Biotechnology Advances xxx (xxxx) xxx–xxx

Contents lists available at ScienceDirect

Biotechnology Advances

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

Research review paper

Nrf2 as regulator of innate immunity: A molecular Swiss army knife!

Maurizio Battino[a], Francesca Giampieri[a], Francesca Pistollato[i], Antoni Sureda[b,*], Marcos Roberto de Oliveira[c], Valeria Pittalà[d], Francesca Fallarino[e], Seyed Fazel Nabavi[f], Atanas Georgiev Atanasov[g,h], Seyed Mohammad Nabavi[f,**]

[a] Dipartimento di Scienze Cliniche Specialistiche ed Odontostomatologiche (DISCO)-Sez. Biochimica, Facoltà di Medicina, Università Politecnica delle Marche, Via Ranieri 65, 60131 Ancona, Italy
[b] Research Group on Community Nutrition and Oxidative Stress (NUCOX) and CIBEROBN (Physiopathology of Obesity and Nutrition CB12/03/30038), University of Balearic Islands, Palma de Mallorca E-07122, Balearic Islands, Spain
[c] Department of Chemistry, ICET, Federal University of Mato Grosso (UFMT), Av. Fernando Corrêa da Costa, 2367, CEP 78060-900, Cuiabá, MT, Brazil
[d] Department of Drug Sciences, University of Catania, V.le A. Doria, 95125 Catania, Italy
[e] Department of Experimental Medicine, University of Perugia, 06132 Perugia, Italy
[f] Applied Biotechnology Research Center, Baqiyatallah University of Medical Sciences, Tehran, Iran
[g] Department of Pharmacognosy, University of Vienna, Althanstrasse 14, Vienna 1090, Austria
[h] Institute of Genetics and Animal Breeding of the Polish Academy of Sciences, Postępu 36A Street, 05-552 Jastrzebiec, Poland
[i] Centre for Nutrition and Health, Universidad Europea Del Atlántico (UEA), Santander, Spain

ARTICLE INFO

Keywords:
Immune system
Inflammation
Natural compounds
Nuclear factor
Oxidative stress

ABSTRACT

Organisms are constantly exposed to a broad range of pathological and stress-inducing agents, allergens and environmental chemicals that can induce infections, toxicity or other undesirable reactions. Our immune system has evolved over time in order to efficiently respond to these exogenous insults and maintain homeostasis. In particular, the innate immune system acts as primary barrier to prevent the entrance of invasive agents or allergens. This system is comprised of a diversity of cell types that are rapidly activated by recognition of common structures present in many potential pathogens known as pathogen-associated molecular patterns (PAMPs). The nuclear factor erythroid 2 related factor 2 (Nrf2) is a relevant basic leucine zipper (bZIP) transcription factor that is essential in the regulation of cell cycle homeostasis, cytoprotection, and innate immunity when cells are under stressful conditions. Although the role of Nrf2 in activating the expression of protective genes – such as antioxidant or anti-inflammatory – is known, its role in innate immunity and immune-related gene expression remains not yet clear. The present review summarizes current knowledge on Nrf2 signaling pathway structure and activity under both physiological state and upon oxidative stress. In addition, the relation between Nrf2 signaling pathway and the innate immune system is discussed, highlighting the potential therapeutic effects of diverse natural and synthetic compounds as Nrf2 regulators.

1. Introduction

The evolutionary process has contributed over time to the development of defense mechanisms against adverse endogenous and exogenous agents. A number of transcription factors are tangled in sustaining cell defense program and, among them, nuclear factor erythroid 2 related factor 2 (Nrf2) is one of the most relevant. Increasingly knowledge-driven data sustain crucial role of Nrf2 in the preservation of the endogenous redox balance upon oxidative stress (OS) and electrophilic stress (Giudice and Montella, 2006; Hayes and Dinkova-Kostova, 2014; Moi et al., 1994; Suzuki et al., 2013). Nrf2 orchestrates the regulation of the network's genes involved in cell cycle homeostasis, cytoprotection, innate immunity, and tumorigenesis, such as Nrf2, besides regulating cellular homeostasis under OS conditions, is strongly involved in the modulation of inflammation-related disorders. Indeed, in murine models the loss of Nrf2 (Nrf2 knock-out mice) sustains the development of different inflammatory disorders, including sepsis (Thimmulappa et al., 2006), pulmonary and cardiovascular diseases (Amata et al., 2017; Rangasamy et al., 2005; Ruotsalainen et al., 2013), and skin allergy (El Ali et al., 2013) In the present models Nrf2 is silenced, a condition accompanied by an increased expression of cytokines, chemokines, and adhesion molecules. These findings suggest that

* Correspondence to: Antoni Sureda, Research Group on Community Nutrition and Oxidative Stress (NUCOX) and CIBEROBN (Physiopathology of Obesity and Nutrition CB12/03/30038), University of Balearic Islands, Ctra. Valldemossa, Km 7.5, E-07122 Palma de Mallorca, Balearic Islands, Spain.
** Correspondence to: Seyed Mohammad Nabavi, Applied Biotechnology Research Center, Baqiyatallah University of Medical Sciences, Tehran, PO Box 19395-5487, Iran.
E-mail addresses: antoni.sureda@uib.es (A. Sureda), Nabavi208@gmail.com (S.M. Nabavi).

https://doi.org/10.1016/j.biotechadv.2017.12.012
Received 1 November 2017; Received in revised form 16 December 2017; Accepted 19 December 2017

Nrf2 can reduce the onset and the severity of inflammatory diseases through a fine-tuning of the immune response (El Ali et al., 2013; Johnson et al., 2010; Morzadec et al., 2014; Rangasamy et al., 2005; Ruotsalainen et al., 2013; Thimmulappa et al., 2006).

Even though the role of Nrf2 in native immunity and immune-related gene expression remains quite elusive, reported findings suggest interesting perspective. Moreover, it has been demonstrated that natural compounds may activate Nrf2 signaling, stimulating redox-related cytoprotective effects and inducing anti-inflammatory responses. The aim of the present review is to discuss the molecular mechanisms underlying Nrf2 signaling activation, and discuss the use of natural compounds as possible modulators of Nrf2 pathway, in an effort to stimulate further studies in the field.

2. Nrf2 signalling pathway

Nrf2 signalling pathway represents the principal cellular defence mechanism against electrophilic and OS and is essential for maintaining cellular redox homeostasis by activating phase II detoxification enzymes at transcriptional level (Chanas et al., 2002; Kumar et al., 2014, 2012; Nguyen et al., 2003a, 2005). Indeed, Nrf2 controls the expression of genes encoding detoxifying enzymes, such as NQO1, GSTs and HO-1, drug-metabolizing enzymes (DMEs), antioxidant enzymes (e.g., SODs, GPx, GSH reductase and UDP-glucuronosyl transferase (UDP-GT)), which present in their promoters the antioxidant response element (ARE), a 41-base-pair *cis*-element containing a core sequence, 5′-TGAXnnnGC-3′ (X = C or G or T, n = any nucleotide) (Nguyen et al., 2003a; Venugopal and Jaiswal, 1998). These enzymes modulate the detoxification processes that take place both at physiological level and in several chronic diseases (Boutten et al., 2011; Kumar et al., 2014; Ramsey et al., 2007a).

Nrf2 is a cap ‘n’ collar (CNC) transcription factor which consists of 605 amino acids and has in the C terminal region a distinctive basic region leucine zipper (bZIP) (Ma, 2013). It comprises 6 functional domains known as Nrf2-ECH (erythroid cell-derived protein with CNC homology) homologies (Neh) indicated as Neh1–6 and highly conserved in humans (Fig. 1) (Baird and Dinkova-Kostova, 2011; Boutten et al., 2011; Ma, 2013).

The Neh1 domain presents both the CNC region, which allows to heterodimerize with small musculoaponeurotic fibrosarcoma (Maf) proteins and bind the DNA, and a functional nuclear localization signal. In the N-terminal region, the Neh2 domain contains the sites DLG and ETGE for the Kelch ECH association protein 1 (Keap1) binding, while in C terminal region Neh3 domain binds to chromodomain helicase DNA-binding protein 6 (CHD6), which is indispensable for the transcriptional activity of Nrf2. The transactivation domains are represented by Neh4 and Neh5, which interact with coactivator proteins CREB-binding protein/p300 (CBP/p300); finally, Neh6 represents a negative regulatory domain which is independent of the cellular redox status (Fig. 1) (Sykiotis and Bohmann, 2010).

Under quiescent (basal) conditions, there are minimal levels of detectable Nrf2 protein, as its main regulator, Keap1, sequesters it in cytoplasm and promotes its ubiquitination through the cullin-3 (Cul3) E3 ubiquitin ligase complex, stimulating its degradation by the 26 S proteasome in a constitutive manner (Fig. 2a). Like Nrf2, Keap1 is expressed largely in the cytoplasm where it is associated with actin, also contributing to retain Nrf2 (Sykiotis and Bohmann, 2010).

Otherwise, in the presence of xenobiotics or free radicals, especially in those tissues where the reactions of detoxification normally occur, such as kidney, intestine and lung, the activity of Keap1-dependent E3 ubiquitin ligase is repressed, leading to the release of Nrf2 from Keap1, which is further stabilized by the protein DJ-1 and translocated into the nucleus (Fig. 2b) (Clements et al., 2006). Inside the nucleus, Nrf2 binds to small Maf proteins, Fra-1, c-Fos (Fos-related antigen-1) and other bZIP transcription factors, activating the transcription factor-4, c-Jun, enrolling in this way the overall transcriptional machinery, and then binds to the ARE sequences present in the promoter regions of target genes in order to upregulate their expression (Boutten et al., 2011). Furthermore, Nrf2 also controls the expression of Keap1 gene, thus providing an autoregulatory loop between these two proteins (Lee et al., 2007). After recovery of cell redox homeostasis, Keap1 transfers into the nucleus to displace Nrf2 from ARE promoters and escorts it back to the cytoplasm (Sun et al., 2007).

Many cellular mechanisms and processes modulate the activation and the inhibition of Nrf2 pathway. For example, electrophilic and oxidant stressors can act directly on Keap1 cysteine residues modifying its spatial conformation and rendering it unable to bind Nrf2, consequently promoting its ubiquitination and degradation (Niture et al., 2010). At the same time, these chemicals can oxidize cysteine residues of Nrf2, fostering its translocation to the nucleus (Jeong et al., 2006). Additionally, in oxidative or autophagy deficient conditions, Nrf2 can be stabilized respectively by the ubiquitin-conjugating enzyme UBCM2 or p21, which compete with Keap1 for the binding to Nrf2, upregulating therefore the activity of its target genes (Chen et al., 2009; Plafker et al., 2010a). Nfr2 can be also negatively regulated by CR6-interacting factor 1 at post-translational levels (Kang et al., 2010). Numerous kinases are involved in the modulation of the Nfr2/ARE pathway through post-transcriptional modifications. For example, phosphatidylinositol 3-kinase (PI3K), c-Jun N-terminal kinase (JNK), and extracellular signal-regulated protein kinase (ERK) phosphorylate Nrf2 threonine and serine residues, promoting its release from Keap1 and its subsequent translocation to the nucleus (Kobayashi and Yamamoto, 2006). Conversely, other protein kinases, such as p38 mitogen-activated protein kinases (MAPK), seem to negatively control this pathway, even if many aspects remain to be investigated (Yu et al., 2000).

Another important regulatory mechanism controlling Nrf2 abundance inside the nucleus, involves its nuclear import/export (Kumar et al., 2014). On one side, early response to stress facilitates the Nrf2 nuclear import by importin or exportin complexes, leading to a synchronized activation of cytoprotective genes (Jain and Jaiswal, 2006); on the other side, a delayed response to stress leads to a decrease of Nrf2 nuclear levels through the phosphorylation of Fyn, promoted by the glycogen synthase kinase-3β (GSK-3 β). Phosphorylated Fyn moves to the nucleus where it phosphorylates Nrf2 at tyrosine 568, thus promoting Nrf2 nuclear export and its degradation in the cytoplasm (Kaspar et al., 2009).

Moreover, under basal conditions, the transcription factor BTB and CNC homology 1 (Bach1) binds to small Maf proteins and to the ARE sequences, suppressing the expression of Nrf2 target genes. Opposite, during oxidative stress Bach1 separates from ARE regions, translocates outside the nucleus and is rapidly degraded by the proteosomal system; in this way Nrf2 can freely bind to the ARE sequences (Kaspar et al., 2009).

Finally, Nrf2 can be also degraded inside the nucleus since the prothymosin-α promotes the nuclear translocation of the complex

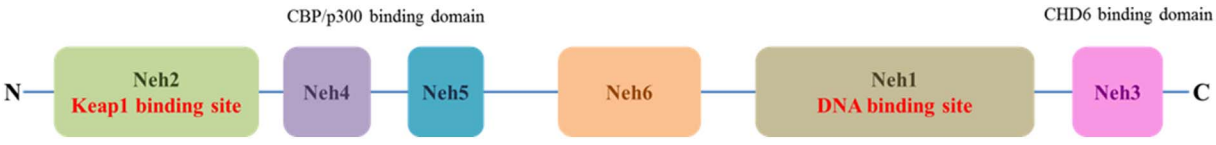

Fig. 1. Simplified illustration of the Nrf2 sequence.