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NMDA receptor signaling: death or survival?

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Abstract

Glutamate-induced neuronal damage is mainly caused by overactivation of N-methyl-D-aspartate (NMDA) receptors. Conversely, normal physiological brain function and neuronal survival require adequate activation of NMDA receptors. Studies have revealed that NMDA receptor-induced neuronal death or survival is mediated through distinct subset of NMDA receptors triggering different intracellular signaling pathways. Here we discuss recent advances in the characterization of NMDA receptors in neuronal protection, emphasizing subunit-specific role, which contributes to temporal-spatial distribution, subcellular localization and diverse channel properties of NMDA receptors.

Keywords

NMDA receptors; glutamate; excitotoxicity; ischemia; neuroprotection

Introduction

NMDA receptors (NMDARs) are ionotropic glutamate receptors that are pivotal in controlling synaptic plasticity and memory function (Collingridge et al., 2004; Nakazawa et al., 2004). They are both voltage-dependent and ligand-gated. Activation of NMDARs requires membrane depolarization to remove the Mg^{2+} block, and binding of two agonists-glutamate and glycine, which induces Na^{+}, K^{+} and Ca^{2+} cation currents. Among these ions, Ca^{2+} signaling accounts for most, if not all, of the NMDAR-mediated functions. Functional NMDARs are heterotetramers, which consist of two obligatory NR1 subunits and two regulatory subunits from either the NR2 (NR2A, NR2B, NR2C and NR2D) or less commonly, the NR3 (NR3A and NR3B) subunits. While splicing variants of NR1 subunit could alter NMDAR properties (Zukin and Bennett, 1995), the two regulatory subunits primarily diversify and determine properties of NMDARs (Cull-Candy et al., 2001). For example, the decay of evoked currents in diheteromeric NMDARs ranged from tens of milliseconds (ms) to several seconds, with NR2A the fastest and NR2D the slowest, indicating differences in NMDAR-mediated excitatory postsynaptic currents (EPSCs) and glutamate binding affinities (Cull-Candy et al., 2001). Single channel studies revealed that NR1/NR2A or NR1/NR2B forms high-conductance channels with a high sensitivity to extracellular Mg^{2+}, while NR1/NR2C or NR1/ NR2D generate low-conductance openings with a low sensitivity to Mg^{2+} (Cull-Candy et al., 2001).

Each NMDAR subunit is a modular structure consisting of four distinct domains: an extracellular N-terminal domain, an extracellular ligand binding domain, a transmembrane domain and an intracellular C-terminal domain of variable length (Dingledine et al., 1999;

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Traynelis et al., 2010). The transmembrane domain, containing three membrane-spanning helices and a membrane re-entrant loop, contributes to the channel pore formation. The cytoplasmic domain is required for protein-protein interactions. It is thought that trafficking and localization of NMDARs depend on intracellular protein-protein interactions. Indeed, the first yeast two-hybrid screen using the NR2B C terminus as bait identified an interaction with PSD-95 (Niethammer et al., 1996), an abundant scaffolding protein present in the postsynaptic density (PSD). This interaction is essential for targeting NMDARs to specific membrane compartments. In addition, the intracellular domain of NMDAR subunits is subject to extensive posttranslational modifications such as phosphorylation, which is essential to regulate trafficking and channel properties of receptors (Chen and Roche, 2007).

The NMDAR subunits are differentially expressed throughout the central nervous system with localization patterns that change during development (Akazawa et al., 1994; Monyer et al., 1994). In the adult brain, NR1 and NR2A are expressed ubiquitously, while expression of NR2B is confined to the forebrain and striatum. NR2C is predominantly expressed in cerebellum. NR2D is mainly expressed early in development and is restricted to brainstem, midbrain and thalamus (Monyer et al., 1994). The NR3A subunit is widely distributed early in development and is localized predominantly in the nucleus of the lateral olfactory tract in the mature rodent brain. It was thought that expression of NR3B is restricted primarily to adult motor neurons. However, a recent study reported immunostaining of NR3B in wider brain regions including the forebrain, cerebellum and spinal cord (Wee et al., 2008).

NMDAR-dependent neuronal cell death and neuroprotective signaling

Glutamate-induced excitotoxicity is neuronal injury and death caused by overactivation of glutamate receptors through excessive glutamate stimulation. It was first reported that high dosage of glutamate caused lesions in the retina of postnatal mice two to ten days of age (P2 to P10) (Lucas and Newhouse, 1957). Similar observation linking excessive glutamate to brain damage was later reported in monkey (Olney and Sharpe, 1969). Choi revealed that excitotoxicity was caused by large amount of calcium (Ca^{2+}) influx induced by high levels of glutamate (Choi, 1987). Subsequent studies suggest that overactivation of NMDARs is necessary and suficient to induce glutamate-mediated calcium toxicity in neurons (Choi et al., 1987; Choi et al., 1988). Under physiological conditions, glutamate can be quickly released to the synaptic cleft with a concentration up to 1 mM, followed by a rapid decrease (Clements et al., 1992). However, during brain trauma or ischemia stroke, reduced blood flow leads to oxygen and glucose shortage, which gradually depletes ATP available for neurons and glial cells to control the extracellular levels of glutamate. Increased accumulation of glutamate in extracellular fluid results in excessive Ca^{2+} influx, which triggers multiple intracellular cascades and causes damage to neuronal cells in the brain. There is growing evidence linking abnormal NMDAR activity to excitotoxicity and brain damage (discussed in NR2A and NR2B section) (Aarts et al., 2002; von Engelhardt et al, 2007; Liu et al., 2007; Tu et al., 2010). In contrast to their role in neuronal death signaling through overactivation, adequate NMDARs are required for neuroprotection. For example, transient blockade of NMDARs in perinatal rat triggered extensive apoptosis in many brain regions (Ikonomidou et al., 1999). Moreover, the *in vitro* survival rate of cerebellar granule cells decreased when rat pups were treated with NMDAR antagonist (Ciani et al., 1997).

Given that the functions of NMDARs often correlate with their subcellular localizations, Hardingham and Bading hypothesized that activation of extrasynatic NMDARs results in cell death, while the activity of synaptic NMDARs promotes neuronal survival (Hardingham and Bading, 2010). The differences in signaling between synaptic and extrasynaptic NMDARs could be due to three factors: the NMDAR signaling complex, receptor subunit composition, and trans-synaptic (synaptic) versus chronic (extrasynaptic) activation of

NMDARs (Hardingham and Bading, 2010). This review focuses on the subunit-specific function of NMDARs in neuronal damage and protection.

NR2A and NR2B

NR2A and NR2B are the major NR2 subunits expressed in cortex and hippocampus. Expression of NR2A and NR2B is developmentally regulated. At nascent hippocampal synapses in culture, majority of NMDARs are located at extrasynaptic sites and mainly composed of NR1/NR2B (Tovar and Westbrook, 1999). During development, the expression level of NR2A is gradually increased, which leads to a switch from NR2B- to primarily NR2A-containing NMDARs (Cull-Candy et al., 2001). This subunit composition change correlates with NMDAR-mediated functions during development, including synaptic plasticity and neuronal survival. In NR2A null mice, the NMDAR channel current and long-term potentiation at the hippocampal CA1 synapses are significantly reduced, and a moderate deficiency in spatial learning is also observed (Sakimura et al., 1995). The NR2B knockout mice shows impairment of suckling response and die shortly after birth (Kutsuwada et al., 1996). Studies have shown that, in mature neurons, NR2A-containing receptors are enriched at synapses, while NR2B is largely localized at extrasynaptic sites (Steigerwald et al., 2000; Groc et al., 2006; Martel et al., 2009). However, synaptic NR2B-containing receptors and extrasynaptic NR2A-containing receptors have also been observed (Tovar and Westbrook, 1999; Thomas et al., 2006). Using subtype-specific antagonists to selectively block NR1/NR2A or NR1/NR2B diheteromeric NMDARs, it has been proposed that NR2A- and NR2B-containing NMDARs promote neuronal survival and death, respectively (Liu et al., 2007). However, there has been much debate on the selectivity of NR2A-specific antagonists (Neyton and Paoletti, 2006). Further complexity comes from the existence of triheteromeric (NR1/NR2A/NR2B) NMDARs, as there is no effective antagonist available. New drugs that selectively block NMDAR subtypes would be critical in defining roles of NR2A and NR2B in cell survival and death.

The extensive intracellular C-terminal domains of NMDARs interact with a network of cytosolic regulatory proteins, which couple receptors to various intracellular signaling pathways. For example, activation of NR2A-containing NMDARs has been linked to survival signaling through anti-apoptotic effects of phosphatidyl inositol 3-kinase (PI3K) dependent pathway (Lee et al., 2002). In contrast, disrupting the interaction of NR2B-containing NMDARs with PSD-95 has been shown to interrupt downstream signaling that leads to neuronal death (Aarts et al., 2002). Consistently, NMDA-induced apoptosis was significantly reduced in mouse cortical neurons cultured from NR2B, but not NR2A, homozygous knockout embryos (Liu et al., 2007). Conceivably, differential roles of NR2A and NR2B in neuronal survival are likely due to their diversified C-terminal domains, which allow distinct signal transductions triggered by calcium influx.

Several signaling pathways are involved in promoting neuronal survival or death. Perhaps the best understood example is the Ca^{2+}/calmodulin-dependent protein (CaM) kinase—cAMP response element binding protein(CREB) signaling pathway. Calcium signals via synaptic NMDARs, mainly NR2A-containing receptors, activate the nuclear CaMK IV and increases phosphorylation of the transcription factor CREB on its crucial regulatory residue serine 133 (Ser133) (Sasaki et al., 2011; Hardingham et al., 2002) (Fig. 1). This phosphorylation of CREB then recruits the CREB coactivator CREB binding protein (CBP) to stabilize the preinitiation complex and increase CRE promoter activity (Mayr and Montminy, 2001). CREB, which is thought to be involved in long-term memory formation, also has a critical role in promoting neuronal survival. Activation of CREB induces the expression of brain-derived neurotrophic factor (BDNF) and potentially other activity-regulated inhibitors of death genes, which protects neurons from NMDAR blockade-induced

neuronal death (Hardingham et al., 2002). Interestingly, CREB can be also activated through transducer of regulated CREB activity (TORC), independent of Ser133 phosphorylation (Fig. 1A). Recent studies revealed that synaptic NMDAR activity triggers phosphorylation of salt-inducible kinase 2 (SIK2) by CaMK I/IV, which would then induce TORC1 phosphorylation and its nuclear translocation (Sasaki et al., 2011). In addition, synaptic NMDAR activity initiates neuroprotective signals by regulating the forkhead box protein O (FOXO) class of transcription factors. FOXO regulates many cellular processes including proliferation, differentiation and apoptosis. It has been shown that activation of FOXO contributes to oxidative stress-induced cell death in cerebellar granule neurons (Lehtinen et al, 2006). Synaptic NMDAR activity suppresses FOXO activity, not only by triggering the nuclear export of FOXO via activation of PI3K-Akt pathway (Brunet et al., 1999; Dick and Bading, 2010), but by reducing the expression of FOXO1 (Al-Mubarak et al., 2009) (Fig. 1A). Suppression of FOXO activity decreases the expression level of pro-death genes, such as Fas ligand, Bcl2-interacting mediator of cell death (Bim) and thioredoxin-interacting protein (Txnip) (Papadia et al., 2008; Al-Mubarak et al., 2009). Taken together, both CREB-activation and FOXO-repression signals are essential in synaptic NMDAR-mediated neuronal protection.

In contrast to sustained CREB phosphorylation by synaptic NMDAR activity, CREB is only transiently phosphorylated when extrasynaptic NMDARs are activated. Subsequent studies have shown that transient phosphorylation of CREB may be resulted from a dominant dephosphorylation of CREB by protein phosphatase 1 (PP1) (Hardingham et al., 2002). Ifenprodil, which is a selective antagonist of NR2B-containing NMDARs, can block the rapid decay of CREB phospharylation on Ser133 triggered by activation of extra-synaptic NMD ARs. The decrease of CREB phosphorylation on Ser133 correlates with the inhibition of CREB function and CREB-target gene expression, such as BDNF. These results suggest that NR1/NR2B is the core NMDARs existing at extrasynaptic sites to shut off the CREB-mediated protective signals induced by synaptic NMDAR activation (Hardingham et al., 2002) (Fig. 1B). In addition, in contrast to synaptic NMDAR activity, extra-synaptic NMDAR activity promotes the nuclear import of FOXO transcription factors and subsequent activation of FOXO target genes (Dick and Bading, 2010), which contributes to neuronal death by the excitotoxic insult (Fig. 1B). Death-associated protein kinase 1 (DAPK1) adds another layer of regulation to the death signaling pathway (Fig. 1B). It has been demonstrated that DAPK1 physically and functionally binds to the NR2B subunit at extrasynaptic sites to trigger the ischemic brain damage (Tu et al., 2010). DAPK1 phosphorylates NR2B at serine 1303, which enhances the channel conductance of NR1/NR2B receptor and causes increased Ca^{2+} influx through NMDAR channels at extrasynaptic sites.

Additional NMDAR-dependent death and survival signaling, exemplified by the extracellular signal-regulated kinases-1/2 (ERK1/2) pathway and the neuronal nitric oxide synthase (nNOs) pathway, are discussed here. ERK1/2 signaling pathway plays an important role in NMDAR-dependent neuronal survival. ERK1/2 is activated by Ca^{2+} influx through NMDARs. Extrasynaptic NMDAR activity inactivates ERK signaling pathway, while activation of synaptic NMDARs induces sustained ERK activation (Chandler et al., 2001; Ivanov et al., 2006). Consistent with being more abundant at extrasynaptic sites, NR2B has been shown to be selectively associated with synaptic Ras GTPase activating protein (SynGAP) (Kim et al., 2005), which represses ERK signaling. Moreover, nNOS, an enzyme that catalyzes the production of highly neurotoxic molecule nitric oxide (NO), is involved in extrasynaptic NMDAR-mediated neuronal death. nNOS is coupled to NMDARs through its interaction with PSD-95 (Sattler et al., 1999), upon which the nNOS activation by NMDAR-mediated calcium influx is also dependent. Studies have shown that disruption of the NR2B

—PSD-95—nNOS signaling complex inhibits NMDAR-mediated NO release and prevents neuronal death (Aarts et al., 2002; Cui et al, 2007; Zhou et al., 2010).

NR2C

NR2C is mostly expressed in cerebellar granule cells, indicating its unique role in cerebellum (Wenzel et al., 1997). Indeed, genetic studies have shown that mice lacking NR2C display an impairment of motor coordination, the central function of cerebellum (Sprengel et al., 1998). In the cerebellum, NR2B-containing NMDARs are predominant early in development, while NR2A- and NR2C-containing receptors prevail at later stages. NR2C is also found in the olfactory bulb and thalamus (Wenzel et al., 1997). Using knock-in mice expressing the β-galactosidase reporter under control of the NR2C promoter, recent studies have shown that NR2C is expressed in retrosplenial cortex, pontine and vestibular nuclei (Karavanova et al., 2007). Unexpectedly, NR2C is also detected in a subset of glial cells, which appears negative for NR1 immunoreactivity (Karavanova et al., 2007), suggesting that NR2C-containing glial cells do not form functional NMDARs.

NR2C confers unique properties on NMDARs producing channels with low conductance openings and exhibiting specific channel kinetics (Stern et al., 1992; Farrant et al., 1994). This is true for both recombinant receptors expressed in heterologous cells and for native NMDARs in cerebellum. The disruption of the NR2C gene eliminates the low conductance channels, indicating that NR2C expression is responsible for the developmental expression of low conductance NMDAR channels (Ebralidze et al., 1996). Whole-cell responses from NR1/NR2C-containing receptors display deactivation kinetics with decay time constants of about 250 ms, which is similar to that of NR2B-containing receptors, but is slow compared with NR2A-containing receptors (Cull-Candy and Leszkiewicz, 2004). The decay time constant of NMDAR-mediated EPSC is markedly reduced in NR2C null mice, reflecting a change in the composition of NMDARs (Ebralidze et al., 1996). Interestingly, the peak amplitudes of NMDAR-EPSC are significantly increased in NR2C null mice, suggesting that NR2C-containing NMDARs have low open probability (Ebralidze et al., 1996). Consistently, single channel analysis reveals that NR1/NR2C receptors have a low open probability (0.011), which is approximately 44-fold and 10-fold less than the peak open probability of NR1/NR2A and NR1/NR2B receptors, respectively (Dravid et al., 2008). In addition, NR2C-containing NMDARs require only modest depolarization to overcome Mg^{2+} blockade. The voltage-dependent Mg^{2+} block of the NMDAR is believed to perform a crucial function in synaptic plasticity. This low sensitivity to Mg^{2+} implies that NMDARs with NR2C can operate at a more negative membrane potential (Cull-Candy et al., 2001).

In addition to its dominant role in cerebellar granule cells, several studies have suggested that NR2C may be critical in other regions of the brain. For instance, studies have shown that oligodendrocytes are damaged during ischemia mediated in part by NMDARs. Among NR2 subunits, NR2C is highly expressed on the processes of oligodendrocytes, which are responsible for myelination (Káradóttir et al., 2005; Salter and Fern, 2005; Micu et al., 2006. In addition, expression of NR2C is significantly increased in rat hippocampus following in vitro oxygen-glucose deprivation (an in vitro ischemia model) although NR2C is not normally detected in hippocampus (Small et al., 1997). Consistently, NR2C-immunoreactive positive cells have been observed in rat hippocampus after prolonged neonatal seizures (Ni et al., 2005). Moreover, gene disruption of NR2C in mice protects the brain from ischemic damage resulting from permanent middle cerebral artery occlusion (Kadotani et al., 1998). In contrast, recent studies have suggested a role for NR2C in neuronal survival (Chen and Roche, 2009). Growth factor stimulation and NMDAR activation lead to a robust increase in both PKB phosphorylation of NR2C and surface expression of cerebellar NMDARs (Fig. 2A). Surface expression of NR2C, unlike NR2A and NR2B, protects neurons from NMDA-

induced excitotoxicity. Taken together, NR2C may contribute to neuronal death or survival after ischemic damage.

NR3A and NR3B

NR3A and NR3B subunits are widely distributed, including cortex, cerebellum, hippocampus in the brain, spinal cord, as well as in glia (Ciabarra et al., 1995; Sucher et al., 1995; Cavara and Hollmann, 2008; Low and Wee, 2010). NR3A is also enriched in the central auditory system. Expression of NR3 subunits is developmentally regulated (Ciabarra et al., 1995; Sucher et al., 1995; Cavara and Hollmann, 2008; Low and Wee, 2010). NR3A expression is largely restricted to a narrow window of postnatal one to two weeks, during which synaptic circuitry is established. Perez-Otano et al. reported that NR3A-containing NMDARs undergo an activity dependent endocytosis during development, implying displacement of NR3 subunit by NR2 at synapses in mature neurons (Pérez-Otaño et al., 2006). Most recently, presynaptic NR3A-containing NMDARs have been found in visual cortical synapses (Larsen et al., 2011). The switch between NR3A and NR2 at presynaptic site regulates glutamate release and spike timing-dependent long term depressiom (LTD) in visual cortex. While level of NR3A is minimal in adolescent, NR3B expression gradually increases and sustains into adult. However, the ultrastructural localization of NR3B within the neuron is not clear.

NR3 subunits bind glycine selectively over glutamate. Compared to the NR1 subunit, NR3 subunits have higher affinity for glycine. Their affinities for other agonists and antagonists are different as well. This is potentially due to differences in hydrogen bonding networks in the ligand binding domains of NR1 and NR3 subunits (Yao et al., 2008). NR3-containing NMDARs exhibit distinct channel properties including reduced current responses, lowered sensitivity to Mg^{2+}, and decreased Ca^{2+} influx (Das et al., 1998; Nishi et al., 2001; Sasaki et al., 2002). In comparison with NR1/ NR2A, NR1/NR2A/NR3 exhibits a roughly 5-fold reduction in relative Ca^{2+} permeability (Cull-Candy et al., 2001). In ionotropic glutamate receptors, two flanking residues designated as N and N + 1 positions in the channel pore forming re-entrant loop are critical for Mg^{2+} sensitivity and Ca^{2+} permeability. The asparagine and arginine residues occupying these two positions in NR1 and NR2 subunits determine the high Ca^{2+} permeability property of conventional NMDARs. In NR3 subunits, the amino acid at the N position is changed to glycine, which conceivably attributes to distinct channel property of NR3 (Cavara et al., 2010).

NR3-containing NMDARs in the form of NR1/NR3 or NR1/NR2/NR3 have been reported using the *X. laevis* oocyte or mammalian cell line heterologous expression systems (Das et al., 1998; Sasaki et al., 2002; Ulbrich and Isacoff, 2008). It is generally considered that NR3A or NR3B interact with NR1 and NR2 subunits to modulate NMDAR activity. Interestingly, the NR1/NR3 composition displays specific glycine evoked currents, indicating the formation of functional channel (Chatterton et al., 2002). A recent study reported the presence of a glycine sensitive and glutamate antagonist insensitive NMDAR in optic nerve myelin, suggesting the existence of a NR1/NR3 NMDAR subtype *in vivo* (Piña-Crespo et al., 2010). However, whether glycine-gated excitatory channels exist in neuron is not clear.

Functional studies revealed that neurons in NR3A transgenic mice exhibit elevated currents and Ca^{2+} permeability in response to glutamate and glycine, while NR3A knockout neurons or mice exhibit opposite trend for channel conductance and Ca^{2+} influx (Ciabarra et al., 1995; Sucher et al., 1995; Tong et al., 2007). This ‘dominant-negative’ effect of NR3 on NMDAR activation is consistent with its distinct channel properties, implying a role of NR3 in neuroprotection upon glutamate-mediated Ca^{2+} neurotoxicity. Indeed, a recent study

reported that cortical neurons with transgenically expressed NR3A exhibited decreased cell death in response to NMDA (Nakanishi et al., 2009) (Fig. 2B). In addition, overexpression of NR3A in a focal cerebral ischemia mice model displaced less damage than wild type mice (Nakanishi et al., 2009). However, it is not known whether the effect is due to direct incorporation of NR3A in synaptic or extrasynaptic of NMDAR.

Prevention of ischemic injury by blocking NMDAR activation induced Ca^{2+} influx has also been found in CNS myelin. Elevated level of Ca^{2+} presumably induces degradation of key enzymes or structural proteins in myelin. Micu et al. reported that Ca^{2+} increase was greatly reduced in a chemical induced ischemia model when a broad-spectrum of NMDAR antagonists MK-801, 7-chlorokynurenic acid and d-AP5 was applied (Micu et al., 2006). Myelin was consequently protected from ischemic injury. Interestingly, reduction of Ca^{2+} was not observed when NR2A and NR2B specific antagonists were applied. These results and the recent finding of NR1/NR3 NMDAR in myelin (Piña-Crespo et al., 2010) suggest that NR3 as well as NR2C and NR2D might play major roles in neuroprotection in CNS white matter such as myelin.

Conclusions

The various compositions of NMDAR subunits provide an important source of diversity for functional regulation of NMDARs. The NR2 subunits each have unique channel properties and are linked to distinct downstream signal transduction pathways, implying different roles of each subunit in neuronal survival. In this review, we have discussed recent advances in the characterization of NR2 and NR3 subunits in neuronal protection. However, several intriguing questions remain. As many of evidence supports the hypothesis that the location of NMDAR directs signaling toward neuronal death or survival, our understanding will benefit from new information on the subcellular distributions of NMDARs with different subunit compositions. Because NMDARs are involved in multiple neuronal functions, directly blocking all NMDARs can cause adverse side effects. Therefore, it is critical to establish new selective antagonists to distinguish different subtypes of NMDARs or to specifically inhibit downstream signaling that cause excitotoxic neuronal death. Given that NMDARs mediate the Ca^{2+} influx in synaptic plasticity and glutamate toxicity, the spatial and temporal regulation of the composition of NMDARs clearly has profound implications in regulating neuronal activity and survival.

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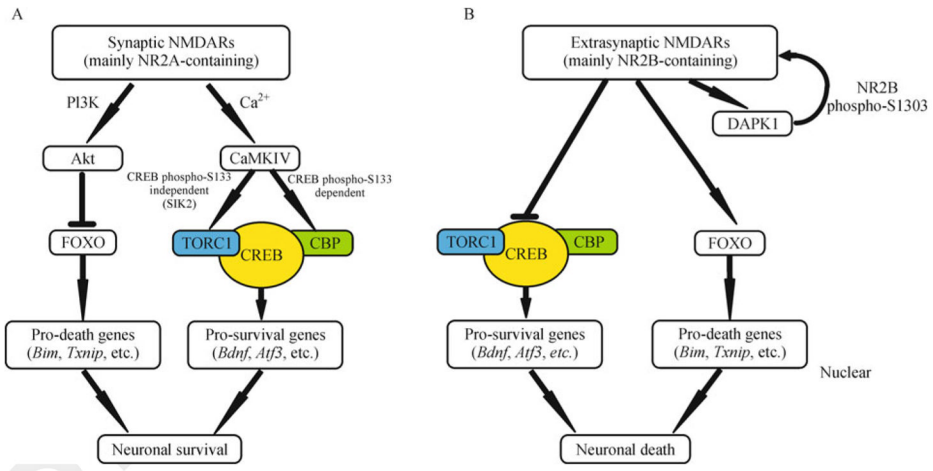

Figure 1.

Distinct signaling pathways of synaptic and extrasynaptic NMDAR in neuron fate. (A) Synaptic NMDAR activity regulates gene expression to promote neuronal survivial. Synaptic NMDAR activity down-regulates the expression of pro-death genes by inhibiting forkhead box protein O (FOXO) activity though PI3K-Akt pathway. Synaptic current can also activate CaMKIV and upregulate a battery of pro-survival genes by activation of cAMP response element binding protein (CREB). (B) Extrasynaptic NMDAR signal has opposing effect on gene expression to induce neuronal death. Extrasynaptic NMDAR activity blocks CREB-dependent pro-survival gene expression. FOXOs are also targets of extrasynaptic NMDAR activity to activate expression of pro-death genes.

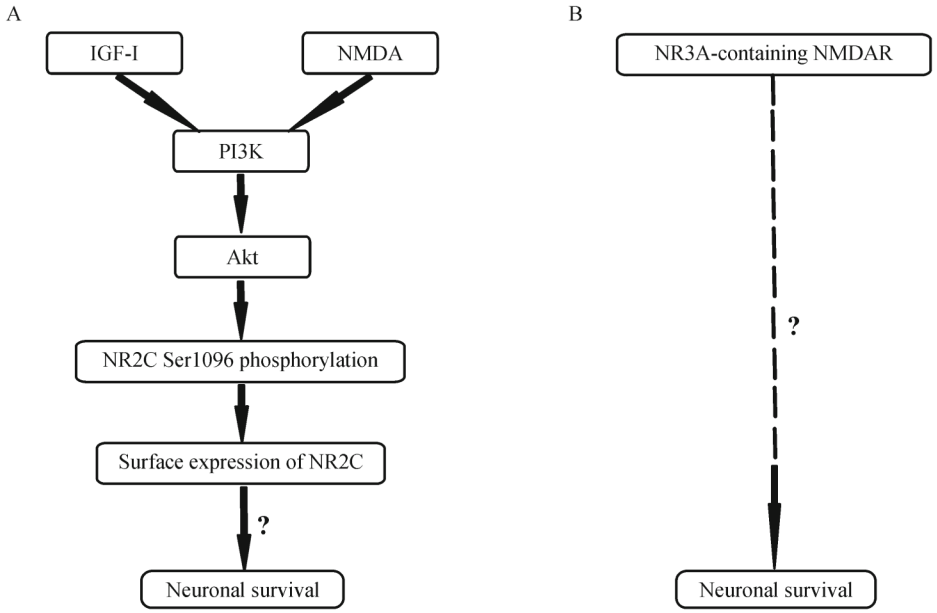

Figure 2.

NR2C and NR3A promote neuronal survival. (A) IGF-1 or NMDA application to neurons phosphorylates serine 1096 of NR2C though PI3K-Akt pathway, which increases the surface expression of NR2C-containing NMDARs and promotes neuronal survival. (B) Overexpression of NR3A-containing NMDARs supports neuronal survival with an unknown mechanism.