in Cancer. *Cancers* **2019**, *11*, 955. [CrossRef] [PubMed]
71. Mitsuishi, Y.; Taguchi, K.; Kawatani, Y.; Shibata, T.; Nukiwa, T.; Aburatani, H.; Yamamoto, M.; Motohashi, H. Nrf2 Redirects Glucose and Glutamine into Anabolic Pathways in Metabolic Reprogramming. *Cancer Cell* **2012**, *22*, 66–79. [CrossRef] [PubMed]
72. Agre, P.; King, L.S.; Yasui, M.; Guggino, W.B.; Ottersen, O.P.; Fujiyoshi, Y.; Engel, A.; Nielsen, S. Aquaporin Water Channels–from Atomic Structure to Clinical Medicine. *J*. *Physiol*. **2002**, *542*, 3–16. [CrossRef] [PubMed]
73. Pimpão, C.; Silva, I.V.; Soveral, G. The Expanding Role of Aquaporin-1, Aquaporin-3 and Aquaporin-5 as Transceptors: Involvement in Cancer Development and Potential Druggability. *Int*. *J*. *Mol*. *Sci*. **2025**, *26*, 1330. [CrossRef] [PubMed]
74. Agre, P. Aquaporin Water Channels. *Biosci*. *Rep*. **2004**, *24*, 127–163. [CrossRef] [PubMed]
75. Verkman, A.S.; Mitra, A.K. Structure and Function of Aquaporin Water Channels. *Am*. *J*. *Physiol*. *Ren*. *Physiol*. **2000**, *278*, 13–28. [CrossRef] [PubMed]
76. Jung, J.S.; Preston, G.M.; Smith, B.L.; Guggino, W.B.; Agre, P. Molecular Structure of the Water Channel through Aquaporin CHIP. The Hourglass Model. *J*. *Biol*. *Chem*. **1994**, *269*, 14648–14654. [CrossRef]
77. Milković, L.; Gašparović, A.Č. AQP3 and AQP5-Potential Regulators of Redox Status in Breast Cancer. *Molecules* **2021**, *26*, 2613. [CrossRef] [PubMed]
78. da Silva, I.V.; Mlinarić, M.; Lourenço, A.R.; Pérez-Garcia, O.; Čipak Gašparović, A.; Soveral, G. Peroxiporins and Oxidative Stress: Promising Targets to Tackle Inflammation and Cancer. *Int*. *J*. *Mol*. *Sci*. **2024**, *25*, 8381. [CrossRef] [PubMed]
79. Medraño-Fernandez, I.; Bestetti, S.; Bertolotti, M.; Bienert, G.P.; Bottino, C.; Laforenza, U.; Rubartelli, A.; Sitia, R. Stress Regulates Aquaporin-8 Permeability to Impact Cell Growth and Survival. *Antioxid*. *Redox Signal*. **2016**, *24*, 1031–1044. [CrossRef] [PubMed]
80. Bestetti, S.; Medraño-Fernandez, I.; Galli, M.; Ghitti, M.; Bienert, G.P.; Musco, G.; Orsi, A.; Rubartelli, A.; Sitia, R. A Persulfidation-Based Mechanism Controls Aquaporin-8 Conductance. *Sci*. *Adv*. **2018**, *4*, eaar5770. [CrossRef] [PubMed]
81. Wang, Q.; Lin, B.; Wei, H.; Wang, X.; Nie, X.; Shi, Y. AQP3 Promotes the Invasion and Metastasis in Cervical Cancer by Regulating NOX4-Derived H_2O_2 Activation of Syk/PI3K/Akt Signaling Axis. *J*. *Cancer* **2024**, *15*, 1124–1137. [CrossRef] [PubMed]
82. Huang, P.; Venskutonytė, R.; Wilson, C.J.; Bsharat, S.; Prasad, R.B.; Gourdon, P.; Artner, I.; de Groot, B.L.; Lindkvist-Petersson, K. Structural Insights into AQP3 Channel Closure upon pH and Redox Changes Reveal an Autoregulatory Molecular Mechanism. *Nat*. *Commun*. **2025**, *16*, 10997. [CrossRef] [PubMed]
83. Rodrigues, C.; Mósca, A.F.; Martins, A.P.; Nobre, T.; Prista, C.; Antunes, F.; Cipak Gasparovic, A.; Soveral, G. Rat Aquaporin-5 Is pH-Gated Induced by Phosphorylation and Is Implicated in Oxidative Stress. *Int*. *J*. *Mol*. *Sci*. **2016**, *17*, 2090. [CrossRef] [PubMed]
84. Markou, A.; Unger, L.; Abir-Awan, M.; Saadallah, A.; Halsey, A.; Balklava, Z.; Conner, M.; Törnroth-Horsefield, S.; Greenhill, S.D.; Conner, A.; et al. Molecular Mechanisms Governing Aquaporin Relocalisation. *Biochim*. *Biophys*. *Acta Biomembr*. **2022**, *1864*, 183853. [CrossRef] [PubMed]
85. Nesverova, V.; Törnroth-Horsefield, S. Phosphorylation-Dependent Regulation of Mammalian Aquaporins. *Cells* **2019**, *8*, 82. [CrossRef] [PubMed]
86. Nakamura, H.; Takada, K. Reactive Oxygen Species in Cancer: Current Findings and Future Directions. *Cancer Sci*. **2021**, *112*, 3945–3952. [CrossRef] [PubMed]
87. Maeda, N.; Funahashi, T.; Hibuse, T.; Nagasawa, A.; Kishida, K.; Kuriyama, H.; Nakamura, T.; Kihara, S.; Shimomura, I.; Matsuzawa, Y. Adaptation to Fasting by Glycerol Transport through Aquaporin 7 in Adipose Tissue. *Proc*. *Natl*. *Acad*. *Sci*. *USA* **2004**, *101*, 17801–17806. [CrossRef] [PubMed]

88. Maeda, N.; Hibuse, T.; Funahashi, T. Role of Aquaporin-7 and Aquaporin-9 in Glycerol Metabolism; Involvement in Obesity. *Handb. Exp. Pharmacol.* **2009**, *190*, 233–249. [CrossRef] [PubMed]
89. Bi, Y.; Pang, S.; Liu, Y.; Cheng, J.; Ma, Q.; Song, A.; Yin, X.; Wang, J. Aquaporins in Lipid Metabolism: Functions and Regulation in Health and Disease. *Lipids Health Dis.* **2025**, *24*, 336. [CrossRef] [PubMed]
90. Galli, M.; Hameed, A.; Żbikowski, A.; Zabielski, P. Aquaporins in Insulin Resistance and Diabetes: More than Channels! *Redox Biol.* **2021**, *44*, 102027. [CrossRef] [PubMed]
91. Chen, L.; Li, Z.; Zhang, Q.; Wei, S.; Li, B.; Lei, X.; Li, Z.; Li, Q.; Xu, H.; Xu, Z. Silencing of AQP3 Induces Apoptosis of Gastric Cancer Cells via Downregulation of Glycerol Intake and Downstream Inhibition of Lipogenesis and Autophagy. *Onco Targets Ther.* **2017**, *10*, 2791–2804. [CrossRef] [PubMed]
92. Li, Z.; Li, B.; Zhang, L.; Chen, L.; Sun, G.; Zhang, Q.; Wang, J.; Zhi, X.; Wang, L.; Xu, Z.; et al. The Proliferation Impairment Induced by AQP3 Deficiency Is the Result of Glycerol Uptake and Metabolism Inhibition in Gastric Cancer Cells. *Tumor Biol.* **2016**, *37*, 9169–9179. [CrossRef] [PubMed]
93. Dai, C.; Charlestin, V.; Wang, M.; Walker, Z.T.; Miranda-Vergara, M.C.; Facchine, B.A.; Wu, J.; Kaliney, W.J.; Dovichi, N.J.; Li, J.; et al. Aquaporin-7 Regulates the Response to Cellular Stress in Breast Cancer. *Cancer Res.* **2020**, *80*, 4071–4086. [CrossRef] [PubMed]
94. Wang, R.; Wang, X.; Zhao, J.; Jin, J.; Fan, W.; Zhu, X.; Chen, Q.; Zhang, B.; Lan, L.; Qu, K.; et al. Clinical Value and Molecular Mechanism of AQGPs in Different Tumors. *Med. Oncol.* **2022**, *39*, 174. [CrossRef] [PubMed]
95. Mlinarić, M.; Lučić, I.; Milković, L.; da Silva, I.V.; Tartaro Bujak, I.; Musani, V.; Soveral, G.; Čipak Gašparović, A. AQP3-Dependent PI3K/Akt Modulation in Breast Cancer Cells. *Int. J. Mol. Sci.* **2023**, *24*, 8133. [CrossRef] [PubMed]
96. Wang, Y.; Chen, D.; Liu, Y.; Zhang, Y.; Duan, C.; Otkur, W.; Chen, H.; Liu, X.; Xia, T.; Qi, H.; et al. AQP3-mediated H_2O_2 Uptake Inhibits LUAD Autophagy by Inactivating PTEN. *Cancer Sci.* **2021**, *112*, 3278. [CrossRef] [PubMed]
97. Satooka, H.; Hara-Chikuma, M. Aquaporin-3 Controls Breast Cancer Cell Migration by Regulating Hydrogen Peroxide Transport and Its Downstream Cell Signaling. *Mol. Cell. Biol.* **2016**, *36*, 1206–1218. [CrossRef] [PubMed]
98. Dong, X.; Wang, Y.; Zhou, Y.; Wen, J.; Wang, S.; Shen, L. Aquaporin 3 Facilitates Chemoresistance in Gastric Cancer Cells to Cisplatin via Autophagy. *Cell Death Discov.* **2016**, *2*, 16087. [CrossRef] [PubMed]
99. Yu, C.; Zhang, X.; Wang, J.; Song, H.; Liu, W.; Luo, B. Molecular Mechanism of Aquaporin 3 (AQP3) Regulating by LMP2A and Its Crosstalk with 4E-BP1 via ERK Signaling Pathway in EBV-Associated Gastric Cancer. *Virus Res.* **2022**, *322*, 198947. [CrossRef] [PubMed]
100. Zhu, Z.; Jiao, L.; Li, T.; Wang, H.; Wei, W.; Qian, H. Expression of AQP3 and AQP5 as a Prognostic Marker in Triple-Negative Breast Cancer. *Oncol. Lett.* **2018**, *16*, 2661–2667. [CrossRef] [PubMed]
101. de Almeida, A.; Parthimos, D.; Dew, H.; Smart, O.; Wiltshire, M.; Errington, R.J. Aquaglyceroporin-3's Expression and Cellular Localization Is Differentially Modulated by Hypoxia in Prostate Cancer Cell Lines. *Cells* **2021**, *10*, 838. [CrossRef] [PubMed]
102. Zhu, L.; Ma, N.; Wang, B.; Wang, L.; Zhou, C.; Yan, Y.; He, J.; Ren, Y. Significant Prognostic Values of Aquaporin mRNA Expression in Breast Cancer. *Cancer Manag. Res.* **2019**, *11*, 1503–1515. [CrossRef] [PubMed]
103. Li, C.F.; Zhang, W.G.; Liu, M.; Qiu, L.W.; Chen, X.F.; Lv, L.; Mei, Z.C. Aquaporin 9 Inhibits Hepatocellular Carcinoma through Up-Regulating FOXO1 Expression. *Oncotarget* **2016**, *7*, 44161–44170. [CrossRef] [PubMed]
104. Gao, C.; Shen, J.; Yao, L.; Xia, Z.; Liang, X.; Zhu, R.; Chen, Z. Low Expression of AQP9 and Its Value in Hepatocellular Carcinoma. *Transl. Cancer Res.* **2021**, *10*, 1826–1841. [CrossRef] [PubMed]
105. Chen, X.-F.; Li, C.-F.; Lü, L.; Mei, Z.-C. Expression and Clinical Significance of Aquaglyceroporins in Human Hepatocellular Carcinoma. *Mol. Med. Rep.* **2016**, *13*, 5283–5289. [CrossRef] [PubMed]
106. Wang, Y.; Wu, G.; Fu, X.; Xu, S.; Wang, T.; Zhang, Q.; Yang, Y. Aquaporin 3 Maintains the Stemness of CD133+ Hepatocellular Carcinoma Cells by Activating STAT3. *Cell Death Dis.* **2019**, *10*, 465. [CrossRef] [PubMed]
107. Wang, Y.; Wu, Y.; Li, X.; Song, W.; Zheng, F.; Mo, X.; Luo, Y.; Li, Y.; Chen, S.; Luo, H. Aquaporin-3 Promotes Proliferation and Inflammation in Hepatocellular Carcinoma. *Genes Dis.* **2024**, *11*, 101029. [CrossRef] [PubMed]
108. Saadoun, S.; Papadopoulos, M.C.; Davies, D.C.; Bell, B.A.; Krishna, S. Increased Aquaporin 1 Water Channel Expression Inhuman Brain Tumours. *Br. J. Cancer* **2002**, *87*, 621–623. [CrossRef] [PubMed]
109. Chen, Y.; Tachibana, O.; Oda, M.; Xu, R.; Hamada, J.-I.; Yamashita, J.; Hashimoto, N.; Takahashi, J.A. Increased Expression of Aquaporin 1 in Human Hemangioblastomas and Its Correlation with Cyst Formation. *J. Neurooncol.* **2006**, *80*, 219–225. [CrossRef] [PubMed]
110. Vacca, A.; Frigeri, A.; Ribatti, D.; Nicchia, G.P.; Nico, B.; Ria, R.; Svelto, M.; Dammacco, F. Microvessel Overexpression of Aquaporin 1 Parallels Bone Marrow Angiogenesis in Patients with Active Multiple Myeloma. *Br. J. Haematol.* **2001**, *113*, 415–421. [CrossRef] [PubMed]
111. Moon, C.; Soria, J.C.; Jang, S.J.; Lee, J.; Hoque, M.O.; Sibony, M.; Trink, B.; Chang, Y.S.; Sidransky, D.; Mao, L. Involvement of Aquaporins in Colorectal Carcinogenesis. *Oncogene* **2003**, *22*, 6699–6703. [CrossRef] [PubMed]

112. Pan, H.; Sun, C.C.; Zhou, C.Y.; Huang, H.F. Expression of Aquaporin-1 in Normal, Hyperplasic, and Carcinomatous Endometria. *Int. J. Gynecol. Obstet.* **2008**, *101*, 239–244. [CrossRef]
113. Yang, J.H.; Shi, Y.F.; Chen, X.D.; Qi, W.J. The Influence of Aquaporin-1 and Microvessel Density on Ovarian Carcinogenesis and Ascites Formation. *Int. J. Gynecol. Cancer* **2006**, *16*, 400–405. [CrossRef]
114. Hoque, M.O.; Soria, J.C.; Woo, J.; Lee, T.; Lee, J.; Jang, S.J.; Upadhyay, S.; Trink, B.; Monitto, C.; Desmaze, C.; et al. Aquaporin 1 Is Overexpressed in Lung Cancer and Stimulates NIH-3T3 Cell Proliferation and Anchorage-Independent Growth. *Am. J. Pathol.* **2006**, *168*, 1345–1353. [CrossRef] [PubMed]
115. Otterbach, F.; Callies, R.; Kimmig, R.; Schmid, K.W.; Bánkfalvi, A. Aquaporin-1-expression bei invasiven mammakarzinomen. *Pathologe* **2008**, *29*, 357–362. [CrossRef]
116. Kang, B.W.; Kim, J.G.; Lee, S.J.; Chae, Y.S.; Jeong, J.Y.; Yoon, G.S.; Park, S.Y.; Kim, H.J.; Park, J.S.; Choi, G.-S.; et al. Expression of Aquaporin-1, Aquaporin-3, and Aquaporin-5 Correlates with Nodal Metastasis in Colon Cancer. *Oncology* **2015**, *88*, 369–376. [CrossRef] [PubMed]
117. MacHida, Y.; Ueda, Y.; Shimasaki, M.; Sato, K.; Sagawa, M.; Katsuda, S.; Sakuma, T. Relationship of Aquaporin 1, 3, and 5 Expression in Lung Cancer Cells to Cellular Differentiation, Invasive Growth, and Metastasis Potential. *Hum. Pathol.* **2011**, *42*, 669–678. [CrossRef] [PubMed]
118. Otterbach, F.; Callies, R.; Adamzik, M.; Kimmig, R.; Siffert, W.; Schmid, K.W.; Bankfalvi, A. Aquaporin 1 (AQP1) Expression Is a Novel Characteristic Feature of a Particularly Aggressive Subgroup of Basal-like Breast Carcinomas. *Breast Cancer Res. Treat.* **2010**, *120*, 67–76. [CrossRef]
119. Sato, K.; Miyamoto, M.; Takano, M.; Furuya, K.; Tsuda, H. Different Prognostic Implications of Aquaporin-1 and Aquaporin-5 Expression among Different Histological Types of Ovarian Carcinoma. *Pathol. Oncol. Res.* **2020**, *26*, 263–271. [CrossRef] [PubMed]
120. Yoshida, T.; Hojo, S.; Sekine, S.; Sawada, S.; Okumura, T.; Nagata, T.; Shimada, Y.; Tsukada, K. Expression of Aquaporin-1 Is a Poor Prognostic Factor for Stage II and III Colon Cancer. *Mol. Clin. Oncol.* **2013**, *1*, 953–958. [CrossRef]
121. Wang, Y.; Yin, J.Y.; Li, X.P.; Chen, J.; Qian, C.Y.; Zheng, Y.; Fu, Y.L.; Chen, Z.Y.; Zhou, H.H.; Liu, Z.Q. The Association of Transporter Genes Polymorphisms and Lung Cancer Chemotherapy Response. *PLoS ONE* **2014**, *9*, e91967. [CrossRef] [PubMed]
122. Zou, L.-B.; Zhang, R.-J.; Tan, Y.-J.; Ding, G.-L.; Shi, S.; Zhang, D.; He, R.-H.; Liu, A.-X.; Wang, T.-T.; Leung, P.C.K.; et al. Identification of Estrogen Response Element in the Aquaporin-2 Gene That Mediates Estrogen-Induced Cell Migration and Invasion in Human Endometrial Carcinoma. *J. Clin. Endocrinol. Metab.* **2011**, *96*, E1399–E1408. [CrossRef] [PubMed]
123. Niu, D.; Kondo, T.; Nakazawa, T.; Yamane, T.; Mochizuki, K.; Kawasaki, T.; Matsuzaki, T.; Takata, K.; Katoh, R. Expression of Aquaporin3 in Human Neoplastic Tissues. *Histopathology* **2012**, *61*, 543–551. [CrossRef] [PubMed]
124. Hara-Chikuma, M.; Verkman, A.S. Prevention of Skin Tumorigenesis and Impairment of Epidermal Cell Proliferation by Targeted Aquaporin-3 Gene Disruption. *Mol. Cell. Biol.* **2008**, *28*, 326–332. [CrossRef] [PubMed]
125. Liu, Y.L.; Matsuzaki, T.; Nakazawa, T.; Murata, S.; Nakamura, N.; Kondo, T.; Iwashina, M.; Mochizuki, K.; Yamane, T.; Takata, K.; et al. Expression of Aquaporin 3 (AQP3) in Normal and Neoplastic Lung Tissues. *Hum. Pathol.* **2007**, *38*, 171–178. [CrossRef] [PubMed]
126. Li, A.; Lu, D.; Zhang, Y.; Li, J.; Fang, Y.; Li, F.; Sun, J. Critical Role of Aquaporin-3 in Epidermal Growth Factor-Induced Migration of Colorectal Carcinoma Cells and Its Clinical Significance. *Oncol. Rep.* **2013**, *29*, 535–540. [CrossRef] [PubMed]
127. Ismail, M.; Bokaee, S.; Davies, J.; Harrington, K.J.; Pandha, H. Inhibition of the Aquaporin 3 Water Channel Increases the Sensitivity of Prostate Cancer Cells to Cryotherapy. *Br. J. Cancer* **2009**, *100*, 1889–1895. [CrossRef] [PubMed]
128. Direito, I.; Paulino, J.; Vigia, E.; Brito, M.A.; Soveral, G. Differential Expression of Aquaporin-3 and Aquaporin-5 in Pancreatic Ductal Adenocarcinoma. *J. Surg. Oncol.* **2017**, *115*, 980–996. [CrossRef] [PubMed]
129. Rubenwolf, P.C.; Otto, W.; Denzinger, S.; Hofstädter, F.; Wieland, W.; Georgopoulos, N.T. Expression of Aquaporin Water Channels in Human Urothelial Carcinoma: Correlation of AQP3 Expression with Tumour Grade and Stage. *World J. Urol.* **2014**, *32*, 991–997. [CrossRef] [PubMed]
130. Ding, T.; Ma, Y.; Li, W.; Liu, X.; Ying, G.; Fu, L.; Gu, F. Role of Aquaporin-4 in the Regulation of Migration and Invasion of Human Glioma Cells. *Int. J. Oncol.* **2011**, *38*, 1521–1531. [CrossRef] [PubMed]
131. Mcferrin, M.B.; Sontheimer, H. A role for ion channels in glioma cell invasion. *Neuron Glia Biol.* **2006**, *2*, 39–49. [CrossRef] [PubMed]
132. Soelberg, K.; Larsen, S.R.; Moerch, M.T.; Thomassen, M.; Brusgaard, K.; Paul, F.; Smith, T.J.; Godballe, C.; Grauslund, J.; Lillevang, S.T.; et al. Aquaporin-4 IgG autoimmune syndrome and immunoreactivity associated with thyroid cancer. *Neurol. Neuroimmunol. Neuroinflammation* **2016**, *3*, e252. [CrossRef] [PubMed]
133. Yamamoto, A.; Shimizu, H.; Takiguchi, K.; Shoda, K.; Shiraishi, K.; Furuya, S.; Akaike, H.; Kawaguchi, Y.; Amemiya, H.; Kawaida, H.; et al. The Expression and Role of Aquaporin 4 in Colon Cancer. *Anticancer Res.* **2024**, *44*, 567–573. [CrossRef] [PubMed]
134. Saadoun, S.; Papadopoulos, M.C.; Davies, D.C.; Krishna, S.; Bell, B.A. Aquaporin-4 Expression Is Increased in Oedematous Human Brain Tumours. *J. Neurol. Neurosurg. Psychiatry* **2002**, *72*, 262–265. [CrossRef]

135. Yang, J.H.; Shi, Y.F.; Cheng, Q.; Deng, L. Expression and Localization of Aquaporin-5 in the Epithelial Ovarian Tumors. *Gynecol. Oncol.* **2006**, *100*, 294–299. [CrossRef]
136. Chae, Y.K.; Woo, J.; Kim, M.J.; Kang, S.K.; Kim, M.S.; Lee, J.; Lee, S.K.; Gong, G.; Kim, Y.H.; Soria, J.C.; et al. Expression of Aquaporin 5 (AQP5) Promotes Tumor Invasion in Human Non Small Cell Lung Cancer. *PLoS ONE* **2008**, *3*, e2162. [CrossRef] [PubMed]
137. Zhang, Z.; Chen, Z.; Song, Y.; Zhang, P.; Hu, J.; Bai, C. Expression of Aquaporin 5 Increases Proliferation and Metastasis Potential of Lung Cancer. *J. Pathol.* **2010**, *221*, 210–220. [CrossRef]
138. Jung, H.J.; Park, J.-Y.; Jeon, H.-S.; Kwon, T.-H. Aquaporin-5: A Marker Protein for Proliferation and Migration of Human Breast Cancer Cells. *PLoS ONE* **2011**, *6*, e28492. [CrossRef] [PubMed]
139. Zhang, T.; Zhao, C.; Chen, D.; Zhou, Z. Overexpression of AQP5 in Cervical Cancer: Correlation with Clinicopathological Features and Prognosis. *Med. Oncol.* **2012**, *29*, 1998–2004. [CrossRef] [PubMed]
140. Huang, Y.H.; Zhou, X.Y.; Wang, H.M.; Xu, H.; Chen, J.; Lv, N.H. Aquaporin 5 Promotes the Proliferation and Migration of Human Gastric Carcinoma Cells. *Tumor Biol.* **2013**, *34*, 1743–1751. [CrossRef] [PubMed]
141. Kang, S.K.; Chae, Y.K.; Woo, J.; Kim, M.S.; Park, J.C.; Lee, J.; Soria, J.C.; Jang, S.J.; Sidransky, D.; Moon, C. Role of Human Aquaporin 5 In Colorectal Carcinogenesis. *Am. J. Pathol.* **2008**, *173*, 518–525. [CrossRef] [PubMed]
142. Peng, R. Auphen and Dibutyryl cAMP Suppress Growth of Hepatocellular Carcinoma by Regulating Expression of Aquaporins 3 and 9 in Vivo. *World J. Gastroenterol.* **2016**, *22*, 3341–3354. [CrossRef] [PubMed]
143. Guo, X.; Sun, T.; Yang, M.; Li, Z.; Li, Z.; Gao, Y. Prognostic Value of Combined Aquaporin 3 and Aquaporin 5 Overexpression in Hepatocellular Carcinoma. *Biomed. Res. Int.* **2013**, *2013*, 206525. [CrossRef] [PubMed]
144. Chen, G.; Shi, Y.; Liu, M.; Sun, J. circHIPK3 Regulates Cell Proliferation and Migration by Sponging miR-124 and Regu-Lating AQP3 Expression in Hepatocellular Carcinoma. *Cell Death Dis.* **2018**, *9*, 175. [CrossRef] [PubMed]
145. He, Z.; Dong, W.; Hu, J.; Ren, X. AQP5 promotes hepatocellular carcinoma metastasis via NF-κB-regulated epitheli-al-mesenchymal transition. *Biochem. Biophys. Res. Commun.* **2017**, *490*, 343–348. [CrossRef] [PubMed]
146. Ma, J.; Zhou, C.; Yang, J.; Ding, X.; Zhu, Y.; Chen, X. Expression of AQP6 and AQP8 in Epithelial Ovarian Tumor. *J. Mol. Histol.* **2016**, *47*, 129–134. [CrossRef] [PubMed]
147. Sega, F.V.D.; Prata, C.; Zambonin, L.; Angeloni, C.; Rizzo, B.; Hrelia, S.; Fiorentini, D. Intracellular Cysteine Oxidation Is Modulated by Aquaporin-8-Mediated Hydrogen Peroxide Channeling in Leukaemia Cells. *BioFactors* **2017**, *43*, 232–242. [CrossRef] [PubMed]
148. Sega, F.V.D.; Zambonin, L.; Fiorentini, D.; Rizzo, B.; Caliceti, C.; Landi, L.; Hrelia, S.; Prata, C. Specific Aquaporins Facilitate Nox-Produced Hydrogen Peroxide Transport through Plasma Membrane in Leukaemia Cells. *Biochim. Biophys. Acta* **2014**, *1843*, 806–814. [CrossRef] [PubMed]
149. Chen, Q.; Zhu, L.; Zheng, B.; Wang, J.; Song, X.; Zheng, W.; Wang, L.; Yang, D.; Wang, J. Effect of AQP9 Expression in Androgen-Independent Prostate Cancer Cell PC3. *Int. J. Mol. Sci.* **2016**, *17*, 738. [CrossRef] [PubMed]
150. Miller, E.W.; Dickinson, B.C.; Chang, C.J. Aquaporin-3 Mediates Hydrogen Peroxide Uptake to Regulate Downstream Intracellular Signaling. *Proc. Natl. Acad. Sci. USA* **2010**, *107*, 15681–15686. [CrossRef] [PubMed]
151. Furfaro, A.L.; Traverso, N.; Domenicotti, C.; Piras, S.; Moretta, L.; Marinari, U.M.; Pronzato, M.A.; Nitti, M. The Nrf2/HO-1 Axis in Cancer Cell Growth and Chemoresistance. *Oxidative Med. Cell. Longev.* **2016**, *2016*, 1958174. [CrossRef] [PubMed]
152. Łukasiewicz, S.; Czeczelewski, M.; Forma, A.; Baj, J.; Sitarz, R.; Stanisławek, A. Breast Cancer—Epidemiology, Risk Factors, Classification, Prognostic Markers, and Current Treatment Strategies—An Updated Review. *Cancers* **2021**, *13*, 4287. [CrossRef] [PubMed]
153. Feng, Y. Breast Cancer Development and Progression: Risk Factors, Cancer Stem Cells, Signaling Pathways, Genomics, and Molecular Pathogenesis. *Genes Dis.* **2018**, *5*, 77–106. [CrossRef] [PubMed]
154. Kannan, K.; Srinivasan, A.; Kannan, A.; Ali, N. The Underlying Mechanisms and Emerging Strategies to Overcome Resistance in Breast Cancer. *Cancers* **2025**, *17*, 2938. [CrossRef] [PubMed]
155. Yates, L.R.; Knappskog, S.; Wedge, D.; Farmery, J.H.R.; Gonzalez, S.; Martincorena, I.; Alexandrov, L.B.; Loo, P.V.; Haugland, H.K.; Lilleng, P.K.; et al. Genomic Evolution of Breast Cancer Metastasis and Relapse. *Cancer Cell* **2017**, *32*, 169–184. [CrossRef] [PubMed]
156. Milković, L.; Mlinarić, M.; Lučić, I.; Čipak Gašparović, A. The Involvement of Peroxiporins and Antioxidant Transcription Factors in Breast Cancer Therapy Resistance. *Cancers* **2023**, *15*, 5747. [CrossRef] [PubMed]
157. Ribatti, D.; Ranieri, G.; Annese, T.; Nico, B. Aquaporins in Cancer. *Biochim. Biophys. Acta (BBA)-Gen. Subj.* **2014**, *1840*, 1550–1553. [CrossRef] [PubMed]
158. Will, M.; Liang, J.; Metcalfe, C.; Chandarlapaty, S. Therapeutic Resistance to Anti-Oestrogen Therapy in Breast Cancer. *Nat. Rev. Cancer* **2023**, *23*, 673–685. [CrossRef] [PubMed]
159. Razavi, P.; Chang, M.T.; Xu, G.; Bandlamudi, C.; Ross, D.S.; Vasan, N. The Genomic Landscape of Endocrine-Resistant Advanced Breast Cancers. *Cancer Cell* **2018**, *34*, 427–438.e6. [CrossRef] [PubMed]

160. Garg, P.; Ramisetty, S.; Nair, M.; Kulkarni, P.; Horne, D.; Salgia, R.; Singhal, S.S. Strategic Advancements in Targeting the PI3K/AKT/mTOR Pathway for Breast Cancer Therapy. *Biochem. Pharmacol.* **2025**, *236*, 116850. [CrossRef] [PubMed]
161. Hanker, A.B.; Sudhan, D.R.; Arteaga, C.L. Overcoming Endocrine Resistance in Breast Cancer. *Cancer Cell* **2020**, *37*, 496–513. [CrossRef] [PubMed]
162. Nayar, U.; Cohen, O.; Kapstad, C.; Cuoco, M.S.; Waks, A.G.; Wander, S.A.; Painter, C.; Freeman, S.; Persky, N.S.; Marini, L.; et al. Acquired HER2 Mutations in ER+ Metastatic Breast Cancer Confer Resistance to Estrogen Receptor-Directed Therapies. *Nat. Genet.* **2019**, *51*, 207–216. [CrossRef] [PubMed]
163. Huang, Y.-T.; Zhou, J.; Shi, S.; Xu, H.-Y.; Qu, F.; Zhang, D.; Chen, Y.-D.; Yang, J.; Huang, H.-F.; Sheng, J.-Z. Identification of Estrogen Response Element in Aquaporin-3 Gene That Mediates Estrogen-Induced Cell Migration and Invasion in Estrogen Receptor-Positive Breast Cancer. *Sci. Rep.* **2015**, *5*, 12484. [CrossRef] [PubMed]
164. Ahmad, A.E.; Khajah, M.A.; Khushaish, S.; Luqmani, Y.A. Aquaporin Expression in Breast Cancer and Their Involvement in Bleb Formation, Cell Motility and Invasion in Endocrine Resistant Variant Cells. *Int. J. Oncol.* **2020**, *56*, 1014–1024. [CrossRef]
165. Fourquet, S.; Guerois, R.; Biard, D.; Toledano, M.B. Activation of NRF2 by Nitrosative Agents and H_2O_2 Involves KEAP1 Disulfide Formation. *J. Biol. Chem.* **2010**, *285*, 8463–8471. [CrossRef]
166. Reinema, F.V.; Sweep, F.C.G.J.; Adema, G.J.; Peeters, W.J.M.; Martens, J.W.M.; Bussink, J.; Span, P.N. Tamoxifen Induces Radioresistance through NRF2-Mediated Metabolic Reprogramming in Breast Cancer. *Cancer Metab.* **2023**, *11*, 3. [CrossRef] [PubMed]
167. Kinnel, B.; Singh, S.K.; Oprea-Ilies, G.; Singh, R. Targeted Therapy and Mechanisms of Drug Resistance in Breast Cancer. *Cancers* **2023**, *15*, 1320. [CrossRef] [PubMed]
168. Robey, R.W.; Pluchino, K.M.; Hall, M.D.; Fojo, A.T.; Bates, S.E.; Gottesman, M.M. Revisiting the Role of ABC Transporters in Multidrug-Resistant Cancer. *Nat. Rev. Cancer* **2018**, *18*, 452–464. [CrossRef] [PubMed]
169. Hembruff, S.L.; Laberge, M.L.; Villeneuve, D.J.; Guo, B.; Veitch, Z.; Cecchetto, M.; Parissenti, A.M. Role of Drug Transporters and Drug Accumulation in the Temporal Acquisition of Drug Resistance. *BMC Cancer* **2008**, *8*, 318. [CrossRef] [PubMed]
170. Gillespie, M.S.; Ward, C.M.; Davies, C.C. DNA Repair and Therapeutic Strategies in Cancer Stem Cells. *Cancers* **2023**, *15*, 1897. [CrossRef] [PubMed]
171. Basourakos, S.P.; Li, L.; Aparicio, A.M.; Corn, P.G.; Kim, J.; Thompson, T.C. Combination Platinum-Based and DNA Damage Response-Targeting Cancer Therapy: Evolution and Future Directions. *Curr. Med. Chem.* **2016**, *24*, 1586–1606. [CrossRef] [PubMed]
172. Lord, C.J.; Ashworth, A. The DNA Damage Response and Cancer Therapy. *Nature* **2012**, *481*, 287–294. [CrossRef] [PubMed]
173. Ryoo, I.-G.; Kim, G.; Choi, B.-H.; Lee, S.-H.; Kwak, M.-K. Involvement of NRF2 Signaling in Doxorubicin Resistance of Cancer Stem Cell-Enriched Colonospheres. *Biomol. Ther.* **2016**, *24*, 482–488. [CrossRef] [PubMed]
174. Dajani, S.; Saripalli, A.; Sharma-Walia, N. Water Transport Proteins-Aquaporins (AQPs) in Cancer Biology. *Oncotarget* **2018**, *9*, 36392–36405. [CrossRef] [PubMed]
175. Hu, D.; Lyu, X.; Li, Z.; Ekambaram, P.; Dongre, A.; Freeman, T.; Joy, M.; Atkinson, J.M.; Brown, D.D.; Cai, Z.; et al. p95HER2, a Truncated Form of the HER2 Oncoprotein, Drives an Immunosuppressive Program in HER2+ Breast Cancer That Limits Trastuzumab Deruxtecan Efficacy. *Nat. Cancer* **2025**, *6*, 1202–1222. [CrossRef] [PubMed]
176. Zeng, H.; Wang, W.; Zhang, L.; Lin, Z. HER3-Targeted Therapy: The Mechanism of Drug Resistance and the Development of Anticancer Drugs. *Cancer Drug Resist.* **2024**, *29*, 14. [CrossRef] [PubMed]
177. Condorelli, R.; Spring, L.; O'Shaughnessy, J.; Lacroix, L.; Bailleux, C.; Scott, V.; Dubois, J.; Nagy, R.J.; Lanman, R.B.; Iafrate, A.J.; et al. Polyclonal RB1 Mutations and Acquired Resistance to CDK 4/6 Inhibitors in Patients with Metastatic Breast Cancer. *Ann. Oncol.* **2018**, *29*, 640–645. [CrossRef] [PubMed]
178. Herrera-Abreu, M.T.; Palafox, M.; Asghar, U.; Rivas, M.A.; Cutts, R.J.; Garcia-Murillas, I.; Pearson, A.; Guzman, M.; Rodriguez, O.; Grueso, J.; et al. Early Adaptation and Acquired Resistance to CDK4/6 Inhibition in Estrogen Receptor–Positive Breast Cancer. *Cancer Res.* **2016**, *76*, 2301–2313. [CrossRef] [PubMed]
179. Yang, C.; Li, Z.; Bhatt, T.; Dickler, M.; Giri, D.; Scaltriti, M.; Baselga, J.; Rosen, N.; Chandarlapaty, S. Acquired CDK6 Amplification Promotes Breast Cancer Resistance to CDK4/6 Inhibitors and Loss of ER Signaling and Dependence. *Oncogene* **2017**, *36*, 2255–2264. [CrossRef] [PubMed]
180. Restifo, N.P.; Marincola, F.M.; Kawakami, Y.; Taubenberger, J.; Yannelli, J.R.; Rosenberg, S.A. Loss of Functional Beta 2-Microglobulin in Metastatic Melanomas from Five Patients Receiving Immunotherapy. *J. Natl. Cancer Inst.* **1996**, *88*, 100–108. [CrossRef] [PubMed]
181. Wen, H.; Suzuki, T.; Zhang, A.; Sato, M.; Matsumoto, M.; Takahashi, Y.; Takahashi, Y.; Yamamoto, M. NRF2 Activation in Cancer Cells Suppresses Immune Infiltration into the Tumor Microenvironment. *iScience* **2025**, *28*, 113519. [CrossRef] [PubMed]
182. Haist, M.; Stege, H.; Grabbe, S.; Bros, M. The Functional Crosstalk between Myeloid-Derived Suppressor Cells and Regulatory T Cells within the Immunosuppressive Tumor Microenvironment. *Cancers* **2021**, *13*, 210. [CrossRef] [PubMed]

183. Tian, T.; Li, Z. Targeting Tim-3 in Cancer With Resistance to PD-1/PD-L1 Blockade. *Front. Oncol.* **2021**, *11*, 731175. [CrossRef] [PubMed]
184. Borgeaud, M.; Sandoval, J.; Obeid, M.; Banna, G.; Michielin, O.; Addeo, A.; Friedlaender, A. Novel Targets for Immune-Checkpoint Inhibition in Cancer. *Cancer Treat. Rev.* **2023**, *120*, 102614. [CrossRef] [PubMed]
185. Jin, X.; Gong, L.; Peng, Y.; Li, L.; Liu, G. Enhancer-Bound Nrf2 Licenses HIF-1α Transcription under Hypoxia to Promote Cisplatin Resistance in Hepatocellular Carcinoma Cells. *Aging* **2020**, *13*, 364–375. [CrossRef] [PubMed]
186. Bel'skaya, V.L.; Dyachenko, E.I. Oxidative Stress in Breast Cancer: A Biochemical Map of Reactive Oxygen Species Production. *Curr. Issues Mol. Biol.* **2024**, *46*, 4646–4687. [CrossRef] [PubMed]
187. Read, G.H.; Bailleul, J.; Vlashi, E.; Kesarwala, A.H. Metabolic Response to Radiation Therapy in Cancer. *Mol. Carcinog.* **2022**, *61*, 200–224. [CrossRef] [PubMed]
188. Morgan, E. Global Burden of Colorectal Cancer in 2020 and 2040: Incidence and Mortality Estimates from GLOBOCAN. *Gut* **2023**, *72*, 338–344. [CrossRef] [PubMed]
189. Chambers, A.C.; Dixon, S.W.; White, P.; Williams, A.C.; Thomas, M.G.; Messenger, D.E. Demographic Trends in the Incidence of Young-Onset Colorectal Cancer: A Population-Based Study. *Br. J. Surg.* **2020**, *107*, 595–605. [CrossRef] [PubMed]
190. Lui, R.N. Global Increasing Incidence of Young-Onset Colorectal Cancer Across 5 Continents: A Joinpoint Regression Analysis of 1,922,167 Cases. *Cancer Epidemiol. Biomark. Prev.* **2019**, *28*, 1275–1282. [CrossRef] [PubMed]
191. Austin, H.; Henley, S.J.; King, J.; Richardson, L.C.; Eheman, C. Changes in Colorectal Cancer Incidence Rates in Young and Older Adults in the United States: What Does It Tell Us about Screening. *Cancer Causes Control* **2014**, *25*, 191–201. [CrossRef] [PubMed]
192. Dey, A.; Mitra, A.; Pathak, S.; Prasad, S.; Zhang, A.S.; Zhang, H.; Sun, X.-F.; Banerjee, A. Recent Advancements, Limitations, and Future Perspectives of the Use of Personalized Medicine in Treatment of Colon Cancer. *Technol. Cancer Res. Treat.* **2023**, *22*, 15330338231178403. [CrossRef] [PubMed]
193. Li, Y.; Luo, Y.; Ran, Y.; Lu, F.; Qin, Y. Biomarkers of Inflammation and Colorectal Cancer Risk. *Front. Oncol.* **2025**, *15*, 1514009. [CrossRef] [PubMed]
194. Veettil, S.K.; Wong, T.Y.; Loo, Y.S.; Playdon, M.C.; Lai, N.M.; Giovannucci, E.L.; Chaiyakunapruk, N. Role of Diet in Colorectal Cancer Incidence: Umbrella Review of Meta-Analyses of Prospective Observational Studies. *JAMA Netw. Open* **2021**, *4*, e2037341. [CrossRef] [PubMed]
195. Kantha, A.; Das, D.; Pai, E.; Kumar, T.; Pandey, M. Consensus Molecular Subtypes (CMS) Classification: A Progress towards Subtype-Driven Treatments in Colorectal Cancer. *World J. Surg. Oncol.* **2025**, *24*, 1. [CrossRef] [PubMed]
196. Guinney, J.; Dienstmann, R.; Wang, X.; de Reyniès, A.; Schlicker, A.; Soneson, C.; Marisa, L.; Roepman, P.; Nyamundanda, G.; Angelino, P.; et al. The Consensus Molecular Subtypes of Colorectal Cancer. *Nat. Med.* **2015**, *21*, 1350–1356. [CrossRef] [PubMed]
197. Siegel, R.L.; Wagle, N.S.; Cercek, A.; Smith, R.A.; Jemal, A. Colorectal Cancer Statistics, 2023. *CA Cancer J. Clin.* **2023**, *73*, 233–254. [CrossRef] [PubMed]
198. Blondy, S.; David, V.; Verdier, M.; Mathonnet, M.; Perraud, A.; Christou, N. 5-Fluorouracil Resistance Mechanisms in Colorectal Cancer: From Classical Pathways to Promising Processes. *Cancer Sci.* **2020**, *111*, 3142–3154. [CrossRef] [PubMed]
199. Malcanlı, S.; Akar, R.O.; Ulukaya, E.; Özkaya, A.B.; Geyik, Ö.G. Enhancing 5-Fluorouracil Efficacy in Colorectal Cancer by Inhibiting Glutathione Antioxidant Mechanisms with an xCT Inhibitor. *Sci. Rep.* **2026**, *16*, 10196. [CrossRef] [PubMed]
200. Kang, K.A. Epigenetic Modification of Nrf2 in 5-Fluorouracil-Resistant Colon Cancer Cells: Involvement of TET-Dependent DNA Demethylation. *Cell Death Dis.* **2014**, *5*, e1183. [CrossRef] [PubMed]
201. Akhdar, H.; Loyer, P.; Rauch, C.; Corlu, A.; Guillouzo, A.; Morel, F. Involvement of Nrf2 Activation in Resistance to 5-Fluorouracil in Human Colon Cancer HT-29 Cells. *Eur. J. Cancer* **2009**, *45*, 2219–2227. [CrossRef] [PubMed]
202. DeNicola, G.M.; Karreth, F.A.; Humpton, T.J.; Gopinathan, A.; Wei, C.; Frese, K.; Mangal, D.; Yu, K.H.; Yeo, C.J.; Calhoun, E.S.; et al. Oncogene-Induced Nrf2 Transcription Promotes ROS Detoxification and Tumorigenesis. *Nature* **2011**, *475*, 106–109. [CrossRef] [PubMed]
203. Tessmann, J.W.; Rocha, M.R.; Morgado-Díaz, J.A. Mechanisms of Radioresistance and the Underlying Signaling Pathways in Colorectal Cancer Cells. *J. Cell. Biochem.* **2023**, *124*, 31–45. [CrossRef] [PubMed]
204. Perše, M. Oxidative Stress in the Pathogenesis of Colorectal Cancer: Cause or Consequence? *Biomed. Res. Int.* **2013**, *2013*, 725710. [CrossRef] [PubMed]
205. Basak, D.; Uddin, M.N.; Hancock, J. The Role of Oxidative Stress and Its Counteractive Utility in Colorectal Cancer (CRC). *Cancers* **2020**, *12*, 3336. [CrossRef] [PubMed]
206. Jiang, Y. Aquaporin-1 Activity of Plasma Membrane Affects HT20 Colon Cancer Cell Migration. *IUBMB Life* **2009**, *61*, 1001–1009. [CrossRef] [PubMed]
207. Dorward, H.S.; Du, A.; Bruhn, M.A.; Wrin, J.; Pei, J.V.; Evdokiou, A.; Price, T.J.; Yool, A.J.; Hardingham, J.E. Pharmacological Blockade of Aquaporin-1 Water Channel by AqB013 Restricts Migration and Invasiveness of Colon Cancer Cells and Prevents Endothelial Tube Formation in Vitro. *J. Exp. Clin. Cancer Res.* **2016**, *35*, 36. [CrossRef] [PubMed]
208. Tan, E.Y. Liver cancer in 2021: Global Burden of Disease study. *J. Hepatol.* **2025**, *82*, 851–860. [CrossRef] [PubMed]

209. Center, M.M.; Jemal, A. International Trends in Liver Cancer Incidence Rates. *Cancer Epidemiol. Biomark. Prev.* **2011**, *20*, 2362–2368. [CrossRef] [PubMed]
210. Xue, Y.; Ruan, Y.; Wang, Y.; Xiao, P.; Xu, J. Signaling Pathways in Liver Cancer: Pathogenesis and Targeted Therapy. *Mol. Biomed.* **2024**, *5*, 20. [CrossRef] [PubMed]
211. Anwanwan, D.; Singh, S.K.; Singh, S.; Saikam, V.; Singh, R. Challenges in Liver Cancer and Possible Treatment Approaches. *Biochim. Biophys. Acta (BBA)-Rev. Cancer* **2020**, *1873*, 188314. [CrossRef] [PubMed]
212. Llovet, J.M. Sorafenib in Advanced Hepatocellular Carcinoma. *N. Engl. J. Med.* **2008**, *359*, 378–390. [CrossRef] [PubMed]
213. Esposti, D.D. Mitochondrial Roles and Cytoprotection in Chronic Liver Injury. *Biochem. Res. Int.* **2012**, *2012*, 387626. [CrossRef] [PubMed]
214. Takaki, A.; Yamamoto, K. Control of Oxidative Stress in Hepatocellular Carcinoma: Helpful or Harmful? *World J. Hepatol.* **2015**, *7*, 968. [CrossRef] [PubMed]
215. Michilli, A. Transcriptomic Analysis Reveals an NRF2-Mediated Redox and Metabolic Reprogramming in Soraf-Enib-Resistant Hepatocellular Carcinoma Cells. *BioTech* **2026**, *15*, 18. [CrossRef] [PubMed]
216. Beretta, G.L.; Cassinelli, G.; Pennati, M.; Zuco, V.; Gatti, L. Overcoming ABC transporter-mediated multidrug resistance: The dual role of tyrosine kinase inhibitors as multitargeting agents. *Eur. J. Med. Chem.* **2017**, *142*, 271–289. [CrossRef] [PubMed]
217. Durmus, S.; Hendrikx, J.J.M.A.; Schinkel, A.H. Apical ABC Transporters and Cancer Chemotherapeutic Drug Dis-Position. *Adv. Cancer Res.* **2015**, *125*, 1–41. [CrossRef] [PubMed]
218. Fornari, F.; Giovannini, C.; Piscaglia, F.; Gramantieri, L. Elucidating the Molecular Basis of Sorafenib Resistance in HCC: Current Findings and Future Directions. *J. Hepatocell. Carcinoma* **2021**, *8*, 741–757. [CrossRef] [PubMed]
219. Ma-on, C. Oxidative Stress Indicated by Elevated Expression of Nrf2 and 8-OHdG Promotes Hepatocellular Carcinoma Progression. *Med. Oncol.* **2017**, *34*, 57. [CrossRef] [PubMed]
220. Niture, S.K.; Jaiswal, A.K. Nrf2-Induced Antiapoptotic Bcl-xL Protein Enhances Cell Survival and Drug Resistance. *Free Radic. Biol. Med.* **2013**, *57*, 119–131. [CrossRef] [PubMed]
221. Sun, X. Activation of the P62-Keap1-NRF2 Pathway Protects against Ferroptosis in Hepatocellular Carcinoma Cells. *Hepatology* **2016**, *63*, 173–184. [CrossRef] [PubMed]
222. Jablonski, E.M. Decreased Aquaporin Expression Leads to Increased Resistance to Apoptosis in Hepatocellular Carcioma. *Cancer Lett.* **2007**, *250*, 36–46. [CrossRef] [PubMed]
223. Kang, M.-I.; Kobayashi, A.; Wakabayashi, N.; Kim, S.-G.; Yamamoto, M. Scaffolding of Keap1 to the Actin Cytoskeleton Controls the Function of Nrf2 as Key Regulator of Cytoprotective Phase 2 Genes. *Proc. Natl. Acad. Sci. USA* **2004**, *101*, 2046–2051. [CrossRef] [PubMed]
224. Sova, M.; Saso, L. Design and Development of Nrf2 Modulators for Cancer Chemoprevention and Therapy: A Review. *Drug Des. Dev. Ther.* **2018**, *12*, 3181–3197. [CrossRef] [PubMed]
225. Robledinos-Antón, N.; Fernández-Ginés, R.; Manda, G.; Cuadrado, A. Activators and Inhibitors of NRF2: A Review of Their Potential for Clinical Development. *Oxid. Med. Cell. Longev.* **2019**, *2019*, 9372182. [CrossRef] [PubMed]
226. Telkoparan-Akillilar, P.; Panieri, E.; Cevik, D.; Suzen, S.; Saso, L. Therapeutic Targeting of the NRF2 Signaling Pathway in Cancer. *Molecules* **2021**, *26*, 1417. [CrossRef] [PubMed]
227. Silva, P.M.; da Silva, I.V.; Sarmento, M.J.; Silva, Í.C.; Carvalho, F.A.; Soveral, G.; Santos, N.C. Aquaporin-3 and Aquaporin-5 Facilitate Migration and Cell-Cell Adhesion in Pancreatic Cancer by Modulating Cell Biomechanical Properties. *Cells* **2022**, *11*, 1308. [CrossRef] [PubMed]
228. Yang, J.; Yan, C.; Zheng, W.; Chen, X. Proliferation Inhibition of Cisplatin and Aquaporin 5 Expression in Human Ovarian Cancer Cell CAOV3. *Arch. Gynecol. Obstet.* **2012**, *285*, 239–245. [CrossRef] [PubMed]
229. Butcher, P.R.; Sykes, D. Strategies for Enhancing Selectivity in Anticancer Metal Complexes. *ACS Omega* **2026**, *11*, 31972–32001. [CrossRef] [PubMed]
230. Małkowska, P.; Tarnowski, M. Natural Modulators of Aquaporins in Cancer Therapy: Functional Mechanisms and Clinical Potential. *Molecules* **2026**, *31*, 1072. [CrossRef] [PubMed]
231. Charlestin, V.; Fulkerson, D.; Arias Matus, C.E.; Walker, Z.T.; Carthy, K.; Littlepage, L.E. Aquaporins: New Players in Breast Cancer Progression and Treatment Response. *Front. Oncol.* **2022**, *12*, 988119. [CrossRef] [PubMed]
232. Zhu, H.; Xie, D.; Yu, Y.; Yao, L.; Xu, B.; Huang, L.; Wu, S.; Li, F.; Zheng, Y.; Liu, X.; et al. KEAP1/NFE2L2 Mutations of Liquid Biopsy as Prognostic Biomarkers in Patients With Advanced Non-Small Cell Lung Cancer: Results From Two Multicenter, Randomized Clinical Trials. *Front. Oncol.* **2021**, *11*, 659200. [CrossRef] [PubMed]
233. Binkley, M.S.; Jeon, Y.-J.; Nesselbush, M.; Moding, E.J.; Nabet, B.Y.; Almanza, D.; Kunder, C.; Stehr, H.; Yoo, C.H.; Rhee, S.; et al. KEAP1/NFE2L2 Mutations Predict Lung Cancer Radiation Resistance That Can Be Targeted by Glutaminase Inhibition. *Cancer Discov.* **2020**, *10*, 1826–1841. [CrossRef] [PubMed]
234. Zhang, G.; Hao, Y.; Chen, L.; Li, Z.; Gao, L.; Tian, J.; Qiao, Q.; Zhang, J. Expression of Aquaporin 1, 3 and 5 in Colorectal Carcinoma: Correlation with Clinicopathological Characteristics and Prognosis. *Pathol. Oncol. Res.* **2023**, *29*, 1611179. [CrossRef] [PubMed]

235. Koelzer, V.H.; Sirinukunwattana, K.; Rittscher, J.; Mertz, K.D. Precision Immunoprofiling by Image Analysis and Artificial Intelligence. *Virchows Arch.* **2019**, *474*, 511–522. [CrossRef] [PubMed]
236. Du, Z.; Lin, J.-R.; Rashid, R.; Maliga, Z.; Wang, S.; Aster, J.C.; Izar, B.; Sorger, P.K.; Santagata, S. Qualifying Antibodies for Image-Based Immune Profiling and Multiplexed Tissue Imaging. *Nat. Protoc.* **2019**, *14*, 2900–2930. [CrossRef] [PubMed]
237. Torrente, L.; Maan, G.; Oumkaltoum Rezig, A.; Quinn, J.; Jackson, A.; Grilli, A.; Casares, L.; Zhang, Y.; Kulesskiy, E.; Saarela, J. High NRF2 Levels Correlate with Poor Prognosis in Colorectal Cancer Patients and with Sensitivity to the Kinase Inhibitor AT9283 In Vitro. *Biomolecules* **2020**, *10*, 1365. [CrossRef] [PubMed]
238. Lee, K.; Kim, S.; Lee, Y.; Lee, H.; Lee, Y.; Park, H.; Nahm, J.H.; Ahn, S.; Yu, S.J.; Lee, K.; et al. The Clinicopathological and Prognostic Significance of Nrf2 and Keap1 Expression in Hepatocellular Carcinoma. *Cancers* **2020**, *12*, 2128. [CrossRef] [PubMed]
239. Hassan, S.; Ferrario, C.; Mamo, A.; Basik, M. Tissue Microarrays: Emerging Standard for Biomarker Validation. *Curr. Opin. Biotechnol.* **2008**, *19*, 19–25. [CrossRef] [PubMed]
240. Camp, R.L.; Charette, L.A.; Rimm, D.L. Validation of Tissue Microarray Technology in Breast Carcinoma. *Lab. Invest.* **2000**, *80*, 1943–1949. [CrossRef] [PubMed]
241. Goldsmith, J.D.; Troxell, M.L.; Roy-Chowdhuri, S.; Colasacco, C.F.; Edgerton, M.E.; Fitzgibbons, P.L.; Fulton, R.; Haas, T.; Kandalaft, P.L.; Kalicanin, T. Principles of Analytic Validation of Immunohistochemical Assays: Guideline Update. *Arch. Pathol. Lab. Med.* **2024**, *148*, 111–153. [CrossRef] [PubMed]
242. Altman, D.G.; McShane, L.M.; Sauerbrei, W.; Taube, S.E. Reporting Recommendations for Tumor Marker Prognostic Studies (REMARK): Explanation and Elaboration. *PLoS Med.* **2012**, *9*, 1001216. [CrossRef] [PubMed]
243. Loughrey, M.B.; Bankhead, P.; Coleman, H.G.; Hagan, R.S.; Craig, S.; McCorry, A.M.B.; Gray, R.T.; McQuaid, S.; Dunne, P.D.; Hamilton, P.W. Validation of the Systematic Scoring of Immunohistochemically Stained Tumour Tissue Microarrays Using QuPath Digital Image Analysis. *Histopathology* **2018**, *73*, 327–338. [CrossRef] [PubMed]
244. Lee, Y.S.; Kang, J.; Jung, E.S.; Lee, A. High Expression of NRF2 and Low Expression of KEAP1 Predict Worse Survival in Patients With Operable Triple-Negative Breast Cancer. *J. Breast Cancer* **2023**, *26*, 461–478. [CrossRef] [PubMed]
245. Jang, S.J.; Moon, C. Aquaporin 5 (AQP5) Expression in Breast Cancer and Its Clinicopathological Characteristics. *PLoS ONE* **2023**, *18*, e0270752. [CrossRef] [PubMed]
246. Onodera, Y.; Motohashi, H.; Takagi, K.; Miki, Y.; Shibahara, Y.; Watanabe, M.; Ishida, T.; Hirakawa, H.; Sasano, H.; Yamamoto, M.; et al. NRF2 Immunolocalization in Human Breast Cancer Patients as a Prognostic Factor. *Endocr. Relat. Cancer* **2014**, *21*, 241–252. [CrossRef] [PubMed]
247. Chang, L.-C.; Fan, C.-W.; Tseng, W.-K.; Chen, J.-R.; Chein, H.-P.; Hwang, C.-C. Immunohistochemical Study of the Nrf2 Pathway in Colorectal Cancer. *Appl. Immunohistochem. Mol. Morphol.* **2013**, *21*, 511–517. [CrossRef] [PubMed]

Disclaimer/Publisher's Note: The statements, opinions and data contained in all publications are solely those of the individual author(s) and contributor(s) and not of MDPI and/or the editor(s). MDPI and/or the editor(s) disclaim responsibility for any injury to people or property resulting from any ideas, methods, instructions or products referred to in the content.