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Exploring the multifaceted role of NRF2 in brain physiology and cancer: A comprehensive review

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Abstract

Chronic oxidative stress plays a critical role in the development of brain malignancies due to the high rate of brain oxygen utilization and concomitant production of reactive oxygen species. The nuclear factor-erythroid-2-related factor 2 (NRF2), a master regulator of antioxidant signaling, is a key factor in regulating brain physiology and the development of age-related neurodegenerative diseases. Also, NRF2 is known to exert a protective antioxidant effect against the onset of oxidative stress-induced diseases, including cancer, along with its pro-oncogenic activities through regulating various signaling pathways and downstream target genes. In glioblastoma (GB), grade 4 glioma, tumor resistance, and recurrence are caused by the glioblastoma stem cell population constituting a small bulk of the tumor core. The persistence and self-renewal capacity of these cell populations is enhanced by NRF2 expression in GB tissues. This review outlines NRF2's dual involvement in cancer and highlights its regulatory role in human brain physiology and diseases, in addition to the development of primary brain tumors and therapeutic potential, with a focus on GB.

Key Points

- NRF2 is vital for optimal functioning and redox homeostasis in brain cells.
- NRF2 contributes to GSC maintenance, GB development, and metabolic reprogramming.
- Targeting NRF2 offers a potential therapeutic target for GB treatment and therapeutic resistance.

Oxidative Stress and Human Cancer

Cellular Oxidation and Cancer Onset

Cellular redox homeostasis is a state of physiological equilibrium between the intracellular reactive oxygen species (ROS), reactive nitrogen species (RNS), thiol-containing compounds, as well as the antioxidants that control their elimination.[1] Endogenous ROS are mainly produced in the mitochondria as byproducts of oxygen metabolism.[2,3] Moreover, ROS are also generated in response to exogenous environmental factors, including ultraviolet (UV) and ionizing radiations (gamma-ray/x-ray), some pollutants and chemicals, heavy metals, as well as xenobiotics.[4] At physiological levels, ROS operate as second messengers in intracellular Ca^{2+} signaling pathways to govern cell proliferation, differentiation, and apoptosis.[5,6] However, sustained elevation of free radicals causes damage to cellular DNA, lipids, and proteins in addition to initiating ROS signaling cascades, which in turn amplify the cellular oxidative stress.[7] Besides, an iron-dependent increase in ROS levels induces p53-dependent cell death,[8,9] autophagy activation, induction of necrosis, and ferroptosis, causing lipid peroxidation-mediated cell death.[10]

Oxidative DNA damage is considered a significant mutagenic and carcinogenic factor by promoting cancer progression through genome instability and chromosomal abnormalities with amplified oncogene activation. In addition, it affects cancer cell metabolism and causes the loss of function in tumor

suppressor genes, leading to DNA damage and altered physiological transcription.[1] Notably, ROS modifies the DNA through guanine to thymine G→T transversions,[11,12] recognized as the most common mutations in the p53 tumor suppressor gene.[13–15] Moreover, tandem CCTT substitution was also noted in DNA exposed to free radicals.[16] Cancer progression and survival are improved by ROS-induced phosphorylation of Jun N-terminal kinase (JNK), enhanced expression of cyclin D1, and mitogen-activated Protein Kinase (MAPK) activation. In addition, ROS regulates cellular proliferation by activating the extracellular-regulated kinase 1/2 (ERK1/2) and ligand-independent receptor tyrosine kinase (RTK). They enhance angiogenesis *via* angiopoietin and vascular endothelial growth factor (VEGF) and facilitate tumor invasion and metastasis *via* the release of metalloproteinase (MMP) into the extracellular matrix.[17] Chronic oxidative stress deactivates p53, phosphatase and tensin homolog (*PTEN*) tumor suppressor genes and induces oncogenes expression, including protein kinase B (*AKT*), *ERK*, and *c-MYC* inhibiting apoptosis and promoting cell proliferation, transformation, and metastasis.[3] It also impacts cancer cell metabolic reprogramming affecting glycolysis, oxidative phosphorylation, and fatty acid metabolism, to support tumor growth and survival.[18,19]

Cellular Antioxidant Systems

Endogenous antioxidant systems include enzymatic antioxidants such as superoxide dismutase (SOD) that decomposes superoxide ion (O_2^-),[20] catalase (CAT) that neutralizes hydrogen peroxide (H_2O_2),[21] glutathione peroxidase (GPx) which utilizes glutathione (GSH) to convert H_2O_2 or organic hydroperoxides to water or corresponding alcohols, respectively.[22] In addition, the thioredoxin (Trx) system is made up of NADPH, thioredoxin reductase (TrxR), and Trx, which operate on DNA and protein mending by inhibiting ribonucleotide reductase and methionine sulfoxide reductase.[23] Other endogenous antioxidants belong to the hydrophilic and lipophilic radical antioxidants. Besides, phenolics, flavonoids, carotenoids, vitamins A, C, and E, and minerals are classified as exogenous nonenzymatic antioxidants usually derived from diets.[24]

Increased ROS stimulate the nuclear factor erythroid 2-related factor 2/ Kelch-like ECH-associated protein 1 (NRF2/KEAP1) pathway, which controls an intracellular antioxidant defense by regulating downstream target genes at their antioxidant response elements (ARE) found in the gene promoters of detoxifying enzymes.[25] NRF2 regulates the expression of glutathione-S-transferases (GST), NAD(P)H quinone dehydrogenase 1 (NQO1), gamma-glutamylcysteine synthase (γ-GCS), ferritin, and heme oxygenase-1 (HO-1), SOD and catalase along with other cytoprotective processes.[26,27]

NRF2: A Double-Barreled Aspect

NRF2 Overview: Architecture, Regulation, and Downstream Targets

NRF2, a cap'n'collar (CNC)-basic region-leucine zipper (bZIP) transcription factor encoded by the *NFE2L2* gene, is a soluble protein primarily localized in the cytoplasm, highly conserved across species, and a major regulator of the cellular antioxidant response.[28,29] Its structure comprises 7 domains, including a bZIP DNA binding domain at the C terminus and 6 highly conserved NRF2-ECH homologies (Neh) domains.[28,29] The bZIP domain, located in the Neh1 domain, mediates NRF2 heterodimerization with small musculoaponeurotic fibrosarcoma proteins (sMafs) in the nucleus.[30] The Neh2 domain, the main regulatory domain of NRF2 located in the N-terminus, contains 7 lysine residues for ubiquitination, and DLG (Asp-Leu-Gly) and ETGE (Glu-Thr-Gly-Glu) motifs that bind to homologous locations on the KEAP1.[31,32] Thereby, the Neh2 domain assists NRF2 in attaching to and regulating its inhibitory cytoplasmic chaperone molecule Keap1.[33] Besides, the C-terminal Neh3 domain is needed to maintain protein stability and transcriptional activation,[34] while Neh4 and Neh5 engage with the CREB binding protein (CBP) to act as transactivation domains.[33] Although the Neh2 domain is required for NRF2 turnover in homeostatic cells, the redox-insensitive serine-rich Neh6 domain, a newly recognized domain, regulates NRF2 ubiquitination and further degradation in oxidatively stressed cells.[35,36] Similarly, the other recently discovered Neh7 domain of NRF2 interacts with retinoic X receptor alpha (RXR), a regulator of NRF2, to reduce NRF2's cytoprotective capacity and sensitizing non-small cell lung cancer cells to therapeutic toxicity.[37] However, further investigations are required to illustrate the role of these 2 newly discovered domains in the context of oxidative stress.

The KEAP1 repressor protein tightly regulates the NRF2 transcription factor.[38] KEAP1, a substrate adaptor protein for the Cul3-Rbx1 E3 ubiquitin ligase complex, primarily localizes in the cytoplasm[39] and drives NRF2 proteasome degradation.[35,40] In response to cellular stress, such as the presence of ROS, disulfide bonds may form on KEAP1 cysteine residues (Cys226, Cys613, Cys622, and Cys624).[41] In addition, when electrophiles are present, KEAP1's cysteine residues bind covalently with these electrophilic compounds through thiol-alkylation.[41] Moreover, KEAP1 has a Zn^{2+} sensor consisting of a group of amino acids, including His-225, Cys-226, and Cys-613, capable of detecting free Zn^{2+} released by damaged proteins. The binding of Zn^{2+} to KEAP1 leads to its structural alteration, disrupting its association with the cullin-3 (Cul3)-RING ubiquitin ligase (CRL) adaptor/scaffold protein.[42] All the above-described modifications affect the KEAP1-based E3 ubiquitin ligase complex and, therefore, prevent the proper alignment and interaction with NRF2. As a consequence, the resulting conformational shift in KEAP1 induces the detachment of the DLG motif from the KEAP1-NRF2 complex, resulting in the inhibition of NRF2 ubiquitination.[31,43] NRF2 is then released, phosphorylated at the Neh2 domain by protein kinase C (PKC)[44] and translocated to the nucleus, where it heterodimerizes sMAFs and binds to antioxidant ARE domains,[32,43] causing transcription of NRF2 targets cytoprotective genes.[45] Once the redox equilibrium is restored, NRF2 is released from the ARE sequence. Then, KEAP1, which acts as an adaptor for Cul3-based E3 ligase, transports NRF2 to the cytoplasmic Cul3-E3 ubiquitin ligase machinery to add Lys-48 linked poly-Ub chain, marking it for 26S proteasome degradation.[46,47] Thereby, a basal level of NRF2 is retained, and the NRF2/KEAP1 signaling pathway is deactivated.[29]

Other regulatory mechanisms of NRF2 activity and expression have been described. On the transcriptional level, the *NFE2L2* gene could be activated by polycyclic aromatic hydrocarbons.[48,49] In addition, NRF2 is activated in response to oncogene stimulation and may be mediated *via KRAS* and *BRAF* induction of JUN and MYC transcription factors.[50] Moreover, transcription factors such as Jun dimerization protein (JDP2), JUN, CREB binding protein (CBP), Brahma-related gene 1 (BRG1), and p21 induce NRF2 activation. In contrast, Fos proto-oncogene, AP-1 transcription factor subunit (cFOS), p53, p65, Fos-related antigen 1 (FRA1), BTB and CNC homology 1 transcription factor (BACH1), CCAAT/enhancer-binding protein (C/EB), activating transcription factor 1 (ATF1), activating transcription factor 3 (ATF3), short-form estrogen-related receptor (SFERR), peroxisome proliferator-activated receptor α (PPAR-α), and retinoic acid receptor (RAR) have been shown to inhibit NRF2 transcription.[51,52] At the post-transcriptional level, microRNAs (miRNAs), endogenous short noncoding RNAs, can suppress gene expression by interacting with target transcript translation or stability. Among miRNAs, miR-507, miR-634, miR-450a, and miR-129-5p inhibit the translation process of NRF2.[53] In addition, it has been documented that hypermethylation of CpG sites in the KEAP1 promoter region occurs in various cancer types,[54–56] and such epigenetic changes result in constitutive activation of the NRF2 pathway. Other NRF2 regulation mechanisms involve the p62-mediated dysfunction of autophagy,[57] electrophilic-mediated inhibition of KEAP1,[56] and hormone-mediated NRF2 activation by gonadotrophins and estrogen, which inhibits KEAP1 *via* oxidation of its multiple cysteine residues.[58]

NRF2 is responsible for regulating the transcription of more than 200 genes that play a role in various cellular processes such as cytoprotection, metabolism, and gene transcription.[59] It activates the transcription of genes involved in the detoxification of reactive species and xenobiotics, such as phase I, II, and III enzymes, including Aldo-keto reductase (*AKR*), NADPH quinine oxidoreductase 1 (*NQO-1*), superoxide dismutase (*SOD*), catalase, multidrug resistance-associated protein (*MRP*), and ATP-binding cassette transporters (*ABC*).[60] In addition, it plays a crucial role in the cellular antioxidant system based on the glutathione molecule. NRF2/KEAP signaling is responsible for regulating the expression of various elements such as the cystine-glutamate antiporter xCT, glutamate cysteine ligase (*GCL*), glutathione peroxidase (*GPX*), and reductase (*GSR*), which are necessary for cysteine import and catalysis of the rate-limiting step in GSH manufacture and ROS detoxification.[61,62] Similar to this, NRF2 upregulates thioredoxin-1 (*TXN1*),[63] thioredoxin reductase 1 (*TRXR1*),[64] peroxiredoxins (*PRXS*),[65] and sulfiredoxin-1 (*SRXN1*),[66] allowing the reduction of oxidized protein thiols and the elimination of peroxides. In addition, NRF2 regulates the transcription of genes involved in metabolism, especially carbohydrate metabolism, and NADPH generation (ie, *G6PD*, glucose-6-phosphate dehydrogenase; *HDK1*, hexokinase domain containing 1; *IDH1*: NADP-dependent isocitrate dehydrogenase), lipid metabolism (ie, *ACOT7*, acetyl-CoA thioesterase 7; *ACOX1*, acetyl-CoA oxidase 1), and heme and iron metabolism (ie *BLVR*, biliverdin reductase; *FTL1*, ferritin, light polypeptide; *HMOX1*, heme oxygenase 1).[59] Therefore, NRF2 plays a crucial role in regulating intracellular redox homeostasis.

NRF2's Dual Role in Cancer

NRF2 tumor suppressive activities.—NRF2 exerts an anti-tumor effect, mainly through sustaining cellular redox homeostasis, regulating cell growth, and exerting anti-inflammatory activities.[29] For instance, the NRF2 signaling pathway detoxifies ROS and RNS by upregulating the expression of numerous phase II drug-metabolizing enzymes, therefore decreasing the oxidative stress that is strongly associated with cancer development.[67] Several in vivo studies have emphasized the role of NRF2 in cancer protection using NRF2-deficient mice that expressed reduced levels of phase II enzymes. In addition, NRF2-knockout (KO) mice were found to be more sensitive to chemical toxicants and carcinogens and resistant to the protective effects of chemopreventive drugs, potent NRF2 inducers. These compounds exert NRF2-dependent adaptive responses against carcinogenic insults. They are either natural molecules such as curcumin and resveratrol or synthetic chemicals such as oltipraz, 2-indol-3-yl-methylenequinuclidin-3-ols, and the synthetic triterpenoid 2-cyano-3,12-dioxooleana-1,9, among others.[68] Besides limiting original tumor development, another study has shown that NRF2 protects against cancer metastasis by maintaining the redox equilibrium in the hematopoietic and immune systems.[69] Paradoxically, NRF2 deficiency renders cancer cells more prone to oxidative cell death but more resistant to chemopreventive compounds. Therefore, targeting the NRF2 pathway presents a critical strategy for developing effective chemopreventive medications.

In terms of inflammation, in NRF2-KO animals, cyclooxygenase 2 (COX2), inducible nitric oxide synthase (iNOS), and tumor necrosis factor (TNF) levels are considerably greater compared to control mice, showing that NRF2 inhibits pro-inflammatory mediators.[70] Besides, NRF2-dependent activation of NQO1 reduces TNF and IL-1 production caused by lipopolysaccharide (LPS), impairing the inflammatory response[71] and subsequent inflammation-induced carcinogenesis. Although ROS elimination is the molecular basis of NRF2-mediated anti-inflammation, NRF2 may also function as an anti-inflammatory mediator in the absence of ROS. This is accomplished by regulating genes encoding for MARCO (macrophage receptor with collagenous structure) and CD36 receptors specific for macrophages, not involved in the oxidative response.[72] In addition, NRF2 protects against H_2O_2-induced damage *via* the p38/MAPK pathway.[73,74] As well, NRF2 inhibits the NF-κB pathway by stabilizing the NF-κB inhibitor (IKK)-α and repressing the degradation of (IKK)-β.[75] On the contrary, the NF-κB p65 subunit competes with NRF2 for the CH1-KIX domain of the transcriptional coactivator CBP, resulting in the inactivation of the NRF2 pathway.[76]

NRF2 oncogenic activities.—Various factors contribute to the constitutive activation of NRF2 in cancer cells, including somatic mutations in *KEAP1* and *NFE2L2*, exon skipping in *NFE2L2*, methylation of the *KEAP1* promoter, accumulation of p62/Sequestosome-1 (SQSTM1), and

mutation in fumarate hydratase. Constitutive NRF2 activation promotes cancer growth, through metabolic alterations, stimulation of proliferation and inhibition of apoptosis, promotion of angiogenesis, invasion, and metastasis in addition to promoting treatment resistance in various cancer types.[29,77] On a molecular level, NRF2 overexpression promotes the transcription of the oncogenes *MYC*, *KRAS*, and *BRAF*.[50] Conversely, the oncogenic activation of NRF2 occurs by inhibiting PTEN/glycogen synthase kinase 3 (GSK-3)/beta-transducin repeat-containing E3 ubiquitin-protein ligase (β-TrCP) activity.[78] Moreover, NRF2 allows for metabolic reprogramming to enhance cancer cell proliferation by upregulating the expression of glycolytic enzymes such as glucose-6-phosphate dehydrogenase [G6PD], phosphogluconate dehydrogenase [PGD], transketolase [TKT], and transaldolase 1 [TALDO1][79]; regulating genes implicated in fatty acid and lipid metabolism,[80] proliferation-associated genes[81] and inhibitory cell-cycle regulators.[82] Interestingly, NRF2 activation participates, through glucose-regulated protein 78 (GRP78)/ phosphorylated protein kinase RNA-like ER kinase (p-PERK)/NRF2 signaling pathway, to glycolytic gene transcription and simultaneous inhibition of the tricarboxylic acid cycle (TCA), which promotes the Warburg effect.[83] Another NRF2-mediated oncogenic activity is the promotion of angiogenesis, mainly by activating heme oxygenase-1 (HO-1),[84] which in turn regulates VEGF to promote angiogenesis.[85]

Besides, regarding cancer cell apoptosis, siRNA-mediated knockdown of NRF2 results in the down-regulation of HO-1-mediated expression and the sensitization to TNF-induced cell death in a model of acute myeloid leukemia. This suggests that NRF2 inhibits cancer cell apoptosis by regulating the levels of the antioxidant enzyme HO-1.[86] Also, NRF2 upregulates the expression of anti-apoptotic protein B-cell lymphoma 2 (BCL-2) while it down-regulates the activity of proapoptotic BAX protein and caspases 3/7 to protect against etoposide/radiation-mediated cell apoptosis that leads to drug resistance.[87] In addition, NRF2 suppresses the activation of proapoptotic c-Jun N-terminal kinases (JNKs)[88] and induces selective autophagy of KEAP1.[89,90] Autophagy is a crucial process for cancer cell growth; however, overexpressed NRF2 renders autophagy-dependent cancer cells to overcome the loss of autophagy and allows them to maintain protein homeostasis.[91]

Regarding cancer stemness, lower levels of endogenous ROS due to the increased antioxidant capacity mediated by the higher NRF2 expression are reported in cancer stem cells (CSCs) compared to non-CSCs, allowing for the enrichment of their stemness phenotype.[92–95] This results in reduced mitochondrial-derived ROS and subsequently maintains CSC stemness-associated properties,[83] such as the ability to initiate an epithelial-to-mesenchymal transition.[96] Similarly, persistent NRF2 activation improves the ability of CSC to self-renew, primarily by maintaining cell quiescence and lowering intracellular ROS.[97,98] In a broader sense, mesenchymal stem cells (MSCs), known to be multipotent stem cells, are present in the tumor niche to encourage cancer cells' ability to spread by promoting their motility and invasiveness.[99,100] NRF2 is needed to maintain MSCs' stemness and prevent their apoptosis under oxidative stress.[101]

Moreover, because NRF2 significantly benefits cancer cells, these cells frequently develop NRF2 addiction.[102,103] Enhanced nuclear accumulation of NRF2 is associated with increased cellular proliferative signals. For instance, phosphoinositide 3-kinase (PI3K)-AKT activation in combination with KEAP1 deficiency in the mouse liver results in a massive accumulation of NRF2 and NRF2-dependent proliferation of hepatocytes and cholangiocytes.[104,105] However, because simple NRF2 stability and accumulation are insufficient to transform NRF2 from cellular defender to cancer driver, the occurrence of additional oncogenic mutations is required.[106–108] *KEAP1* mutations paired with activating mutations of *KRAS/HRAS* and *TP53* loss of function are needed to establish NRF2-addicted cancer models.[109–111] Furthermore, NRF2-dependent malignancies with somatic *KEAP1* or *NFE2L2* mutations differ depending on the specific tissue and species. For example, the mutations of *KRAS/KEAP1* in the human lung tissue induce tumors with aggressive proliferation,[109] whereas *KRAS/KEAP1* mutations in the mice pancreas cause fibrosis rather than malignancy.[112] As a result, tissue-specific variables are another factor likely to influence the requirements for developing NRF2-dependent cancer.

In therapy resistance, NRF2-regulated drug efflux transporters are significant predictors of therapy resistance in many tumors. Multidrug resistance protein 1 (MDR1), multidrug resistance-associated protein 1-5 (MRP1-5), and breast cancer resistance protein (BCRP) are overexpressed as a result of abnormal NRF2 activation leading to widespread chemoresistance.[113–117]

NRF2 Biology in the Brain

Brain Cellular Composition and NRF2 Expression

Quantifying the cellular makeup of the human brain is highly challenging because of the brain's huge size, cell composition, and limited access to human postmortem brain samples.[118] In addition to approximately 100 billion neurons, glial cells (astrocytes, oligodendrocytes, and microglia) are present with a median of 0.85 glia-neuron ratio.[119,120] In the brain, neutralization of ROS or electrophilic xenobiotics is usually mediated by the glutathione system, thioredoxin/peroxiredoxin system, superoxide dismutases, and catalase.[121,122] It is interesting to note that the *NFE2L2* gene displays varying expression levels across different brain regions. It exhibits the highest expression primarily in the medulla oblongata, regulating hub of homeostatic functions of the nervous system, and basal ganglia, responsible for motor control, executive functions and emotions.[123] On the other hand, the hippocampus shows the lowest level of *NFE2L2* expression (Figure 1A). Similarly, the expression of the *NFE2L2* gene varies among different types of brain cells. It is most highly expressed in oligodendrocytes, while neurons exhibit the lowest level of expression (Figure 1B). Being a master regulator of antioxidant defenses, NRF2 exhibits distinct activities in the brain in addition to its cytoprotective effects.[124,125] Herein, we will discuss the expression of NRF2 regarding brain biology and the function of different brain cells.

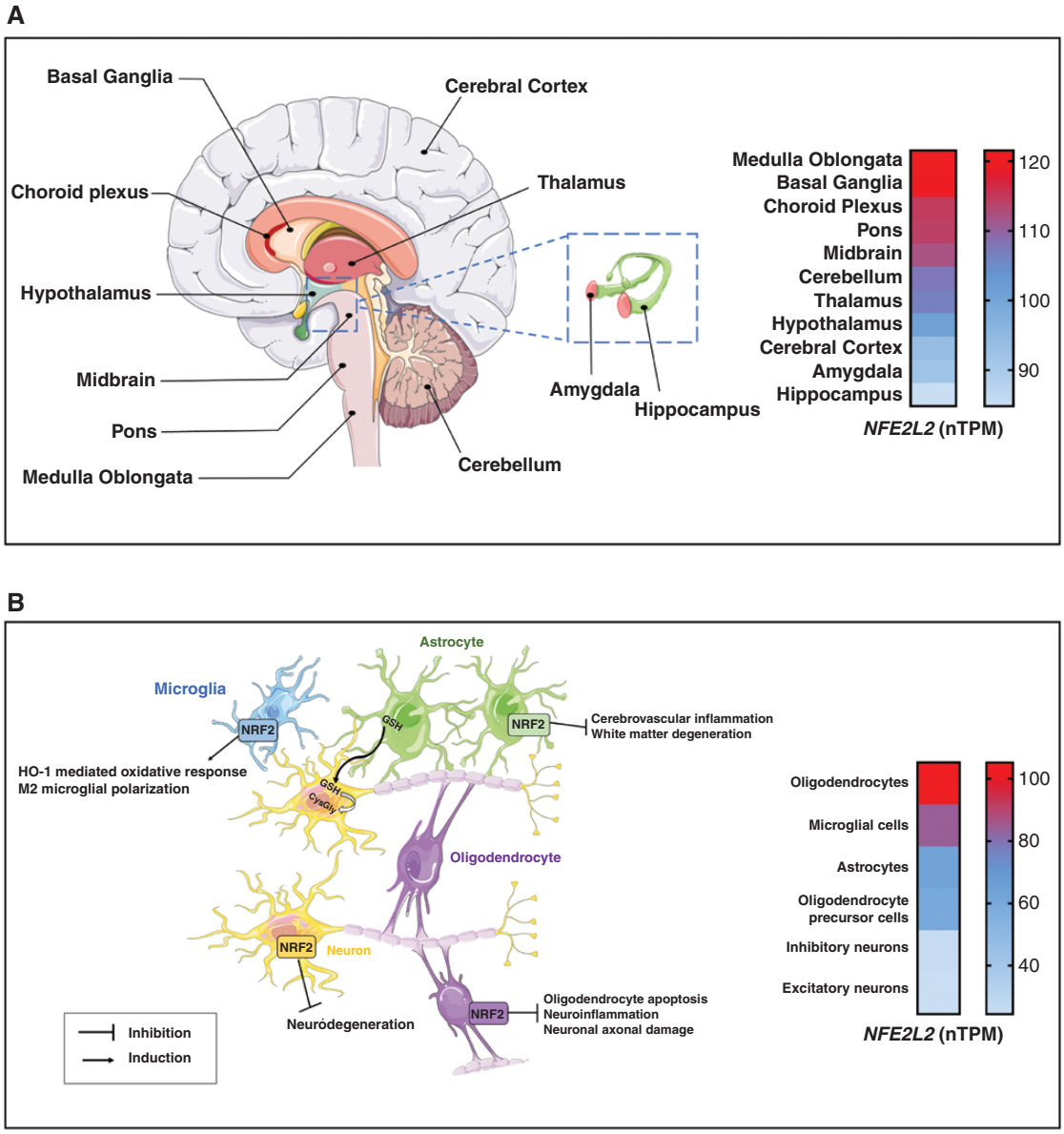

Figure 1. Overview of *NFE2L2* gene expression and the role of NRF2 in brain physiology. (A) Human brain regions are visually represented on the left side, while the accompanying heat map on the right side displays *NFE2L2* gene expression across the various human brain regions. The data were sourced from the human protein atlas (HPA) dataset, available at https://www.proteinatlas.org/ from version 23.0, accessed from the following URL: https://www.proteinatlas.org/ENSG00000116044-NFE2L2/brain. (B) Graphical summary of NRF2's role in brain physiology among the different brain cells on the left side, while the accompanying heat map on the right side displays *NFE2L2* gene expression across the different brain cells. The data were sourced from the RNA single cell type data, available at https://www.proteinatlas.org/ from version 23.0, accessed from the following URL: https://www.proteinatlas.org/about/download. The figure was partly generated using Servier Medical Art, provided by Servier, licensed under a Creative Commons Attribution 3.0 unported license. nTPM, normalized transcript per million; HO-1, heme oxygenase 1; CysGly, cysteinylglycine dipeptide; GSH, glutathione.

In the adult brain, *astrocytes* are the most abundant glial cell type.[126] Morphologically, protoplasmic astrocytes possess small irregular branching in a globoid distribution and are located in gray matter tissue, whereas fibrous astrocytes have numerous uniform cylindrical fibers and are broadly distributed across white matter tissue.[127,128] In terms of function, astrocytes facilitate synaptic transmission and information processing, govern the migration of growing axons and neurons, and connect with blood vessels.[129,130] In addition, the proportion of astrocytes to neurons differs greatly between species and correlates with cognitive ability.[131]

Neurons are fundamental units of the brain and electrically excitable cells responsible for information processing and performing various functions within the brain.[132] They are highly susceptible to oxidative stress mainly due to

their high reliance on oxidative phosphorylation for energy and enrichment in metal ions (catalyst for oxidative species formation), possess membranes rich in polyunsaturated fatty acids, and exhibit low levels of antioxidants.[133] The NRF2-ARE pathway in neurons is noticeably weak both in inhibitory and excitatory neurons (Figure 1B). Stimulation with tert-butylhydroquinone (tBHQ), an NRF2 activator, successfully induces the expression of NRF2 target genes in astrocytes, while no such induction is observed in cerebellar granule neurons.[134] The lower neuronal NRF2-ARE pathway activation is explained by the fact that basal NRF2 expression is lower in neurons, along with a greater Cul3-dependent NRF2 degradation capability than astrocytes.[134–136] Also, hypo-expression of NRF2 in neurons results from epigenetic repression caused by NRF2 promoter hypo-acetylation compared to astrocytes.[134] Furthermore, maturing neurons require fewer antioxidant defenses to facilitate redox signaling involved in their development.[137,138] Indeed, ectopic expression of NRF2 in neurons exerts a protective role against oxidative insults[66]; however, it retards structural and electrophysiological maturation[134] and suppresses the activity of c-Jun N-terminal kinase (JNK) and Wnt signaling pathways required for neuronal development.[139–142] On the other hand, astrocytes usually mature even when they express high amounts of NRF2, indicating that the signaling mechanisms involved in their maturation are less sensitive to the redox state.[121,143] On the contrary, neurons that present repressed NRF2 expression for their maturation require astrocytic assistance to avoid oxidative damage.[134] Nearby astrocytes provide cysteine and/or glutathione to neurons, as well as other metabolites, to support neurons' activity.[144]

Oligodendrocytes, another type of glial cell, provide structural support and a myelin coating around the neuronal axon to allow for a fast impulse transmission.[145] Evidence suggests that ROS drives the oligodendrocytes differentiation from precursors cells,[146] but oxidative stress is implied in demyelinating diseases.[147,148] Similarly to neurons, oligodendrocytes receive antioxidant assistance from astrocytes.[149] Conversely, oxidative stress in oligodendrocytes activates an endoplasmic reticulum stress response in an NRF2-dependent manner in response to chemical hypoxia.[150] In the same context, oligodendrocyte apoptosis is more pronounced in addition to neuroinflammation and axonal damage in cuprizone-fed NRF2-deficient mice than in wild-type controls. Also, NRF2-deficient mice exhibited increased vulnerability to cuprizone-induced damage within the commissure anterior white matter tract, a region typically less affected by cuprizone in wild-type animals.[151] However, NRF2 activation in oligodendrocytes in the context of other neurological disorders has yet to be thoroughly investigated.[152]

Microglial cells are brain-resident immune cells[153] found in 5% of the cerebral cortex and up to 12% of the substantia nigra.[154] These cells are responsible for neuronal proliferation and differentiation, as well as removing debris and rebuilding synapses.[155] Microglia exhibit more NRF2 transcripts and ARE promotor activity than neurons in the brain,[156] indicating higher NRF2 expression than neurons. NRF2, which is actively produced by microglia in response to oxidative stress, promotes the activation of the M2-like pro-inflammatory microglial phenotype.[157] However, its absence increases microgliosis, primarily characterized by the activation and proliferation of microglial cells. This absence also promotes the polarization of microglia towards an M1-like anti-inflammatory phenotype, which contributes to neuronal demise.[158] Knowing that glial activation associated with various neurodegenerative disorders,[159] NRF2-mediated modulation of microglial dynamics regulates neurodegeneration.[160] In contrast, microglia activation in reaction to atrazine-induced neuroinflammation boosts the production of inflammatory factors and inhibits the KEAP1/NRF2-ARE signaling cascade, resulting in increased dopaminergic neuron cell death and neurotoxicity.[161] As a result, it appears prudent to conduct further research into the KEAP1/NRF2-ARE signaling pathway in microglia, as it may be a therapeutic target for NRF2 activation in neurodegenerative diseases. It is worth noting that astrocytes induce microglial NRF2 activation and the subsequent microglial HO-1 expression to decrease microglial intracellular ROS levels together with excessive microglial brain inflammation.[162]

NRF2 in Neurological Diseases

Regarding human health, age-related NRF2 system impairment is a significant risk factor for almost all oxidative stress-related neurological diseases. Neurons are non-regenerative and postmitotic; therefore, significant oxidative damage should be avoided or reversed. Neuronal oxidative damage rises with age and is linked to neurodegenerative illnesses.[151,152] Reduced NRF2 activity is related to both the development of chronic diseases like Parkinson's disease (PD), Alzheimer's disease (AD), and amyotrophic lateral sclerosis (ALS), as well as increased susceptibility to acute insults like oxidative stress and chronic inflammation in the brain.[163] In the hippocampus, where neurodegeneration in AD begins, astrocytes from AD patients' brains have lower levels of NRF2.[163] NRF2 expression is decreased in the motor neurons of the spinal cord and cortex, as shown in the postmortem brains of ALS patients.[164] Supporting the evidence that the NRF2 system is dysfunctional in PD, olfactory neurosphere-derived cells from patients with sporadic PD express low GSH levels, which an NRF2 inducer agent could restore.[165] Hence, age-related reduction in NRF2 contributes to the development of neurodegenerative diseases and other age-related pathologies. Mainly, reduced neural stem cell (NSC) counts due to aging,[166] along with NSCs' clonogenic, proliferative, and differentiating capacities, are associated with NRF2 deficiency.[167] However, the transplantation of NSCs with high expression content of NRF2 lessens age-related declines in dentate gyrus stem cell regeneration.[168] Besides, ROS plays a role in regulating the fate of NSCs by inhibiting self-renewal and promoting differentiation through NRF2-mediated signaling.[169]

Moreover, Dang et al. discussed NRF2 expression and its role in oxidative stress-related pathogenesis under acute ischemic stroke-like conditions.[170] Their results show that after the initiation of the stroke, NRF2 was not expressed in the core ischemic zone. However, its expression was elevated in the ischemic penumbra in both glial and neuronal cells. This suggests that NRF2 activation in

the penumbra results from enormous ROS generation owing to reoxygenation, whereas NRF2 activation in the undamaged cortical areas represents a preadaptation to oxidative stress. Surprisingly, compared to other cell types in the unaffected contralateral area, NRF2 expression was elevated in neurons. This phenomenon could also be attributed to the possible ROS independent-NRF2 activation in response to the growth factors, cyto- and chemokines, neurochemical mediators, and cross-hemispheral neural connections. Hence, NRF2 represents a therapeutic target that possesses a cytoprotective role in the brain after the initiation of injury.[170] The activation of endogenous NRF2 has been reported in oligodendrocytes in multiple sclerosis (MS)[171]; however, it is expressed in actively demyelinating lesions but not in late-stage active lesions.[172] Moreover, in MS, reduced NRF2 expression is reported in oligodendrocytes compared to other central nervous systems (CNS) cell types, suggesting an impaired oxidative stress response.[173]

NRF2 in Brain Metabolic and Mitochondrial Functions

Regarding mitochondrial bioenergetics, it has been shown that KEAP1-knockdown (KD) increases the glucose uptake in neurons and astrocytes compared to NRF2-KO and WT cells. Activation of NRF2 increases cytoplasmic NADPH and NADH levels in neurons and astrocytes; however, it favors energy production over antioxidant defense when glucose availability is limited in astrocytes.[174]

In neurodegenerative diseases such as ALS, mutation of SOD1 produces motor neuron injury associated with NRF2 dysregulation coupled with reduced pentose phosphate pathway (PPP) activity and decreased generation of NADPH.[175] In PD, acute and chronic astrocyte exposure to dopamine enhanced PPP activity *via* the KEAP1/NRF2 system.[176] NRF2 eliminates oxidative stress in dopaminergic neurons by supplying NADPH to support the activity of NQO1, which is another target of NRF2.[177,178] Moreover, NRF2-KO mice were rendered more sensitive to neurotoxicity caused by 1-methyl-4-phenyl-1,2,3,6-tetrahydropyridine, complex I inhibitor, in animal models of Parkinson's disease.[179]

Moreover, knocking out NRF2 negatively affects the mitochondrial NADH redox index, which is the ratio between NADH consumption by complex I and its production in the TCA cycle. Also, a slower NADH and FADH2 generation is obtained after the inhibition of complex IV in NRF2 mutant neurons.[180] NRF2 is also crucial to maintain mitochondrial integrity, particularly the mitochondria isolated from the brain of rats that were administered a single dose of isothiocyanate sulforaphane, an NRF2 activator, were resistant to the opening of the mitochondrial permeability transition pore.[181,182]

Regarding mitochondrial biogenesis, treatment with the α7 acetylcholine nicotinic receptor (nAChR) agonist PNU282987 increases the mitochondrial mass and oxygen consumption in primary glial cultures without increasing oxidative stress. However, these results were abolished in the absence of NRF2. This result indicates that NRF2, through the stimulation of HO-1 or binding with peroxisome proliferator-activated receptor gamma coactivator-1 alpha (PCG-1α), modulates glial mitochondrial mass.[183]

However, it is important to highlight that NRF2 is an essential player in maintaining mitochondrial homeostasis and structural integrity via various mechanisms that are not exclusive to the brain but extend to various other tissues. Consequently, since oxidative stress, inflammation, and mitochondrial integrity contribute to the development of diseases, the pharmaceutical activation of NRF2 might be a key for both disease prevention and treatment.[184]

NRF2 in Brain Cancer

Given the high oxygen consumption of the brain compared to other organs, the implication of oxidative stress in the development of brain tumors is of particular interest.[185] Primary brain tumors (PBT) grow from brain tissue and its surroundings and can be glial or non-glial. In this section, we will discuss the modulation of NRF2 in various types of PBTs and its potential therapeutic applications. We will also focus on glioblastoma, the most common type of glioma in adults, which has a very poor prognosis.

NRF2 in Pediatric Brain Tumors

After hematologic malignancies, CNS tumors are the second most common neoplasm in children.[186] Unfortunately, despite the extensive studies on the dual role of NRF2 in cancer, little is known regarding NRF2's function in most pediatric CNS malignancies. Among pediatric brain tumors, medulloblastomas (MB) are the most prevalent CNS embryonal tumor. MB, classified as a grade 4 cancer, comprises 4 subgroups: WNT, sonic hedgehog (SHH), Group 3, and Group 4; each is associated with different genetic alterations, age at onset, and prognosis.[187] When MB cases are compared to peritumoral control brain tissues, higher expression of NRF2 and HO-1 suggests that the NRF2/HO-1 pathway contributes to the progression of MB and hence might be a therapeutic target for the disease.[188] Others have shown that nifurtimox, an antiprotozoal compound, and tetrathiomolybdate, a copper chelator, act synergistically to induce oxidative stress and subsequent upregulation of NRF2 target genes, including HO-1, GCLM, solute carrier family 7 member 11 (SLC7A11), and SRXN1 in D2 and DAOY MB cell lines.[189] It is worth noting that although the drug combination effectively lowered medulloblastoma cell viability and triggered cellular death,[189] the rise in NRF2, which might exert a protumoral role, should be carefully assessed.

Peroxiredoxins (Prxs) are linked to cell apoptosis,[190] differentiation,[191] and resistance to radiation or chemotherapy.[192,193] In ependymomas, another type of pediatric brain cancer where a tumor arises from ependymal cells,[194] all Prxs (except Prx IV) are upregulated. However, Prx I expression is substantially related to the upregulated cytoplasmic and nuclear NRF2 expression, suggesting that NRF2 plays a role in Prx I production in ependymomas.[195] Additionally, there are no functional studies of NRF2 on

pilocytic astrocytoma, another frequent pediatric CNS cancer. It can likely play a minor role in the development of this tumor, given its low expression compared to higher WHO-grade gliomas. Therefore, the current evidence on the role of the NRF2 pathway in pediatric CNS tumors is limited, necessitating further investigation to enhance our understanding of its significance.

NRF2 in Adult Glioma

In 2021, the World Health Organization (WHO) published a new edition of the classification of tumors of the central nervous system, incorporating molecular and histological pathogenesis, to improve the diagnosis and determination of optimal treatment.[196] This classification separates pediatric and adult gliomas. Gliomas are the most prevalent type of adult brain tumor, comprising approximately 78% of malignant brain tumors. Three types of adult gliomas: oligodendrocytomas and astrocytomas which are isocitrate dehydrogenase (*IDH*) mutated and glioblastomas which are *IDH* wild type were classified.[196] Frequently, brain tumors are also classified according to the WHO grade from grade 1 to grade 4, with grade 1 being the least aggressive and grade 4 being the most aggressive.[197]

Overall, in gliomas, the NRF2-KEAP1 pathway acts as a switch for malignancy, mainly through amplifying glutamate secretion and xCT augmentation.[198] Similarly, NRF2 overexpression or KEAP1 knockdown in glioma cells promotes proliferation and oncogenic transformation.[198] However, some discrepancies can be noted according to the type and grade of glioma, particularly regarding prognosis. Indeed, in contrast to other types of cancer, there are relatively few studies that have explored the relationship between NRF2 expression and brain cancer prognosis. NRF2 overexpression is shown to be positively correlated with WHO grades in gliomas.[199] *In silico* analysis, using the Rembrandt glioma dataset, shows that the upregulated *NFE2L2* RNA expression levels are associated with the poor prognosis in grade 2-4 gliomas.[200]

IDH-mutant glioma: oligodendrocytomas and astrocytomas.—It is well-established that *IDH*-mutated tumors generally have a more favorable disease outcome and give rise to low-grade gliomas.[201] Somatic mutation in *IDH1*, and less commonly in *IDH2*, are considered as early events. Next, during glioma development, additional subclonal mutations are added leading to higher-grade *IDH*-mutant gliomas. For instance, oligodendrocytoma, arising from oligodendroglial precursors, is classified as grades 2 or 3 while astrocytoma, arising from astrocytic precursors, can be found as grades 2, 3, or 4.[196]

Examining the NRF2 pathway in the context of *IDH* mutations, in gliomas with mutated *IDH1/2*, the expression levels of NRF2 target genes, *NQO1* and *GCLM*, were notably elevated and were significantly linked to poorer patient survival, whereas the expression of NRF2 itself did not exhibit such an association.[202] However, in primary astrocytomas, an increase in both cytoplasmic and nuclear expression of NRF2, as well as nuclear DJI, a multifunctional protein involved in oxidative stress response, is associated with *IDH1* mutation.[200] These results suggest that the association between NRF2 expression and *IDH* mutation depends on the *IDH*-mutated glioma type but more studies are needed. Interestingly, it has been shown that IDH1-mutated cells develop a dependency on the NRF2 antioxidant pathways and, therefore, using NRF2 inhibitors, such as brusatol, suppresses cancer progression.[203]

Glioblastomas.—Glioblastoma (GB), classified as grade 4 *IDH1* wild-type glioma, is the most prevalent primary brain tumor with a median survival rate of 15 months[204–206] and a median age of detection of 65 years.[207] GB is detected in the forebrain almost exclusively but may develop in the brain stem, cerebellum, and spinal cord.[205,208] Despite the therapeutic options, such as surgery with maximal safe resection followed by concurrent radiotherapy and temozolomide (TMZ) and 6-monthly rounds of adjuvant TMZ, recurrent GB management remains a problem with limited treatment options.[209,210]

NRF2 oncogenic activity has been more studied in GB than in other glial tumors and has recently been reviewed.[211] Evidence shows that knocking down NRF2 attenuates tumor growth by inhibiting cell proliferation, increasing cell apoptosis, and suppressing angiogenesis.[113,212] Also, the NRF2 pathway is shown to be activated by a positive feedback loop involving p62/SQSTM1, a stress-inducible and multifunctional protein, whereas NRF2 and p62 enhance proliferation, invasion, and mesenchymal transition in GB.[213] Finally, NRF2 overexpression partly reversed the ERK and PI3K inhibitor-induced reduction of human GB cell viability,[214] suggesting that signaling cascades for NRF2 activation may offer new treatments for glioblastoma.

NRF2 Expression in GB Prognosis

It is now widely accepted that NRF2 expression is higher in GB than in normal brain tissue or other types of brain cancer. However, the relationship between NRF2 expression and GB patient survival is still controversial due to conflicting results in published studies, noting that most studies are *in silico* analyses using available databases.

On one side, studies have shown that high NRF2 expression is associated with lowered survival in GB patients. For example, Fan et al. have demonstrated that GB tissues exhibit a significant elevation in *NFE2L2* mRNA expression compared to normal brain tissue samples using the Oncomine database. Moreover, using the Rembrandt database, they showed that patients with *NFE2L2* expression upregulated by 2-folds or more had significantly poorer overall survival rates compared to those with lower NRF2 expression profiles.[198] Another example is using the SurvExpress tool and the data from 538 GB patients, higher expression of the *NFE2L2* gene and related genes were associated with higher risk for the patient.[215] In an interesting study, the TCGA GBM prognostic clinical data (520 cases) were stratified by the NRF2 activity status. The authors found no difference in the overall survival of patients with high NRF2 activity but the progression-free survival was strongly decreased.[213] However, contradictory studies can be highlighted. For example, *NFE2L2* expression was

not associated with overall survival in GB patients in the Rembrandt database, and an IHC analysis done on 213 GB patients further revealed that nuclear NRF2 expression was a predictor of better survival.[200] In another study based on a cohort of 52 GB patients, the expression of 2 NRF2 target genes, *NQO1* and *GCLM*, was not associated with progression-free or overall survival.[202] To compare with the existing literature, we analyzed another database, the GEPIA2 database,[216] and found that *NFE2L2* gene expression was elevated in GB tumors compared to normal tissue (Figure 2A). However, the variation in the overall survival or disease-free survival rates among GB patients with low or high *NFE2L2* gene expression did not achieve statistical significance (Figure 2B-C).

Moreover, GB is classified into subtypes: mesenchymal, classical, proneural, and G-CIMP.[217,218] The high invasiveness of the mesenchymal subtype is indicated by recurrence and worst survival rates compared to others.[213,219] The overexpression of *NFE2L2* has been reported in the mesenchymal subtype of GB tumors.[213] In our *in silico* analysis, we observed that *NFE2L2* gene expression is significantly elevated not only in the mesenchymal but also in the classical subtype of GB, which is not the case with the proneural subtype compared to normal tissue (Figure 2D).

In light of these findings, it is reasonable to conclude that the divergence observed in the context of NRF2 and GB patients' survival among the different database tools may be attributed to the limitations inherent in the database's methodology, sample size, and selection criteria used. It is important to clarify the link between NRF2 expression and GB prognosis by using cohort patient tissues associated with clinicobiological data. In addition, NRF2 activity is regulated by numerous post-transcriptional and post-translational modifications. Therefore, it is crucial to correlate patients' prognosis with NRF2 protein level expression and its sublocalization since nuclear localization is associated with its activity.

NRF2 in Glioblastoma Stem Cells

The tumorigenic potential of glioblastoma stem cells (GSCs) in GB owes to the progression and therapeutic resistance to chemotherapy and radiation.[220,221] GSCs constitute a small fraction of the tumor bulk. Yet, they possess high self-renewal capacity, allowing them to sustain tumor growth, neurosphere forming capacity, and therapeutic resistance.[221] In GB, under hypoxic conditions, increased necrosis favors the maintenance of GSCs responsible for the tumor's initiation, resistance, and recurrence.[222,223]

Despite the limited studies conducted on the role of NRF2 in GSC, NRF2 has been shown not only to maintain the self-renewal capacity of GSCs despite the anti-cancer treatment[224] but also to enhance neurosphere proliferation in NSCs.[225] Interestingly, differential NRF2 expression exists between glioma stem cells and non-stem-like cells. For instance, NRF2 is overexpressed in CD133 + GSCs compared to CD133- GB cells,[226] and downregulation of NRF2 improves GSC differentiation as it lowers the number of sphere-like colonies.[227] Also, knocking down NRF2 in GSCs using RNA interference technology resulted in decreased expression of pluripotency-associated transcription factors, increased expression of markers associated with astrocyte development, caused a significant reduction in S-phase cells, reduced expression of SRY-box transcription factor 2 (SOX2), B-cell-specific moloney murine leukemia virus integration site 1 (BMI-1), and Cyclin E proteins responsible for cell self-renewal.[228] Furthermore, the transcriptional coactivator with PDZ-binding motif (TAZ)-dependent growth, encoded by the gene *WWTR1*, is a crucial element of the Hippo signaling pathway, which regulates the development and stemness in multiple human cancers through the yes-associated protein (YAP) and transcriptional coactivator with PDZ-binding motif (TAZ) coactivators of the TEA domain (TEAD) transcription factors 1–4.[229] Interestingly, the upregulation of NRF2 induces the expression of TAZ, which acts as an effector of NRF2-induced tumorigenicity in GBs. TAZ ectopic expression also rescues neurosphere growth of NRF2-KD glioma stem cells and, along with NRF2 expression, accelerates GB tumor formation.[230]

Cluster of differentiation 90 (CD90), cluster of differentiation 15 (CD15), A2B5, aldehyde dehydrogenase 1 (ALDH1), nestin, and ATP-binding cassette (ABC) transporters are frequently recognized as markers of GSCs.[231–233] These markers help elucidate the tumorigenic process and serve as an effective diagnostic and therapeutic tool for GB. However, the precise mechanisms and functions of these putative markers have not yet been fully clarified. Therefore, identifying various biomarkers rather than just one marker and their correlation with NRF2 expression in the context of GB stem cell self-renewal capacity and maintenance may enable tailored targeting of GSC treatments and further tumor relapse.

NRF2 in GB Metabolism

The role of NRF2 in GB metabolism still needs to be fully elucidated, and a comprehensive understanding of its specific mechanisms and implications in GB metabolism necessitates further investigation. The NRF2-driven human telomerase reverse transcriptase (hTERT) loop mediates the NRF2-PPP regulation. Mainly, hTERT knockdown abrogated the NRF2 level, while overexpression of NRF2 increased hTERT expression. GB patient tumors bearing hTERT promoter mutations associated with increased telomerase activity had an increased NRF2 and transketolase (TKT) expression and decreased glycogen accumulation. Overexpression of NRF2 rescued the Costunolide, a telomerase inhibitor, mediated decrease in G6PD and TKT levels, while the inhibition of hTERT abolished not only the expression of G6PD and TKT but also the phosphorylation of glycogen synthase (GS) and increased glycogen accumulation.[234] The physical interaction of cytochrome B-245 beta chain (CYBB), a major catalytic subunit of NADPH oxidase (NOX) with NRF2, allows for the promotion of a mesenchymal GB phenotype, increased cancer stemness, and the development of resistance in GB.

NRF2 in Therapeutic Resistance

In GB, methylation of the O6-methylguanine-DNA methyltransferase (MGMT) promoter has been demonstrated to predict responsiveness to alkylating drugs such

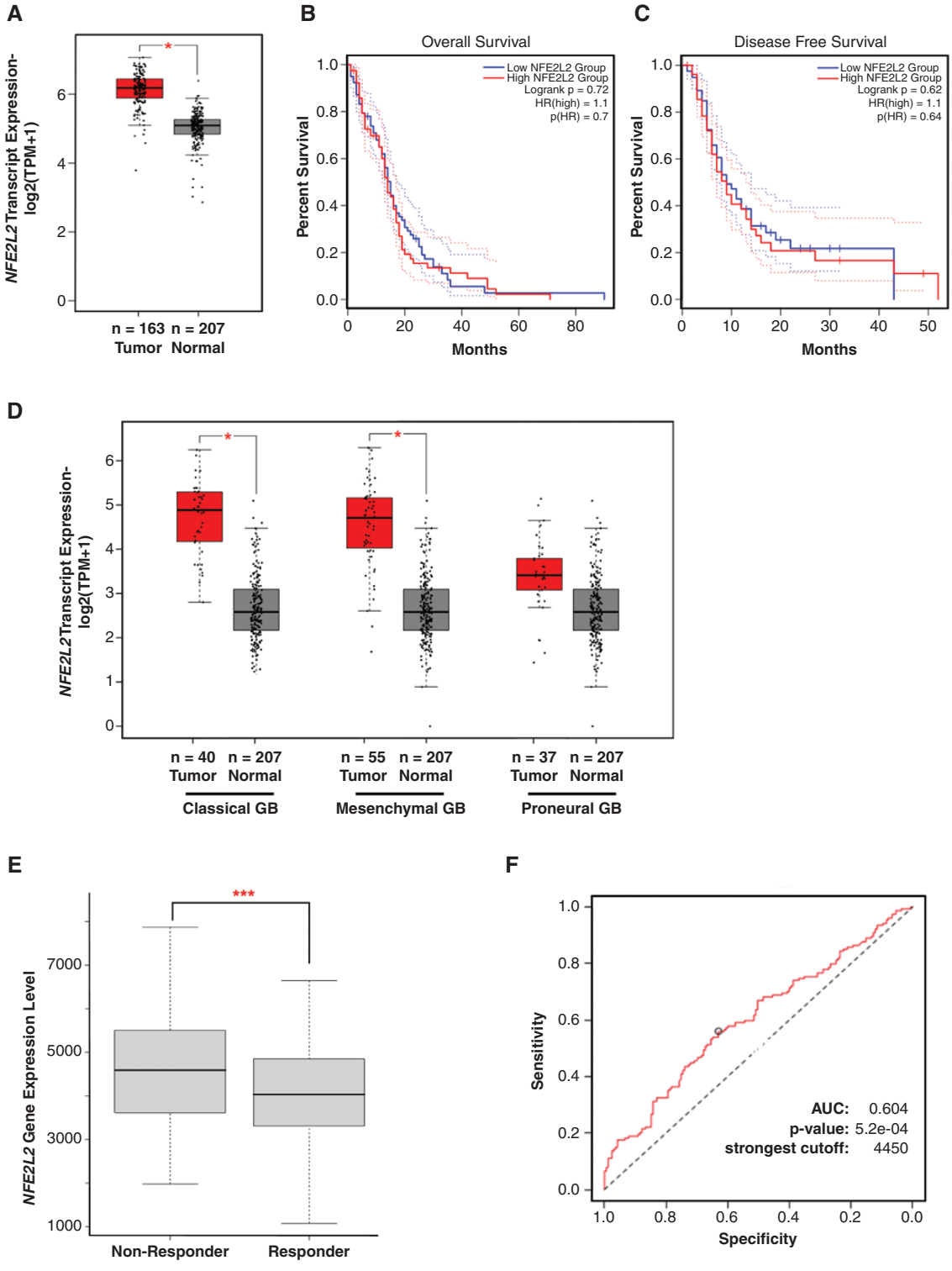

Figure 2. *NFE2L2* gene expression levels in GB and the impact on the clinical outcome. (A) Tissue-wise expression profile of the *NFE2L2* gene expression in GB tumors compared to normal tissue. Data is sourced from GEPEIA2 for GB patient databases. (B) Kaplan–Meier survival curves of overall survival and (C) disease-free survival of patients with GB based on the high (red) and low (blue) expression of the *NFE2L2* gene, respectively. (D) Tissue-wise expression profile of *NFE2L2* gene expression in GB subtypes compared to normal tissues. Data is sourced from GEPEIA2 for GB patient databases. (E) ROC plotter showing the *NFE2L2* gene expression in patients classified as responders (165 patients) and nonresponders (154 patients) to TMZ treatment (*P*-value = .0013). (F) ROC curve analysis shows the validity of *NFE2L2* gene expression in discriminating responders and nonresponders, with the sensitivity representing the true positive rate and the specificity representing the false positive rate. Data is sourced from ROC Plotter—Online ROC analysis for GB patient data. The red star denotes statistical significance. AUC, area under the curve; TPM, transcripts per million reads; n, number of tissue samples; HR, hazards ratio; TMZ, temozolomide.

as TMZ, which has become a cornerstone of GB treatment.[235] Mechanistically, at physiological pH, TMZ is activated to produce methyl diazonium ions with methyl groups, which are transported to DNA at the N7 position of guanine, O3 position of adenine, and O6 position of guanine,[235–237] resulting in numerous DNA adducts and the formation of single- and double-stranded DNA breaks, ultimately causing cell cytotoxicity.[237] However, because of broad TMZ exposure and the very heterogeneous and mutation-prone character of GB, it is quite usual for these deadly tumors to develop TMZ resistance. Unfortunately, over half of GB patients treated with TMZ do not respond to the medication.[237] As a result, TMZ resistance is a significant challenge that must be overcome for the effective treatment of GB.

A recent study has revealed, using a CRISPR activation library, that the NRF2 pathway is involved in TMZ resistance.[238] Moreover, inhibiting the NRF2/ARE pathway sensitizes GB cells to TMZ treatment,[239] implying that targeting NRF2 activation could be a promising strategy to enhance chemoradiation sensitivity in GB. In response to the treatment with TMZ coupled with the suppression of NRF2, the RAS/RAF/MEK signaling pathway was inhibited, leading to a decrease in the proliferation of U251 glioma cells. In addition, the subsequent downregulated HO-1, GSH, TRX, and other oxidative enzymes, along with the elevated Keap1 levels, inhibited the anti-oxidative stress mechanism in glioma cells.[240] Three-dimensional tumor models such as spheroid and organoid systems confer an advantage over other culturing methods by mimicking the in vivo characteristics of CNS malignancies.[241] Knowing that TMZ induces DNA damage, the DNA repair pathways, including O6-methylguanine-DNA methyltransferase (O6-MGMT), base excision repair, and mismatch repair, are implicated in TMZ resistance and other identified mechanisms.[237,242,243] In an elegant study, Rocha et al. highlighted essential mechanisms involved in TMZ resistance.[243] Briefly, TMZ therapy increases ROS production, which causes NRF2 to be activated, resulting in increased expression of 2 glutathione (GSH) synthesis enzymes, GCLM and glutamate-cysteine ligase, catalytic subunit (GCLC). Consequently, increased GSH availability mediates TMZ resistance by maintaining cancer cells' low ROS content and subsequent reduction of TMZ cytotoxicity.[243] However, GSH depletion mimicked by L-buthionine [S, R]-sulfoximine (BSO) in glioma cells is responsible for overcoming TMZ drug resistance.[243] In a similar context, increased NRF2 expression improves ferroptosis sensitivity in TMZ-resistant GB by increasing the expression of its pro-ferroptosis target ATP-binding cassette sub-family C member 1 (ABCC1), which contributes to GSH depletion. Thus, inducing ferroptosis could be a proper therapeutic method for reversing drug resistance in gliomas with high NRF2 and ABCC1 expression.[244] The activation of NRF2 and its downstream target, SOD2, prevented ferroptosis and excessive production of ROS. In contrast, inhibiting SOD2, combined with tolerable ferroptosis-inducing agents like erastin, sensitizes GB cells, overcoming TMZ resistance in mesenchymal GB.[245] However, further research is needed to confirm the effectiveness of the disruption of the NRF2/SOD2 antioxidant circuitry approach in developing GB therapeutic strategies.

Moreover, knocking down the *NRF2* gene in glioma neurospheres followed by gamma rays' irradiation resulted in less self-renewal, more differentiated cells, and less proliferative potential.[246] Consequently, this suggests that NRF2 suppression enhances cellular sensitivity to radiation-induced oxidative stress. In comparison, a compelling association between *NFE2L2* gene expression and patient response to TMZ is demonstrated using the receiver operating characteristic (ROC) analysis for GB patient database[247] (Figure 2E), in addition to the fact that *NFE2L2* gene seems to exhibit a predictive power and of potential clinical utility (Figure 2F). Overall, evidence suggests that NRF2 is a crucial player to be employed in therapeutic strategies involved in GB-TMZ resistance.

Conclusions

This review highlights the pivotal role of cellular redox homeostasis within the intricate landscape of cancer biology. The delicate interplay between reactive oxygen species (ROS), antioxidants, and diverse cellular processes is central to comprehending cancer's genesis, progression, and therapeutic interventions. Moreover, NRF2 emerges as a master regulator, orchestrating an extensive array of cytoprotective genes to maintain redox equilibrium and ensure proper cellular function.

However, NRF2 exhibits a dual role in cancer. It acts as a guardian by preserving redox homeostasis and serving as an anti-inflammatory mediator while simultaneously harboring the potential to fuel cancer growth, drug resistance, metabolic adaptations, and the activation of various oncogenes. Understanding the context-dependent nature of NRF2's actions in cancer is pivotal for developing precise and efficient cancer therapies, thereby shedding light on the intricate landscape of cancer biology.

Within the human brain, NRF2 exhibits diverse expression patterns among different brain cell types, including astrocytes, microglia, oligodendrocytes, and neurons. Its activation is critical in maintaining redox homeostasis, executing distinct functions in neurons and astrocytes, thereby preserving brain health. NRF2's involvement extends to preserving brain mitochondrial function and integrity, offering promising prospects for interventions in brain health maintenance.

Finally, NRF2 in cancer prognosis is a subject of significant interest, yet more studies are needed to explain the intricate relationship between NRF2 expression and brain cancer prognosis, considering various tumor types, grades, and characteristics. In glioblastoma, NRF2 emerges as a prominent player, significantly influencing malignancy, oncogenic transformation, and the development of therapeutic resistance. Noteworthy is NRF2's role in the maintenance of GSCs, which contributes to temozolomide resistance and tumor recurrence. Nonetheless, there remains a need for a comprehensive understanding of the molecular mechanisms underlying NRF2-mediated GSC maintenance and the metabolic pathways implicated in glioblastoma.

Overall, these discoveries highlight how NRF2 is involved in many aspects of cancer and various cell functions. This

knowledge sets a solid basis for further research and the development of precisely targeted therapies, including NRF2 silencing approaches, within the domains of cancer biology and brain health.

Keywords

Brain physiology | NRF2 | glioblastoma stem cells | oxidative stress | therapeutic resistance

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Conflict of interest

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References

1. Ghoneum A, Abdulfattah AY, Warren BO, Shu J, Said N. Redox homeostasis and metabolism in cancer: a complex mechanism and potential targeted therapeutics. *Int J Mol Sci* . 2020;21(9):3100.
2. Pizzino G, Irrera N, Cucinotta M, et al. Oxidative stress: harms and benefits for human health. *Oxid Med Cell Longev.* 2017;2017:8416763.
3. Sajadimajd S, Khazaei M. Oxidative stress and cancer: the role of Nrf2. *Curr Cancer Drug Targets.* 2018;18(6):538–557.
4. Trachootham D, Lu W, Ogasawara MA, Nilsa RD, Huang P. Redox regulation of cell survival. *Antioxid Redox Signal.* 2008;10(8):1343–1374.
5. Milkovic L, Cipak Gasparovic A, Cindric M, Mouthuy PA, Zarkovic N. Short overview of ROS as cell function regulators and their implications in therapy concepts. *Cells.* 2019;8(8):793–807.
6. Sauer H, Wartenberg M, Hescheler J. Reactive oxygen species as intracellular messengers during cell growth and differentiation. *Cell Physiol Biochem.* 2001;11(4):173–186.
7. Brieger K, Schiavone S, Miller FJ, Krause KH. Reactive oxygen species: from health to disease. *Swiss Med Wkly.* 2012;142(3334):w13659.
8. Parvez S, Long MJC, Poganik JR, Aye Y. Redox signaling by reactive electrophiles and oxidants. *Chem Rev.* 2018;118(18):8798–8888.
9. Shi Y, Nikulenkov F, Zawacka-Pankau J, et al. ROS-dependent activation of JNK converts p53 into an efficient inhibitor of oncogenes leading to robust apoptosis. *Cell Death Differ.* 2014;21(4):612–623.
10. Stockwell BR, Friedmann Angeli JP, Bayir H, et al. Ferroptosis: A regulated cell death nexus linking metabolism, redox biology, and disease. *Cell.* 2017;171(2):273–285.
11. Higinbotham KG, Rice JM, Diwan BA, et al. GGT to GTT transversions in codon 12 of the K-ras oncogene in rat renal sarcomas induced with nickel subsulfide or nickel subsulfide/iron are consistent with oxidative damage to DNA. *Cancer Res.* 1992;52(17):4747–4751.
12. Du MQ, Carmichael PL, Phillips DH. In duction of activating mutations in the human c-Ha-ras-1 proto-oncogene by oxygen free radicals. *Mol Carcinog.* 1994;11(3):170–175.
13. Harris CC, Hollstein M. Clinical implications of the p53 tumor-suppressor gene. *N Engl J Med.* 1993;329(18):1318–1327.
14. Hollstein M, Sidransky D, Vogelstein B, Harris CC. p53 mutations in human cancers. *Science.* 1991;253(5015):49–53.
15. Brash DE, Rudolph JA, Simon JA, et al. A role for sunlight in skin cancer: UV-induced p53 mutations in squamous cell carcinoma. *Proc Natl Acad Sci U S A.* 1991;88(22):10124–10128.
16. Reid TM, Loeb LA. Effect of DNA-repair enzymes on mutagenesis by oxygen free radicals. *Mutat Res.* 1993;289(2):181–186.
17. Sosa V, Moliné T, Somoza R, et al. Oxidative stress and cancer: an overview. *Ageing Res Rev.* 2013;12(1):376–390.
18. He F, Antonucci L, Karin M. NRF2 as a regulator of cell metabolism and inflammation in cancer. *Carcinogenesis.* 2020;41(4):405–416.
19. Wang YY, Chen J, Liu XM, Zhao R, Zhe H. Nrf2-mediated metabolic reprogramming in cancer. *Oxid Med Cell Longev.* 2018;2018:9304091.
20. Buettner GR. Superoxide dismutase in redox biology: the roles of superoxide and hydrogen peroxide. *Anticancer Agents Med Chem.* 2011;11(4):341–346.
21. Nandi A, Yan LJ, Jana CK, Das N. Role of catalase in oxidative stress- and age-associated degenerative diseases. *Oxid Med Cell Longev.* 2019;2019:9613090.
22. Margis R, Dunand C, Teixeira FK, Margis-Pinheiro M. Glutathione peroxidase family—an evolutionary overview. *FEBS J.* 2008;275(15):3959–3970.
23. Lu J, Holmgren A. The thioredoxin antioxidant system. *Free Radic Biol Med.* 2014;66:75–87.
24. He L, He T, Farrar S, et al. Antioxidants maintain cellular redox homeostasis by elimination of reactive oxygen species. *Cell Physiol Biochem.* 2017;44(2):532–553.
25. Calvani M, Subbiani A, Vignoli M, Favre C. Spotlight on ROS and beta3-adrenoreceptors fighting in cancer cells. *Oxid Med Cell Longev.* 2019;2019:6346529.
26. Chen XL, Kunsch C. Induction of cytoprotective genes through Nrf2/antioxidant response element pathway: a new therapeutic approach for the treatment of inflammatory diseases. *Curr Pharm Des.* 2004;10(8):879–891.
27. Zhu H, Itoh K, Yamamoto M, Zweier JL, Li Y. Role of Nrf2 signaling in regulation of antioxidants and phase 2 enzymes in cardiac fibroblasts: protection against reactive oxygen and nitrogen species-induced cell injury. *FEBS Lett.* 2005;579(14):3029–3036.
28. Rojo de la Vega M, Chapman E, Zhang DD. NRF2 and the hallmarks of cancer. *Cancer Cell.* 2018;34(1):21–43.
29. Wu S, Lu H, Bai Y. Nrf2 in cancers: a double-edged sword. *Cancer Medicine.* 2019;8(5):2252–2267.
30. Motohashi H, Katsuoka F, Engel JD, Yamamoto M. Small Maf proteins serve as transcriptional cofactors for keratinocyte differentiation in the Keap1-Nrf2 regulatory pathway. *Proc Natl Acad Sci U S A.* 2004;101(17):6379–6384.
31. Tong KI, Padmanabhan B, Kobayashi A, et al. Different electrostatic potentials define ETGE and DLG motifs as hinge and latch in oxidative stress response. *Mol Cell Biol.* 2007;27(21):7511–7521.
32. Moon EJ, Giaccia A. Dual roles of NRF2 in tumor prevention and progression: possible implications in cancer treatment. *Free Radic Biol Med.* 2015;79:292–299.
33. Motohashi H, Yamamoto M. Nrf2-Keap1 defines a physiologically important stress response mechanism. *Trends Mol Med.* 2004;10(11):549–557.
34. Nioi P, Nguyen T, Sherratt PJ, Pickett CB. The carboxy-terminal Neh3 domain of Nrf2 is required for transcriptional activation. *Mol Cell Biol.* 2005;25(24):10895–10906.

35. McMahon M, Thomas N, Itoh K, Yamamoto M, Hayes JD. Redox-regulated turnover of Nrf2 is determined by at least two separate protein domains, the redox-sensitive Neh2 degron and the redox-insensitive Neh6 degron. *J Biol Chem.* 2004;279(30):31556–31567.
36. Chowdhry S, Zhang Y, McMahon M, et al. Nrf2 is controlled by two distinct beta-TrCP recognition motifs in its Neh6 domain, one of which can be modulated by GSK-3 activity. *Oncogene.* 2013;32(32):3765–3781.
37. Wang H, Liu K, Geng M, et al. RXRalpha inhibits the NRF2-ARE signaling pathway through a direct interaction with the Neh7 domain of NRF2. *Cancer Res.* 2013;73(10):3097–3108.
38. Furukawa M, Xiong Y. BTB protein Keap1 targets antioxidant transcription factor Nrf2 for ubiquitination by the Cullin 3-Roc1 ligase. *Mol Cell Biol.* 2005;25(1):162–171.
39. Sun Z, Wu T, Zhao F, et al. KPNA6 (Importin {alpha}7)-mediated nuclear import of Keap1 represses the Nrf2-dependent antioxidant response. *Mol Cell Biol.* 2011;31(9):1800–1811.
40. Bryan HK, Olayanju A, Goldring CE, Park BK. The Nrf2 cell defence pathway: Keap1-dependent and -independent mechanisms of regulation. *Biochem Pharmacol.* 2013;85(6):705–717.
41. Suzuki T, Takahashi J, Yamamoto M. Molecular basis of the KEAP1-NRF2 signaling pathway. *Mol Cells.* 2023;46(3):133–141.
42. McMahon M, Swift SR, Hayes JD. Zinc-binding triggers a conformational-switch in the cullin-3 substrate adaptor protein KEAP1 that controls transcription factor NRF2. *Toxicol Appl Pharmacol.* 2018;360:45–57.
43. Zhang DD, Hannink M. Distinct cysteine residues in Keap1 are required for Keap1-dependent ubiquitination of Nrf2 and for stabilization of Nrf2 by chemopreventive agents and oxidative stress. *Mol Cell Biol.* 2003;23(22):8137–8151.
44. Huang HC, Nguyen T, Pickett CB. Phosphorylation of Nrf2 at Ser-40 by protein kinase C regulates antioxidant response element-mediated transcription. *J Biol Chem.* 2002;277(45):42769–42774.
45. Kansanen E, Kuosmanen SM, Leinonen H, Levonen AL. The Keap1-Nrf2 pathway: mechanisms of activation and dysregulation in cancer. *Redox Biol.* 2013;1(1):45–49.
46. Villeneuve NF, Lau A, Zhang DD. Regulation of the Nrf2-Keap1 antioxidant response by the ubiquitin proteasome system: an insight into cullin-ring ubiquitin ligases. *Antioxid Redox Signal.* 2010;13(11):1699–1712.
47. Kobayashi A, Kang MI, Okawa H, et al. Oxidative stress sensor Keap1 functions as an adaptor for Cul3-based E3 ligase to regulate proteasomal degradation of Nrf2. *Mol Cell Biol.* 2004;24(16):7130–7139.
48. Ma Q, Kinneer K, Bi Y, Chan JY, Kan YW. Induction of murine NAD(P)H:quinone oxidoreductase by 2,3,7,8-tetrachlorodibenzo-p-dioxin requires the CNC (cap "n" collar) basic leucine zipper transcription factor Nrf2 (nuclear factor erythroid 2-related factor 2): cross-interaction between AhR (aryl hydrocarbon receptor) and Nrf2 signal transduction. *Biochem J.* 2004;377(Pt 1):205–213.
49. Miao W, Hu L, Scrivens PJ, Batist G. Transcriptional regulation of NF-E2 p45-related factor (NRF2) expression by the aryl hydrocarbon receptor-xenobiotic response element signaling pathway: direct cross-talk between phase I and II drug-metabolizing enzymes. *J Biol Chem.* 2005;280(21):20340–20348.
50. DeNicola GM, Karreth FA, Humpton TJ, et al. Oncogene-induced Nrf2 transcription promotes ROS detoxification and tumorigenesis. *Nature.* 2011;475(7354):106–109.
51. Sanderson LM, Boekschoten MV, Desvergne B, Muller M, Kersten S. Transcriptional profiling reveals divergent roles of PPARalpha and PPARbeta/delta in regulation of gene expression in mouse liver. *Physiol Genomics.* 2010;41(1):42–52.
52. Basak P, Sadhukhan P, Sarkar P, Sil PC. Perspectives of the Nrf-2 signaling pathway in cancer progression and therapy. *Toxicol Rep.* 2017;4:306–318.
53. Yamamoto S, Inoue J, Kawano T, et al. The impact of miRNA-based molecular diagnostics and treatment of NRF2-stabilized tumors. *Mol Cancer Res.* 2014;12(1):58–68.
54. Muscarella LA, Barbano R, D'Angelo V, et al. Regulation of KEAP1 expression by promoter methylation in malignant gliomas and association with patient's outcome. *Epigenetics.* 2011;6(3):317–325.
55. Zhang P, Singh A, Yegnasubramanian S, et al. Loss of Kelch-like ECH-associated protein 1 function in prostate cancer cells causes chemoresistance and radioresistance and promotes tumor growth. *Mol Cancer Ther.* 2010;9(2):336–346.
56. Hanada N, Takahata T, Zhou Q, et al. Methylation of the KEAP1 gene promoter region in human colorectal cancer. *BMC Cancer.* 2012;12:66.
57. Fan W, Tang Z, Chen D, et al. Keap1 facilitates p62-mediated ubiquitin aggregate clearance via autophagy. *Autophagy.* 2010;6(5):614–621.
58. Liao H, Zhou Q, Zhang Z, et al. NRF2 is overexpressed in ovarian epithelial carcinoma and is regulated by gonadotrophin and sex-steroid hormones. *Oncol Rep.* 2012;27(6):1918–1924.
59. Song M-Y, Lee D-Y, Chun K-S, Kim E-H. The Role of NRF2/KEAP1 signaling pathway in cancer metabolism. *Int J Mol Sci .* 2021;22(9):4376.
60. Zhang M, An C, Gao Y, et al. Emerging roles of Nrf2 and phase II antioxidant enzymes in neuroprotection. *Prog Neurobiol.* 2013;100:30–47.
61. Habib E, Linher-Melville K, Lin HX, Singh G. Expression of xCT and activity of system xc(-) are regulated by NRF2 in human breast cancer cells in response to oxidative stress. *Redox Biol.* 2015;5:33–42.
62. Solis WA, Dalton TP, Dieter MZ, et al. Glutamate-cysteine ligase modifier subunit: mouse Gclm gene structure and regulation by agents that cause oxidative stress. *Biochem Pharmacol.* 2002;63(9):1739–1754.
63. Kim YC, Masutani H, Yamaguchi Y, et al. Hemin-induced activation of the thioredoxin gene by Nrf2 A differential regulation of the antioxidant responsive element by a switch of its binding factors. *J Biol Chem.* 2001;276(21):18399–18406.
64. Sakurai A, Nishimoto M, Himeno S, et al. Transcriptional regulation of thioredoxin reductase 1 expression by cadmium in vascular endothelial cells: role of NF-E2-related factor-2. *J Cell Physiol.* 2005;203(3):529–537.
65. Kim YJ, Ahn JY, Liang P, et al. Human prx1 gene is a target of Nrf2 and is up-regulated by hypoxia/reoxygenation: implication to tumor biology. *Cancer Res.* 2007;67(2):546–554.
66. Soriano FX, Leveille F, Papadia S, et al. Induction of sulfiredoxin expression and reduction of peroxiredoxin hyperoxidation by the neuroprotective Nrf2 activator 3H-1,2-dithiole-3-thione. *J Neurochem.* 2008;107(2):533–543.
67. Menegon S, Columbano A, Giordano S. The dual roles of NRF2 in cancer. *Trends Mol Med.* 2016;22(7):578–593.
68. Lau A, Villeneuve NF, Sun Z, Wong PK, Zhang DD. Dual roles of Nrf2 in cancer. *Pharmacol Res.* 2008;58(5–6):262–270.
69. Satoh H, Moriguchi T, Taguchi K, et al. Nrf2-deficiency creates a responsive microenvironment for metastasis to the lung. *Carcinogenesis.* 2010;31(10):1833–1843.
70. Boyanapalli SS, Paredes-Gonzalez X, Fuentes F, et al. Nrf2 knockout attenuates the anti-inflammatory effects of phenethyl isothiocyanate and curcumin. *Chem Res Toxicol.* 2014;27(12):2036–2043.
71. Saha S, Buttari B, Panieri E, Profumo E, Saso L. An overview of Nrf2 signaling pathway and its role in inflammation. *Molecules.* 2020;25(22):5474–5505.
72. Kobayashi EH, Suzuki T, Funayama R, et al. Nrf2 suppresses macrophage inflammatory response by blocking proinflammatory cytokine transcription. *Nat Commun.* 2016;7:11624.
73. Kong X, Thimmulappa R, Craciun F, et al. Enhancing Nrf2 pathway by disruption of Keap1 in myeloid leukocytes protects against sepsis. *Am J Respir Crit Care Med.* 2011;184(8):928–938.
74. Chen XL, Dodd G, Thomas S, et al. Activation of Nrf2/ARE pathway protects endothelial cells from oxidant injury and inhibits inflammatory

gene expression. *Am J Physiol Heart Circ Physiol.* 2006;290(5):H1862–H1870.
75. Kim JE, You DJ, Lee C, et al. Suppression of NF-kappaB signaling by KEAP1 regulation of IKKbeta activity through autophagic degradation and inhibition of phosphorylation. *Cell Signal.* 2010;22(11):1645–1654.
76. Gao W, Guo L, Yang Y, et al. Dissecting the crosstalk between Nrf2 and NF-kappaB response pathways in drug-induced toxicity. *Front Cell Dev Biol.* 2021;9:809952.
77. Zimta A-A, Cenariu D, Irimie A, et al. The role of Nrf2 activity in cancer development and progression. *Cancers.* 2019;11(11):E1755.
78. Rojo AI, Rada P, Mendiola M, et al. The PTEN/NRF2 axis promotes human carcinogenesis. *Antioxid Redox Signal.* 2014;21(18):2498–2514.
79. Mitsuishi Y, Taguchi K, Kawatani Y, et al. Nrf2 redirects glucose and glutamine into anabolic pathways in metabolic reprogramming. *Cancer Cell.* 2012;22(1):66–79.
80. Kitteringham NR, Abdullah A, Walsh J, et al. Proteomic analysis of Nrf2 deficient transgenic mice reveals cellular defence and lipid metabolism as primary Nrf2-dependent pathways in the liver. *J Proteomics.* 2010;73(8):1612–1631.
81. Malhotra D, Portales-Casamar E, Singh A, 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(17):5718–5734.
82. Reddy NM, Kleeberger SR, Bream JH, et al. Genetic disruption of the Nrf2 compromises cell-cycle progression by impairing GSH-induced redox signaling. *Oncogene.* 2008;27(44):5821–5832.
83. Chang CW, Chen YS, Tsay YG, et al. ROS-independent ER stress-mediated NRF2 activation promotes Warburg effect to maintain stemness-associated properties of cancer-initiating cells. *Cell Death Dis.* 2018;9(2):194.
84. Zhou S, Ye W, Zhang M, Liang J. The effects of nrf2 on tumor angiogenesis: a review of the possible mechanisms of action. *Crit Rev Eukaryot Gene Expr.* 2012;22(2):149–160.
85. Bussolati B, Mason JC. Dual role of VEGF-induced heme-oxygenase-1 in angiogenesis. *Antioxid Redox Signal.* 2006;8(7–8):1153–1163.
86. Rushworth SA, MacEwan DJ. HO-1 underlies resistance of AML cells to TNF-induced apoptosis. *Blood.* 2008;111(7):3793–3801.
87. Niture SK, Jaiswal AK. Nrf2 protein up-regulates antiapoptotic protein Bcl-2 and prevents cellular apoptosis. *J Biol Chem.* 2012;287(13):9873–9886.
88. Elsby R, Kitteringham NR, Goldring CE, et al. Increased constitutive c-Jun N-terminal kinase signaling in mice lacking glutathione S-transferase Pi. *J Biol Chem.* 2003;278(25):22243–22249.
89. Jain A, Lamark T, Sjottem E, et al. p62/SQSTM1 is a target gene for transcription factor NRF2 and creates a positive feedback loop by inducing antioxidant response element-driven gene transcription. *J Biol Chem.* 2010;285(29):22576–22591.
90. Komatsu M, Kurokawa H, Waguri S, et al. The selective autophagy substrate p62 activates the stress responsive transcription factor Nrf2 through inactivation of Keap1. *Nat Cell Biol.* 2010;12(3):213–223.
91. Towers CG, Fitzwalter BE, Regan D, et al. Cancer cells upregulate NRF2 signaling to adapt to autophagy inhibition. *Dev Cell.* 2019;50(6):690–703.e6.
92. Chang CW, Chen YS, Chou SH, et al. Distinct subpopulations of head and neck cancer cells with different levels of intracellular reactive oxygen species exhibit diverse stemness, proliferation, and chemosensitivity. *Cancer Res.* 2014;74(21):6291–6305.
93. Ryoo I, Lee S, Kwak M-K. Redox modulating NRF2: a potential mediator of cancer stem cell resistance. *Oxid Med Cell Longevity.* 2016;2016:1–14.
94. Kumar H, Kumar RM, Bhattacharjee D, Somanna P, Jain V. Role of Nrf2 signaling cascade in breast cancer: strategies and treatment. *Front Pharmacol.* 2022;13:720076.
95. Singh A, Boldin-Adamsky S, Thimmulappa RK, et al. RNAi-mediated silencing of nuclear factor erythroid-2-related factor 2 gene expression in non-small cell lung cancer inhibits tumor growth and increases efficacy of chemotherapy. *Cancer Res.* 2008;68(19):7975–7984.
96. Yasuda T, Ishimoto T, Baba H. Conflicting metabolic alterations in cancer stem cells and regulation by the stromal niche. *Regen Ther.* 2021;17:8–12.
97. Gao L, Morine Y, Yamada S, et al. Nrf2 signaling promotes cancer stemness, migration, and expression of ABC transporter genes in sorafenib-resistant hepatocellular carcinoma cells. *PLoS One.* 2021;16(9):e0256755.
98. Kahroba H, Shirmohamadi M, Hejazi MS, Samadi N. The role of Nrf2 signaling in cancer stem cells: from stemness and self-renewal to tumorigenesis and chemoresistance. *Life Sci.* 2019;239:116986.
99. Ridge SM, Sullivan FJ, Glynn SA. Mesenchymal stem cells: key players in cancer progression. *Mol Cancer.* 2017;16(1):31.
100. Mohammadzadeh-Vardin M, Habibi Roudkenar M, Jahanian-Najafabadi A. Adenovirus-mediated over-expression of Nrf2 within mesenchymal stem cells (MSCs) protected rats against acute kidney injury. *Adv Pharm Bull.* 2015;5(2):201–208.
101. Yuan Z, Zhang J, Huang Y, et al. NRF2 overexpression in mesenchymal stem cells induces stem-cell marker expression and enhances osteoblastic differentiation. *Biochem Biophys Res Commun.* 2017;491(1):228–235.
102. Kitamura H, Motohashi H. NRF2 addiction in cancer cells. *Cancer Sci.* 2018;109(4):900–911.
103. Okazaki K, Papagiannakopoulos T, Motohashi H. Metabolic features of cancer cells in NRF2 addiction status. *Biophys Rev.* 2020;12(2):435–441.
104. Taguchi K, Hirano I, Itoh T, et al. Nrf2 enhances cholangiocyte expansion in Pten-deficient livers. *Mol Cell Biol.* 2014;34(5):900–913.
105. Shirasaki K, Taguchi K, Unno M, Motohashi H, Yamamoto M. NF-E2-related factor 2 promotes compensatory liver hypertrophy after portal vein branch ligation in mice. *Hepatology.* 2014;59(6):2371–2382.
106. Suzuki T, Seki S, Hiramoto K, et al. Hyperactivation of Nrf2 in early tubular development induces nephrogenic diabetes insipidus. *Nat Commun.* 2017;8:14577.
107. Murakami S, Suzuki T, Harigae H, et al. NRF2 activation impairs quiescence and bone marrow reconstitution capacity of hematopoietic stem cells. *Mol Cell Biol.* 2017;37(19):e00086–17.
108. Taguchi K, Maher JM, Suzuki T, et al. Genetic analysis of cytoprotective functions supported by graded expression of Keap1. *Mol Cell Biol.* 2010;30(12):3016–3026.
109. Romero R, Sayin VI, Davidson SM, et al. Keap1 loss promotes Kras-driven lung cancer and results in dependence on glutaminolysis. *Nat Med.* 2017;23(11):1362–1368.
110. Kitamura H, Onodera Y, Murakami S, Suzuki T, Motohashi H. IL-11 contribution to tumorigenesis in an NRF2 addiction cancer model. *Oncogene.* 2017;36(45):6315–6324.
111. Jeong Y, Hoang NT, Lovejoy A, et al. Role of KEAP1/NRF2 and TP53 mutations in lung squamous cell carcinoma development and radiation resistance. *Cancer Discov.* 2017;7(1):86–101.
112. Hamada S, Shimosegawa T, Taguchi K, et al. Simultaneous K-ras activation and Keap1 deletion cause atrophy of pancreatic parenchyma. *Am J Physiol Gastrointest Liver Physiol.* 2018;314(1):G65–G74.
113. Ji L, Li H, Gao P, et al. Nrf2 pathway regulates multidrug-resistance-associated protein 1 in small cell lung cancer. *PLoS One.* 2013;8(5):e63404.
114. 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(4):541–567.
115. Ryoo I, Kim G, Choi B, Lee S, Kwak M-K. Involvement of NRF2 signaling in doxorubicin resistance of cancer stem cell-enriched colonospheres. *Biomolecules & Therapeutics.* 2016;24(5):482–488.

116. Sasaki H, Shitara M, Yokota K, et al. MRP3 gene expression correlates with NRF2 mutations in lung squamous cell carcinomas. *Mol Med Rep.* 2012;6(4):705–708.
117. Gao AM, Ke ZP, Wang JN, et al. Apigenin sensitizes doxorubicin-resistant hepatocellular carcinoma BEL-7402/ADM cells to doxorubicin via inhibiting PI3K/Akt/Nrf2 pathway. *Carcinogenesis.* 2013;34(8):1806–1814.
118. von Bartheld CS. Myths and truths about the cellular composition of the human brain: A review of influential concepts. *J Chem Neuroanat.* 2018;93:2–15.
119. Mondello S, Jeromin A, Buki A, et al. Glial neuronal ratio: a novel index for differentiating injury type in patients with severe traumatic brain injury. *J Neurotrauma.* 2012;29(6):1096–1104.
120. von Bartheld CS, Bahney J, Herculano-Houzel S. The search for true numbers of neurons and glial cells in the human brain: A review of 150 years of cell counting. *J Comp Neurol.* 2016;524(18):3865–3895.
121. Baxter PS, Hardingham GE. Adaptive regulation of the brain's antioxidant defences by neurons and astrocytes. *Free Radic Biol Med.* 2016;100:147–152.
122. Dringen R, Pawlowski PG, Hirrlinger J. Peroxide detoxification by brain cells. *J Neurosci Res.* 2005;79(1–2):157–165.
123. Kandel RE, Koester DJ, Mack HS, Siegelbaum AS. *Principles of Neural Science. 6th ed.* McGraw Hill/ Medical.; 2021.
124. Shih AY, Johnson DA, Wong G, et al. Coordinate regulation of glutathione biosynthesis and release by Nrf2-expressing glia potently protects neurons from oxidative stress. *J Neurosci.* 2003;23(8):3394–3406.
125. Kraft AD, Johnson DA, Johnson JA. Nuclear factor E2-related factor 2-dependent antioxidant response element activation by tert-butylhydroquinone and sulforaphane occurring preferentially in astrocytes conditions neurons against oxidative insult. *J Neurosci.* 2004;24(5):1101–1112.
126. Freeman MR. Specification and morphogenesis of astrocytes. *Science.* 2010;330(6005):774–778.
127. Vaughn JE, Peters A. Electron microscopy of the early postnatal development of fibrous astrocytes. *Am J Anat.* 1967;121(1):131–152.
128. Oberheim NA, Goldman SA, Nedergaard M. Heterogeneity of astrocytic form and function. *Methods Mol Biol.* 2012;814:23–45.
129. Powell EM, Geller HM. Dissection of astrocyte-mediated cues in neuronal guidance and process extension. *Glia.* 1999;26(1):73–83.
130. Sofroniew MV, Vinters HV. Astrocytes: biology and pathology. *Acta Neuropathol.* 2010;119(1):7–35.
131. Budday S, Steinmann P, Kuhl E. Physical biology of human brain development. *Front Cell Neurosci.* 2015;9:257.
132. Stiles J, Jernigan TL. The basics of brain development. *Neuropsychol Rev.* 2010;20(4):327–348.
133. Singh A, Kukreti R, Saso L, Kukreti S. Oxidative stress: a key modulator in neurodegenerative diseases. *Molecules.* 2019;24(8):1583.
134. Bell KF, Al-Mubarak B, Martel MA, et al. Neuronal development is promoted by weakened intrinsic antioxidant defences due to epigenetic repression of Nrf2. *Nat Commun.* 2015;6:7066.
135. Ahlgren-Beckendorf JA, Reising AM, Schander MA, Herdler JW, Johnson JA. Coordinate regulation of NAD(P)H:quinone oxidoreductase and glutathione-S-transferases in primary cultures of rat neurons and glia: role of the antioxidant/electrophile responsive element. *Glia.* 1999;25(2):131–142.
136. Jimenez-Blasco D, Santofimia-Castano P, Gonzalez A, Almeida A, Bolanos JP. Astrocyte NMDA receptors' activity sustains neuronal survival through a Cdk5-Nrf2 pathway. *Cell Death Differ.* 2015;22(11):1877–1889.
137. Kennedy KA, Sandiford SD, Skerjanc IS, Li SS. Reactive oxygen species and the neuronal fate. *Cell Mol Life Sci.* 2012;69(2):215–221.
138. Vieira HL, Alves PM, Vercelli A. Modulation of neuronal stem cell differentiation by hypoxia and reactive oxygen species. *Prog Neurobiol.* 2011;93(3):444–455.
139. Funato Y, Michiue T, Asashima M, Miki H. The thioredoxin-related redox-regulating protein nucleoredoxin inhibits Wnt-beta-catenin signalling through dishevelled. *Nat Cell Biol.* 2006;8(5):501–508.
140. Rharass T, Lemcke H, Lantow M, et al. Ca^{2+}-mediated mitochondrial reactive oxygen species metabolism augments Wnt/beta-catenin pathway activation to facilitate cell differentiation. *J Biol Chem.* 2014;289(40):27937–27951.
141. Yu X, Malenka RC. Beta-catenin is critical for dendritic morphogenesis. *Nat Neurosci.* 2003;6(11):1169–1177.
142. Rosso SB, Sussman D, Wynshaw-Boris A, Salinas PC. Wnt signaling through dishevelled, Rac and JNK regulates dendritic development. *Nat Neurosci.* 2005;8(1):34–42.
143. Yang Y, Higashimori H, Morel L. Developmental maturation of astrocytes and pathogenesis of neurodevelopmental disorders. *J Neurodev Disord.* 2013;5(1):22.
144. Wang XF, Cynader MS. Astrocytes provide cysteine to neurons by releasing glutathione. *J Neurochem.* 2000;74(4):1434–1442.
145. Simons M, Nave KA. Oligodendrocytes: myelination and axonal support. *Cold Spring Harb Perspect Biol.* 2015;8(1):a020479.
146. Accetta R, Damiano S, Morano A, et al. Reactive oxygen species derived from NOX3 and NOX5 drive differentiation of human oligodendrocytes. *Front Cell Neurosci.* 2016;10:146.
147. Fetisova E, Chernyak B, Korshunova G, Muntyan M, Skulachev V. Mitochondria-targeted antioxidants as a prospective therapeutic strategy for multiple sclerosis. *Curr Med Chem.* 2017;24(19):2086–2114.
148. Islam MT. Oxidative stress and mitochondrial dysfunction-linked neurodegenerative disorders. *Neurol Res.* 2017;39(1):73–82.
149. Lundgaard I, Osorio MJ, Kress BT, Sanggaard S, Nedergaard M. White matter astrocytes in health and disease. *Neuroscience.* 2014;276:161–173.
150. Teske N, Liessem A, Fischbach F, et al. Chemical hypoxia-induced integrated stress response activation in oligodendrocytes is mediated by the transcription factor nuclear factor (erythroid-derived 2)-like 2 (NRF2). *J Neurochem.* 2018;144(3):285–301.
151. Nellessen A, Nyamoya S, Zendedel A, et al. Nrf2 deficiency increases oligodendrocyte loss, demyelination, neuroinflammation and axonal damage in an MS animal model. *Metab Brain Dis.* 2020;35(2):353–362.
152. Liddell JR. Are astrocytes the predominant cell type for activation of Nrf2 in aging and neurodegeneration? *Antioxidants (Basel).* 2017;6(3):65.
153. Dudvarski Stankovic N, Teodorczyk M, Ploen R, Zipp F, Schmidt MHH. Microglia-blood vessel interactions: a double-edged sword in brain pathologies. *Acta Neuropathol.* 2016;131(3):347–363.
154. Ochocka N, Kaminska B. Microglia diversity in healthy and diseased brain: insights from single-cell omics. *Int J Mol Sci .* 2021;22(6):3027.
155. Harry GJ. Microglia during development and aging. *Pharmacol Ther.* 2013;139(3):313–326.
156. He F, Ru X, Wen T. NRF2, a transcription factor for stress response and beyond. *Int J Mol Sci .* 2020;21(13):4777.
157. Hu L, Cao Y, Chen H, et al. The novel Nrf2 activator omaveloxolone regulates microglia phenotype and ameliorates secondary brain injury after intracerebral hemorrhage in mice. *Oxid Med Cell Longev.* 2022;2022:4564471.
158. Vilhardt F, Haslund-Vinding J, Jaquet V, McBean G. Microglia antioxidant systems and redox signalling. *Br J Pharmacol.* 2017;174(12):1719–1732.
159. Colonna M, Butovsky O. Microglia function in the central nervous system during health and neurodegeneration. *Annu Rev Immunol.* 2017;35:441–468.
160. Rojo AI, Innamorato NG, Martin-Moreno AM, et al. Nrf2 regulates microglial dynamics and neuroinflammation in experimental Parkinson's disease. *Glia.* 2010;58(5):588–598.
161. Ma K, Wu HY, Wang SY, Li BX. The Keap1/Nrf2-ARE signaling pathway is involved in atrazine induced dopaminergic neurons degeneration via microglia activation. *Ecotoxicol Environ Saf.* 2021;226:112862.

162. Min KJ, Yang MS, Kim SU, Jou I, Joe EH. Astrocytes induce hemeoxygenase-1 expression in microglia: a feasible mechanism for preventing excessive brain inflammation. *J Neurosci.* 2006;26(6):1880–1887.

163. Ramsey CP, Glass CA, Montgomery MB, et al. Expression of Nrf2 in neurodegenerative diseases. *J Neuropathol Exp Neurol.* 2007;66(1):75–85.

164. Sarlette A, Krampfl K, Grothe C, et al. Nuclear erythroid 2-related factor 2-antioxidative response element signaling pathway in motor cortex and spinal cord in amyotrophic lateral sclerosis. *J Neuropathol Exp Neurol.* 2008;67(11):1055–1062.

165. Cook AL, Vitale AM, Ravishankar S, et al. NRF2 activation restores disease related metabolic deficiencies in olfactory neurosphere-derived cells from patients with sporadic Parkinson's disease. *PLoS One.* 2011;6(7):e21907.

166. Ahlenius H, Visan V, Kokaia M, Lindvall O, Kokaia Z. Neural stem and progenitor cells retain their potential for proliferation and differentiation into functional neurons despite lower number in aged brain. *J Neurosci.* 2009;29(14):4408–4419.

167. Robledinos-Anton N, Rojo AI, Ferreiro E, et al. Transcription factor NRF2 controls the fate of neural stem cells in the subgranular zone of the hippocampus. *Redox Biol.* 2017;13:393–401.

168. Ray S, Corenblum MJ, Anandhan A, et al. A role for Nrf2 expression in defining the aging of hippocampal neural stem cells. *Cell Transplant.* 2018;27(4):589–606.

169. Khacho M, Clark A, Svoboda DS, et al. Mitochondrial dynamics impacts stem cell identity and fate decisions by regulating a nuclear transcriptional program. *Cell Stem Cell.* 2016;19(2):232–247.

170. Dang J, Brandenburg LO, Rosen C, et al. Nrf2 expression by neurons, astroglia, and microglia in the cerebral cortical penumbra of ischemic rats. *J Mol Neurosci.* 2012;46(3):578–584.

171. Licht-Mayer S, Wimmer I, Traffehn S, et al. Cell type-specific Nrf2 expression in multiple sclerosis lesions. *Acta Neuropathol.* 2015;130(2):263–277.

172. Lee DH, Gold R, Linker RA. Mechanisms of oxidative damage in multiple sclerosis and neurodegenerative diseases: therapeutic modulation via fumaric acid esters. *Int J Mol Sci* . 2012;13(9):11783–11803.

173. Spaas J, van Veggel L, Schepers M, et al. Oxidative stress and impaired oligodendrocyte precursor cell differentiation in neurological disorders. *Cell Mol Life Sci.* 2021;78(10):4615–4637.

174. Esteras N, Blacker TS, Zherebtsov EA, et al. Nrf2 regulates glucose uptake and metabolism in neurons and astrocytes. *Redox Biol.* 2023;62:102672.

175. Kirby J, Halligan E, Baptista MJ, et al. Mutant SOD1 alters the motor neuronal transcriptome: implications for familial ALS. *Brain.* 2005;128(Pt 7):1686–1706.

176. Mashima K, Takahashi S, Minami K, et al. Neuroprotective role of astroglia in Parkinson disease by reducing oxidative stress through dopamine-induced activation of pentose-phosphate pathway. *ASN Neuro.* 2018;10:1759091418775562.

177. Parga JA, Rodriguez-Perez AI, Garcia-Garrote M, Rodriguez-Pallares J, Labandeira-Garcia JL. Angiotensin II induces oxidative stress and upregulates neuroprotective signaling from the NRF2 and KLF9 pathway in dopaminergic cells. *Free Radic Biol Med.* 2018;129:394–406.

178. Zafar KS, Inayat-Hussain SH, Siegel D, et al. Overexpression of NQO1 protects human SK-N-MC neuroblastoma cells against dopamine-induced cell death. *Toxicol Lett.* 2006;166(3):261–267.

179. Kaidery NA, Banerjee R, Yang L, et al. Targeting Nrf2-mediated gene transcription by extremely potent synthetic triterpenoids attenuate dopaminergic neurotoxicity in the MPTP mouse model of Parkinson's disease. *Antioxid Redox Signal.* 2013;18(2):139–157.

180. Holmstrom KM, Baird L, Zhang Y, et al. Nrf2 impacts cellular bioenergetics by controlling substrate availability for mitochondrial respiration. *Biol Open.* 2013;2(8):761–770.

181. Greco T, Fiskum G. Brain mitochondria from rats treated with sulforaphane are resistant to redox-regulated permeability transition. *J Bioenerg Biomembr.* 2010;42(6):491–497.

182. Greco T, Shafer J, Fiskum G. Sulforaphane inhibits mitochondrial permeability transition and oxidative stress. *Free Radic Biol Med.* 2011;51(12):2164–2171.

183. Navarro E, Gonzalez-Lafuente L, Perez-Liebana I, et al. Heme-oxygenase I and PCG-1alpha regulate mitochondrial biogenesis via microglial activation of alpha7 nicotinic acetylcholine receptors using PNU282987. *Antioxid Redox Signal.* 2017;27(2):93–105.

184. Dinkova-Kostova AT, Abramov AY. The emerging role of Nrf2 in mitochondrial function. *Free Radic Biol Med.* 2015;88(Pt B):179–188.

185. Watts ME, Pocock R, Claudianos C. Brain energy and oxygen metabolism: emerging role in normal function and disease. *Front Mol Neurosci.* 2018;11:216.

186. Zahnreich S, Schmidberger H. Childhood cancer: occurrence, treatment and risk of second primary malignancies. *Cancers (Basel).* 2021;13(11):2607.

187. Northcott PA, Robinson GW, Kratz CP, et al. Medulloblastoma. *Nat Rev Dis Primers.* 2019;5(1):11.

188. Li Tang YD. Nrf-2 and HO-1 expression in medulloblastoma: a clinicopathological analysis. *J Biosci Med.* 2017;5:142–147.

189. Koto KS, Lescault P, Brard L, et al. Antitumor activity of nifurtimox is enhanced with tetrathiomolybdate in medulloblastoma. *Int J Oncol.* 2011;38(5):1329–1341.

190. Kim H, Lee TH, Park ES, et al. Role of peroxiredoxins in regulating intracellular hydrogen peroxide and hydrogen peroxide-induced apoptosis in thyroid cells. *J Biol Chem.* 2000;275(24):18266–18270.

191. Sasagawa I, Matsuki S, Suzuki Y, et al. Possible involvement of the membrane-bound form of peroxiredoxin 4 in acrosome formation during spermiogenesis of rats. *Eur J Biochem.* 2001;268(10):3053–3061.

192. Chung YM, Yoo YD, Park JK, Kim YT, Kim HJ. Increased expression of peroxiredoxin II confers resistance to cisplatin. *Anticancer Res.* 2001;21(2A):1129–1133.

193. Park SH, Chung YM, Lee YS, et al. Antisense of human peroxiredoxin II enhances radiation-induced cell death. *Clin Cancer Res.* 2000;6(12):4915–4920.

194. Zamora EA, Alkherayf F. Ependymoma. In: *StatPearls.* Treasure Island (FL): StatPearls Publishing; 2022.

195. Haapasalo T, Nordfors K, Jarvela S, et al. Peroxiredoxins and their expression in ependymomas. *J Clin Pathol.* 2013;66(1):12–17.

196. Berger TR, Wen PY, Lang-Orsini M, Chukwueke UN. World health organization 2021 classification of central nervous system tumors and implications for therapy for adult-type gliomas: a review. *JAMA Oncol.* 2022;8(10):1493–1501.

197. Marquet G, Dameron O, Saikali S, Mosser J, Burgun A. Grading glioma tumors using OWL-DL and NCI thesaurus. *AMIA.* 2007;2007:508–512.

198. Fan Z, Wirth AK, Chen D, et al. Nrf2-Keap1 pathway promotes cell proliferation and diminishes ferroptosis. *Oncogenesis.* 2017;6(8):e371.

199. Tsai WC, Hueng DY, Lin CR, Yang TC, Gao HW. Nrf2 expressions correlate with WHO grades in gliomas and meningiomas. *Int J Mol Sci* . 2016;17(5):722.

200. Haapasalo J, Nordfors K, Granberg KJ, et al. NRF2, DJ1 and SNRX1 and their prognostic impact in astrocytic gliomas. *Histol Histopathol.* 2018;33(8):791–801.

201. Han S, Liu Y, Cai SJ, et al. IDH mutation in glioma: molecular mechanisms and potential therapeutic targets. *Br J Cancer.* 2020;122(11):1580–1589.

202. Kanamori M, Higa T, Sonoda Y, et al. Activation of the NRF2 pathway and its impact on the prognosis of anaplastic glioma patients. *Neuro-Oncology.* 2015;17(4):555–565.

203. Liu Y, Lu Y, Celiku O, et al. Targeting IDH1-mutated malignancies with NRF2 blockade. *J Natl Cancer Inst.* 2019;111(10):1033–1041.

204. Wu W, Klockow JL, Zhang M, et al. Glioblastoma multiforme (GBM): An overview of current therapies and mechanisms of resistance. *Pharmacol Res.* 2021;171:105780.

205. Davis ME. Glioblastoma: overview of disease and treatment. *Clin J Oncol Nurs.* 2016;20(5 Suppl):S2–S8.

206. Thakkar JP, Dolecek TA, Horbinski C, et al. Epidemiologic and molecular prognostic review of glioblastoma. *Cancer Epidemiol Biomarkers Prev.* 2014;23(10):1985–1996.

207. Ostrom QT, Cioffi G, Gittleman H, et al. CBTRUS statistical report: primary brain and other central nervous system tumors diagnosed in the United States in 2012-2016. *Neuro-Oncology.* 2019;21(Suppl 5):v1–v100.

208. Nakada M, Kita D, Watanabe T, et al. Aberrant signaling pathways in glioma. *Cancers (Basel).* 2011;3(3):3242–3278.

209. Stupp R, Mason WP, van den Bent MJ, et al; European Organisation for Research and Treatment of Cancer Brain Tumor and Radiotherapy Groups. Radiotherapy plus concomitant and adjuvant temozolomide for glioblastoma. *N Engl J Med.* 2005;352(10):987–996.

210. Birzu C, French P, Caccese M, et al. Recurrent glioblastoma: from molecular landscape to new treatment perspectives. *Cancers (Basel).* 2020;13(1):47.

211. Awuah WA, Toufik A-R, Yarlagadda R, et al. Exploring the role of Nrf2 signaling in glioblastoma multiforme. *Discover. Oncology.* 2022;13(1):94.

212. Ji X, Wang H, Zhu J, et al. Knockdown of Nrf2 suppresses glioblastoma angiogenesis by inhibiting hypoxia-induced activation of HIF-1alpha. *Int J Cancer.* 2014;135(3):574–584.

213. Polonen P, Jawahar Deen A, Leinonen HM, et al. Nrf2 and SQSTM1/p62 jointly contribute to mesenchymal transition and invasion in glioblastoma. *Oncogene.* 2019;38(50):7473–7490.

214. Cong ZX, Wang HD, Wang JW, et al. ERK and PI3K signaling cascades induce Nrf2 activation and regulate cell viability partly through Nrf2 in human glioblastoma cells. *Oncol Rep.* 2013;30(2):715–722.

215. Rocha CRR, Reily Rocha A, Molina Silva M, et al. Revealing temozolomide resistance mechanisms via genome-wide CRISPR libraries. *Cells.* 2020;9(12):2573.

216. Tang Z, Li C, Kang B, et al. GEPIA: a web server for cancer and normal gene expression profiling and interactive analyses. *Nucleic Acids Res.* 2017;45(W1):W98–W102.

217. Verhaak RG, Hoadley KA, Purdom E, et al; Cancer Genome Atlas Research Network. Integrated genomic analysis identifies clinically relevant subtypes of glioblastoma characterized by abnormalities in PDGFRA, IDH1, EGFR, and NF1. *Cancer Cell.* 2010;17(1):98–110.

218. Noushmehr H, Weisenberger DJ, Diefes K, et al; Cancer Genome Atlas Research Network. Identification of a CpG island methylator phenotype that defines a distinct subgroup of glioma. *Cancer Cell.* 2010;17(5):510–522.

219. Behnan J, Finocchiaro G, Hanna G. The landscape of the mesenchymal signature in brain tumours. *Brain.* 2019;142(4):847–866.

220. Johannessen TC, Bjerkvig R, Tysnes BB. DNA repair and cancer stem-like cells--potential partners in glioma drug resistance? *Cancer Treat Rev.* 2008;34(6):558–567.

221. Ahmed AU, Auffinger B, Lesniak MS. Understanding glioma stem cells: rationale, clinical relevance and therapeutic strategies. *Expert Rev Neurother.* 2013;13(5):545–555.

222. Venere M, Fine HA, Dirks PB, Rich JN. Cancer stem cells in gliomas: identifying and understanding the apex cell in cancer's hierarchy. *Glia.* 2011;59(8):1148–1154.

223. Lathia JD, Heddleston JM, Venere M, Rich JN. Deadly teamwork: neural cancer stem cells and the tumor microenvironment. *Cell Stem Cell.* 2011;8(5):482–485.

224. Singer E, Judkins J, Salomonis N, et al. Reactive oxygen species-mediated therapeutic response and resistance in glioblastoma. *Cell Death Dis.* 2015;6(1):e1601.

225. Karkkainen V, Pomeshchik Y, Savchenko E, et al. Nrf2 regulates neurogenesis and protects neural progenitor cells against Abeta toxicity. *Stem Cells.* 2014;32(7):1904–1916.

226. Zhu J, Wang H, Ji X, et al. Differential Nrf2 expression between glioma stem cells and non-stem-like cells in glioblastoma. *Oncol Lett.* 2014;7(3):693–698.

227. Zhu J, Wang H, Fan Y, et al. Knockdown of nuclear factor erythroid 2-related factor 2 by lentivirus induces differentiation of glioma stem-like cells. *Oncol Rep.* 2014;32(3):1170–1178.

228. Zhu J, Wang H, Sun Q, et al. Nrf2 is required to maintain the self-renewal of glioma stem cells. *BMC Cancer.* 2013;13:380.

229. Moroishi T, Hansen CG, Guan KL. The emerging roles of YAP and TAZ in cancer. *Nat Rev Cancer.* 2015;15(2):73–79.

230. Escoll M, Lastra D, Pajares M, et al. Transcription factor NRF2 uses the Hippo pathway effector TAZ to induce tumorigenesis in glioblastomas. *Redox Biol.* 2020;30:101425.

231. Ludwig K, Kornblum HI. Molecular markers in glioma. *J Neurooncol.* 2017;134(3):505–512.

232. He J, Liu Y, Zhu T, et al. CD90 is identified as a candidate marker for cancer stem cells in primary high-grade gliomas using tissue microarrays. *Mol Cell Proteomics.* 2012;11(6):M111.010744.

233. Tang X, Zuo C, Fang P, et al. Targeting glioblastoma stem cells: a review on biomarkers, signal pathways and targeted therapy. *Front Oncol.* 2021;11:701291.

234. Ahmad F, Dixit D, Sharma V, et al. Nrf2-driven TERT regulates pentose phosphate pathway in glioblastoma. *Cell Death Dis.* 2016;7(5):e2213.

235. Stupp R, Tonn JC, Brada M, Pentheroudakis G. Esmo guidelines working group high-grade malignant glioma: ESMO clinical practice guidelines for diagnosis, treatment and follow-up. *Ann Oncol.* 2010;21(Suppl 5):vv190–vv193.

236. Friedman HS, Kerby T, Calvert H. Temozolomide and treatment of malignant glioma. *Clin Cancer Res.* 2000;6(7):2585–2597.

237. Singh N, Miner A, Hennis L, Mittal S. Mechanisms of temozolomide resistance in glioblastoma - a comprehensive review. *Cancer Drug Resist.* 2021;4(1):17–43.

238. Ribeiro Reily Rocha C, Reily Rocha A, Molina Silva M, et al. Revealing temozolomide resistance mechanisms via genome-wide CRISPR libraries. *Cells.* 2020;9(12):2573.

239. Zhang L, Wang H. FTY720 inhibits the Nrf2/ARE pathway in human glioblastoma cell lines and sensitizes glioblastoma cells to temozolomide. *Pharmacol Rep.* 2017;69(6):1186–1193.

240. Sun W, Zhang W, Yu J, Lu Z, Yu J. Inhibition of Nrf2 might enhance the anti-tumor effect of temozolomide in glioma cells via inhibition of Ras/Raf/MEK signaling pathway. *Int J Neurosci.* 2021;131(10):975–983.

241. Abou-Mrad Z, Bou Gharios J, Moubarak MM, et al. Central nervous system tumors and three-dimensional cell biology: Current and future perspectives in modeling. *World J Stem Cells.* 2021;13(8):1112–1126.

242. Johannessen TC, Bjerkvig R. Molecular mechanisms of temozolomide resistance in glioblastoma multiforme. *Expert Rev Anticancer Ther.* 2012;12(5):635–642.

243. Rocha CR, Kajitani GS, Quinet A, Fortunato RS, Menck CF. NRF2 and glutathione are key resistance mediators to temozolomide in glioma and melanoma cells. *Oncotarget.* 2016;7(30):48081–48092.

244. de Souza I, Monteiro LKS, Guedes CB, et al. High levels of NRF2 sensitize temozolomide-resistant glioblastoma cells to ferroptosis via ABCC1/MRP1 upregulation. *Cell Death Dis.* 2022;13(7):591.

245. Su I-C, Su Y-K, Setiawan SA, et al. NADPH oxidase subunit CYBB confers chemotherapy and ferroptosis resistance in mesenchymal glioblastoma via Nrf2/SOD2 Modulation. *Int J Mol Sci.* 2023;24(9):7706.

246. Godoy P, Pour Khavari A, Rizzo M, Sakamoto-Hojo ET, Haghdoost S. Targeting NRF2, regulator of antioxidant system, to sensitize glioblastoma neurosphere cells to radiation-induced oxidative stress. *Oxid Med Cell Longev.* 2020;2020(1):2534643.

247. Fekete JT, Győrffy B. ROCplotorg: validating predictive biomarkers of chemotherapy/hormonal therapy/anti-HER2 therapy using transcriptomic data of 3,104 breast cancer patients. *Int J Cancer.* 2019;145(11):3140–3151.