The Potential Therapeutic Target Nrf2 in Childhood Brain Tumors

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Abstract: Nuclear factor erythroid-2 related factor-2 (Nrf2) is a transcription factor, widely considered the master regulator of the antioxidant response. Under normal condition, its inhibitor, the Kelch-like ECH-associated protein 1 (Keap1), binds Nrf2, causing its cytoplasmatic retention and mediating Nrf2 ubiquitination followed by proteasomal degradation; under oxidative stress, Nrf2 dissociates from Keap1, translocates into the nucleus, and induces the expression of genes bearing Antioxidant Responsive Element (ARE) in their promoters, such as phase II detoxifying enzymes and antioxidant genes. In these years several studies confirmed that Nrf2 has a cancer-protective activity via its cytoprotective functions; however, growing body of evidence showed that it is frequently activated in many resistant tumors, as an adaptative response to the increased oxidative stress. This activation makes cancer cells more resistant to drug treatments. Thus, its inhibition could sensitize tumor cells to pro-oxidant therapies, suggesting that the Nrf2 inhibition, through chemical inhibitors or RNA interference strategies, is a promising tool in cancer therapy.

As observed in many cancers, Nrf2 plays a role in the progression of brain tumors, such as astrocytomas, multiforme glioblastomas, gliosarcomas, medulloblastomas, oligodendroglial and ependymal tumors. However, most of the research focusing on the mechanisms in the regulation of Nrf2, as well as on its potential value in the cancer treatment, have been carried out in adult glioblastomas. Thus, little is known about the role of Nrf2 in most of the common childhood brain tumors. Here, we summarize and discuss findings on Nrf2 in common pediatric CNS tumors, as well as the clinical perspectives in using Nrf2 inhibitors for cancer CNS treatment.

Keywords: Nrf2, Childhood Brain Tumors.

1. NRF2: THE MASTER REGULATOR OF ANTIOXIDANT RESPONSE

Nuclear factor erythroid-2 related factor-2 (Nrf2) is a Cap'n'collar basic leucine zipper transcription factor, widely considered the master regulator of the antioxidant response [1] Nrf2 acts by binding and positively regulating the expression of genes bearing Antioxidant Responsive Element (ARE) in their promoter. The Nrf2-regulated transcriptional program essentially controls the genes involved in the redox homeostasis, xenobiotics detoxification and multidrug-resistance [2]. Although Nrf2 has been found constitutively expressed in various tissues and cell lines [3], its activity is strictly controlled by posttranslational mechanisms. In particular Kelch-like ECH-associated protein 1 (Keap1) binds Nrf2, causing its cytoplasmatic retention and mediating Nrf2 ubiquitination followed by proteasomal degradation [4]. Keap1 is a cysteine-rich and redox-sensitive protein containing five functional domains, which include an N-terminal region (NTR), a broad-complex, tramtrack, bric a' brac (BTB) homodimerization domain, a cysteine rich intervening region (IVR), a kelch/double glycine repeat (DGR) domain (harboring six Kelch repeats), and a C-terminal region (CTR) [5]. The IVR domain contains highly reactive cysteine residues, such as Cys273, Cys288, and Cys297, which are easily oxidized and are thus responsible for sensing oxidative stress [6]. Under oxidative stress, these highly reactive cysteine residues in Keap1 are oxidized, and Keap1 releases Nrf2 which translocates in the nucleus, where it heterodimerizes with the small Musculo Aponeurotic Fibrosarcoma protein (sMAF), and binds to ARE, inducing the transcription of a battery of cytoprotective proteins [7] (Figure **1**). These include several Phase II detoxify-cation enzymes, such as glutathione-S transferases (GSTs), NADPH quinine oxidoreductase (NQO1), glutathione peroxidases (GPx), catalase, superoxide dismutases (SODs), epoxide hydrolase, heme oxygenase (HO1), UDP-glucuronosyl transferases (UGTs), and gamma-glutamylcysteine synthetase (GCL), as well as many antioxidant genes such as the Glutamate-cysteine ligase, catalytic subunit (GCLC), glutaredoxin 1 (GLRX), glutathione reductase (GRS1), and peroxiredoxins 1 and 6 (PRDX1-6) [8-9]. Nrf2 induces the expression of several enzymes involved in the synthesis of glutathione (GSH), the most important low-molecular weight antioxidant synthesized in cells [10]. The detoxification and the antioxidant enzymes protect normal cells under stress conditions, have the ability to inhibit damage to DNA, proteins, and lipids [11] and contribute to the generation of new tissue [12].

2. NRF2 AND CANCER

The interest on the Nrf2 role in cancer has grown exponentially over the past eight years (Figure **2**). Only in January 2018, we found 82 publications. If this

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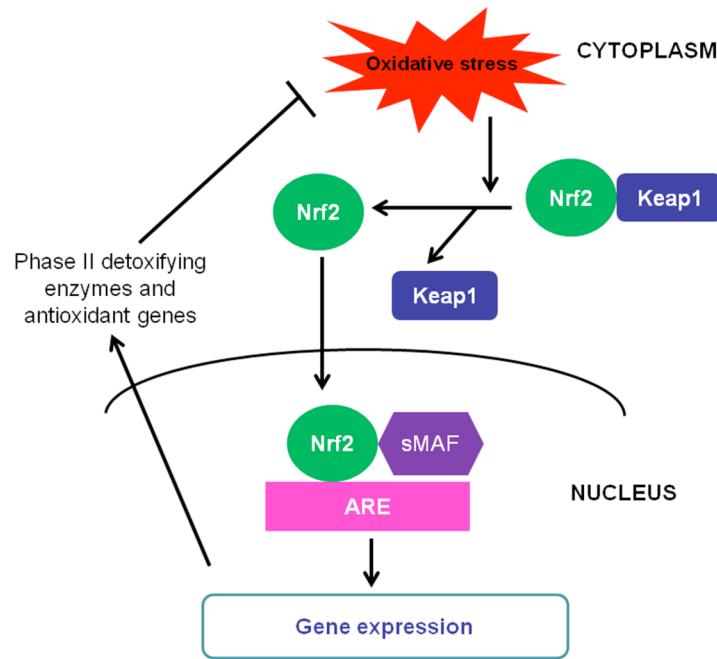
Figure 1: Schematic representation of Nrf2 function and regulation. Under normal condition, Keap1 binds Nrf2, causing its cytoplasmatic retention and mediating Nrf2 ubiquitination followed by proteasomal degradation; under oxidative stress, Nrf2 dissociates from Keap1, translocates into the nucleus, binds to ARE, and transactivates phase II detoxifying enzymes and antioxidant genes.
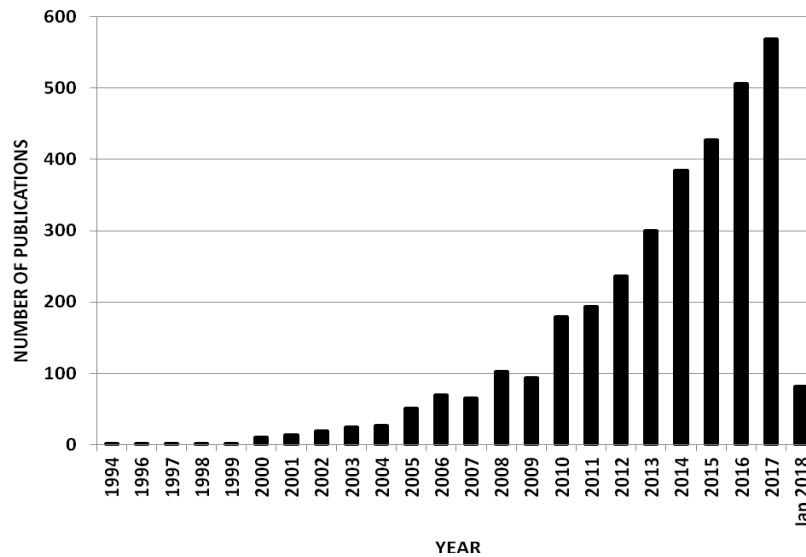

Figure 2: Number of publications per year on Nrf2 and Cancer. Number of publications per year obtained on PubMed with the following query: "(cancer OR Tumor) and Nrf2". The last bar represents the number of publications relative to January 2018.

number was kept constant monthly for the rest of the year, we would reach about 1000 publications on this subject for 2018.

In these years several studies confirmed that Nrf2 has a cancer-protective activity via its cytoprotective functions, since it protects normal cells exposed to oxidative stress and xenobiotics, from DNA damage

and destruction of the lipids, carbohydrates, and proteins [13]. It has been demonstrated that Nrf2-null mice also had decreased levels of phase II detoxifying enzymes and exhibited elevated sensitivity to the carcinogens and chemical toxicants [14]. On the contrary, recent studies have demonstrated that aberrant activation of NRF2, due to genetic and/or epigenetic mutations in tumor, contributes to the

decreased therapeutic efficacy of anticancer drugs through biotransformation or extrusion during chemotherapy [9, 15]. The increase of Nrf2 expression can be related to the increased levels of reactive oxygen species (ROS), which has been observed in many types of cancer cells [16]. A moderate increase in ROS can promote cell proliferation and differentiation [17], whereas excessive amounts of ROS can cause oxidative damage to lipids, proteins and DNA [18]. An increase in ROS, associated with abnormal cancer cell growth, reflects a disruption of redox homeostasis and an increase of oxidative stress. Under persistent intrinsic oxidative stress, during cancer progression, many cancer cells become well-adapted to such stress and develop an enhanced, endogenous antioxidant capacity, which makes the malignant cells resistant to exogenous stress [19]. These intrinsically resistant cancer cells display high levels of Nrf2 expression which provides a growth advantage and diminishes the cytotoxic effects of anticancer agents [20]. Analogously, high expression of Nrf2 was also found in numerous cancer cell lines, which develop drug-resistant phenotypes during chemotherapy [9, 15]. These resistant phenotypes can be induced to a great extent by anticancer drugs, such as such as alkylating agents and platinum compounds [21- 22]. Indeed, in chemo resistance induction, Nrf2 may act in different ways. Besides mediating stress-stimulated induction of antioxidant and detoxification genes, Nrf2 contributes to cancer cell resistance by up regulating the repair and degradation of damaged macromolecules, and by modulating intermediary metabolism [23]. Moreover, Nrf2 controls some genes belonging to the ABC (ATP-binding cassette) super family of drug transporters. Bao *et al*. demonstrated that ABCF2, a cytosolic member of the ABC super family, involved in chemo resistance in clear cell ovarian cancer, contains ARE sequences in its promoter region, establishing ABCF2 as an NRF2 target gene [24]. Canet and collaborators [25] identified a functional Nrf2 response element within the eighth intron of the ABCC3 gene, which may provide mechanistic insight to the Nrf2 induction of ABCC3, during antioxidant response stimuli. Finally, Ishikawa *et al*. demonstrated that Nrf2 interacts with the antioxidant responsive element (ARE) located in the promoter region of the human ABCG2 gene [26].

Finally, several studies identified cross-talk between Nrf2 and other signaling pathways involved in tumor progression (i.e. Yap, NF-kB Notch,) [22, 27-28], which provides extended functions of Nrf2 in regulating normal homeostasis as well as its contribution in the cancer dual role.

3. NRF2 IN CHILDHOOD BRAIN TUMOR

Central Nervous System (CNS) tumors are the second most common childhood cancers, after hematologic malignancies, representing 20 to 25% of all childhood cancers [29-31]. Although progress has been made in the treatment of pediatric tumors, significant mortality is still associated with malignant brain tumors.

The universally adopted WHO classification of CNS tumors is based not only on histopathologic appearance, but also on well-established molecular parameters, such as the presence of mutated genes (i.e. isocitrate dehydrogenase- IDH, or WNT) [32]. The grading system, besides the classical morphological features (number of mitotic figures, the presence of necrosis, giant cells, vascular proliferation, hyperchromatic nuclei, the degree of pleomorphism), also includes these new molecular parameters [32]. Low-grade CNS cancers encompass WHO grade I and grade II tumors, whereas WHO grade III and IV are considered high-grade CNS malignancies [32].

The largest subgroup, in pediatric CNS tumor, is represented by gliomas. They account for 40-50% of all primary CNS tumors in children and they show an extremely broad range of clinical behavior. Most of the gliomas are diagnosed as low-grade and they are frequently indolent and do not undergo malignant transformation. Thus, they show excellent overall survival under current treatment strategies. However, some of these gliomas develop over a short period, progress rapidly and are therefore classified as WHO grade III or IV high-grade gliomas. Despite all therapeutic efforts, the mortality rate is very high, with the most aggressive forms being lethal within months [33].

The low-grade pilocytic astrocytoma represents the 33.2% of gliomas, followed by other low-grade gliomas (27.1%), high-grade gliomas (i.e. anaplastic astrocytoma, and glioblastoma multiforme, GBM) (21.0%), and ependymal tumors (10.4%). The second most common group is represented by the embryonal tumors (15% of all childhood brain and CNS tumors). Most of these are medulloblastomas, followed by atypical teratoid / rhabdoid tumors (ATRT) (15.0%), and primitive neuroectodermal tumors (PNET) (14.9%) [29, 34].

The dual role of Nrf2 has been elucidated in cells of nervous system similarly to that observed in several types of tissues. From one hand, Nrf2 has a neuroprotective action [35-36], from the other hand it can contribute to the cancer progression and chemoresistance in several CNS tumors [37-39]. Moreover, Tsai and collaborators observed that higher scores of Nrf2 immunostaining in primary CNS tumors are significantly correlated with more advanced World Health Organization (WHO) grades [40] (Figure **3**). These authors analyzed 72 cases of astrocytic tumors of various WHO grades, and they found that the average expression scores of Nrf2 were 19.29 in pilocytic astrocytomas (WHO grade I), 75.98 in diffuse astrocytomas (WHO grade II), 114.00 in anaplastic astrocytomas (WHO grade III), 160.56 in glioblastomas multiforme (WHO grade IV), and 42.50 in gliosarcomas (WHO grade IV). Statistical analysis revealed a positive correlation between the Nrf2 expression score and WHO grade for astrocytic tumors. Similar results were obtained for oligodendroglial and ependymal tumors, where the average expression score of Nrf2 in anaplastic type (WHO grade III) was significantly higher than that in the corresponding lower WHO grade II. Among the neuroepithelial tumors, the WHO grade IV medulloblastoma showed the highest score of Nrf staining (170), similar to the values obtained for anaplastic ependymoma (171.11). More interestingly, patients whose glioma had higher Nrf2 expression tended to have lower overall survival (OS), although the association did not reach statistical significance [40].

The pro-tumoral role of Nrf2 has been extensively studied in glioblastoma multiforme (GMB), the most common and aggressive among all gliomas in adults, as recently reviewed [38]. Several reports have elucidated its contribution to chemo- and radio-resistance, proliferation, invasion, migration, apoptosis, differentiation, and autophagy in GMB. Moreover, Nrf2 modulation can influence the tumor microenvironment. For instance it has been demonstrated that Nrf2 enhances angiogenesis; its activation induced up-regulation of HO-1, and mediated the cellular adaptive survival response to a hypoxic conditions; moreover, it may regulate tumor immunosurveillance through regulation of the secretion of cytokines (IL-4, IL-5, and IL-13), affecting the function of immune cells and contributing to the immune escape [38]. For these reasons, several authors proposed Nrf2 as a promising therapeutic target in GMB.

In contrast to the large literature on adult GMB, little is known about the role of Nrf2 in most of the common pediatric CNS tumors. For instance, few experimental papers are dealing with medulloblastoma and Nrf2. One of these confirmed that the expression levels of Nrf-2 and HO-1 in medulloblastoma young patients were significantly increased, compared with those observed in peritumoral control brain tissues, according to the studies on GMB [41]. A different approach was prosecuted by Koto and collaborators [42], who evaluated the anti-tumoral activity of two Nrf2 activators, nifurtimox and Tetrathiomolybdate (TM), in two medulloblastoma cell lines, D283 and DAOY. The results obtained with a combined treatments were encouraging: these two drugs synergistically decreased medulloblastoma cell viability and induced cellular apoptosis. Consistent with the robust reactive oxygen species (ROS) production, these authors demonstrated
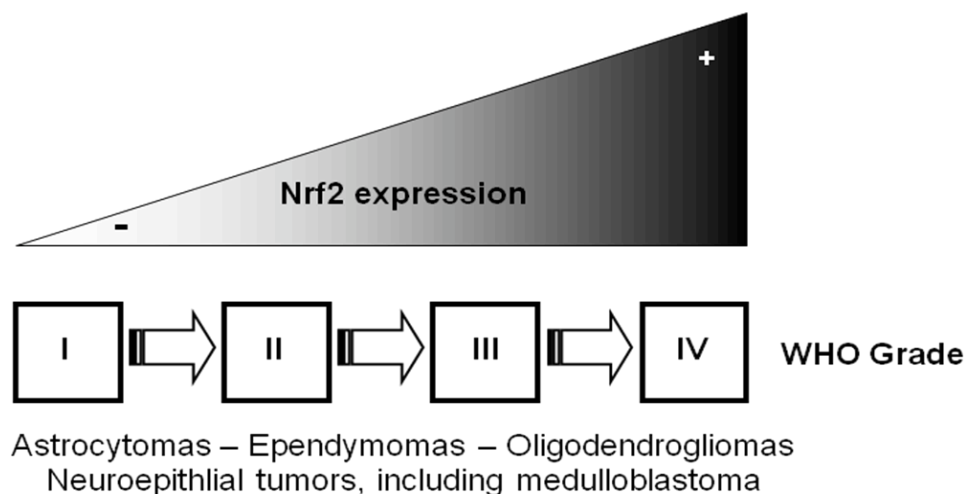

Figure 3: Schematic representation of Nrf2 immunostaining in primary CNS tumors that significantly correlated with more advanced World Health Organization (WHO) grades, as demonstrated by Tsai and collaborators [38].

that the drug combination caused the up-regulation of several target genes of the Nrf2 pathway. Although the encouraging results of this therapeutic approach, we believe that the activation of Nrf2 might be considered with caution. Indeed, as mentioned above, Nrf2 is involved in chemo-radio resistance, thereby its up-regulation might promote tumor recurrence [9, 15]. At this regards, there is an interesting paper by Cong and collaborators [43], demonstrating that the standard care for newly diagnosed glioblastoma, including radio-therapy plus concomitant and adjuvant temozolomide (TMZ, may improve the survival rated of patients, but it leads to the activation of Nrf2, which in turn might decrease chemo-radio sensitivity and promote glioblastoma recurrence.

At the moment there are not specific studies on Nrf2 in pediatric ependymoma. However, a very recent paper investigated, in adult ependymoma patients (median age 33 years), the immunohistochemical Nrf2 expression, together with peroxiredoxins (Prxs), a very large and highly conserved family of antioxidant enzymes, able to reduce peroxides [44]. In particular, they studied 17 myxopapillary ependymomas (WHO grade I), 45 grade II ependymomas and 14 anaplastic ependymomas (WHO grade III) [45]. Strong nuclear and cytoplasmic expression of Nrf2 could be detected in these tumors and Prx I, a Nrf2 target, expression was significantly associated with cytoplasmic and nuclear Nrf2 expression. In contrast to what reported by Tsai and collaborators [40], neither nuclear Nrf2 nor Prxs expressions did not correlate with histological grade.

Functional studies of Nrf2 on pilocytic astrocytoma, one of the most common pediatric CNS malignancies, are also missing. In consideration of its low expression with respect to higher WHO grade gliomas [40], it is very likely that it can have a marginal role in the progression of this tumor.

Although the Nrf2 pathway in pediatric CNS tumors needs to be further investigated, a growing body of evidence suggests that it is frequently activated in many CNS tumors, and that its inhibition generally could sensitize tumor cells to pro-oxidant therapies, suggesting that Nrf2 inhibition is a promising strategy for cancer therapy.

4. CLINICAL PERSPECTIVES

Although consistent literature data highlight the possibility to use Nrf inhibitors to treat cancers [9, 46-

48], there are not ongoing clinical trials addressing this specific issue. One of the reasons is that although several anti-tumor agents that display activity as Nrf2 inhibitor (i.e. ascorbic acid AA, all-trans-retinoic acid ATRA, metformin, glucocorticoids, brusatol, trigonelline, luteolin), they have poor specificity, low bioactivity, and toxicity, which represent major obstacles in developing clinical therapies. Moreover, the treatment of CNS tumors needs to consider an adjunctive crucial feature: the ability of the drug to cross the brain blood barrier (BBB). In this context, nanomedicine can play an important role by overcoming the above limitations [49]. Indeed, nanoparticles can penetrate and facilitate the drug delivery through the BBB, because in is possible to modulate their shape, size, hydrophobicity, coating, chemistry and surface charge. They can transport a variety of natural or synthetic compounds [50], and they represent an interesting therapeutical opportunity for the treatment of brain tumors [51].

Recently, nanoparticles have received attention for their use in RNA interference (RNAi) strategies, based on the highly specific and efficient silencing of a target gene. Indeed, these nanovectors can carry small RNA oligonucleotides, such as microRNA (miRNA) or small interfering RNA (siRNA) [52], and they have also been proposed for the treatment of glioblastoma [53]. Thus, in the absence of a specific Nrf2 inhibitor, the *in vivo* delivery of a specific siRNA targeting this gene, can represent an interesting strategy for treating CNS tumors.

In conclusion, although little is known about the role of Nrf2 in most of the common pediatric CNS cancers, there is a strong suggestion for a clinical application of Nrf2 inhibitors in these type of tumors. Research program needs to fill the gaps and deepen the role of this gene in childhood brain tumor and to consider the development of new specific Nrf2 inhibitors able to cross the BBB.

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REFERENCES

[1]   Moi P, Chan K, Asunis I, Cao A and Kan YW. Isolation of NF-E2-related factor 2 (Nrf2), a NF-E2-like basic leucine zipper transcriptional activator that binds to the tandem NF-E2/AP1 repeat of the β-globin locus control region. Proc Natl Acad Sci USA 1994; 91: 9926-30.
https://doi.org/10.1073/pnas.91.21.9926

[2] Hayes JD and Dinkova-Kostova AT. The Nrf2 regulatory network provides an interface between redox and intermediary metabolism. Trends Biochem Sci 2014; 39: 199-218.
https://doi.org/10.1016/j.tibs.2014.02.002

[3] Nguyen T, Yang CS and Pickett CB. The pathways and molecular mechanisms regulating Nrf2 activation in response to chemical stress. Free Radic Biol Med 2004; 37(4): 433-41.
https://doi.org/10.1016/j.freeradbiomed.2004.04.033

[4] Katoh Y, Iida K, Kang MI, Kobayashi A, Mizukami M, Tong KI, et al. Evolutionary conserved N-terminal domain of Nrf2 is essential for the Keap1-mediated degradation of the protein by proteasome. Arch Biochem Biophys 2005; 433: 342-50.
https://doi.org/10.1016/j.abb.2004.10.012

[5] Itoh K, Mimura J and Yamamoto M. Discovery of the negative regulator of Nrf2, Keap1: a historical overview. Antioxid Redox Signal 2010; 13: 1665-78.
https://doi.org/10.1089/ars.2010.3222

[6] Dinkova-Kostova AT, Holtzclaw WD, Cole RN, Itoh K, Wakabayashi N, Katoh Y, et al. Direct evidence that sulfhydryl groups of Keap1 are the sensors regulating induction of phase 2 enzymes that protect against carcinogens and oxidants. Proc Natl Acad Sci USA 2002; 99: 11908-13.
https://doi.org/10.1073/pnas.172398899

[7] Taguchi K, Motohashi H and Yamamoto M. Molecular mechanisms of the Keap1–Nrf2 pathway in stress response and cancer evolution. Genes Cells 2011; 16: 123-40.
https://doi.org/10.1111/j.1365-2443.2010.01473.x

[8] Pool-Zobel B, Veeriah S and Böhmer FD. Modulation of xenobiotic metabolising enzymes by anticarcinogens—focus on glutathione S-transferases and their role as targets of dietary chemoprevention in colorectal carcinogenesis Mutat Res Fund Mol Mech Mut 2005; 591(1): 74-92.

[9] Furfaro AL, Traverso N, Domenicotti C, Piras S, Moretta L, Marinari UM, et al. The Nrf2/HO-1 Axis in Cancer Cell Growth and Chemoresistance. Oxid Med Cell Longev 2016; 2016: 1958174.
https://doi.org/10.1155/2016/1958174

[10] Chan JY and Kwong M. Impaired expression of glutathione synthetic enzyme genes in mice with targeted deletion of the Nrf2 basic-leucine zipper protein. Biochim Biophys Acta: Gene Struct Expr 2000; 1517: 19-26.
https://doi.org/10.1016/S0167-4781(00)00238-4

[11] Schäfer M, Dütsch S, auf dem Keller U and Werner S. Nrf2: a central regulator of UV protection in the epidermis. Cell Cycle 2010; 9: 2917-8.
https://doi.org/10.4161/cc.9.15.12701

[12] Beyer TA, Xu W, Teupser D, auf dem Keller U, Bugnon P, Hildt E, et al. Impaired liver regeneration in Nrf2 knockout mice: role of ROS-mediated insulin/IGF-1 resistance. EMBO J 2008; 27: 212-23.
https://doi.org/10.1038/sj.emboj.7601950

[13] Kwak MK and Kensler TW. Targeting NRF2 signaling for cancer chemoprevention. Toxicol Appl Pharmacol 2010; 244: 66-76.
https://doi.org/10.1016/j.taap.2009.08.028

[14] Chanas SA, Jiang Q, McMahon M, McWalter GK, McLellan LI, Elcombe CR, et al. Loss of the Nrf2 transcription factor causes a marked reduction in constitutive and inducible expression of the glutathione S-transferase Gsta1, Gsta2, Gstm1, Gstm2, Gstm3 and Gstm4 genes in the livers of male and female mice. Biochem J 2002; 365(Pt 2): 405-16.
https://doi.org/10.1042/bj20020320

[15] Bai X, Chen Y, Hou X, Huang M and Jin J. Emerging role of NRF2 in chemoresistance by regulating drug-metabolizing enzymes and efflux transporters. Drug Metab Rev 2016; 48: 541-67.
https://doi.org/10.1080/03602532.2016.1197239

[16] Szatrowski TP and Nathan CF. Production of large amounts of hydrogen peroxide by human tumor cells. Cancer Res 1991; 51: 794-8.

[17] Boonstra J and Post JA. Molecular events associated with reactive oxygen species and cell cycle progression in mammalian cells. Gene 2004; 337: 1-13.
https://doi.org/10.1016/j.gene.2004.04.032

[18] Perry G, Raina AK, Nunomura A, Wataya T, Sayre LM and Smith MA. How important is oxidative damage? Lessons from Alzheimer's disease. Free Radic Biol Med 2000; 28: 831-4.
https://doi.org/10.1016/S0891-5849(00)00158-1

[19] Pervaiz S and Clement MV. Tumor intracellular redox status and drug resistance-serendipity or a causal relationship? Curr Pharm Des 2004; 10: 1969-77.
https://doi.org/10.2174/1381612043384411

[20] Zhang DD. The Nrf2-Keap1-ARE signaling pathway: the regulation and dual function of Nrf2 in cancer. Antioxid Redox Signal 2010; 13: 1623-26.
https://doi.org/10.1089/ars.2010.3301

[21] Wang XJ, Hayes JD and Wolf CR. Generation of a stable antioxidant response element-driven reporter gene cell line and its use to show redox-dependent activation of nrf2 by cancer chemotherapeutic agents. Cancer Res 2006; 66: 10983-94.
https://doi.org/10.1158/0008-5472.CAN-06-2298

[22] Ciamporcero E, Daga M, Pizzimenti S, Roetto A, Dianzani C, Compagnone A, et al. Crosstalk between Nrf2 and YAP contributes to maintaining the antioxidant potential and chemoresistance in bladder cancer. Free Radic Biol Med 2018; 115: 447-57.
https://doi.org/10.1016/j.freeradbiomed.2017.12.005

[23] Hayes JD and Dinkova-Kostova AT. The Nrf2 regulatory network provides an interface between redox and intermediary metabolism. Trends Biochem Sci 2014; 39: 199-218.
https://doi.org/10.1016/j.tibs.2014.02.002

[24] Bao L, Wu J, Dodson M, Rojo de la Vega EM, Ning Y, Zhang Z, et al. ABCF2, an Nrf2 target gene, contributes to cisplatin resistance in ovarian cancer cells. Mol Carcinog 2017; 56: 1543-53.
https://doi.org/10.1002/mc.22615

[25] Canet MJ, Merrell MD, Harder BG, Maher JM, Wu T, Lickteig AJ, et al. Identification of a functional antioxidant response element within the eighth intron of the human ABCC3 gene. Drug Metab Dispos 2015; 43: 93-9.
https://doi.org/10.1124/dmd.114.060103

[26] Ishikawa T, Kajimoto Y, Sun W, Nakagawa H, Inoue Y, Ikegami Y, et al. Role of Nrf2 in cancer photodynamic therapy: regulation of human ABC transporter ABCG2. J Pharm Sci 2013; 102: 3058-69.
https://doi.org/10.1002/jps.23563

[27] Wardyn JD, Ponsford AH and Sanderson CM. Dissecting molecular cross-talk between Nrf2 and NF-κB response pathways. Biochem Soc Trans 2015; 43(4): 621-6.
https://doi.org/10.1042/BST20150014

[28] Wakabayashi N, Chartoumpekis DV and Kensler TW. Crosstalk between Nrf2 and Notch signaling. Free Radic Biol Med 2015; 88(Pt B): 158-167.

[29] Ostrom QT, de Blank PM, Kruchko C, Petersen CM, Liao P, Finlay JL, et al. Alex's Lemonade Stand Foundation Infant and Childhood Primary Brain and Central Nervous System Tumors Diagnosed in the United States in 2007-2011. Neuro Oncol 2015; 16 Suppl 10: x1-x36.
https://doi.org/10.1093/neuonc/nou327

[30] Linabery AM and Ross JA. Trends in childhood cancer incidence in the U.S. (1992-2004). Cancer 2008; 112: 416-32.
https://doi.org/10.1002/cncr.23169

[31]   Kaatsch P. Epidemiology of childhood cancer. Cancer Treat Rev 2010; 36(4): 277-85.
https://doi.org/10.1016/j.ctrv.2010.02.003

[32]   Louis DN, Perry A, Reifenberger G, von Deimling A, Figarella-Branger D, Cavenee WK, et al. The 2016 World Health Organization Classification of Tumors of the Central Nervous System: a summary. Acta Neuropathol 2016; 131: 803-20.
https://doi.org/10.1007/s00401-016-1545-1

[33]   Waanders AJ, Minturn JE, Fisher MJ. Pediatric Low-Grade Gliomas: Diagnosis, Treatment, and Future Directions. In: Hayat M. (eds) Tumors of the Central Nervous System, Volume 14. Tumors of the Central Nervous System, Springer, Dordrecht 2015; 14: 13-24.
https://doi.org/10.1007/978-94-017-7224-2_2

[34]   Johnson KJ, Cullen J, Barnholtz-Sloan JS, Ostrom QT, Langer CE, Turner MC, et al. Childhood brain tumor epidemiology: a brain tumor epidemiology consortium review. Cancer Epidemiol Biomarkers Prev 2014; 23: 2716-36.
https://doi.org/10.1158/1055-9965.EPI-14-0207

[35]   González-Burgos E, Carretero ME and Gómez-Serranillos MP. Involvement of Nrf2 signaling pathway in the neuroprotective activity of natural kaurane diterpenes. Neuroscience 2013; 231: 400-12.
https://doi.org/10.1016/j.neuroscience.2012.10.018

[36]   Lawal AO and Ellis EM. Nrf2-mediated adaptive response to cadmium-induced toxicity involves protein kinase C delta in human 1321N1 astrocytoma cells. Environ Toxicol Pharmacol 2011; 32(1): 54-62.
https://doi.org/10.1016/j.etap.2011.03.010

[37]   Pan H, Wang H, Zhu L, Wang X, Cong Z, Sun K, et al. The involvement of Nrf2-ARE pathway in regulation of apoptosis in human glioblastoma cell U251. Neurol Res 2013; 35: 71-8.
https://doi.org/10.1179/1743132812Y.0000000094

[38]   Zhu J, Wang H, Fan Y, Lin Y, Zhang L, Ji X, et al. Targeting the NF-E2-related factor 2 pathway: a novel strategy for glioblastoma (review). Oncol Rep 2014; 32(2): 443-50.
https://doi.org/10.3892/or.2014.3259

[39]   Ji X, Wang H, Zhu J, Zhu L, Pan H, Li W, et al. Knockdown of Nrf2 suppresses glioblastoma angiogenesis by inhibiting hypoxia-induced activation of HIF-1α. Int J Cancer 2014; 135: 574-84.
https://doi.org/10.1002/ijc.28699

[40]   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): pii: E722.
https://doi.org/10.3390/ijms17050722

[41]   Tang L, DengY, Gao M.N, Lin X, Zhu J, Li Y. Nrf-2 and HO-1 Expression in Medulloblastoma: A Clinicopathological Analysis. Journal of Biosciences and Medicines 2017; 5: 142-47.
https://doi.org/10.4236/jbm.2017.53015

[42]   Koto KS, Lescault P, Brard L, Kim K, Singh RK, Bond J, et al. Antitumor activity of nifurtimox is enhanced with tetrathiomolybdate in medulloblastoma. Int J Oncol 2011; 38: 1329-41.

[43]   Cong ZX, Wang HD, Zhou Y, Wang JW, Pan H, Zhang DD, et al. Temozolomide and irradiation combined treatment-induced Nrf2 activation increases chemoradiation sensitivity in human glioblastoma cells. J Neurooncol 2014; 116(1): 41-8.
https://doi.org/10.1007/s11060-013-1260-x

[44]   Rhee SG. Overview on Peroxiredoxin. Molecules and Cells 2016; 39(1): 1-5.
https://doi.org/10.14348/molcells.2016.2368

[45]   Haapasalo T, Nordfors K, Järvelä S, Kok E, Sallinen P, Kinnula VL, et al. Peroxiredoxins and their expression in ependymomas. J Clin Pathol 2013; 66(1): 12-7.
https://doi.org/10.1136/jclinpath-2012-201048

[46]   Jung BJ, Yoo HS, Shin S, Park YJ and Jeon SM. Dysregulation of NRF2 in Cancer: from Molecular Mechanisms to Therapeutic Opportunities. Biomol Ther (Seoul) 2018; 26(1): 57-68.
https://doi.org/10.4062/biomolther.2017.195

[47]   Zhu J, Wang H, Chen F, Fu J, Xu Y, Hou Y, et al. An overview of chemical inhibitors of the Nrf2-ARE signalling pathway and their potential applications in cancer therapy. Free Radic Biol Med 2016; 99: 544-56.
https://doi.org/10.1016/j.freeradbiomed.2016.09.010

[48]   Catanzaro E, Calcabrini C, Turrini E, Sestili P and Fimognari C. Nrf2: a potential therapeutic target for naturally occurring anticancer drugs? Expert Opin Ther Targets 2017; 21: 781-793.
https://doi.org/10.1080/14728222.2017.1351549

[49]   Jain KK. Nanobiotechnology-based strategies for crossing the blood-brain barrier. Nanomedicine (Lond) 2012; 7: 1225-33.
https://doi.org/10.2217/nnm.12.86

[50]   Xin Y, Yin M, Zhao L, Meng F and Luo L. Recent progress on nanoparticle-based drug delivery systems for cancer therapy. Cancer Biology & Medicine 2017; 14(3): 228-41.
https://doi.org/10.20892/j.issn.2095-3941.2017.0052

[51]   Chakroun RW, Zhang P, Lin R, Schiapparelli P, Quinones-Hinojosa A and Cui H. Nanotherapeutic systems for local treatment of brain tumors. Wiley Interdiscip Rev Nanomed Nanobiotechnol 2018; 10(1).
https://doi.org/10.1002/wnan.1479

[52]   Xin Y, Huang M, Guo WW, Huang Q, Zhang LZ and Jiang G. Nano-based delivery of RNAi in cancer therapy. Molecular Cancer 2017; 16: 134.
https://doi.org/10.1186/s12943-017-0683-y

[53]   Lozada-Delgado EL, Grafals-Ruiz N and Vivas-Mejía PE. RNA interference for glioblastoma therapy: Innovation ladder from the bench to clinical trials. Life Sci. 2017; 188: 26-36.
https://doi.org/10.1016/j.lfs.2017.08.027