MINIREVIEW

The cross-talk of nuclear factor kappaB and nitric oxide in the brain

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Abstract

In the brain, NF-κB has been implicated in both normal processes of synaptic plasticity and memory as well as in pathological mechanisms of neurodegenerative diseases such as Parkinson's disease, Huntington's disease or Alzheimer's disease. The involvement of nitric oxide in the learning, memory, behavioral processes, cognition but also in neurodegenerative processes and disorders was clearly described. Since the similar mechanisms including increase in glutamate and intracellular calcium level lead to activation of both NF-κB and nitric oxide, the mutual regulation of these factors is suggested. It seems that concerning the neurons, the increase in NF-κB expression and activation is accompanied by increased inducible and neuronal NOS isoform expressions and vice versa.

Introduction

In general, NF-κB (nuclear factor kappa-light-chain-enhancer of activated B cells) is a protein complex that controls the transcription of DNA. Five mammalian NF-κB family members have been identified till now: NF-κB1 (known as p50), NF-κB2 (named p52), RelA (called as p65), RelB and c-Rel. They all share a highly conserved Rel homology domain, responsible for their dimerization and binding to DNA and IκB (inhibitor of NF-κB). The transcription factor NF-κB works only when two members form a dimer. The most abundant activated form consists of a p50 or p52 subunit and a p65 subunit (Chen *et al* 2001).

NF-κB has been found in almost all animal cell types and is involved in cellular responses to stimuli such as stress, cytokines, ultraviolet irradiation, different antigens, free radicals, oxidized LDL (Gilmore 2006; Perkins 2007) and also in cell cycle regulation and apoptosis (Chen *et al* 2001). NF-κB plays a key role in regulating the immune response to infection. Conversely, incorrect regulation of NF-κB has been linked to cancer, inflammatory and autoimmune diseases, septic shock, viral infection, and improper immune development. Concerning the brain, NF-κB has been implicated in normal processes of synaptic plasticity and memory (Albensi & Mattson 2000) but also in molecular mechanisms of neurodegenerative diseases such as Parkinson's disease, Huntington's disease or Alzheimer's disease (Malek *et al* 2007).

Regulation of NF-κB and nitric oxide in neurons

Though NF-κB is nearly ubiquitous in distribution, its role in neurons of the central nervous system is controversial. Since we are still unable to determine the exact role of NF-κB in the organism, its contribution to neu-

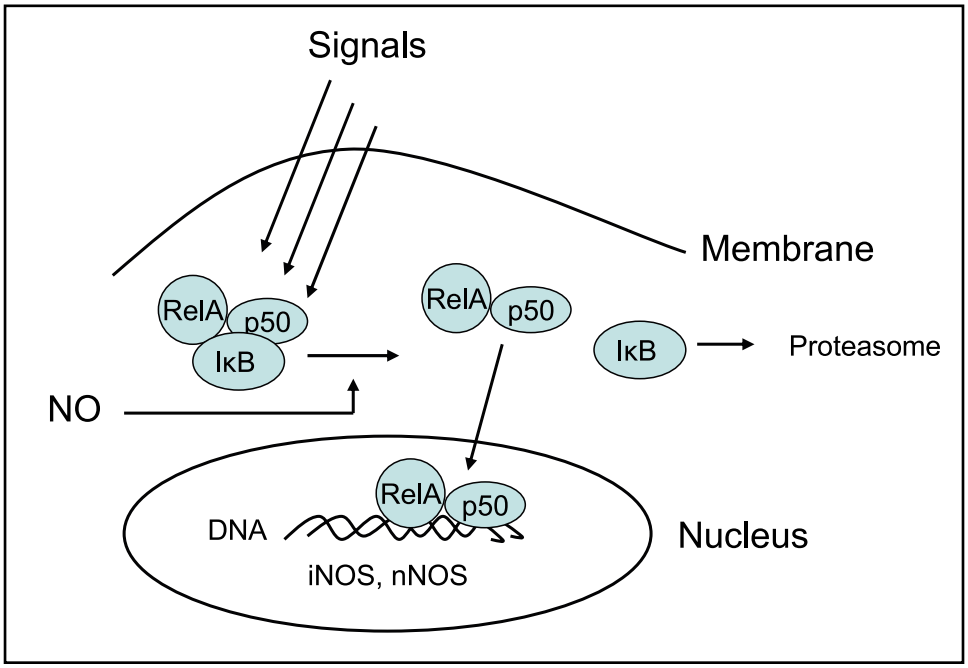

Fig. 1. In CNS neurons, increased level of nitric oxide (NO) may led to dissociation of NF-κB subunits (p50 and Rel A) from its inhibitor IκB which transfer to nucleus and activate number of genes including nNOS and iNOS. Following, IκB degradation is mediated via proteasome mechanism.

roprotection or neurodegeneration is unclear. According to Clemens (2000), the complex role of NF-κB is due to the fact that, depending on the interaction with other factors, it may induce genes encoding either death or survival proteins. Therefore, numerous studies have shown that under specific circumstances it may have proapoptotic or antiapoptotic action or in general it could induce protective or degenerative processes.

Several studies have shown that NF-κB is either constitutively active in neurons or activated by excitatory amino acid, neurotransmitters or both (Massa *et al* 2006). Although nitric oxide (NO) belongs to the untraditional neurotransmitters, the role of NO in the neuron NF-κB activation could be suggested. Similarly as NF-κB, nitric oxide is involved in both neuroprotective and neurodegenerative processes. In neurons, nitric oxide may be synthesized by neuronal NO synthase (nNOS) and inducible NO synthase (iNOS) isoforms. Neuronal NOS is constitutively expressed and is activated by increased intracellular Ca^{2+} level (Pecháňová & Bernátová 1998). In the brain, activation of N-methyl-D-aspartate (NMDA) receptors by glutamate leads to increased calcium in the postsynaptic neurons. Calcium binds to calmodulin and activates nNOS, increasing nitric oxide, which activates soluble guanylate cyclase, increasing cGMP. Part of this cGMP is released to the extracellular space. Several reports indicate that impairment of this glutamate-NO-cGMP pathway reduces the ability to learn (Llansola *et al* 2009). On the other hand, iNOS is independent of agonist stimulation and intracellular calcium levels and is regulated by cytokines primarily via the transcriptional factor NF-κB. Studies performed in rodents mostly imply that iNOS activity plays a detrimental role in autoimmune or chronically inflammatory processes (Pecháňová & Bernátová 1998). Several analyses indicate that NF-κB mediates expression of iNOS and vice versa NO can regulate transcriptional activity of NF-κB. In general, nitric oxide can regulate DNA-binding and transcriptional activity of NF-κB either by directly interacting with the factor itself, or with its endogenous inhibitors represented by IκB proteins or by activating upstream mechanisms that indirectly modulate the transcriptional activity in both cGMP-dependent or independent way (Contestabile 2008).

Reports on constitutively active NF-κB transcription factors provided an evidence that endogenous synaptic transmission might be capable of activating NF-κB. Application of excitatory neurotransmitters and their analogs, such as glutamate, N-methyl-D-aspartate and kainate, were shown to activate NF-κB in cerebellar granular neurons (Guerrini *et al* 1995; Kaltschmidt *et al* 1995). Subsequently, it has been found that even basal levels of synaptic activity are sufficient to activate NF-κB family members in a variety of neurons. Blockade of NMDA receptors and L-type Ca^{2+} channels can effectively reduce basal synaptic activation of NF-κB (Guerrini *et al* 1995; Meffert *et al* 2003). A physiologically active synaptosomal preparation has been used to demonstrate that also glutamate can activate NF-κB transcription factors in the isolated synaptic compartment through a Ca^{2+}-responsive pathway. Experiments using intracellular Ca^{2+} chelators in intact neurons show that local elevation of Ca^{2+} level is sufficient to activate NF-κB and NF-κB-dependent transcription (Meffert *et al* 2003). On contrast, several other activ-

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ity dependent neuronal transcription factors, such as cAMPresponsive-element-binding protein (CREB), are not fully transcriptionally active without global or sustained elevation in intracellular Ca^{2+} level (Meffert & Baltimore 2005).

Since the nanomolar concentrations of glutamate were proved to activate NF-κB in cerebellar neurons *in vitro* and this effect involved particularly NMDA receptor activation (Guerrini *et al* 1995) and elevation in intracellular Ca^{2+} level, it could implicate the cooperative role of NF-κB and nitric oxide in the normal brain functions.

Positive cross-talk of NF-κB and nitric oxide

The involvement of nitric oxide in the learning, memory, behavioral processes and cognition was clearly described (Llansola *et al* 2009; Meffert *et al* 2003; Pecháňová *et al* 2009; Jagla *et al* 2009; Kream *et al* 2009). This review tries to suggest a cooperative role of NO and NF-κB in these functions. Meffert *et al* (2003) demonstrated that mice lacking a subunit of NF-κB - p65 shown a selective learning deficit in the spatial version of the radial arm maze. These observations suggest that long-term changes to adult neuronal function caused by synaptic stimulation can be regulated by NF-κB nuclear translocation and gene, like nNOS, activation. O'Sullivan *et al* (2009) suggest that NF-kappaB activity following hippocampal learning may contribute to consolidation-associated synaptic reorganisation. Moreover, the possible role of nitric oxide and NF-κB in the locomotor activity of hypertensive rats have been described (Pecháňová *et al* 2006).

Kaltschmidt *et al* (1995) have speculated that changes during cerebellar development could be controlled by glutamate-induced gene expression involving NF-κB. Also Guerrini *et al* (1997) have observed glutamate-induced NF-κB activation during mouse cerebellum development but not in adult mice. Similarly, the important role of NO in the cell development was described in several animal models and humans (Török 2008).

Negative cross-talk of NF-κB and nitric oxide

The pathological activation, abnormal either overexpression or impairment of NF-κB may result in many diseases. As those molecular mechanisms have not been yet thoroughly studied, we can only roughly name some facts. NF-κB regulates a vast number of genes including those encoding cytokines, death and survival proteins, adhesion molecules, cyclooxygenase-2, manganesesuperoxide dismutase, and of course inducible nitric oxide synthase (Llansola *et al* 2009). Both increased expression of iNOS and NF-κB have been shown to participate in neurodegenerative diseases such as Parkinson's disease, Huntington's disease or Alzheimer's disease (Malek *et al* 2007). Inhibition of NF-κB decreased inducible nitric oxide synthase and cyclooxygenase-2 expression, and restored working memory in the mice model of Alzheimer's disease (Echeverria *et al* 2009). Recently the pathological action of nNOS was described as well. Concerning Parkinson's disease the relevant data demonstrate a role for NF-κB in selective induction of nNOS during early inflammatory activation of astrocytes stimulated by low-dose of inflammatory cytokines (Carbone *et al* 2008). Thus, the cross-talk of iNOS, nNOS and NF-κB is suggested in neurodegenerative processes and disorders.

Conclusion

In the brain, NF-κB and nitric oxide have been implicated in normal processes of synaptic plasticity and memory but also in molecular mechanisms of neurodegenerative diseases and disorders. The neurotransmitter glutamate was proved to activate both NF-κB and nitric oxide in the neurons, which could implicate the cooperative role of NF-κB and nitric oxide in the brain functions. Nitric oxide may, however, induce opposite responses, i.e. activation or inhibition of NF-κB, depending not only on NO concentration, but on the cell type as well. It seems that concerning the neurons, the increase in NF-κB expression and activation is accompanied by increased inducible and neuronal NOS isoform expressions and vice versa. Further studies are urgently needed to prove this hypothesis.

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