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Lithium neuroprotection: molecular mechanisms and clinical implications

Michael K. Rowe and De-Maw Chuang

Lithium has emerged as a neuroprotective agent efficacious in preventing apoptosis-dependent cellular death. Lithium neuroprotection is provided through multiple, intersecting mechanisms, although how lithium interacts with these mechanisms is still under investigation. Lithium increases cell survival by inducing brain-derived neurotrophic factor and thereby stimulating activity in anti-apoptotic pathways, including the phosphatidylinositol 3-kinase/Akt and the mitogen-activated protein kinase pathways. In addition, lithium reduces pro-apoptotic function by directly and indirectly inhibiting glycogen synthase kinase-3β activity and indirectly inhibiting *N*-methyl-D-aspartate (NMDA)-receptor-mediated calcium influx. Lithium-induced regulation of anti- and pro-apoptotic pathways alters a wide variety of downstream effectors, including β-catenin, heat shock factor 1, activator protein 1, cAMP-response-element-binding protein, and the Bcl-2 protein family. Lithium neuroprotection has a wide variety of clinical implications. Beyond its present use in bipolar mood disorder, lithium's neuroprotective abilities imply that it could be used to treat or prevent brain damage following traumatic injury, such as stroke, and neurodegenerative diseases such as Huntington's and Alzheimer's diseases.

Lithium treatment has a long clinical history. Initially, lithium was used to treat gout, but advantage was quickly taken of its mood-altering properties. Its popularity rose to such an extent that during the late 1800s and early 1900s water purported to be high in lithium was sold as cure-alls. Scientific documentation of lithium's mood-altering properties began in 1949 with the work of Cade (Ref. 1). This was systematically continued by Schou (Ref. 2), and in the early 1970s

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lithium was approved by the US Food and Drug Administration for the treatment of bipolar mood disorder (Ref. 3). Despite long-standing use of lithium, its mechanisms of action are still not fully understood. Recently, it has been proposed that lithium might exert its mood-stabilising effects at least in part through neuroprotection against apoptosis (Refs 4, 5, 6).

Bipolar mood disorder is a complex and devastating disease whose symptoms include alternating depressive and manic episodes. Depressive episodes are characterised by sadness, loss of interest in activities, feelings of worthlessness, and decreased energy, whereas manic episodes are characterised by elated mood, increased activity, decreased sleep, inflated self-esteem, racing thought, and disregard for negative consequences (Ref. 7). Bipolar disorder is both prevalent, affecting 1.3–1.5% of the US population of both sexes (Ref. 8), and serious, with the World Health Organization ranking bipolar disorder fourth among the neuropsychiatric disorders in disability-adjusted life years (Ref. 9).

An overview of apoptosis

Cell death occurs through two morphologically distinct processes: apoptosis and necrosis. In apoptosis, the cellular contents are packaged into apoptotic bodies, which are then consumed by phagocytes. Necrosis leads to cellular disintegration, plasma membrane rupture, and the leakage of cellular contents. Lithium appears to be neuroprotective primarily against the apoptotic pathway (Refs 4, 10). Apoptosis is a programmed and highly controlled form of cell death. Apoptosis can be triggered by a wide variety of stimuli, all of which lead to the same cell death machinery. As shown in Figure 1, apoptosis is regulated by three general methods: the pro-apoptotic receptor-mediated or extrinsic pathway, the pro-apoptotic intrinsic pathway, and an anti-apoptotic survival signalling pathway.

The extrinsic pathway is activated by death receptors, such as Fas and tumour necrosis factor receptor (TNFR), which have a cytosolic death domain. When the receptors are activated by the appropriate ligand, the death domains bind adaptor proteins and recruit initiator caspases to form death-inducing signalling complexes (DISCs). For example, FasL binds to the death receptor Fas. The adaptor protein's Fas-associated death domain (FADD) then binds the death domain of Fas. FADD also contains a death effector domain (DED), which recruits caspase 8. This complex forms the DISC (Ref. 11).

The intrinsic, or mitochondrial, pathway is activated by a wide variety of pro-apoptotic stimuli, which cause the release of cytochrome *c* from mitochondria, generation of reactive oxygen species, and subsequent activation of caspases. Key regulators of this pathway are the Bcl-2 family, which determine the integrity of the mitochondria. Proteins in the Bcl-2 family can be anti- or pro-apoptotic, depending on their Bcl-2 homology (BH) domains: proteins possessing the BH1–4 domains (e.g. Bcl-2, Bcl-X_L) are anti-apoptotic, whereas other Bcl-2 family proteins, having either the BH1–3 domains (e.g. Bax, Bak,

Figure 1. An overview of pathways regulating apoptosis. *(Legend; see next page for figure.)* Three pathways that regulate cell fate are the extrinsic (green), the intrinsic (blue), and cell survival (red) pathways. (a) The extrinsic pathway centres around death receptors, exemplified by Fas in this figure. Ligand (FasL) binding results in binding of the cytosolic death domain of the receptor to an adaptor protein (FADD) containing a death domain. The adaptor protein also has a death effector domain to which a caspase can bind. All four molecules (ligand, death receptor, adaptor protein, and caspase) form a death-inducing signalling complex that activates the effector caspases. (b) The intrinsic pathway centres on the integrity of the mitochondria, which is maintained by the Bcl-2 family. Some Bcl-2 family members increase membrane integrity (anti-apoptotic) and some decrease it (pro-apoptotic). The ratio of the pro- and anti-apoptotic Bcl-2 family members is tightly regulated by p53 among other molecules. If the ratio of the Bcl-2 family members tips towards the pro-apoptotic side, the mithochondria's membrane intergrity decreases and the pro-apoptotic factors, such as cytochrome *c*, are released. Cytochrome *c* binds to Apaf-1, and this complex activates initiator caspases that in turn activate effector caspases. (c) The cell-survival pathway begins with the activation of cell-surface receptors such as TrkB, which activate anti-apoptotic pathways including PI3K/Akt and MEK/ERK. Both of these pathways inhibit GSK-3β activity. When regulated via the cell-survival signalling pathway, PI3K/Akt, MEK/ERK and GSK-3β alter activity in a large number of effector systems, including the Bcl-2 family members, in an anti-apoptotic fashion. Abbreviations: Apaf-1, apoptosis-protease-activating factor 1; DISC, death-inducing signalling complex; ERK, extracellular-signal-regulated kinase; FADD, Fas-associated death domain protein; GSK3-β, glycogen synthase kinase-3β; MEK, mitogen-activated protein kinase kinase; PI3K/Akt, phosphatidylinositol 3-kinase/Akt.

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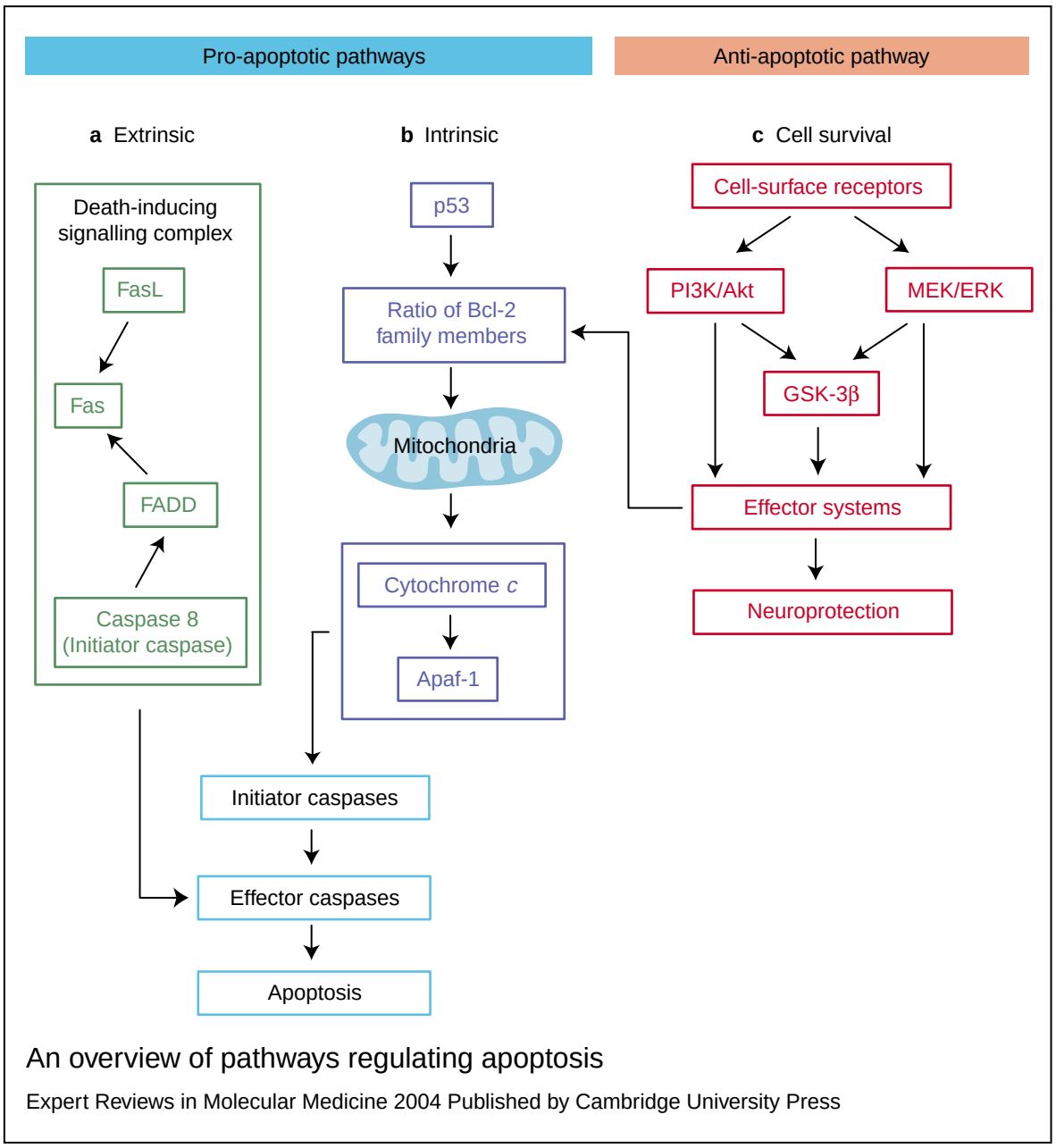


Figure 1. An overview of pathways regulating apoptosis. *(See previous page for legend.)*

Bok) or just the BH3 interaction domain (e.g. Bid, Bik, Bad, Bim, Noxa and Puma) are pro-apoptotic (Ref. 12). The ratio of the pro- versus anti-apoptotic Bcl-2 proteins is critical and their gene expression is tightly regulated by p53 (Ref. 13). If pro-apoptotic Bcl-2 family members are activated, they decrease the integrity of the mitochondria, which release several pro-apoptotic factors, the best characterised of which is cytochrome *c*. Cytochrome *c* binds apoptosis-protease-activating factor 1 (Apaf-1), and this complex activates procaspase 9 (Ref. 14). The end result of the intrinsic and extrinsic pathways is the activation of the initiator procaspases 2, 8, 9 and 10. The initiator caspases cleave and hence activate the effector caspases 3, 6 and 7, which in turn cleave cellular proteins such as cytoskeletal and nuclear proteins (Ref. 15).

In addition to the two pro-apoptotic pathways, apoptosis is regulated by a survival signalling

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pathway. This pathway begins with the activation of cell-surface receptors, including tyrosine receptor kinase B (TrkB) (Ref. 16). The stimulation of these receptors leads to activation of multiple pathways/kinases including the phosphatidylinositol 3-kinase/Akt (PI3K/Akt) pathway (Ref. 17) and the mitogen-activated protein (MAP) kinase kinase/extracellular-signal-regulated kinase (MEK/ERK) pathway (Ref. 18). These pathways regulate the phosphorylation state of glycogen synthase kinase-3β (GSK-3β) (Ref. 19). Combined, the PI3K/Akt pathway, the MEK/ERK pathway and GSK-3β regulate a staggeringly large number of effector systems related to neuroprotection, including β-catenin, heat shock factor 1 (HSF-1), activator protein 1 (AP-1), and cAMP-response-element-binding protein (CREB). Many of these effector systems in turn regulate the Bcl-2 protein family, which thus serves as an intersection between survival and death receptors.

Lithium is neuroprotective

A large number of studies, encompassing in vitro, in vivo, and clinical analyses, have concluded that lithium is neuroprotective. Given the vastly different methodologies and parameters employed by these studies, the basic finding – that lithium is neuroprotective – is very strong.

The in vitro studies have shown that lithium has neuroprotective effects in multiple cell lines, including cerebellar granule cells, cerebral cortical cells, hippocampal neurons (Ref. 4), PC12 cells (Ref. 20) and neuroblastoma cells (Ref. 21) from both humans and rodents. Furthermore, in vitro lithium treatment has successfully provided neuroprotection against a wide variety of insults, such as oubain (Refs 22, 23), anticonvulsants (Ref. 24), potassium deprivation (Refs 25, 26), α-amino-3-hydroxy-5-methylisoxazole-4-propionate (AMPA) and kainate (Ref. 27), β-amyloid peptide (Refs 28, 29), and β-bungarotoxin (Ref. 30). Glutamate is the primary excitatory neurotransmitter in the brain (Ref. 31) as well as a potent excitotoxin (Ref. 32), and hence of particular interest in studies of neuroprotection. Pre-incubation with therapeutic levels of lithium protects brain neurons from glutamate excitotoxicity, which is mediated by the *N*-methyl-D-aspartate (NMDA) receptor (Ref. 4).

Lithium has also proven to be neuroprotective in vivo. As with in vitro neuroprotection, this has been found following a wide variety of insults, including focal cerebral ischaemia (Ref. 33), transient ischaemia (Ref. 34), quinolinic acid striatal infusion [an excitotoxic model of Huntington's disease (Ref. 35)], excitotoxic lesions of the cholinergic system (Ref. 36), and kainic-acid-induced brain lesions (Ref. 37). A particularly exciting extension of the in vivo ischaemia studies has shown that lithium applied after the ischaemic event is neuroprotective, suggesting that lithium has therapeutic use in treating stroke (Ref. 38).

The final set of evidence supporting lithium's neuroprotective abilities comes from clinical studies. In the clinical setting, it is difficult to separate neuroprotective from neurotrophic effects; however, with that qualification, several studies have shown alterations in cell numbers and/or volume of grey matter believed to be due to the neuroprotective/neurotrophic effects of this drug. Patients with bipolar mood disorder not taking lithium had significant reductions in glial cell counts as compared with patients taking lithium (Ref. 39). In addition, four weeks of lithium treatment increased the volume of grey matter (Ref. 40) as well as brain levels of *N*-acetyl-aspartate (NAA), a marker of neuron viability (Ref. 41). Finally, studies have shown a correlation between grey matter volume and lithium use (Refs 42, 43).

Neuroprotective mechanisms

Lithium produces its neuroprotective effects not only via multiple pathways, but possibly through direct influence at several levels of the same pathway. While many of these points hold interest in terms of future research, this review focuses on the neuroprotective effects of lithium that have gained wide acceptance. These effects include lithium's induction of the survival signalling pathway and lithium's inhibition of NMDA-receptor-mediated calcium influx. The survival signalling pathway is the primary focus here, and is discussed from upstream factors (brain-derived neurotrophic factor; BDNF) through activated pathways (PI3K/Akt and MEK/ERK) and central regulators (GSK-3β), to effector systems (β-catenin, HSF-1, AP-1 and CREB) and finally to the Bcl-2 family and the caspases. Figure 2 summarises these pathways and their connections.

We would be remiss if we failed to mention that lithium has multiple mechanisms of action beyond those outlined in the previous paragraph. Included in the list of enyzmes directly altered by lithium is inositol monophosphate phosphatase

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(IMPase), polyphosphate 1-phosphatase, fructose 1,6-bisphosphatase, bisphosphate nucleotidase, and phosphoglucomutase (Ref. 44). Although pathways involving these enzymes have not been shown to be related to lithium neuroprotection, they have been implicated in other lithium-induced effects. For example, lithium inhibits the collapse of sensory growth cones and increases the area of sensory growth cones. Inositol reverses these lithium-induced effects, suggesting that they are dependent on inositol depletion via lithium's effect on IMPase (Ref. 45). A second example is pilocarpine-induced reciprocal hind limb scratching. Lithium's ability to inhibit this behaviour is dependent on inositol depletion (Ref. 46). The mechanism of action of lithium, besides neuroprotection, which has gained widespread attention, is the inositol depletion hypothesis. In short, this hypothesis claims that lithium inhibits IMPase, thus decreasing the level of inositol and phosphatidylinositol (4,5)-bisphosphate (Ref. 47). However, the initial experiments investigating lithium's neuroprotection abilities found that neuroprotection was independent of inositol depletion. These experiments found that a potent IMPase inhibitor did not mimic lithium's neuroprotective effect and/or that excess inositol did not prevent lithium's neuroprotective effect (Refs 4, 24, 48).

Upstream survival events: BDNF, PI3K/Akt, MAP kinases

The survival pathway begins at cell-surface trophic factor receptors. Following lithium treatment, these receptors are activated by BDNF. Functionally, BDNF is a neurotrophin known to regulate neural development, survival and plasticity (Refs 49, 50). Molecularly, BDNF binds to the TrkB receptor and this binding causes the phosphorylation of Tyr490 of the receptor (Ref. 51). The TrkB receptor in turn has several effector systems, including phospholipase Cγ (PLCγ) signalling, MEK/ERK signalling and PI3K/Akt signalling (Ref. 16).

Two pieces of evidence strongly link BDNF to lithium-induced neuroprotection. First, lithium increases BDNF levels in vivo and in vitro (Refs 52, 53), although the exact underlying mechanism is still under investigation. Second, BDNF activity is necessary for lithium-induced neuroprotection. If the biological activity of BDNF is blocked at the TrkB receptor level, either by the TrkB antagonist K252a or by utilising BDNF knockout mice as experimental subjects, lithium's neuroprotective abilities against glutamate-induced excitotoxic insult are completely blocked (Ref. 53). Taken together, this provides strong evidence that BDNF is a necessary component of lithium's neuroprotective effect.

Once BDNF binds to the TrkB receptor, two pathways – PI3K/Akt and MEK/ERK – critical for the neuroprotective effects of lithium are activated. The PI3K/Akt pathway is an important regulator of cell survival (Ref. 54). In this pathway, phosphorylation of the TrkB receptor leads to phosphorylation of adaptor protein Shc, which in turn recruits the Grb2–SOS complex. At this point the pathway splits. In one direction, the Grb2–SOS complex activates Ras, which then activates PI3K; in the other, Grb2 recruits Gab1 and Gab2, which in turn recruit and activate PI3K. Once activated, PI3K produces 3-phosphoinositides, which then recruit phospholipid-dependent kinases (PDKs) to activate Akt-1 by phosphorylation of Ser473 and/or Thr308 (Refs 55, 56). Finally, Akt-1 acts on several targets in an anti-apoptotic fashion, including GSK-3β, the Bcl-2 family member BAD, CREB, members of the forkhead family, and procaspase 9 (Refs 16, 57, 58). The activation of Akt-1 through increased PI3K signalling, rather than direct activation, was shown by the action of PI3K inhibitors: these block the lithium-induced increase in Akt-1 activation (Refs 59, 60). Lithium's ability to induce Akt-1 activity is supported by work showing that glutamate excitotoxicity reduces Akt-1 phosphorylation, while lithium treatment normalises Akt-1 and provides neuroprotection (Ref. 59). It should be noted that lithium-induced stimulation of the PI3K/Akt pathway is cell-type-specific, as some studies have failed to detect changes in Akt-1 phosphorylation levels following lithium application in certain cell lines (Refs 61, 62).

Other major pathways with a role in apoptosis/neuroprotection activated by lithium treatment involve the MAP kinases. Major members of the MAP kinase family include c-Jun N-terminal kinase (JNK), p38 kinase, and ERK. ERK is regulated through the MEK/ERK pathway and is, in general, neuroprotective (Ref. 63). In the MEK/ERK pathway, BDNF phosphorylation of the TrkB receptor leads to Ras activation (Ref. 64). Ras binds the Raf proteins Raf-1 and B-Raf. Raf in turn targets MEK, which in turn phosphorylates ERK. Finally, ERK directly and indirectly regulates several effector systems, such as the NF-κB

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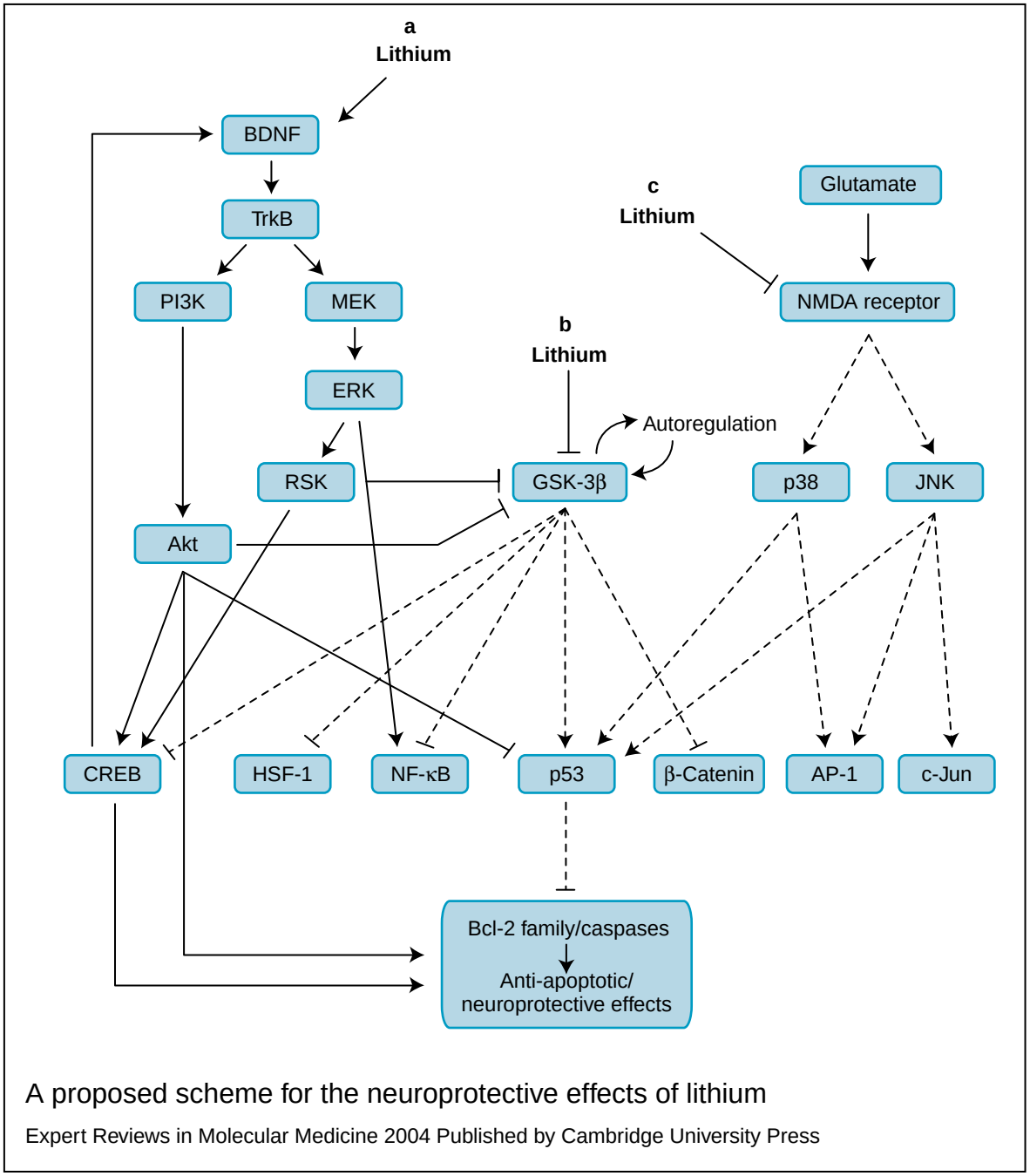


Figure 2. A proposed scheme for the neuroprotective effects of lithium. *(See next page for legend.)*

pathway, and ribosomal S6 kinase (RSK), which in turn regulates CREB and GSK-3β. Downstream targets of the MEK/ERK pathway include the Bcl-2 family and AP-1 transcription factor (Refs 18, 65). Chronic lithium treatment increases activity in the MEK/ERK pathway in vivo and it has been suggested that lithium's neuroprotection is dependent on MEK/ERK induction (Ref. 66). However, this effect might be much more complex, as other reports have shown that lithium can enhance or block the MEK/ERK pathway, depending on the cell type studied (Ref. 67).

JNK and p38 are activated by stress stimuli and are generally apoptotic. In the JNK and p38 pathways, stress signals activate families of GTPases (Rho, Rac, Cdc42) which in turn

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Figure 2. A proposed scheme for the neuroprotective effects of lithium. *(Legend; see previous page for figure.)* Lines with solid arrows represent stimulatory connections; lines with flattened ends represent inhibitory connections. Pathways with reduced activity as a result of lithium treatment are represented by dashed lines. Lithium's interaction with the cell survival/apoptotic machinery occurs at three points: indirectly at BDNF and NMDA receptors, and directly through GSK-3β. (a) Lithium induces BDNF, which in turn activates TrkB and the downstream PI3K/Akt and MEK/ERK pathways. Both of these pathways are strongly associated with neuroprotective effects. The PI3K/Akt pathway inhibits the level and/or activity of GSK-3β, p53 and caspase 9. The MEK/ERK pathway, via RSK, stimulates CREB and inhibits GSK-3β. (b) Second, lithium directly inhibits GSK-3β. This signal might be amplified by GSK-3β autoregulation. GSK-3β constitutively inhibits a large number of anti-apoptotic factors, including CREB, HSF-1, NF-κB, and β-catenin. By removing the GSK-3β-induced inhibition, lithium is in effect stimulating all of these factors. (c) Third, lithium inhibits NMDA-receptor-mediated calcium influx. The calcium influx stresses cells, activating the MAPK pathways including the pro-apoptotic factors p38 and JNK. Lithium, by inhibiting calcium influx, suppresses this activation. By targeting the cell survival machinery at multiple points, lithium regulates a wide variety of pathways and effectors. Together, these changes shift the Bcl-2 family member ratio towards the anti-apoptotic members, prevent cytochrome *c* release, and inhibit caspase activation (see Fig. 1). Abbreviations: AP-1, activator protein 1; BDNF, brain-derived neurotrophic factor; CREB, cAMP-response-element-binding protein; ERK, extracellular-signal-regulated kinase; GSK-3β, glycogen synthase kinase-3β; HSF-1, heat shock factor 1; JNK, c-Jun N-terminal kinase; MAPK, mitogen-activated protein kinase; MEK, mitogen-activated protein kinase kinase; NMDA, *N*-methyl-D-aspartate; PI3K/Akt, phosphatidylinositol 3-kinase/Akt; RSK, ribosomal S6 kinase; TrkB, tyrosine receptor kinase B.

activate sets consisting of a MAP kinase kinase kinase (MEKK1/2/3/4), a MAP kinase kinase (MKK3/4/6/7) and a MAP kinase (JNK or p38) (Refs 68, 69). The MAP kinase translocates to the nucleus where it activates, via phosphorylation, transcription factors including AP-1 and p53 (Refs 70, 71). Lithium's interaction with the JNK and p38 pathways has been shown following glutamate excitotoxicity (discussed further below) and trophic deprivation. These challenges induce JNK and p38 activity, which increases AP-1 binding activity. Lithium-induced neuroprotection is associated with inhibition of glutamate-induced increases in JNK, p38 and AP-1 (Ref. 72) and trophic-deprivation-induced increases in JNK (Ref. 73).

GSK-3β: a centrally located mechanism

GSK-3β is a pro-apoptotic protein that serves as an intersection point for apoptotic pathways and plays a crucial role in lithium's neuroprotective capabilities (Ref. 74). A wide variety of kinases and systems regulate GSK-3β activity, including Akt, protein kinase A (PKA), protein kinase C (PKC), MAP kinases and the Wnt pathway. As the list of substrates phosphorylated by GSK-3β is over 40 proteins long and includes metabolic, signalling and structural proteins as well as transcription factors, regulating GSK-3β activity has great impact (Refs 75, 76). Some of the more important substrates in terms of lithium-induced neuroprotection include β-catenin, HSF-1, AP-1, c-Jun, Myc and CREB.

Lithium inhibits GSK-3β through three mechanisms: directly, indirectly through the induction of survival signalling pathways, and through autoregulation. Lithium also inhibits the GSK isozyme GSK-3α (Ref. 77); this review focuses on GSK-3β, because it has been rigorously investigated and characterised, but inhibition of GSK-3α might prove to have relevance for lithium's neuroprotective capabilities.

Lithium directly inhibits GSK-3β activity possibly by competitively inhibiting Mg^{2+} binding to the active site of the enzyme (Refs 78, 79). Although undoubtedly important, there are two indications that direct inhibition of GSK-3β alone cannot fully explain lithium's neuroprotective ability nor its ability to inhibit GSK-3β. First, direct inhibition of GSK-3β by lithium is rapid, whereas many of lithium's neuroprotective effects require chronic treatment. Second, the IC_{50} for direct inhibition, 1.5–2 mM, is greater than the therapeutic doses (e.g. 1 mM) typically used in neuroprotective experiments, indicating we can see neuroprotective effects when GSK-3β inhibition is under 50%.

Regulation of the phosphorylation status of GSK-3β might underlie lithium's indirect inhibition of the protein. Activity levels of GSK-3β are regulated by the phosphorylation states of Ser9 and Tyr216: Ser9 phosphorylation decreases GSK-3β activity and Tyr216 phosphorylation increases it (Ref. 80). Lithium treatment increases Ser9 phosphorylation and hence inhibits GSK-3β activity (Refs 78, 81). Lithium treatment similarly

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regulates GSK-3α by increasing Ser21 phosphorylation (Refs 59, 61). Lithium might affect GSK-3β phosphorylaiton through the previously discussed survival signalling pathways. In this scheme, lithium treatment induces BDNF, which then stimulates the PI3K/Akt and MEK/ERK pathways, which in turn inhibit GSK-3β. However, the exact mechanism by which this inhibition occurs is still under debate and it is likely that multiple components of the survival signalling pathway contribute to lithium-induced GSK-3β inhibition (Ref. 26).

The third mechanism by which lithium inhibits GSK-3β is via GSK-3β autoregulation. GSK-3β autoregulates its phosphorylation status and activity level via the protein phosphatase 1/inhibitor 2 (PP-1/I-2) complex. In short, GSK-3β activates PP-1 by phosphorylating I-2. PP-1 then regulates GSK-3β by dephosphorylating Ser9. In this system, if GSK-3β is inhibited, then PP-1 is inhibited, and as PP-1 stimulates GSK-β, its inhibition results in GSK-3β inhibition. In essence, this system will amplify inhibitory signals received by GSK-3β. Thus, when lithium directly inhibits GSK-3β, it would indirectly inhibit the PP-1/I-2 complex, which would then produce further inhibition on GSK-3β (Ref. 62). This system provides a theoretical explanation to counter the criticisms offered against the clinical significance of lithium's direct inhibition of GSK-3β (Ref. 76).

Downstream factors

Inhibition of GSK-3β or stimulation of PI3K/Akt and the MEK/ERK pathway regulates the activity of a large number of pathways, proteins and transcription factors. Rather than the complete list, this discussion focuses on β-catenin, HSF-1, AP-1 and CREB. These targets are widely believed to have a strong neuroprotective effects and have been shown to undergo alteration following lithium treatment, as discussed below. All these mechanisms are potentially important in lithium-derived neuroprotection; however, there is little evidence as to their relative contributions at this time.

β-Catenin is a proto-oncogene product and part of the canonical Wnt-1 pathway. In this pathway Wnt proteins bind to a family of extracellular receptors known as Frizzleds. Frizzleds activate the intracellular protein 'dishevelled 1', which in turn inhibits GSK-3β. GSK-3β constitutively forms part of a β-catenin destruction complex. The presence of this complex results in the phosphorylation and degradation of β-catenin. Thus, inhibition of GSK-3β leads to increased β-catenin levels. As β-catenin accumulates, it interacts with the transcription factor Tcf/Lef (T-cell factor/lymphoid enhancer factor), which is translocated to the nucleus. The Tcf/Lef–β-catenin complex activates the transcription of diverse genes that include several involved in the inhibition of apoptosis (Refs 82, 83, 84, 85, 86). As previously described, lithium inhibits GSK-3β and hence will interact with the Wnt/β-catenin pathway at this level. As would be expected from this interaction, lithium treatment increases β-catenin levels both in vitro (Refs 77, 87) and in vivo (Ref. 88).

HSF-1 is a transcription factor activated by many conditions that place stress on the cell. Once activated, HSF-1 triggers the expression of chaperone proteins, including heat shock protein 70 (Hsp70). Hsp70 has a wide variety of neuroprotective functions (Ref. 89), such as binding with and stabilising Akt (Ref. 90), interfering with Apaf-1, a key link between mitochondrial cytochrome *c* release and caspase activation (Ref. 91), and suppressing JNK activation (Ref. 92). HSF-1 activity is suppressed by serial phosphorylation. ERK phosphorylates HSF-1 at Ser307, which then allows GSK-3β to phosphorylate HSF-1 at Ser303 (Ref. 93) . GSK-3β negatively regulates HSF-1 as both DNA binding by HSF-1 and HSF-1-dependent transcription are negatively correlated with GSK-3β activity (Refs 94, 95). Studies have shown that lithium treatment leads to alterations in Hsp levels both in vitro (Ref. 94) and recently in vivo, where it has been demonstrated that lithium's neuroprotective abilities are correlated with an increase in Hsp70 levels in the brain following ischaemic insult (Ref. 38).

AP-1 is a transcription factor and a dimeric complex that contains members of the Jun, Fos, CREB and ATF families. AP-1 is activated by a wide variety of stress factors and cellular signals. Typically, these signals activate the MAP kinases, primarily JNK and p38, which results in AP-1 binding to a variety of genes (Refs 69, 70, 96). A secondary regulator of AP-1 is GSK-3β, which phosphorylates c-Jun so as to reduce AP-1 binding activity (Ref. 97). AP-1 plays a complex role in neuroprotection/apoptosis in that its activation can be either neuroprotective or neurodegenerative, depending upon the

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particular molecules that form the AP-1 complex and their target genes (Refs 98, 99). The fact that AP-1 can be either neuroprotective/apoptotic correlates well with the evidence that lithium can both induce and inhibit AP-1 activity. Lithium inhibits stimulus-induced AP-1 activity, whereas it increases basal activity levels of AP-1 (Refs 72, 73, 100, 101).

CREB is a transcription factor that regulates the expression of genes with a promoter containing a cAMP-response element (CRE). Functionally, CREB plays a role in many diverse processes, from memory formation (Ref. 102) to apoptosis. CREB is sensitive to several stimuli including growth factors and stress, resulting in its regulation by several of the previously discussed pathways, including BDNF (Ref. 103), MEK/ERK (Ref. 104) and GSK-3β (Ref. 105). CREB's neuroprotective properties reside in its ability to increase expression of anti-apoptotic proteins, including members of the Bcl-2 family (Refs 106, 107) and BDNF (Ref. 108). As BDNF induces CREB activation and CREB upregulates BDNF expression, this creates a feedback loop throughout the cell survival signalling pathway. Chronic lithium treatment alters CREB activity in excitotoxic situations by preventing glutamate-induced loss of phosphorylated CREB and CRE-driven gene expression (Ref. 104).

As previously discussed, the Bcl-2 family plays a key role in apoptosis. All of the previously discussed pathways and molecules modify the Bcl-2 family to some extent. Lithium treatment alters the Bcl-2 family in an anti-apoptotic fashion by decreasing the expression of Bax and increasing the expression of Bcl-2 (Refs 109, 110), although this effect might be specific to neurons (Ref. 111).

NMDA receptors

A second upstream mechanism by which lithium provides neuroprotection is through NMDA receptors. The NMDA receptor plays a vital role in a variety of brain functions including development, neuroplasticity and excitotoxicity. NMDA receptors are ionotrophic glutamate receptors that, when stimulated, allow the passage of calcium and sodium into the cell. When NMDA receptors are overstimulated, the calcium influx becomes excessive. Excessive calcium is taken up by the mitochondria, resulting in the collapse of the mitochondrial membrane potential and subsequent cell death (Ref. 112). This excessive calcium influx is a primary component of glutamate excitotoxicity (Ref. 113). Chronic treatment of cultured brain neurons with lithium decreases glutamate-induced NMDA-receptor-mediated calcium influx. As calcium influx is necessary for NMDA-receptor-mediated excitotoxic damage to occur, its reduction likely correlates with neuroprotection (Ref. 27). Additionally, the NMDA-receptor-mediated neurotoxicity is partially dependent on the MAP kinases. Glutamate treatment results in activation of JNK, p38 and p53, and an increase in AP-1. Pretreatment with lithium prevents these changes and this action correlates with lithium's neuroprotective effects (Ref. 72).

The NMDA receptor is made up of heteromeric assemblies of NR1 and NR2 subunits. The NR1 subunit is mandatory for receptor function, and one or more NR2 (NR2A–D) subunits regulate NMDA-receptor function. In mature neurons NMDA receptor activity, and hence calcium influx and potential excitotoxic damage, is regulated by the phosphorylation states of the NR2A and NR2B subunits (Refs 114, 115). Primary regulators of these subunits are the Src family tyrosine kinases that bind to NMDA scaffolding proteins and enhance receptor activity (Ref. 116). The lithium-induced decrease in calcium influx might be due to a decrease in phosphorylation of NR2B at Tyr1472. The phosphorylation of NR2B Tyr1472 is necessary for NMDA-receptor-mediated calcium influx (Ref. 117) and its phosphorylation is believed to regulate NMDA-receptor internalisation/endocytosis (Ref. 118). Chronic lithium treatment results in a 40% decrease in Tyr1472 phosphorylation (Ref. 27). In addition, lithium decreases Src activation as measured by a reduction in the level of Tyr416-phosphorylated Src kinase (Ref. 119). As the total levels of Src kinase and NMDA scaffolding proteins are unaffected by lithium, and the decrease in Src activation parallels the reduction in Tyr1472 phosphorylation, decreased Src activation is likely responsible for lithium's regulation of Tyr1472 phosphorylation. However, the mechanism by which lithium decreases Src activation is presently unknown.

Lithium has also been shown to reduce excitotoxic-related brain damage following ischaemic events. After an ischaemic incident, two events occur that enhance NMDA-receptor function and ischaemia-induced excitotoxicity. First, Src-mediated tyrosine phosphorylation of NR2A is increased (Refs 120, 121). Second, the interaction of NR2A with Src and Fyn, mediated

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by postsynaptic density protein 95 kDa (PSD-95), is increased (Ref. 122). Lithium blocks both the ischaemia-induced increase in NR2A phosphorylation and the PSD-95 interaction (Ref. 123). This could well be regulated by the same decrease in Src function that regulates NR2B tyrosine phosphorylation.

Clinical implications/applications

Scientists have investigated lithium's mood-stabilising properties for over 50 years. After such an extended period, one would expect lithium's clinical capabilities to be well delineated and lithium to be a 'stock' drug that generates little new interest. In fact, the opposite is true: lithium's mechanisms of action are still under investigation and just starting to become understood. In addition, new findings suggest that lithium has a wide variety of clinical applications beyond bipolar disorder. First, in bipolar disorder lithium's neuroprotective properties imply a new aetiology and hence treatment paradigm. Second, lithium's neuroprotective properties suggest that lithium might have use in preventing or treating brain damage after an insult has occurred. Finally, the neuroprotective properties of long-term lithium administration suggest that this drug might have a clinical use in treating chronic and progressive neurodegenerative diseases.

Bipolar disorder is a devastating disease and an increased understanding of its underlying causes has great significance not only for research purposes, but also for a reduction of human suffering. As lithium shows great efficacy in treating bipolar disorder, new discoveries on lithium's methods of action provide new insights into the potential causes of this disease. In this case, since lithium is neuroprotective, it is possible that neuroprotection is important to the treatment of bipolar disorder. This in turn implies that there must be something against which to protect. As more details are discovered about the mechanisms of lithium neuroprotection, more specific hypotheses of what lithium is protecting against can be tested. It should be noted that this argument does not prove that neuroprotection is relevant to bipolar disorder. It is possible that lithium's other properties could be of greater relevance or that the pathways discussed as neuroprotective could also independently regulate mood. This argument does imply that neuroprotection is an area worth investigating in bipolar disorder.

Supporting evidence for the hypothesis that lithium's neuroprotection might play a role in its ability to effectively manage bipolar disorder comes from two sources. First, neuroprotection appears to be a common mechanism of action of the drugs prescribed to treat bipolar disorder (e.g. lithium, valproate, lamotrigine), with the exception of carbamazepine, which fails to protect against apoptotic insults (Refs 6, 124, 125). Second, magnetic resonance imaging and post-mortem studies on bipolar patients indicate a loss in brain tissue volume and the number of neuronal and glial cells, consistent with said hypothesis (Refs 126, 127, 128). Third, as previously discussed, lithium treatment reduces this tissue and cell loss.

The second potential clinical use for lithium's neuroprotection is in the prevention of brain damage following acute trauma. The most common form of acute brain trauma is ischaemic stroke, which is among the leading causes of death. Following a stroke there is a region surrounding the core of the insult known as the penumbra zone in which the tissue can survive for a period of time. If that tissue could be kept alive by neuroprotective agents, such as lithium, it would drastically improve a patient's chance of functional recovery (Refs 129, 130, 131). A large portion of this brain damage is induced by excessive glutamate acting on NMDA receptors (Ref. 132). Theoretically, lithium should provide excellent therapeutic protection: it is not only neuroprotective in general, but acts specifically against the primary cause of excitotoxic death following a stroke. This theoretical possibility is starting to bear fruit, as lithium is not only effective at reducing ischaemia-induced brain damage (Refs 33, 34), both in terms of decreasing infarct volume and increasing functional recovery, but more importantly, it is effective in a time frame at which it could realistically be administered to human stroke victims (Ref. 38). Stroke is just one example of an entire category of acute brain injuries, and, although it has yet to be investigated, similar implications hold for lithium's ability to prevent brain damage following other types of acute neural injury.

A third clinical use of lithium suggested by its neuroprotective properties is in the treatment of neurodegenerative diseases (Ref. 133). Neurodegenerative diseases are hereditary or sporadic diseases in which, among their defining characteristics, there is the progressive apoptotic death of specific neuronal populations;

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thus, one would expect a positive effect from neuroprotective agents that could prevent or delay said death. Two of the best-known neurodegenerative diseases are Alzheimer's disease and Huntington's disease. Beyond its general neuroprotective function, lithium, and/or molecules regulated by lithium, have been shown to inhibit pathways by which Alzheimer's disease and Huntington's disease produce toxicity.

Alzheimer's disease is the most common form of dementia. It is characterised by the accumulation of β-amyloid peptide (Aβ) and tau protein hyperphosphorylation. These events are critical to Alzheimer's-induced neurotoxicity (Ref. 134). Lithium is believed to antagonise Alzheimer's toxicity through inhibition of GSK-3β. Whereas Aβ increases GSK-3β activity and GSK-3β in turn phosphorylates tau, lithium inhibits both these phenomena and reduces Aβ-induced toxicity (Refs 135, 29, 136). This suggests that lithium will have great therapeutic benefit in Alzheimer's, and initial reports on current investigations look promising. Of further importance, it has also been shown that through inhibition of GSK-3α, not GSK-3β, lithium reduces the production of Aβ from its precursor protein (Ref. 137).

Huntington's disease is one of nine diseases caused by mutated expanded repeats of CAG, which codes for expanded polyglutamine. In Huntington's disease, the expanded area is in exon 1 of the huntingtin gene. The expansion results in cell death, particularly in the medium-sized spiny neurons of the striatum (Ref. 138). Initial research has found that lithium is neuroprotective against animal models of Huntington's disease (Ref. 139) both through Bcl-2 induction (Ref. 35) and GSK inhibition (Ref. 140). Huntington's disease might cause neurotoxicity both through a toxic gain-of-function and a loss of BDNF due to the loss of wild-type huntingtin protein (Ref. 141). The toxic gain-of-function might be ameliorated by Akt, as Akt inactivates mutant huntingtin (Ref. 142). Lithium's molecular properties seem ideally suited to treat Huntington's disease because it increases both the production of BDNF and the activity of Akt (Refs 52, 53, 59, 60, 66, 143).

Future research

In recent years, research into the neuroprotective effects of lithium has exploded and this trend will undoubtedly continue. This research falls into three broad categories. The first area is the exact mechanism and pathways by which lithium exerts its neuroprotective effects. Some of the more pressing questions include identifying molecules with which lithium directly interacts, and understanding the relative contributions of direct GSK-3β inhibition versus other pathways. It must be acknowledged that before this area can be fully understood, more basic research into the cellular machinery of apoptosis is necessary.

A second area of research concerns the role that lithium neuroprotection plays in its efficacy in treating bipolar disorder. Of particular interest are longitudinal studies and the comparison of lithium with other neuroprotective drugs used to treat this syndrome. Not all patients respond to all drugs: identifying the underlying cause of this discrepancy, and defining subtypes of bipolar disorder, will provide great advances in the efficacy of treatment.

The third area of research concerns the numerous other potential therapeutic uses of lithium. Stroke, Alzheimer's and Huntington's diseases have been specifically discussed here, but many other disease states bear investigation. As these diseases and injuries exact a great toll every year, finding treatments for them must be a research priority.

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References

1 Cade, J.F.J. (1949) Lithium salts in the treatment of psychotic excitement. Medical Journal of Australia 36, 349-352
2 Schou, M. (2001) Lithium treatment at 52. J Affect Disord 67, 21-32, PubMed: 11869750
3 Johnson, F.N. (1984) The History of Lithium Treatment, McMillan, London
4 Nonaka, S., Hough, C.J. and Chuang, D.M. (1998) Chronic lithium treatment robustly protects neurons in the central nervous system against excitotoxicity by inhibiting N-methyl-D-aspartate receptor-mediated calcium influx. Proc Natl Acad Sci U S A 95, 2642-2647, PubMed: 9482940
5 Jope, R.S. (1999) Anti-bipolar therapy: mechanism of action of lithium. Mol Psychiatry

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
Published by Cambridge University Press

https://doi.org/10.1017/S1462399404008385 Published online by Cambridge University Press

4, 117-128, PubMed: 10208444
6 Li, X., Ketter, T.A. and Frye, M.A. (2002) Synaptic, intracellular, and neuroprotective mechanisms of anticonvulsants: are they relevant for the treatment and course of bipolar disorders? J Affect Disord 69, 1-14, PubMed: 12103447
7 American Psychiatric Association (1994) Diagnosis and Statistical Manual of Mental Disorders (4th edn), American Psychiatric Association, Washington, DC
8 Narrow, W.E. et al. (2002) Revised prevalence estimates of mental disorders in the United States: using a clinical significance criterion to reconcile 2 surveys' estimates. Arch Gen Psychiatry 59, 115-123, PubMed: 11825131
9 World Health Organization (2003) World Health Report 2003, WHO, Geneva, Switzerland
10 Kang, H.J. et al. (2003) Calcium-dependent prevention of neuronal apoptosis by lithium ion: essential role of phosphoinositide 3-kinase and phospholipase Cgamma. Mol Pharmacol 64, 228-234, PubMed: 12869627
11 Lawen, A. (2003) Apoptosis-an introduction. Bioessays 25, 888-896, PubMed: 12938178
12 Cory, S. and Adams, J.M. (2002) The Bcl2 family: regulators of the cellular life-or-death switch. Nat Rev Cancer 2, 647-656, PubMed: 12209154
13 Miyashita, T. et al. (1994) Tumor suppressor p53 is a regulator of bcl-2 and bax gene expression in vitro and in vivo. Oncogene 9, 1799-1805, PubMed: 8183579
14 Loeffler, M. and Kroemer, G. (2000) The mitochondrion in cell death control: certainties and incognita. Exp Cell Res 256, 19-26, PubMed: 10739647
15 Earnshaw, W.C., Martins, L.M. and Kaufmann, S.H. (1999) Mammalian caspases: structure, activation, substrates, and functions during apoptosis. Annu Rev Biochem 68, 383-424, PubMed: 10872455
16 Huang, E.J. and Reichardt, L.F. (2003) Trk receptors: roles in neuronal signal transduction. Annu Rev Biochem 72, 609-642, PubMed: 12676795
17 Franke, T.F. et al. (2003) PI3K/Akt and apoptosis: size matters. Oncogene 22, 8983-8998, PubMed: 14663477
18 Chang, F. et al. (2003) Regulation of cell cycle progression and apoptosis by the Ras/Raf/MEK/ERK pathway (Review). Int J Oncol 22, 469-480, PubMed: 12579299
19 Doble, B.W. and Woodgett, J.R. (2003) GSK-3: tricks of the trade for a multi-tasking kinase. J Cell Sci 116, 1175-1186, PubMed: 12615961
20 Bournat, J.C., Brown, A.M. and Soler, A.P. (2000) Wnt-1 dependent activation of the survival factor NF-kappaB in PC12 cells. J Neurosci Res 61, 21-32, PubMed: 10861796
21 Bijur, G.N., De Sarno, P. and Jope, R.S. (2000) Glycogen synthase kinase-3beta facilitates staurosporine- and heat shock-induced apoptosis. Protection by lithium. J Biol Chem 275, 7583-7590, PubMed: 10713065
22 Li, R., Shen, Y. and El-Mallakh, R.S. (1994) Lithium protects against ouabain-induced cell death. Lithium 5, 211-216
23 Hennion, J.P. et al. (2002) Evaluation of neuroprotection by lithium and valproic acid against ouabain-induced cell damage. Bipolar Disord 4, 201-206, PubMed: 12180275
24 Nonaka, S., Katsube, N. and Chuang, D.M. (1998) Lithium protects rat cerebellar granule cells against apoptosis induced by anticonvulsants, phenytoin and carbamazepine. J Pharmacol Exp Ther 286, 539-547, PubMed: 9655900
25 D'Mello, S.R., Anelli, R. and Calissano, P. (1994) Lithium induces apoptosis in immature cerebellar granule cells but promotes survival of mature neurons. Exp Cell Res 211, 332-338, PubMed: 8143780
26 Mora, A. et al. (2001) Lithium inhibits caspase 3 activation and dephosphorylation of PKB and GSK3 induced by K+ deprivation in cerebellar granule cells. J Neurochem 78, 199-206, PubMed: 11432986
27 Hashimoto, R. et al. (2002) Lithium protection against glutamate excitotoxicity in rat cerebral cortical neurons: involvement of NMDA receptor inhibition possibly by decreasing NR2B tyrosine phosphorylation. J Neurochem 80, 589-597, PubMed: 11841566
28 Alvarez, G. et al. (1999) Lithium protects cultured neurons against beta-amyloid-induced neurodegeneration. FEBS Lett 453, 260-264, PubMed: 10405156
29 Wei, H. et al. (2000) beta-amyloid peptide-induced death of PC 12 cells and cerebellar granule cell neurons is inhibited by long-term lithium treatment. Eur J Pharmacol 392, 117-123, PubMed: 10762662
30 Tseng, W.P. and Lin-Shiau, S.Y. (2002) Long-term lithium treatment prevents neurotoxic effects of beta-bungarotoxin in primary cultured neurons. J Neurosci Res 69, 633-641, PubMed: 12210829
31 Watkins, J. and Collingridge, G. (1994)

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
Published by Cambridge University Press

https://doi.org/10.1017/S1462399404008385 Published online by Cambridge University Press

Phenylglycine derivatives as antagonists of metabotropic glutamate receptors. Trends Pharmacol Sci 15, 333-342, PubMed: 7992387
32 Choi, D.W. (1992) Excitotoxic cell death. J Neurobiol 23, 1261-1276, PubMed: 1361523
33 Nonaka, S. and Chuang, D.M. (1998) Neuroprotective effects of chronic lithium on focal cerebral ischemia in rats. Neuroreport 9, 2081-2084, PubMed: 9674597
34 Xu, J. et al. (2003) Chronic treatment with a low dose of lithium protects the brain against ischemic injury by reducing apoptotic death. Stroke 34, 1287-1292, PubMed: 12677021
35 Senatorov, V.V. et al. (2004) Short-term lithium treatment promotes neuronal survival and proliferation in rat striatum infused with quinolinic acid, an excitotoxic model of Huntington's disease. Mol Psychiatry 9, 371-385, PubMed: 14702090
36 Pascual, T. and Gonzalez, J.L. (1995) A protective effect of lithium on rat behaviour altered by ibotenic acid lesions of the basal forebrain cholinergic system. Brain Res 695, 289-292, PubMed: 8556348
37 Sparapani, M. et al. (1997) Effects of chronic lithium treatment on ornithine decarboxylase induction and excitotoxic neuropathology in the rat. Brain Res 765, 164-168, PubMed: 9310408
38 Ren, M. et al. (2003) Postinsult treatment with lithium reduces brain damage and facilitates neurological recovery in a rat ischemia/reperfusion model. Proc Natl Acad Sci U S A 100, 6210-6215, PubMed: 12732732
39 Bowley, M.P. et al. (2002) Low glial numbers in the amygdala in major depressive disorder. Biol Psychiatry 52, 404-412, PubMed: 12242056
40 Moore, G.J. et al. (2000) Lithium-induced increase in human brain grey matter. Lancet 356, 1241-1242, PubMed: 11072948
41 Moore, G.J. et al. (2000) Lithium increases N-acetyl-aspartate in the human brain: in vivo evidence in support of bcl-2's neurotrophic effects? Biol Psychiatry 48, 1-8, PubMed: 10913502
42 Sassi, R.B. et al. (2002) Increased gray matter volume in lithium-treated bipolar disorder patients. Neurosci Lett 329, 243-245, PubMed: 12165422
43 Kieseppa, T. et al. (2003) Reduced left hemispheric white matter volume in twins with bipolar I disorder. Biol Psychiatry 54, 896-905, PubMed: 14573317
44 Gould, T.D. et al. (2004) Emerging experimental therapeutics for bipolar disorder: insights from the molecular and cellular actions of current mood stabilizers. Mol Psychiatry 9, 734-755, PubMed: 15136794
45 Williams, R.S. et al. (2002) A common mechanism of action for three mood-stabilizing drugs. Nature 417, 292-295, PubMed: 12015604
46 Martinez, R.P. and Raffa, R.B. (2002) LiCl attenuates M(1)AChR-mediated intrathecal pilocarpine-induced reciprocal hindlimb scratching in mice. Pharmacology 65, 210-214, PubMed: 12119451
47 Berridge, M.J., Downes, C.P. and Hanley, M.R. (1989) Neural and developmental actions of lithium: a unifying hypothesis. Cell 59, 411-419, PubMed: 2553271
48 Centeno, F. et al. (1998) Partial lithium-associated protection against apoptosis induced by C2-ceramide in cerebellar granule neurons. Neuroreport 9, 4199-4203, PubMed: 9926873
49 Huang, E.J. and Reichardt, L.F. (2001) Neurotrophins: roles in neuronal development and function. Annu Rev Neurosci 24, 677-736, PubMed: 11520916
50 Russo-Neustadt, A. (2003) Brain-derived neurotrophic factor, behavior, and new directions for the treatment of mental disorders. Semin Clin Neuropsychiatry 8, 109-118, PubMed: 12728410
51 Klein, R. et al. (1991) The trkB tyrosine protein kinase is a receptor for brain-derived neurotrophic factor and neurotrophin-3. Cell 66, 395-403, PubMed: 1649702
52 Fukumoto, T. et al. (2001) Chronic lithium treatment increases the expression of brain-derived neurotrophic factor in the rat brain. Psychopharmacology (Berl) 158, 100-106, PubMed: 11685390
53 Hashimoto, R. et al. (2002) Lithium induces brain-derived neurotrophic factor and activates TrkB in rodent cortical neurons: an essential step for neuroprotection against glutamate excitotoxicity. Neuropharmacology 43, 1173-1179, PubMed: 12504924
54 Brunet, A., Datta, S.R. and Greenberg, M.E. (2001) Transcription-dependent and -independent control of neuronal survival by the PI3K-Akt signaling pathway. Curr Opin Neurobiol 11, 297-305, PubMed: 11399427
55 Chan, T.O., Rittenhouse, S.E. and Tsichlis, P.N. (1999) AKT/PKB and other D3 phosphoinositide-regulated kinases: kinase activation by phosphoinositide-dependent phosphorylation. Annu Rev Biochem 68, 965-

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
Published by Cambridge University Press

https://doi.org/10.1017/S1462399404008385 Published online by Cambridge University Press

1014, PubMed: 10872470
56 Scheid, M.P. and Woodgett, J.R. (2003) Unravelling the activation mechanisms of protein kinase B/Akt. FEBS Lett 546, 108-112, PubMed: 12829245
57 Neri, L.M. et al. (2002) The nuclear phosphoinositide 3-kinase/AKT pathway: a new second messenger system. Biochim Biophys Acta 1584, 73-80, PubMed: 12385889
58 Nicholson, K.M. and Anderson, N.G. (2002) The protein kinase B/Akt signalling pathway in human malignancy. Cell Signal 14, 381-395, PubMed: 11882383
59 Chalecka-Franaszek, E. and Chuang, D.M. (1999) Lithium activates the serine/threonine kinase Akt-1 and suppresses glutamate-induced inhibition of Akt-1 activity in neurons. Proc Natl Acad Sci U S A 96, 8745-8750, PubMed: 10411946
60 Kumari, S. et al. (2001) Distinct phosphorylation patterns underlie Akt activation by different survival factors in neurons. Brain Res Mol Brain Res 96, 157-162, PubMed: 11731021
61 De Sarno, P., Li, X. and Jope, R.S. (2002) Regulation of Akt and glycogen synthase kinase-3 beta phosphorylation by sodium valproate and lithium. Neuropharmacology 43, 1158-1164, PubMed: 12504922
62 Zhang, F. et al. (2003) Inhibitory phosphorylation of glycogen synthase kinase-3 (GSK-3) in response to lithium. Evidence for autoregulation of GSK-3. J Biol Chem 278, 33067-33077, PubMed: 12796505
63 Segal, R.A. and Greenberg, M.E. (1996) Intracellular signaling pathways activated by neurotrophic factors. Annu Rev Neurosci 19, 463-489, PubMed: 8833451
64 Yan, J. et al. (1998) Ras isoforms vary in their ability to activate Raf-1 and phosphoinositide 3-kinase. J Biol Chem 273, 24052-24056, PubMed: 9727023
65 Steelman, L.S. et al. (2004) JAK/STAT, Raf/MEK/ERK, PI3K/Akt and BCR-ABL in cell cycle progression and leukemogenesis. Leukemia 18, 189-218, PubMed: 14737178
66 Einat, H. et al. (2003) The role of the extracellular signal-regulated kinase signaling pathway in mood modulation. J Neurosci 23, 7311-7316, PubMed: 12917364
67 Pardo, R. et al. (2003) Opposed effects of lithium on the MEK-ERK pathway in neural cells: inhibition in astrocytes and stimulation in neurons by GSK3 independent mechanisms. J Neurochem 87, 417-426, PubMed: 14511119
68 Tibbles, L.A. and Woodgett, J.R. (1999) The stress-activated protein kinase pathways. Cell Mol Life Sci 55, 1230-1254, PubMed: 10487205
69 Mielke, K. and Herdegen, T. (2000) JNK and p38 stresskinases—degenerative effectors of signal-transduction-cascades in the nervous system. Prog Neurobiol 61, 45-60, PubMed: 10759064
70 Whitmarsh, A.J. and Davis, R.J. (1996) Transcription factor AP-1 regulation by mitogen-activated protein kinase signal transduction pathways. J Mol Med 74, 589-607, PubMed: 8912180
71 Sionov, R.V. and Haupt, Y. (1999) The cellular response to p53: the decision between life and death. Oncogene 18, 6145-6157, PubMed: 10557106
72 Chen, R.W. et al. (2003) Regulation of c-Jun N-terminal kinase, p38 kinase and AP-1 DNA binding in cultured brain neurons: roles in glutamate excitotoxicity and lithium neuroprotection. J Neurochem 84, 566-575, PubMed: 12558976
73 Hongisto, V. et al. (2003) Lithium blocks the c-Jun stress response and protects neurons via its action on glycogen synthase kinase 3. Mol Cell Biol 23, 6027-6036, PubMed: 12917327
74 Jope, R.S. and Bijur, G.N. (2002) Mood stabilizers, glycogen synthase kinase-3beta and cell survival. Mol Psychiatry 7 Suppl 1, S35-45, PubMed: 11986994
75 Grimes, C.A. and Jope, R.S. (2001) CREB DNA binding activity is inhibited by glycogen synthase kinase-3 beta and facilitated by lithium. J Neurochem 78, 1219-1232, PubMed: 11579131
76 Jope, R.S. (2003) Lithium and GSK-3: one inhibitor, two inhibitory actions, multiple outcomes. Trends Pharmacol Sci 24, 441-443, PubMed: 12967765
77 Stambolic, V., Ruel, L. and Woodgett, J.R. (1996) Lithium inhibits glycogen synthase kinase-3 activity and mimics wingless signalling in intact cells. Curr Biol 6, 1664-1668, PubMed: 8994831
78 Klein, P.S. and Melton, D.A. (1996) A molecular mechanism for the effect of lithium on development. Proc Natl Acad Sci U S A 93, 8455-8459, PubMed: 8710892
79 Ryves, W.J. and Harwood, A.J. (2001) Lithium inhibits glycogen synthase kinase-3 by competition for magnesium. Biochem Biophys Res Commun 280, 720-725, PubMed: 11162580
80 Wang, Q.M. et al. (1994) Glycogen synthase kinase-3 beta is a dual specificity kinase differentially regulated by tyrosine and serine/

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
Published by Cambridge University Press

https://doi.org/10.1017/S1462399404008385 Published online by Cambridge University Press

threonine phosphorylation. J Biol Chem 269, 14566-14574, PubMed: 7514173
81 Li, X., Bijur, G.N. and Jope, R.S. (2002) Glycogen synthase kinase-3beta, mood stabilizers, and neuroprotection. Bipolar Disord 4, 137-144, PubMed: 12071511
82 Eastman, Q. and Grosschedl, R. (1999) Regulation of LEF-1/TCF transcription factors by Wnt and other signals. Curr Opin Cell Biol 11, 233-240, PubMed: 10209158
83 Roose, J. and Clevers, H. (1999) TCF transcription factors: molecular switches in carcinogenesis. Biochim Biophys Acta 1424, M23-37, PubMed: 10528152
84 Seidensticker, M.J. and Behrens, J. (2000) Biochemical interactions in the wnt pathway. Biochim Biophys Acta 1495, 168-182, PubMed: 10656974
85 Huelsken, J. and Behrens, J. (2002) The Wnt signalling pathway. J Cell Sci 115, 3977-3978, PubMed: 12356903
86 Moon, R.T. et al. (2002) The promise and perils of Wnt signaling through beta-catenin. Science 296, 1644-1646, PubMed: 12040179
87 Chen, G. et al. (1999) The mood-stabilizing agent valproate inhibits the activity of glycogen synthase kinase-3. J Neurochem 72, 1327-1330, PubMed: 10037507
88 Gould, T.D., Chen, G. and Manji, H.K. (2004) In vivo evidence in the brain for lithium inhibition of glycogen synthase kinase-3. Neuropsychopharmacology 29, 32-38, PubMed: 12942141
89 Takayama, S., Reed, J.C. and Homma, S. (2003) Heat-shock proteins as regulators of apoptosis. Oncogene 22, 9041-9047, PubMed: 14663482
90 Gao, T. and Newton, A.C. (2002) The turn motif is a phosphorylation switch that regulates the binding of Hsp70 to protein kinase C. J Biol Chem 277, 31585-31592, PubMed: 12080070
91 Beere, H.M. et al. (2000) Heat-shock protein 70 inhibits apoptosis by preventing recruitment of procaspase-9 to the Apaf-1 apoptosome. Nat Cell Biol 2, 469-475, PubMed: 10934466
92 Mosser, D.D. et al. (2000) The chaperone function of hsp70 is required for protection against stress-induced apoptosis. Mol Cell Biol 20, 7146-7159, PubMed: 10982831
93 Chu, B. et al. (1996) Sequential phosphorylation by mitogen-activated protein kinase and glycogen synthase kinase 3 represses transcriptional activation by heat shock factor-1. J Biol Chem 271, 30847-30857, PubMed: 8940068
94 Bijur, G.N. and Jope, R.S. (2000) Opposing actions of phosphatidylinositol 3-kinase and glycogen synthase kinase-3beta in the regulation of HSF-1 activity. J Neurochem 75, 2401-2408, PubMed: 11080191
95 Xavier, I.J. et al. (2000) Glycogen synthase kinase 3beta negatively regulates both DNA-binding and transcriptional activities of heat shock factor 1. J Biol Chem 275, 29147-29152, PubMed: 10856293
96 Shaulian, E. and Karin, M. (2002) AP-1 as a regulator of cell life and death. Nat Cell Biol 4, E131-136, PubMed: 11988758
97 Boyle, W.J. et al. (1991) Activation of protein kinase C decreases phosphorylation of c-Jun at sites that negatively regulate its DNA-binding activity. Cell 64, 573-584, PubMed: 1846781
98 Karin, M. (1998) Mitogen-activated protein kinase cascades as regulators of stress responses. Ann N Y Acad Sci 851, 139-146, PubMed: 9668616
99 Wisdom, R. (1999) AP-1: one switch for many signals. Exp Cell Res 253, 180-185, PubMed: 10579922
100 Ozaki, N. and Chuang, D.M. (1997) Lithium increases transcription factor binding to AP-1 and cyclic AMP-responsive element in cultured neurons and rat brain. J Neurochem 69, 2336-2344, PubMed: 9375664
101 Asghari, V. et al. (1998) Differential effects of mood stabilizers on Fos/Jun proteins and AP-1 DNA binding activity in human neuroblastoma SH-SY5Y cells. Brain Res Mol Brain Res 58, 95-102, PubMed: 9685595
102 Impey, S. et al. (1996) Induction of CRE-mediated gene expression by stimuli that generate long-lasting LTP in area CA1 of the hippocampus. Neuron 16, 973-982, PubMed: 8630255
103 Bonni, A. et al. (1999) Cell survival promoted by the Ras-MAPK signaling pathway by transcription-dependent and -independent mechanisms. Science 286, 1358-1362, PubMed: 10558990
104 Kopnisky, K.L. et al. (2003) Chronic lithium treatment antagonizes glutamate-induced decrease of phosphorylated CREB in neurons via reducing protein phosphatase 1 and increasing MEK activities. Neuroscience 116, 425-435, PubMed: 12559097
105 Hansen, T.O., Rehfeld, J.F. and Nielsen, F.C. (2004) GSK-3beta reduces cAMP-induced cholecystokinin gene expression by inhibiting CREB binding. Neuroreport 15, 841-845, PubMed: 15073527

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
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106 Finkbeiner, S. (2000) CREB couples neurotrophin signals to survival messages. Neuron 25, 11-14, PubMed: 10707967
107 Freeland, K., Boxer, L.M. and Latchman, D.S. (2001) The cyclic AMP response element in the Bcl-2 promoter confers inducibility by hypoxia in neuronal cells. Brain Res Mol Brain Res 92, 98-106, PubMed: 11483246
108 Shieh, P.B. et al. (1998) Identification of a signaling pathway involved in calcium regulation of BDNF expression. Neuron 20, 727-740, PubMed: 9581764
109 Chen, R.W. and Chuang, D.M. (1999) Long term lithium treatment suppresses p53 and Bax expression but increases Bcl-2 expression. A prominent role in neuroprotection against excitotoxicity. J Biol Chem 274, 6039-6042, PubMed: 10037682
110 Manji, H.K., Moore, G.J. and Chen, G. (2000) Lithium up-regulates the cytoprotective protein Bcl-2 in the CNS in vivo: a role for neurotrophic and neuroprotective effects in manic depressive illness. J Clin Psychiatry 61 Suppl 9, 82-96, PubMed: 10826666
111 Corson, T.W. et al. (2004) Cell-type specific regulation of calreticulin and Bcl-2 expression by mood stabilizer drugs. Eur Neuropsychopharmacol 14, 143-150, PubMed: 15013030
112 Stout, A.K. et al. (1998) Glutamate-induced neuron death requires mitochondrial calcium uptake. Nat Neurosci 1, 366-373, PubMed: 10196525
113 Sattler, R. and Tymianski, M. (2001) Molecular mechanisms of glutamate receptor-mediated excitotoxic neuronal cell death. Mol Neurobiol 24, 107-129, PubMed: 11831548
114 Yu, X.M. et al. (1997) NMDA channel regulation by channel-associated protein tyrosine kinase Src. Science 275, 674-678, PubMed: 9005855
115 Salter, M.W. (1998) Src, N-methyl-D-aspartate (NMDA) receptors, and synaptic plasticity. Biochem Pharmacol 56, 789-798, PubMed: 9774140
116 Ali, D.W. and Salter, M.W. (2001) NMDA receptor regulation by Src kinase signalling in excitatory synaptic transmission and plasticity. Curr Opin Neurobiol 11, 336-342, PubMed: 11399432
117 Takasu, M.A. et al. (2002) Modulation of NMDA receptor-dependent calcium influx and gene expression through EphB receptors. Science 295, 491-495, PubMed: 11799243
118 Lavezzari, G. et al. (2003) Differential binding of the AP-2 adaptor complex and PSD-95 to the C-terminus of the NMDA receptor subunit NR2B regulates surface expression. Neuropharmacology 45, 729-737, PubMed: 14529712
119 Hashimoto, R. et al. (2003) Lithium-induced inhibition of Src tyrosine kinase in rat cerebral cortical neurons: a role in neuroprotection against N-methyl-D-aspartate receptor-mediated excitotoxicity. FEBS Lett 538, 145-148, PubMed: 12633868
120 Takagi, N. et al. (1997) Transient ischemia differentially increases tyrosine phosphorylation of NMDA receptor subunits 2A and 2B. J Neurochem 69, 1060-1065, PubMed: 9282928
121 Liu, Y. et al. (2001) NMDA receptor activation results in tyrosine phosphorylation of NMDA receptor subunit 2A(NR2A) and interaction of Pyk2 and Src with NR2A after transient cerebral ischemia and reperfusion. Brain Res 909, 51-58, PubMed: 11478920
122 Hou, X.Y. et al. (2002) Activation of NMDA receptors and L-type voltage-gated calcium channels mediates enhanced formation of Fyn-PSD95-NR2A complex after transient brain ischemia. Brain Res 955, 123-132, PubMed: 12419528
123 Ma, J. and Zhang, G.Y. (2003) Lithium reduced N-methyl-D-aspartate receptor subunit 2A tyrosine phosphorylation and its interactions with Src and Fyn mediated by PSD-95 in rat hippocampus following cerebral ischemia. Neurosci Lett 348, 185-189, PubMed: 12932824
124 Manji, H.K. and Duman, R.S. (2001) Impairments of neuroplasticity and cellular resilience in severe mood disorders: implications for the development of novel therapeutics. Psychopharmacol Bull 35, 5-49, PubMed: 12397885
125 Ketter, T.A., Manji, H.K. and Post, R.M. (2003) Potential mechanisms of action of lamotrigine in the treatment of bipolar disorders. J Clin Psychopharmacol 23, 484-495, PubMed: 14520126
126 Drevets, W.C. (2001) Neuroimaging and neuropathological studies of depression: implications for the cognitive-emotional features of mood disorders. Curr Opin Neurobiol 11, 240-249, PubMed: 11301246
127 Manji, H.K. et al. (2003) Enhancing neuronal plasticity and cellular resilience to develop novel, improved therapeutics for difficult-to-treat depression. Biol Psychiatry 53, 707-742, PubMed: 12706957

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
Published by Cambridge University Press

https://doi.org/10.1017/S1462399404008385 Published online by Cambridge University Press

128 Post, R.M. et al. (2003) Neurobiology of bipolar illness: implications for future study and therapeutics. Ann Clin Psychiatry 15, 85-94, PubMed: 12938866
129 Friedlander, R.M. (2003) Apoptosis and caspases in neurodegenerative diseases. N Engl J Med 348, 1365-1375, PubMed: 12672865
130 Janardhan, V. and Qureshi, A.I. (2004) Mechanisms of ischemic brain injury. Curr Cardiol Rep 6, 117-123, PubMed: 14759356
131 Wahlgren, N.G. and Ahmed, N. (2004) Neuroprotection in cerebral ischaemia: facts and fancies—the need for new approaches. Cerebrovasc Dis 17 Suppl 1, 153-166, PubMed: 14694293
132 Lipton, P. (1999) Ischemic cell death in brain neurons. Physiol Rev 79, 1431-1568, PubMed: 10508238
133 Chuang, D.M. et al. (2002) Neuroprotective effects of lithium in cultured cells and animal models of diseases. Bipolar Disord 4, 129-136, PubMed: 12071510
134 Selkoe, D.J. (2001) Alzheimer's disease: genes, proteins, and therapy. Physiol Rev 81, 741-766, PubMed: 11274343
135 Hong, M. et al. (1997) Lithium reduces tau phosphorylation by inhibition of glycogen synthase kinase-3. J Biol Chem 272, 25326-25332, PubMed: 9312151
136 Alvarez, G. et al. (2002) Regulation of tau phosphorylation and protection against beta-amyloid-induced neurodegeneration by lithium. Possible implications for Alzheimer's disease. Bipolar Disord 4, 153-165, PubMed: 12180271
137 Phiel, C.J. et al. (2003) GSK-3alpha regulates production of Alzheimer's disease amyloid-beta peptides. Nature 423, 435-439, PubMed: 12761548
138 Hickey, M.A. and Chesselet, M.F. (2003) Apoptosis in Huntington's disease. Prog Neuropsychopharmacol Biol Psychiatry 27, 255-265, PubMed: 12657365
139 Wei, H. et al. (2001) Lithium suppresses excitotoxicity-induced striatal lesions in a rat model of Huntington's disease. Neuroscience 106, 603-612, PubMed: 11591460
140 Carmichael, J. et al. (2002) Glycogen synthase kinase-3beta inhibitors prevent cellular polyglutamine toxicity caused by the Huntington's disease mutation. J Biol Chem 277, 33791-33798, PubMed: 12097329
141 Zuccato, C. et al. (2001) Loss of huntingtin-mediated BDNF gene transcription in Huntington's disease. Science 293, 493-498, PubMed: 11408619
142 Humbert, S. et al. (2002) The IGF-1/Akt pathway is neuroprotective in Huntington's disease and involves Huntingtin phosphorylation by Akt. Dev Cell 2, 831-837, PubMed: 12062094
143 Chuang, D.M. Lithium protection from excitotoxicity: therapeutic implications. Clin Neurosci Res (in press)

Accession information: DOI: 10.1017/S1462399404008385; Vol. 6; Issue 21; 8 October 2004
Published by Cambridge University Press

https://doi.org/10.1017/S1462399404008385 Published online by Cambridge University Press

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Figures

Figure 1. An overview of pathways regulating apoptosis.
Figure 2. A proposed scheme for the neuroprotective effects of lithium.

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