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Targeting molecules to medicine with mTOR, autophagy and neurodegenerative disorders

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Neurodegenerative disorders are significantly increasing in incidence as the age of the global population continues to climb with improved life expectancy. At present, more than 30 million individuals throughout the world are impacted by acute and chronic neurodegenerative disorders with limited treatment strategies. The mechanistic target of rapamycin (mTOR), also known as the mammalian target of rapamycin, is a 289 kDa serine/threonine protein kinase that offers exciting possibilities for novel treatment strategies for a host of neurodegenerative diseases that include Alzheimer's disease, Parkinson's disease, Huntington's disease, epilepsy, stroke and trauma. mTOR governs the programmed cell death pathways of apoptosis and autophagy that can determine neuronal stem cell development, precursor cell differentiation, cell senescence, cell survival and ultimate cell fate. Coupled to the cellular biology of mTOR are a number of considerations for the development of novel treatments involving the fine control of mTOR signalling, tumourigenesis, complexity of the apoptosis and autophagy relationship, functional outcome in the nervous system, and the intimately linked pathways of growth factors, phosphoinositide 3-kinase (PI 3-K), protein kinase B (Akt), AMP activated protein kinase (AMPK), silent mating type information regulation two homologue one (*Saccharomyces cerevisiae*) (SIRT1) and others. Effective clinical translation of the cellular signalling mechanisms of mTOR offers provocative avenues for new drug development in the nervous system tempered only by the need to elucidate further the intricacies of the mTOR pathway.

Introduction

Neurodegenerative disorders progressively lead to dysfunction of the nervous system and include more than 600 disease entities [1]. Acute and chronic neurodegenerative disorders result in disability and death for greater than 30 million individuals worldwide and the number of individuals afflicted is expected to increase [2]. One factor that may be fueling this rise is the improvements in clinical care that have fostered the increased life span of the global population. The age of the global population continues to increase with life expectancy approaching 80 years for individuals [3]. In conjunction, there has been a 1 % decrease in the age-adjusted death rate from the years 2000 through 2011 [4]. The number of individuals over the age of 65 years also has doubled during the previous 50 years [5]. It is expected that the number of elderly individuals in large developing countries such as India and China also will increase from 5 % to 10 % over the next several decades [6, 7].

In tandem with an increase in the life span of the global population has been a rise in non-communicable diseases (NCDs) [8]. According to the World Health Organization, more than 60 % of the 57 million global deaths are caused by NCDs [9]. Of the NCDs, the five leading causes of death are cardiac disease, cancer, chronic lower respiratory disease, stroke and traumatic accidents [10]. In regards to cardiovascular disease, high cholesterol and hypertension are important risk factors for this disorder with hypertension alone contributing to 13 % of all deaths [11].

Interestingly, cardiovascular disease in combination with hypercholesterolaemia and hypertension can significantly lead to disorders of the nervous system that include cognitive loss and stroke. In relation to dementia, 10 % of the world's population over the age of 65 years is affected with the sporadic form of Alzheimer's disease (AD) [12]. In

the United States (US) alone, greater than 5 million individuals have AD and another 3.5 million are under treatment at an annual cost of 4 billion US dollars. With the rise in the age of the population, it is expected that the number of individuals with AD will increase to 30 million individuals over the next 15 to 20 years [2, 12, 13]. In contrast, familial cases of AD (FAD) affect a significantly smaller proportion of the population. FAD is an autosomal dominant form of a mutated amyloid precursor protein (APP) gene, presents in approximately 200 families worldwide [14], occurs most often prior to age 55 years [15] and results from variable single gene mutations on chromosomes 1, 14 and 21 [16]. Other neurodegenerative disorders also will place a severe burden on the global population [3, 17]. By the year 2030, epilepsy will affect over 50 million people and peripheral neuropathies are estimated to afflict almost 300 million individuals [18]. Parkinson's disease (PD), also a progressive disorder and the second most common neurodegenerative disease [19, 20], affects 1 to 4 % of individuals over the age of 60 years in the world and this number of individuals is expected to double by the year 2030. Currently, 50 000 new cases of PD present each year in the US.

Clinical treatments for neurodegenerative disorders have progressed over the years with some promising results. For example, stroke is no longer ranked as the third leading cause of death among cardiac disease, cancer, chronic lower respiratory disease and traumatic accidents. Preventive care has fostered a reduction in disability and death with stroke. Reduction in tobacco consumption, improved control of hypertension and low density lipoprotein cholesterol disorders, heightened public education for the need for rapid treatment of stroke, and focused care on metabolic disorders such as diabetes mellitus (DM) most likely have led to this lower ranking for stroke [3, 21]. Treatment with recombinant tissue plasminogen activator has resulted in a reduction in mortality and morbidity in patients presenting with stroke, but this is only applicable to a sub-group of patients who require a narrow therapeutic window [22, 23]. Stroke, similar to other neurodegenerative disorders, continues to remain as a significant cause of death [10] and affects 15 million individuals every year [3]. Overall, the availability of treatments that can limit or halt neurodegenerative disorders continues to remain limited.

mTOR components and signalling pathways

One promising avenue to target and advance the development of therapeutic strategies for neurodegenerative disorders is the mechanistic target of rapamycin (mTOR) [8, 24]. mTOR also is known as the mammalian target of rapamycin and the FK506-binding protein 12-rapamycin complex-associated protein 1. mTOR controls multiple functions in cells that govern the transcription of genes, protein formation, proliferation and senescence of cells, cellular metabolism and cellular longevity.

Initially, the target of rapamycin (TOR) was observed in *Saccharomyces cerevisiae* with the genes *TOR1* and *TOR2* [25]. These genes were found to encode two isoforms in yeast, Tor1 and Tor2, through the use of rapamycin-resistant TOR mutants [26]. Rapamycin itself is a macrolide antibiotic [27] originating from *Streptomyces hygroscopicus* that inhibits TOR and mTOR activity [8]. mTOR is a primary component of the protein complexes mTOR complex one (mTORC1) and mTOR complex two (mTORC2) (Figure 1) [12, 19, 28, 29]. Rapamycin blocks mTORC1 activity by binding to immunophilin FK-506-binding protein 12 (FKBP12) that attaches to the FKBP12-rapamycin-binding domain (FRB) at the carboxy (C)-terminal of mTOR to prevent the phosphorylation of mTOR [30]. Overall, mTORC1 is more sensitive to inhibition by rapamycin than mTORC2 [31]. However, chronic administration of rapamycin can inhibit mTORC2 activity that can arise from the disruption of the assembly of mTORC2 [31].

The terminal domains of mTOR control catalytic activity, binding and phosphorylation of this protein. The C-terminal domain has a sequence homology to the catalytic domain of the phosphoinositide 3-kinase (PI 3-K) family [32] and contains several phosphorylation sites that regulate mTOR. The C-terminal domain of mTOR regulates catalytic activity and includes the FAT (FKBP12-rapamycin-associated protein (FRAP), ataxia-telangiectasia (ATM) and the transactivation/transformation domain-associated protein) domain, the FRB domain, the catalytic PI 3/PI 4-related kinase domain and the FATC (the FAT-C-terminal) domain [25]. The FAT domain is adjacent to the FRB domain that promotes the association of mTOR and FKBP12 when bound to rapamycin. Following these domains are the PI 3/PI 4-related kinase domain and the small FATC domain. The N-terminal domain of mTOR contains at least a 20 HEAT (huntingtin, elongation factor 3, a subunit of protein phosphatase-2A and TOR1) repeat that leads to binding with the regulatory proteins Raptor (Regulatory-associated protein of mTOR) and Rictor (rapamycin-insensitive companion of mTOR) [33].

mTORC1 is composed of Raptor, the proline rich Akt substrate 40 kDa (PRAS40), Deptor (DEP domain-containing mTOR interacting protein) and mammalian lethal with Sec13 protein 8, termed mLST8 or G protein β subunit-like (GβL) (Figure 1) [25]. Phosphorylation of Raptor can occur through several pathways. One pathway that allows mTORC1 to bind to constituents involves the protein Ras homologue enriched in brain (Rheb) that phosphorylates the Raptor residue serine^{863} and other residues that include serine^{859}, serine^{855}, serine^{877}, serine^{696} and threonine^{706} [34]. Once active, mTOR also can control Raptor activity that can be blocked by rapamycin [35]. As

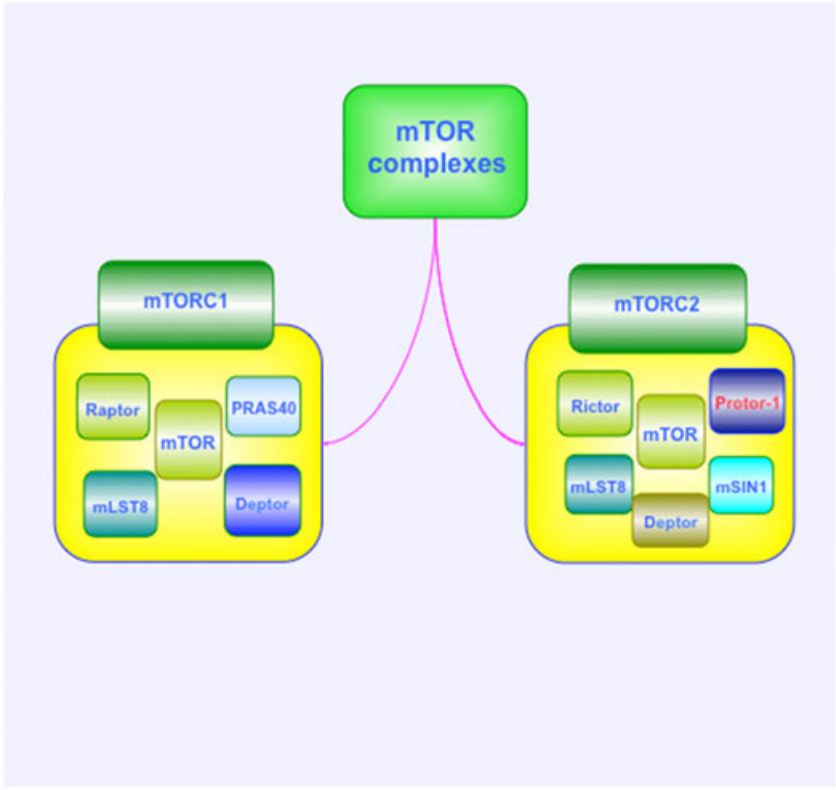

Figure 1

mTOR is a significant component of mTORC1 and mTORC2. The mechanistic target of rapamycin (mTOR) is significant for the protein complexes of mTOR complex one (mTORC1) and mTOR complex two (mTORC2). mTORC1 consists of Raptor (regulatory-associated protein of mTOR), Deptor (DEP domain-containing mTOR interacting protein), mammalian lethal with Sec13 protein 8 (mLST8/GβL) and the proline rich Akt substrate 40 kDa (PRAS40). mTORC2 is composed of Rictor (rapamycin-insensitive companion of mTOR), Deptor, the mammalian stress-activated protein kinase interacting protein (mSIN1), mLST8, and the protein observed with Rictor-1 (Protor-1)

an inhibitor of this system, PRAS40 prevents the binding of mTORC1 to Raptor [36]. PRAS40 can inhibit mTORC1 activity by preventing the association of p70 ribosomal S6 kinase (p70S6K) and the eukaryotic initiation factor 4E (eIF4E)-binding protein one (4EBP1) with Raptor [2, 36]. If PRAS40 is phosphorylated by protein kinase B (Akt), PRAS40 disengages from Raptor and is then sequestered in the cytoplasm by the docking protein 14–3-3 [36–40]. This allows activation of mTORC1. Deptor also inhibits mTORC1 activity by binding to the FAT domain of mTOR. If Deptor activity is limited, Akt, mTORC1, and mTORC2 activity are increased [41]. mLST8 is a promoter of mTOR kinase activity through p70S6K and 4EBP1 that bind to Raptor [42], controls insulin signalling through the transcription factor FoxO3 [43], is necessary for Akt and protein kinase C-α (PKCα) phosphorylation [43] and is required for Rictor to associate with mTOR [43].

mTORC2 is composed of Rictor, mLST8, Deptor, the mammalian stress-activated protein kinase interacting protein (mSIN1) and the protein observed with Rictor-1 (Protor-1) (Figure 1) [3, 33, 44–47]. The integrity of mSIN1 and prevention of its lysosomal degradation is controlled by the kinase domain of mTOR that phosphorylates mSIN1 [48]. In addition, Rictor [49] and mSIN1 [50] can activate Akt at serine^{473} and facilitate threonine^{308} phosphorylation by phosphoinositide-dependent kinase one (PDK1) to promote cell survival [2, 44, 50]. mTORC2 oversees cytoskeleton remodelling through PKCα and cell migration through the Rac guanine nucleotide exchange factors P-Rex1 and P-Rex2 and through Rho signalling [51]. In regards to Protor-1, this protein is a Rictor-binding subunit of mTORC2 that activates serum and glucocorticoid induced protein kinase one (SGK1) [52]. mTORC2 also can activate SGK1, a member of the protein kinase A/protein kinase G/protein kinase C (AGC) family of protein kinases [53].

The pathways of PI 3-K, Akt, and AMP activated protein kinase (AMPK) play a significant role in mTOR signalling (Figure 2). In regards to AMPK, this protein can inhibit mTORC1 activity through activation of the hamartin (tuberous sclerosis 1)/tuberin (tuberous sclerosis 2) (TSC1/TSC2) complex. In contrast, mTORC2 activity is increased during activation of the TSC1/TSC2 complex through the N-terminal region of TSC2 and the C-terminal region of Rictor [54]. Removal of the TSC1/TSC2 complex leads to the loss of mTORC2 kinase activity *in vitro* [54]. Of note, TSC2 functions as a GTPase-activating protein (GAP) converting G protein

Figure 2

Targeting mTOR for neurodegenerative disorders. The mechanistic target of rapamycin (mTOR) and the signalling pathways of phosphoinositide three-kinase (PI 3-K), AMP activated protein kinase (AMPK), protein kinase B (Akt) and the hamartin (tuberous sclerosis 1)/tuberin (tuberous sclerosis 2) (TSC1/TSC2) complex are vital determinants of cell survival in the nervous system. mTOR governs programmed cell death pathways of apoptosis and autophagy that can affect stem cell development and maintenance, growth factors (erythropoietin (EPO), epidermal growth factor (EGF), fibroblast growth factor (FGF), brain derived neurotrophic growth factor (BDNF)), silent mating type information regulation two homologue one (*Saccharomyces cerevisiae*) (SIRT1), Wnt signalling, and Wnt1 inducible signalling pathway protein one (WISP1). Ultimately, mTOR signalling can impact the onset and progression of multiple disorders in the nervous system that include Alzheimer's disease (AD), Parkinson's disease (PD), Huntington's disease (HD), epilepsy, stroke and trauma

Rheb (Rheb-GTP) into the inactive GDP-bound form (Rheb-GDP). If Rheb-GTP is active, Rheb-GTP can then associate with Raptor to control the binding of 4EBP1 to mTORC1 and increase mTORC1 activity [55]. AMPK phosphorylates TSC2 to increase the activity of GAP to change Rheb-GTP into the inactive Rheb-GDP to block mTORC1 activity [56]. AMPK also can increase RTP801 (REDD1/product of the *Ddit4* gene) expression during hypoxia to control TSC1/TSC2 activity and suppress mTORC1 activity by releasing TSC2 from its inhibitory binding to protein 14–3-3 [57]. AMPK also has a relationship with the sirtuin silent mating type information regulation two homologue one (*Saccharomyces cerevisiae*) (SIRT1) that can be important for cellular survival and stem cell proliferation [6]. AMPK increases nicotinamide phosphoribosyltransferase (NAMPT) activity. This process catalyzes the conversion of nicotinamide to nicotinamide mononucleotide [58], increases nicotinamide adenine dinucleotide (NAD^+) levels, decreases levels of the SIRT1 inhibitor nicotinamide and leads to SIRT1 transcription [59–61]. As a result of increasing the intracellular NAD^+ : NADH ratio, AMPK leads to deacetylation of the SIRT1 targets peroxisome proliferator-activated receptor-gamma co-activator one (PGC-1α) and forkhead transcription factors that include FoxO1 [62] and FoxO3a [63]. Although SIRT1 may function similar to AMPK as an inhibitor of mTOR to block its activity [64], SIRT1 and AMPK may be protective in combination since they can foster the induction of autophagy that can protect endothelial cells exposed to oxidized low density lipoproteins [65].

Akt is another component of the the mTOR signalling pathway that can block activity of the TSC1/TSC2 complex that inhibits mTORC1 [66–69]. Control of the TSC1/TSC2 complex is primarily mediated though the phosphorylation of TSC2 by Akt. However, extracellular signal-regulated kinases (ERKs), activating protein p90 ribosomal S6 kinase one (RSK1), glycogen synthase kinase-3β (GSK-3β) and, as previously described, AMPK, can modulate the TSC1/TSC2 complex. Akt can phosphorylate TSC2 on several sites that leads to TSC2 destabilization and disruption of TSC2 association with

TSC1. Phosphorylation of TSC2 at serine[939], serine[981] and threonine[1462] results in the binding of TSC2 to cytoplasmic protein 14–3-3, disengagement of the TSC1/TSC2 complex and activation of Rheb and mTORC1 [70]. It should be noted that during some instances to promote cell survival, a limited activity of TSC2 and AMPK [71] is required since complete knockdown of TSC2 can result in cellular injury [72].

mTOR pathways of apoptosis and autophagy

Critical for mTOR to affect the onset and progression of neurodegenerative disorders is the ability of the mTOR pathway to alter programmed cell death through apoptosis and autophagy (Figure 2). Apoptosis has two distinct phases that consist of an early phase that involves the loss of plasma membrane phosphatidylserine (PS) asymmetry and a subsequent phase that leads to genomic DNA degradation [73–75]. Apoptosis is the result of a series of cascade activation of nucleases and proteases that involve caspases [59, 76]. Membrane PS externalization activates inflammatory cells to engulf and remove injured cells [77–80]. If this process is blocked since the loss of PS asymmetry can be reversible [7, 81, 82], loss of functional cells expressing membrane PS residues can be prevented. If PS asymmetry is not reversed, genomic DNA degradation can result [73, 74, 77, 78, 83–85]. The destruction of cellular DNA is usually not considered to be a reversible process [86–89].

Under most conditions, mTOR activation prevents apoptotic cell death in the nervous system [19, 47]. If mTOR activity is lost, this leads to apoptotic neuronal cell death [90] and aggravation of oxidative stress pathways [91]. mTOR activation can protect against neuronal injury during ischaemic preconditioning [92], permanent cerebral ischaemia [93], loss of neurite outgrowth [94], cervical spinal cord injury [95], memory loss [96] and β-amyloid (Aβ) toxicity [39, 72, 97–99]. Components of the mTOR pathway, such as p70S6K, promote progenitor cell neovascularization in ischaemic tissue [100], block cortical ischaemic injury [101, 102], provide growth factor neuronal cell protection during apoptosis [37, 39] and block apoptosis during oxidative stress exposure [103, 104].

Protection against apoptotic cell injury with mTOR is closely associated with trophic factor activity [105]. The growth factors epidermal growth factor (EGF) and fibroblast growth factor (FGF) in mice are protective of stem cells and neurons [106–110]. EGF and FGF also rely upon mTOR to maintain the proliferation of neural stem and progenitor cells [111]. Through PI 3-K, Akt and mTOR, EGF prevents cell injury during metabolic stress [112] and restores memory function [113]. Brain derived neurotrophic factor (BDNF) also relies upon mTOR activation for memory consolidation [114]. Erythropoietin (EPO) uses mTOR signalling to prevent apoptotic cell death in the nervous system [115, 116]. Through mTOR, EPO prevents cerebral microglial injury [117], Aβ toxicity [97], sepsis-associated encephalopathy [118], cerebral microglial injury [117], oxidative stress injury [37, 119, 120] and retinal progenitor cell loss [121].

EPO can oversee mTOR signalling through several downstream pathways that include PRAS40 function to increase neuronal cell survival [37]. EPO works with other proliferative pathways with mTOR, such as Wnt signalling, to prevent caspase activation and apoptosis [97]. Wnt signalling involves cysteine-rich glycosylated proteins that oversee cell development and survival [122, 123]. EPO requires Wnt signalling to block 'pro-apoptotic' forkhead transcription factor activity that can result in cell death [124, 125]. A downstream target in the Wnt pathway also linked to mTOR is the Wnt1 inducible signalling pathway protein one (WISP1) [3, 19], a member of the six secreted extracellular matrix associated CCN family of proteins [126, 127]. WISP1 activates mTOR, inhibits PRAS40 [39] and limits TSC2 activity [72] to increase microglial cell survival during oxidative stress and Aβ toxicity. WISP1 fosters nuclear trafficking and activity of SIRT1 that results in neuronal protection [128] and blocks apoptotic injury [78, 129, 130]. WISP1 also regulates the post-translational phosphorylation of AMPK [64, 131–133]. WISP1 modulates AMPK activation by differentially decreasing phosphorylation of TSC2 at serine[1387], a target of AMPK, and increasing phosphorylation of TSC2 at threone[1462], a target of Akt [72]. As a result, WISP1 provides a minimal but not excessive level of TSC2 and AMPK activity to promote cellular survival during toxic insults [72]. Interestingly, the ability of EPO to control neuroinflammation also is linked to AMPK activity [134]. EPO appears to require a specific level of AMPK activity to alleviate detrimental effects of oxidative stress [135].

Yet, growth factor protection in the nervous system may sometimes necessitate inhibition of mTOR activity with the promotion of autophagy. Cortical neurons can be protected by BDNF through the induction of autophagy and the inhibition of mTOR during oxygen deprivation [136]. In other scenarios, glial cell-derived neurotrophic factor (GDNF) and nerve growth factor (NGF) protect against photo-oxidative stress with inhibition of mTOR activity [137]. The concentrations of growth factors also may affect mTOR signalling and apoptotic cell death. For example, elevated concentrations of EPO can lead to cellular damage and lessen the activity of mTOR [138].

Given that under some scenarios, trophic factors require limited mTOR activity to promote cell survival suggests that the programmed cell death pathway of autophagy also has a critical role for cell survival in the nervous system [2]. Autophagy recycles components of the cell cytoplasm to eliminate non-functional organelles for disposal and tissue remodelling [17, 139–141]. Macroautophagy is the autophagy classification most often responsible for the recycling

of organelles [12, 19, 142]. This process consists of the sequestration of cytoplasmic proteins and organelles into autophagosomes that combine with lysosomes for degradation and recycling [7, 141, 143–145].

There exist at least 33 autophagic related genes (*Atg*) that have been identified in yeast with TOR and can affect metabolic disorders, cardiovascular disease, cancer and neurodegenerative disease [139–141, 146–151]. Atg1, Atg13 (also known as Apg13), and Atg17 are tied to the PI 3-K, Akt and TOR pathways. Either the Atg1 complex in yeast or the UNC-51 like kinase one (ULK1) complex in mammals requires activation for the induction of autophagy [140, 152]. Atg13 as well as Atg17 can determine Atg1 activity [153]. Phosphorylation of Atg13 through TOR dependent pathways releases Atg13 from Atg1 and reduces Atg1 activity [154]. In contrast, dephosphorylation of Atg13 that can occur during starvation and reduced mTOR activity results in the activation of Atg1 and the induction of autophagy. In mammals, several ULK isoforms exist [147]. The homologues of Atg1 are UNC-51 like kinase one (ULK1) and ULK2 [19]. Mammalian Atg13 binds to ULK1, ULK2 and FIP200 (focal adhesion kinase family interacting protein of 200 kDa) to activate ULKs, promote the phosphorylation of FIP200 by ULKs and results in the induction of autophagy [155]. mTOR activity blocks the induction of autophagy by phosphorylating Atg13 and ULKs to inhibit the ULK-Atg13-FIP200 complex [155].

In the nervous system, induction of autophagy with the inhibition of mTOR activity can be neuroprotective [156]. mTOR blockade with the induction of autophagy increases cell survival in neonatal models of ischaemia [157] and during excitotoxicity [158]. Inhibition of mTOR signalling with autophagy activation results in neural tissue protection and functional improvement in models of spinal cord injury [159]. Autophagy is protective during prion protein disease [160] and in models of Huntington's disease (HD) [25, 161]. Consumption of a high calorie diet has been observed to inhibit autophagy and facilitate hippocampal neuronal loss in mice [162]. In experimental models of AD, disease progression and duration appears to be associated with dysfunctional autophagic processes as well as inhibition of mTOR activity [148]. In contrast, reduction in Aβ production and improved memory function in animal models of AD has been associated with autophagy activation [163]. Vascular cells that may protect during periods of cerebral ischaemia also are lost if autophagy is blocked [164].

Autophagy is impacted not only by mTOR but also by SIRT1. SIRT1 increases lifespan in higher organisms and offers protection against oxidative stress in neuronal cells [165]. SIRT1 promotes cell survival in the nervous system through the blockade of mammalian forkhead transcription factor activity, such as FoxOs [59, 166–169]. SIRT1 also can work in conjunction with FoxOs to protect neurons. Mammalian forkhead transcription factors can bind to the SIRT1 promoter region that contains a cluster of five putative core binding repeat motifs (IRS-1) and a forkhead-like consensus-binding site (FKHD-L) [125]. FoxO proteins, such as FoxO1, can subsequently regulate SIRT1 transcription and increase SIRT1 expression [170]. Expression of FoxO1 can lead to induction of autophagy and the clearance of toxic mutant Huntingtin (mHtt) protein neurons in experimental models of full length mutant mHtt transgenic mice [171]. In regards to the relationship of mTOR with SIRT1, SIRT1 is protective [172] in embryonic stem cells and has an inverse relationship with mTOR [6]. SIRT1 can inhibit mTOR pathways and promote autophagy to protect human embryonic stem cells against oxidative stress [173]. SIRT1 inhibits mTOR signalling to promote neuronal growth [174] and can oversee cellular metabolism during caloric restriction [175]. In endothelial cells exposed to oxidized low density lipoproteins that can lead to atherosclerosis, SIRT1 up-regulation in combination with AMPK activity results in autophagy that is necessary for cell protection [65].

In some cases, limitations in the induction of autophagy may be required to promote cell survival in the nervous system. A reduction in autophagy combined with the activation of mTOR in animal models of traumatic spinal cord injury leads to improvement in function and increased survival of motor neurons [95]. During ischaemic stroke in rodents, inhibition of autophagy reduces infarct size and protects cerebral neurons [176]. Blockade of autophagy and activation of mTOR protects dopaminergic neurons during oxidative stress exposure [91]. In tri-cultures of neurons, astrocytes and microglia that are exposed to inflammatory stressors and Aβ, cell injury increases during the induction of autophagy [177]. Other studies suggest that autophagy is not always beneficial for cell survival. Autophagy can impair endothelial progenitor cells, lead to mitochondrial oxidative stress and prevent new blood vessel formation during elevated glucose exposure [144]. Trophic factors, such as EPO, promote protection against hypoxia and oxidative stress in retinal progenitor cells by limiting the induction of autophagy [121]. EPO can limit excessive autophagy that precedes apoptosis during experimental neonatal necrotizing enterocolitis [178] and can prevent neonatal brain damage in the developing rodent during hyperoxia exposure and oxygen toxicity by blocking autophagy [179]. In Purkinje neurons, insulin growth factor-1 prevents neuronal injury by preventing the induction of autophagy [180].

Conditions that lead to detrimental effects with the induction of autophagy may be the result of a fine balance required between apoptotic and autophagic pathways. Under some experimental conditions in neuronal cell line models, EPO suppresses apoptotic cell injury through the increased activity of AMPK and limited autophagy activity [181]. In addition, WISP1 has been shown to prevent neuronal cell death through the

modulation of both apoptotic and autophagic pathways [182].

mTOR oversight of stem cells in the nervous system

One mechanism that mTOR may influence disease progression in the nervous system is through stem cell development and maintenance (Figure 2) [8]. Trophic factors that rely upon mTOR can direct stem cell development. EPO regulates Wnt signalling to maintain the survival of mesenchymal stem cells [183]. EPO uses mTOR to control bone homeostasis with osteoblastogenesis and osteoclastogenesis [184]. For the growth factors insulin [185] or EPO [119], mTOR activity is necessary for neuronal precursor cell differentiation to ensue.

Without mTORC1, neural stem cells succumb to reduced lineage expansion, lack of differentiation and reduced neuronal production [186]. Furthermore, the degree of activity of mTOR can lead to the modification of stem cell differentiation. Blockade of mTOR activity can result in stem cell differentiation into astrocytic cells [111]. Inhibition of both Akt and mTORC1 can result in reduced neuronal stem cell self-renewal and earlier neuronal and astroglial differentiation [187].

mTOR activity is necessary for the onset and progression of neurogenesis. mTOR controls the expression of RTP801/REDD1 during neuronal maturation. During development, levels of RTP801/REDD1 in newborn and mature neurons become diminished and mTOR activity is increased to promote the maturation of neurons [188]. Neuronal phenotype also is controlled by mTOR. mTOR activity is required for the expression of the neuronal phenotype of post mortem neuronal precursors [119]. In addition, mTOR activity appears necessary for the maintenance of neuronal stem cells during ageing. mTOR signalling in the aged brain is reduced and is accompanied by a reduced proliferation of active neural stem cells [189]. mTOR in conjunction with Akt has been shown to prevent mesenchymal stem cell ageing [190]. Non-neuronal neighbouring cells also can influence the growth of neuronal stem cells. Endothelial cells can foster mTOR activity and lead to the expansion of long term glioblastoma stem-like cells [191].

mTOR and neurodegenerative disorders

Alzheimer's disease

mTOR has a critical role during cognitive function and memory [12] and affects genetic pathways that lead to cognitive loss [113]. In models of vascular dementia, recovery of memory and learning are dependent upon the up-regulation of mTOR and eIF4E [96]. During periods of cerebral ischaemia in rodents, increased expression of mTOR is associated with hippocampal neuron preservation and memory improvement [92]. In models of experimental autoimmune encephalomyelitis (EAE) for multiple sclerosis, cognitive deficits are associated with impaired myelin growth in the parahippocampal cortex that can be reversed with mTOR activation [192]. Memory formation with mTOR also may require metabotropic glutamate receptor (mGluR) signalling [193]. Environmental enrichment for improvements in learning and memory relies upon long term potentiation with mGluR5 activation and sustained phosphorylation of the mTOR pathway p70S6K [194]. Loss of mTOR signalling can impair long term potentiation and synaptic plasticity in animal models of AD that can be restored by mTOR activation [195]. Loss of Akt, mTOR and p70S6K activity also can injure newborn neurons and limit dendritic density formation during Aβ exposure [196]. mTOR activation can control dendritic protein synthesis in hippocampal neurons [197] and the formation of long term memory in the amygdala [198].

However, under conditions that require autophagy induction for enhanced neuronal survival, limitations in the activation of mTOR have been shown to be more fruitful. In murine models of AD, autophagic clearance of cortical Aβ that also involves reduced production of this protein can afford protection against memory impairment though the inhibition of mTOR signalling [163]. Similar studies in cells also show that Aβ clearance requires induction of autophagy with mTOR inhibition, processes that may be associated with memory improvement in disorders such as AD [199]. In animal models of caloric restriction, learning and memory were significantly improved under conditions that promoted the induction of autophagy with a decrease in mTOR signalling [162].

The role of mTOR and its signalling pathways in memory formation highlights the potential for targeting mTOR for the treatment of cognitive disorders, such as AD (Figure 2). Early studies have observed that a decrease in mTOR activity is present in peripheral lymphocytes of patients with AD and this loss of mTOR appears to correlate with the progression of the disease [200]. Loss of mTOR activity also may result in neuronal atrophy in AD. Absence of the retinoblastoma-1 (RB1) inducible coiled-coil one (RB1CC1) tumour suppressor is present in the brains of patients with AD and may be linked to neuronal loss. RB1CC1 maintains mTOR signalling and appears to be necessary for neurite growth, since reduced expression of RB1CC1 leads to loss of mTOR activity, neuronal apoptosis and neuronal atrophy [201]. Reduction in Aβ accumulation in AD also may require mTOR activity. Increased Rheb GTPase activity that is known to increase mTORC1 activity can reduce β-site amyloid precursor protein (APP)-cleaving enzyme one (β-secretase, BACE1) activity to limit Aβ accumulation

in AD [202]. Aβ exposure independently can reduce mTOR activity [203] and may lead to apoptotic neuronal death through the loss of activity in the Akt and mTOR axis [39, 72, 97–99, 199]. In longitudinal studies, mTOR signalling is reduced in aged models of AD with the progression of Aβ accumulation [148], suggesting that the loss of mTOR activity may ultimately lead to Aβ toxicity and cognitive loss. In a number of scenarios, protection against Aβ toxicity requires the activation of the mTOR pathway [97–99, 196, 203].

Given the role mTOR holds in memory formation and the fine balance that may be necessary for mTOR activity to promote cognitive function, it is not surprising to observe that excessive activation of mTOR also may function as a pathological mechanism for cognitive loss and the progression of AD [204]. Down-stream components of mTOR that include active p70S6K can co-localize with hyper-phosphorylated tau deposition in the brains of patients with AD, indicating that p70S6K may assist with the accumulation of hyper-phosphorylated tau [205]. SIRT1 can prevent neuronal degeneration during Aβ exposure and involve the down-regulation of mTOR and p70S6K [174] while suppression of mTOR expression can reduce Aβ deposition [206]. In related studies that pertain to cognitive loss and metabolic dysfunction, high levels of mTOR activity can result in elevated protein expression of acetylcholinesterase (AChE) in central neurons [207] that may impair memory formation. Treatment with temsirolimus, an inhibitor of mTOR and approved by the US Federal Drug Administration (FDA) for the treatment of renal cell carcinoma, can improve spatial learning in animal models of AD and promote the clearance of Aβ [208]. Temsirolimus treatment in mutant tau mouse models also can stimulate macroautophagy, inhibit Akt, mTOR, and p70S6K signalling, decrease abnormal tau accumulation and improve motor deficits [209]. Interestingly, some studies illustrate that reductions in Aβ deposition can be achieved with autophagy that is not dependent upon mTOR, suggesting that memory dysfunction in disorders such as AD may not be tied to high levels of mTOR. Blockade of acetyl-coA cholesterol acyltransferase one (ACAT1) has recently been shown to decrease amyloidopathy in experimental models of AD through induction of autophagy in microglia with the sequestration of Aβ that is independent of mTOR signalling [210].

Parkinson's disease and Huntington's disease

PD is a movement disorder that leads to resting tremor, rigidity and bradykinesia. It is characterized by the loss of dopaminergic neurons in the substantia nigra. Recent work suggests that during disease progression in PD, mTOR signalling is altered [211]. For the treatment of PD, mTOR activation can offer protection against this neurodegenerative disorder (Figure 2). In cell culture with dopaminergic cells, activation of mTOR and p70S6K or down-regulation of 4EBP1 can offer protection against oxidative stress [212]. Persistent activation of 4EBP1 by leucine-rich repeat kinase two (LRRK2), a site for dominant mutations in PD, has been reported to alter protein translation and result in the loss of dopaminergic neurons [213]. Yet, over-expression of 4EBP1 in other experimental studies that include the degeneration of dopaminergic neurons in *Drosophila* indicates that 4EBP1 may suppress pathologic experimental phenotypes of PD [214]. In addition to mTOR, activation of the Akt pathway with mTOR can block methamphetamine neurotoxicity in dopaminergic neurons [215]. Experimental PD toxic mimetics have been shown to suppress mTOR and p70S6K activity [216]. Over-expression of wild-type mTOR or constitutively active p70S6K can prevent neuronal cell death in the presence of PD toxins [216].

Modulation of autophagy through mTOR also represents another mechanism to address the treatment of PD and progression of neurodegenerative disorders [2]. In some studies, inhibition of autophagy with mTOR activation has been shown to prevent dopaminergic neuronal injury during oxidative stress exposure [91], suggesting that autophagy may be detrimental to dopamine neurons. Yet, a number of studies that address α-synuclein toxicity in PD support the premise that induction of autophagy degrades and eliminates α-synuclein to protect dopaminergic neurons [20, 217]. In models of α-synucleinopathy, defects in autophagic pathways can result in neurodegeneration [218]. Mutation of α-synuclein and accumulation of wild-type α-synuclein in dopaminergic neurons have been associated with the onset and progression of PD [219]. Pathways such as those that involve mTOR inhibition or modulation of FoxO proteins [125] can prevent nigral neuron cell death in the presence of human α-synuclein accumulation by reducing the amount of α-synuclein and promoting the accumulation of autophagic vacuoles containing lipofuscin [220]. The growth factor EPO has been shown to protect against dopaminergic neurotoxicity through induction of autophagy and mTOR inhibition [181]. Autophagy also may protect neurons in PD through the maintenance of mitochondrial homeostasis [160, 221] and may not require mTOR inhibition [222].

Similar to PD, autophagy and the modulation of mTOR activity may offer promising therapeutic strategies to treat other disorders that involve toxic intracellular aggregates such as the mutant aggregates that occur in HD (Figure 2). HD is the result of mutations resulting in the polyglutamine tract expansion of CAG in the *huntingtin* (*Htt*) *gene*. mTOR is one of several proteins that can interact with mHtt and affect the course of HD [223]. Blockade of mTOR activity can result in the induction of autophagy and the removal of proteins with long polyglutamine or polyalanine expansions [224]. Other pathways, such as FoxO proteins, also can increase clearance of mHtt through autophagic pathways [171]. mHtt can increase the activity of mTORC1. Enhanced mTORC1 activity has

been shown to accelerate the onset of the loss of motor coordination and premature death in murine models of HD [225]. Yet, inhibition of mTORC1 alone may not be sufficient to alter autophagy or mHtt accumulation. Some studies suggest that combined inhibition of mTORC1 and mTORC2 is required for autophagy and the reduction of mHtt accumulation [226]. In addition, work in mouse models of HD indicate that prevention of motor performance decline may be more associated with decreasing the activity of p70S6K that improves muscle function rather than changes in cerebral mHtt accumulation and neuronal protection [227]. Induction of macroautophagy that is independent of mTOR signalling to remove cellular aggregates also may play a role in HD [222]. Complete elimination of mTOR activity during HD may not be beneficial and a careful modulation of this pathway may be required for optimal clinical efficacy. In HD patients and in rodent models of HD, the expression of Rhes, an mTOR activator in the striatum, is reduced [228]. Activation of mTORC1 protects against striatal atrophy, impaired dopamine signalling, mitochondrial dysfunction and results in the induction of autophagy [228].

Epilepsy

In regards to disorders such as epilepsy and tuberous sclerosis complex (TSC), a disorder that leads to epilepsy in more than 80% of patients [229], activation of mTOR signalling is associated with epileptic pathology (Figure 2) [25, 230]. Limiting cerebral mTOR activity has been advocated for the treatment of these disorders [231, 232]. In models of pilocarpine-induced status epilepticus, animals develop both seizures and aggressive behaviour. Aggressive behaviour precedes seizure development and both are controlled with the inhibition of mTOR activity [233]. Inhibition of mTOR also prevents the development of absence seizures in animal models [234]. In hypoxia-induced neonatal seizures in rodents, mTORC1 activity has been observed to spike within 3 weeks following birth and to be associated with the induction of seizures [235]. Models of status epilepticus suggest that the components of autophagy are also altered during seizure activity with increase activation of mTOR [236].

Experimental mouse models of TSC that delete the *TSC2* gene from mouse Purkinje cells develop increases in Purkinje cell size with eventual apoptotic cell death of these cells, indicating that mTORC1 increased activity in the absence of TSC2 oversight leads to cerebellar disease pathology [237] and eventual epilepsy [33]. In comparison, TSC patients were found to have significant Purkinje cell loss as well [237]. Furthermore, in astrocytes of TSC patients in the mesial temporal lobe, a region that can lead to mesial temporal lobe epilepsy, mTOR signalling with p70S6K was found to have increased activity [238]. Glutamine may be one method to control p70S6K activity and reduce the risk of seizures as indicated in studies with TSC2 knockout mice [239]. For the clinical treatment of epilepsy, lamotrigine, an approved Food and Drug Administration (FDA) anti-epileptic agent, leads to the induction of autophagy that may be associated with limitation in mTOR activity to reduce seizure frequency [151]. The FDA has approved the use of everolimus (RAD-001), an analogue of rapamycin and an inhibitor of mTOR, for the treatment of subependymal giant cell astrocytoma. In TSC, everolimus limits giant cell astrocytoma cell growth [240, 241]. Seizure frequency was reduced in approximately 60 % of the patients studied, but some patients experienced increased seizure frequency with the drug [241]. In addition, toxic side effects were present that included respiratory infections, stomatitis and leukopenia [241]. Additional work has reported that everolimus in patients with TSC and refractory epilepsy reduced seizure frequency by a median reduction of 73 % in 17 of 20 patients examined [242].

Stroke and trauma

mTOR and its signalling pathways markedly influence acute neurodegenerative disorders, such as stroke and trauma (Figure 2) [3, 69]. Following traumatic rodent spinal cord injury, reduced motor neuron death and enhanced forelimb function occur with treatments that increase Akt and mTOR activity with a reduction in autophagy [95]. mTOR activation fosters axonal growth following injury in the peripheral nervous system, but proper target innervation may require fine control of mTOR activity [243]. During ischaemic pre-conditioning in animal models of stroke, mTOR activation can lead to neuronal cell protection. During remote ischaemic preconditioning in the hippocampus prior to global brain ischaemia, mTOR activation prevents neuronal injury and leads to improvements in memory function [92]. In models of ischaemic post-conditioning, activation of the Akt/mTOR pathway reduce long term cerebral focal ischaemic damage and neurological disability [244]. During ischaemic-reperfusion injury, Golgi phosphoprotein-3 and mTOR were found to be necessary for the reduction in stroke volume [245]. Middle cerebral artery infarction in rodent models is also reduced with activation of mTOR signalling that includes mTOR and p70S6K [102]. During oxygen-glucose deprivation in neuronal cells, induction of EPO expression prevents cell death through activation of Akt and mTOR pathways [120, 246]. Non-coding micro RNAs (miRNAs) may offer treatment options for stroke that require mTOR activation [141, 247]. A cohort of circulating miRNAs related to PI 3-K, Akt and mTOR were found to be associated with a greater degree of neuronal cell protection in adult rodent females [248]. Exercise following injury in the spinal cord may lead to improved neuronal plasticity though miRNA expression and activation of p70S6K [249].

Non-neuronal cells also may play a critical role in treating acute neurodegenerative disorders through

mTOR. Activation of mTOR is necessary to maintain the integrity of cerebral microglia that can be reparative [117, 250, 251]. mTOR signalling with a reduction in PRAS40 activity in neuronal cell lines and microglia during oxidative stress can prevent apoptotic cell death [37, 39]. Interestingly, mice with a PRAS40 gene knockout that were subjected to cerebral ischaemia were observed to suffer increased infarctions, but lacked significant p70S6K activity, suggesting that a specific level of mTOR may be required for cortical protection [40]. Over-expression of PRAS40 in this model resulted in a potential feed-back pathway that resulted in a reduction in cerebral ischaemic injury with phosphorylation and increased activity of Akt and mTOR [40]. mTORC1 and mTORC2 also have been demonstrated to be necessary in astrocytes for glutamate transporter subtype two (GLT-1) expression that would promote glutamate uptake during brain ischaemia and limit ischaemic injury [252]. During cerebral ischaemia, leucine-rich repeat in Flightless-1 interaction protein one (Lrrfip1) has recently been shown to be up-regulated as a neuroprotective mechanism to increase mTOR activity in astrocytes and enhance GLT-1 activity [101].

Similar to chronic neurodegenerative disorders, mTOR activation with a reduction in autophagy is not consistently protective in the nervous system [69, 127]. The induction of autophagy and mTOR inhibition is necessary with histamine H_3-receptor antagonism during stroke to result in neuronal cell protection [253]. Excitotoxicity in hippocampal neurons is also prevented with activation of autophagy and mTOR inhibition [158]. During oxygen-glucose deprivation in human umbilical vein endothelial cells, mTOR inhibition and induction of autophagy protect vascular cells from injury [164]. Application of rapamycin blocks mTORC1 and mTORC2 activation and leads to increased cortical neuronal survival [254]. During ischaemic brain tolerance studies, rapamycin can promote autophagy induction, reduce brain damage and result in improved neurological scores that are associated with TSC1 activity [255]. In murine models of spinal cord injury, inhibition of mTOR and activation of autophagy lead to reduced neuronal tissue damage and improved locomotor function [159].

Future perspectives

Neurodegenerative disorders impact a significant proportion of the global population and are increasing in incidence as a result of the extended lifespan enjoyed by individuals throughout the world. Currently, acute and chronic neurodegenerative diseases impact more than 30 million individuals globally, but treatments to prevent death or disability for these disorders are limited. mTOR, a 289-kDa serine/threonine protein kinase, offers stimulating possibilities to provide novel treatment strategies for multiple neurodegenerative disorders that include AD, PD, HD, epilepsy, stroke and trauma. In particular, strategies that can modulate mTOR activity with lamotrigine for the treatment of epilepsy and everolimus for the treatment of subependymal giant cell astrocytoma are already in play and approved by the FDA.

Yet, a number of considerations must be addressed for the development of efficacious and safe strategies that employ mTOR signalling. First, mTOR has an important role in stem cell development and proliferation that may be critical for reparative processes in the nervous system, but close modulation of mTOR pathways is required to optimize clinical utility. Neuronal stem cell production and proliferation are closely dependent upon mTOR activity [8, 186, 188–190]. Loss of mTOR activity can lead to impaired stem cell self-renewal. In addition, ageing appears to deplete mTOR stores that results in the loss of neural stem cells. Maintaining the activation of mTOR under these conditions could assist with the repair of injured tissue as well as assist with treatments for cognitive loss. Yet, some levels of mTOR activity can promote the differentiation of stem cells into non-neuronal cells that may be an undesired clinical outcome and could further tissue injury.

As a second consideration, mTOR signalling has the potential to promote tumourigenesis, an outcome that must be avoided when considering repair and regeneration in the nervous system. The role of mTOR during tumour growth is complex and involves not only the level of mTOR activation, but also the association of mTOR with other proliferative pathways that either can foster tumour growth or function to block metastatic disease. Considering that mTOR is a proliferative agent, mTOR activity is usually restricted for the treatment of cancer. mTOR activity can result in neurofibromatosis type 1, TSC, Lhermitte-Duclos disease and glioblastoma multiforme [19]. The FDA has approved 'rapalogues' that inhibit mTOR to treat tumours associated with TSC, renal cancer and neuroendocrine pancreatic tumours [25, 68]. Recent work also suggests that mTOR activity in neurons may foster high grade glioma proliferation in neural and oligodendroglial precursor cells [256, 257]. mTOR signalling can promote cancer stem cell growth [258] and has been correlated with chemotherapy resistance of breast cancer cells with stem cell characteristics [259]. In regards to the association of mTOR with other proliferative mechanisms, growth factors that rely upon mTOR signalling, such as EPO, may lead to cancer [260–262] and blood–brain barrier injury [263] during prolonged treatment regimens. Down-stream signalling pathways of EPO, such as Wnt and WISP1, also can lead to unchecked cellular growth. Wnt signalling may result in glioma proliferation [264, 265], metastatic disease [266–269] and malignant melanoma [270]. Variants of WISP1 can promote distant metastatic disease [271]. However, in potential association with mTOR, non-variant WISP1 expression can block tumour cell invasion,

motility and metastatic disease [272]. Under these conditions, mTOR may limit the excessive cell growth that is promoted by Wnt signalling. mTOR activation has been shown to maintain cell senescence and prevent tumour cell growth that is promoted by Wnt [273].

The role of mTOR in programmed cell death pathways also is a challenge for the development of new treatment options. Under many conditions, mTOR activation is protective against apoptotic pathways for both early membrane PS externalization and later DNA degradation. Through these pathways, mTOR can regulate immune cell activity and prevent the removal of functional cells that may be marked for disposal by nervous system microglia [72, 97, 210, 230, 250, 274]. Acute and chronic injury in the nervous system can be ameliorated through activation of mTOR signalling pathways. Yet, pathways tied to mTOR, such as EPO, WISP1 and SIRT1, can require a controlled level of mTOR activity to protect neurons through inhibition of apoptosis in conjunction with autophagic pathways [39, 97, 117, 174, 175, 275]. For disorders in the nervous system, toxic cellular and extracellular accumulations that can involve AD, PD and HD may require the induction of autophagy to halt progression of these disorders [7, 136, 162, 209]. These circumstances may argue for the inhibition of mTOR activity to promote autophagic pathways. However, it is sometimes unclear which particular components of the mTOR pathway are required for clinical success. Some work suggests that combined inhibition of mTORC1 and mTORC2 is required for autophagy for the treatment of disorders, such as HD [226]. mTORC1 and mTORC2 also may be necessary in astrocytes for GLT-1 expression that would promote glutamate uptake during brain ischaemia and limit ischaemic injury [252]. In addition, autophagy is not consistently protective [144, 178–180]. In some cases, induction of autophagy may proceed independently of mTOR [210], suggesting that the development of treatment strategies for progressive neurodegeneration may require careful control of mTOR activity and the induction of autophagy. Alternatively, severely limiting mTOR activity in disorders such as AD, PD and HD to promote autophagy may yield unexpected detrimental effects that involve new or progressive cognitive dysfunction. Loss of mTOR signalling can impair long term potentiation, synaptic plasticity, dendritic density and memory formation [92, 96, 113].

Consideration also must be given for the role mTOR holds within the PI 3-K, Akt and mTOR axis. In many scenarios of nervous system dysfunction, mTOR does not function independently and new clinical strategies may need to seek modulation of family pathways linked to mTOR. For example, mTOR in conjunction with PI 3-K and Akt are necessary for growth factors, such as EGF and EPO, to maintain memory function and prevent cellular injury [37, 97, 112, 113, 117]. mTOR in combination with Akt is necessary to prevent mesenchymal stem cell ageing [190]. Loss of Akt and mTOR signalling can injure newborn neurons and limit dendritic density formation [196]. During either trauma or ischaemia to the brain, both Akt and mTOR may be required to preserve neuronal survival and restore neurological function [120, 244, 245]. Autophagy also is tied to combined signalling with PI 3-K, Akt, and mTOR [140]. In a similar fashion, nervous system tumours may require therapies that adequately address the PI 3-K, Akt and mTOR axis. Tumours of the nervous system can develop resistance to agents that inhibit mTOR signalling [276], since these cancers may have an abnormal increased basal activity of PI 3-K, Akt and mTOR [277]. Inhibition of only the mTOR pathway without controlling the activity of PI 3-K or Akt can lead to a poor clinical response and ineffective blockade of cell tumour cell proliferation [277]. Other studies indicate that blockade of mTORC1 can result in the feedback activation of PI 3-K, Akt and Ras-mitogen activated protein kinase (MAPK) signalling that can lead to further neoplastic growth [278, 279]. Treatments with current strategies that block mTOR, such as rapamycin or its derivative compounds, also have additional limitations with toxicity that include oral and respiratory infections, stomatitis, hypertriglyceridaemia, hypercholesterolaemia, leukopenia and immunosuppression [241, 280, 281]. mTOR offers great promise and exciting avenues for drug development for disorders of the nervous system, but future focus will need to target several critical considerations for the generation of rewarding clinical outcomes.

Competing Interests

The author has completed the Unified Competing Interest form at www.icmje.org/coi_disclosure.pdf (available on request from the corresponding author) and declares support from American Diabetes Association, American Heart Association, NIH NIEHS, NIH NIA, NIH NINDS and NIH ARRA for the submitted work, no financial relationships with any organizations that might have an interest in the submitted work in the previous 3 years and no other relationships or activities that could appear to have influenced the submitted work.

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REFERENCES

1 Chong ZZ, Li F, Maiese K. Oxidative stress in the brain: Novel cellular targets that govern survival during neurodegenerative disease. Prog Neurobiol 2005; 75: 207–46.

2 Maiese K. Driving neural regeneration through the mammalian target of rapamycin. Neural Rege Res 2014; 9: 1413–7.

3 Maiese K. Cutting through the complexities of mTOR for the treatment of stroke. Curr Neurovasc Res 2014; 11: 177–86.

4 Minino AM. Death in the United States, 2011. NCHS Data Brief 2013; March: 1–8.

5 Hayutin A. Global demographic shifts create challenges and opportunities. PREA Quarterly 2007; Fall: 46–53.

6 Maiese K. SIRT1 and stem cells: In the forefront with cardiovascular disease, neurodegeneration and cancer. World J Stem Cells 2015; 7: 235–42.

7 Maiese K. Programming apoptosis and autophagy with novel approaches for diabetes mellitus. Curr Neurovasc Res 2015; 12: 173–88.

8 Maiese K. Stem cell guidance through the mechanistic target of rapamycin. World J Stem Cells 2015; Aug 26;7: 999–1009.

9 World Health Organization. Description of the global burden of NCDs, their risk factors and determinants. Global status report on noncommunicable diseases 2010. 2011; 1–176.

10 Minino AM, Murphy SL. Death in the United States, 2010. NCHS Data Brief 2012; July: 1–8.

11 Sivaraman V, Yellon DM. Pharmacologic therapy that simulates conditioning for cardiac ischemic/reperfusion injury. J Cardiovasc Pharmacol Ther 2014; 19: 83–96.

12 Maiese K. Taking aim at Alzheimer's disease through the mammalian target of rapamycin. Ann Med 2014; 46: 587–96.

13 Schluesener JK, Zhu X, Schluesener HJ, Wang GW, Ao P. Key network approach reveals new insight into Alzheimer's disease. IET Syst Biol 2014; 8: 169–75.

14 Maiese K, Chong ZZ, Hou J, Shang YC. New strategies for Alzheimer's disease and cognitive impairment. Oxid Med Cell Longev 2009; 2: 279–89.

15 Agis-Torres A, Solhuber M, Fernandez M, Sanchez-Montero JM. Multi-target-directed ligands and other therapeutic strategies in the search of a real solution for Alzheimer's disease. Curr Neuropharmacol 2014; 12: 2–36.

16 Filley CM, Rollins YD, Anderson CA, Arciniegas DB, Howard KL, Murrell JR, Boyer PJ, Kleinschmidt-DeMasters BK, Ghetti B. The genetics of very early onset Alzheimer disease. Cognit Behav Neurol 2007; 20: 149–56.

17 Nakka VP, Prakash-Babu P, Vemuganti R. Crosstalk between endoplasmic reticulum stress, oxidative stress, and autophagy: potential therapeutic targets for acute CNS injuries. Mol Neurobiol 2014; 9.

18 World Health Organization. Neurological Disorders: Public Health Challenges. WHO Library Cataloguing-in Publication Data. 2006; 1–232.

19 Chong ZZ, Shang YC, Wang S, Maiese K. Shedding new light on neurodegenerative diseases through the mammalian target of rapamycin. Prog Neurobiol 2012; 99: 128–48.

20 Mishra AK, Ur Rasheed MS, Shukla S, Tripathi MK, Dixit A, Singh MP. Aberrant autophagy and Parkinsonism: does correction rescue from disease progression? Mol Neurobiol 2015 Jun; 51: 893–908.

21 Pergola PE, White CL, Szychowski JM, Talbert R, Brutto OD, Castellanos M, Graves JW, Matamala G, Pretell EJ, Yee J, Rebello R, Zhang Y, Benavente OR. Achieved blood pressures in the secondary prevention of small subcortical strokes (SPS3) study: challenges and lessons learned. Am J Hypertens 2014; 27: 1052–60.

22 Chen H, Zhu G, Liu N, Zhang W. Low-dose tissue plasminogen activator is as effective as standard tissue plasminogen activator administration for the treatment of acute ischemic stroke. Curr Neurovasc Res 2014; 11: 62–7.

23 Pineda D, Ampurdanes C, Medina MG, Serratosa J, Tusell JM, Saura J, Planas AM, Navarro P. Tissue plasminogen activator induces microglial inflammation via a noncatalytic molecular mechanism involving activation of mitogen-activated protein kinases and Akt signaling pathways and AnnexinA2 and Galectin-1 receptors. Glia 2012; 60: 526–40.

24 Johnson SC, Sangesland M, Kaeberlein M, Rabinovitch PS. Modulating mTOR in aging and health. Interdiscip Top Gerontol 2015; 40: 107–27.

25 Maiese K, Chong ZZ, Shang YC, Wang S. mTOR: on target for novel therapeutic strategies in the nervous system. Trends Mol Med 2013; 19: 51–60.

26 Heitman J, Movva NR, Hall MN. Targets for cell cycle arrest by the immunosuppressant rapamycin in yeast. Science 1991; 253: 905–9.

27 Singh K, Sun S, Vezina C. Rapamycin (AY-22,989), a new antifungal antibiotic. IV. Mechanism of action. J Antibiot (Tokyo) 1979; 32: 630–45.

28 Gulhati P, Bowen KA, Liu J, Stevens PD, Rychahou PG, Chen M, Lee EY, Weiss HL, O'Connor KL, Gao T, Evers BM. mTORC1 and mTORC2 regulate EMT, motility, and metastasis of colorectal cancer via RhoA and Rac1 signaling pathways. Cancer Res 2011; 71: 3246–56.

29 Zoncu R, Efeyan A, Sabatini DM. mTOR: from growth signal integration to cancer, diabetes and ageing. Nat Rev Mol Cell Biol 2011; 12: 21–35.

30 Chen J, Zheng XF, Brown EJ, Schreiber SL. Identification of an 11-kDa FKBP12-rapamycin-binding domain within the 289-kDa FKBP12-rapamycin-associated protein and characterization of a critical serine residue. Proc Natl Acad Sci U S A 1995; 92: 4947–51.

31 Sarbassov DD, Ali SM, Sengupta S, Sheen JH, Hsu PP, Bagley AF, Markhard AL, Sabatini DM. Prolonged rapamycin treatment inhibits mTORC2 assembly and Akt/PKB. Mol Cell 2006; 22: 159–68.

32 Acosta-Jaquez HA, Keller JA, Foster KG, Ekim B, Soliman GA, Feener EP, Ballif BA, Fingar DC. Site-specific mTOR phosphorylation promotes mTORC1-mediated signaling and cell growth. Mol Cell Biol 2009; 29: 4308–24.

33 Chong ZZ, Shang YC, Zhang L, Wang S, Maiese K. Mammalian target of rapamycin: hitting the bull's-eye for neurological disorders. Oxid Med Cell Longev 2010; 3: 374–91.

34 Bai X, Ma D, Liu A, Shen X, Wang QJ, Liu Y, Jiang Y. Rheb activates mTOR by antagonizing its endogenous inhibitor, FKBP38. Science 2007; 318: 977–80.

35 Wang L, Lawrence JC Jr, Sturgill TW, Harris TE. Mammalian target of rapamycin complex one (mTORC1) activity is associated with phosphorylation of raptor by mTOR. J Biol Chem 2009; 284: 14693–7.

36 Wang H, Zhang Q, Wen Q, Zheng Y, Philip L, Jiang H, Lin J, Zheng W. Proline-rich Akt substrate of 40kDa (PRAS40): a novel downstream target of PI3k/Akt signaling pathway. Cell Signal 2012; 24: 17–24.

37 Chong ZZ, Shang YC, Wang S, Maiese K. PRAS40 Is an Integral Regulatory Component of Erythropoietin mTOR Signaling and Cytoprotection. PLoS One 2012; 7: e45456.

38 Fonseca BD, Smith EM, Lee VH, MacKintosh C, Proud CG. PRAS40 is a target for mammalian target of rapamycin complex one and is required for signaling downstream of this complex. J Biol Chem 2007; 282: 24514–24.

39 Shang YC, Chong ZZ, Wang S, Maiese K. WNT1 Inducible Signaling Pathway Protein one (WISP1) Targets PRAS40 to Govern beta-Amyloid Apoptotic Injury of Microglia. Curr Neurovasc Res 2012; 9: 239–49.

40 Xiong X, Xie R, Zhang H, Gu L, Xie W, Cheng M, Jian Z, Kovacina K, Zhao H. PRAS40 plays a pivotal role in protecting against stroke by linking the Akt and mTOR pathways. Neurobiol Dis 2014 Jun; 66: 43–52.

41 Peterson TR, Laplante M, Thoreen CC, Sancak Y, Kang SA, Kuehl WM, Gray NS, Sabatini DM. DEPTOR is an mTOR inhibitor frequently overexpressed in multiple myeloma cells and required for their survival. Cell 2009; 137: 873–86.

42 Kim DH, Sarbassov DD, Ali SM, Latek RR, Guntur KV, Erdjument-Bromage H, Tempst P, Sabatini DM. GbetaL, a positive regulator of the rapamycin-sensitive pathway required for the nutrient-sensitive interaction between raptor and mTOR. Mol Cell 2003; 11: 895–904.

43 Guertin DA, Stevens DM, Thoreen CC, Burds AA, Kalaany NY, Moffat J, Brown M, Fitzgerald KJ, Sabatini DM. Ablation in mice of the mTORC components raptor, rictor, or mLST8 reveals that mTORC2 is required for signaling to Akt-FOXO and PKCalpha, but not S6K1. Dev Cell 2006; 11: 859–71.

44 Glidden EJ, Gray LG, Vemuru S, Li D, Harris TE, Mayo MW. Multiple site acetylation of Rictor stimulates mammalian target of rapamycin complex two (mTORC2)-dependent phosphorylation of Akt protein. J Biol Chem 2012; 287: 581–8.

45 James MF, Stivison E, Beauchamp R, Han S, Li H, Wallace MR, Gusella JF, Stemmer-Rachamimov AO, Ramesh V. Regulation of mTOR complex two signaling in neurofibromatosis 2-deficient target cell types. Mol Cancer Res 2012; 10: 649–59.

46 Kamarudin MN, Mohd Raflee NA, Syed Hussein SS, Lo JY, Supriady H, Abdul Kadir H. (R)-(+)-alpha-Lipoic acid protected NG108-15 cells against H2O2-induced cell death through PI3K-Akt/GSK-3beta pathway and suppression of NF-kappabeta-cytokines. Drug Des Devel Ther 2014; 8: 1765–80.

47 Tang Z, Baykal AT, Gao H, Quezada HC, Zhang H, Bereczki E, Serhatli M, Baykal B, Acioglu C, Wang S, Ioja E, Ji X, Zhang Y, Guan Z, Winblad B, Pei JJ. mTor is a signaling hub in cell survival: a mass-spectrometry-based proteomics investigation. J Proteome Res 2014; 13: 2433–44.

48 Chen CH, Sarbassov dos D. The mTOR (mammalian target of rapamycin) kinase maintains integrity of mTOR complex 2. J Biol Chem 2011; 286: 40386–94.

49 Sarbassov DD, Guertin DA, Ali SM, Sabatini DM. Phosphorylation and regulation of Akt/PKB by the rictor-mTOR complex. Science 2005; 307: 1098–101.

50 Frias MA, Thoreen CC, Jaffe JD, Schroder W, Sculley T, Carr SA, Sabatini DM. mSin1 is necessary for Akt/PKB phosphorylation, and its isoforms define three distinct mTORC2s. Curr Biol 2006; 16: 1865–70.

51 Jacinto E, Loewith R, Schmidt A, Lin S, Ruegg MA, Hall A, Hall MN. Mammalian TOR complex two controls the actin cytoskeleton and is rapamycin insensitive. Nat Cell Biol 2004; 6: 1122–8.

52 Pearce LR, Sommer EM, Sakamoto K, Wullschleger S, Alessi DR. Protor-1 is required for efficient mTORC2-mediated activation of SGK1 in the kidney. Biochem J 2011; 436: 169–79.

53 Garcia-Martinez JM, Alessi DR. mTOR complex two (mTORC2) controls hydrophobic motif phosphorylation and activation of serum- and glucocorticoid-induced protein kinase one (SGK1). Biochem J 2008; 416: 375–85.

54 Huang J, Dibble CC, Matsuzaki M, Manning BD. The TSC1-TSC2 complex is required for proper activation of mTOR complex 2. Mol Cell Biol 2008; 28: 4104–15.

55 Sato T, Nakashima A, Guo L, Tamanoi F. Specific activation of mTORC1 by Rheb G-protein *in vitro* involves enhanced recruitment of its substrate protein. J Biol Chem 2009; 284: 12783–91.

56 Inoki K, Zhu T, Guan KL. TSC2 mediates cellular energy response to control cell growth and survival. Cell 2003; 115: 577–90.

57 DeYoung MP, Horak P, Sofer A, Sgroi D, Ellisen LW. Hypoxia regulates TSC1/2-mTOR signaling and tumor suppression through REDD1-mediated 14-3-3 shuttling. Genes Dev 2008; 22: 239–51.

58 Maiese K, Chong ZZ, Hou J, Shang YC. The vitamin nicotinamide: translating nutrition into clinical care. Molecules 2009; 14: 3446–85.

59 Chong ZZ, Shang YC, Wang S, Maiese K. SIRT1: New avenues of discovery for disorders of oxidative stress. Expert Opin Ther Targets 2012 Feb; 16: 167–78.

60 Maiese K. New insights for oxidative stress and diabetes mellitus. Oxid Med Cell Longev 2015; 2015 (Article ID 875961).

61 Wang Y, Liang Y, Vanhoutte PM. SIRT1 and AMPK in regulating mammalian senescence: a critical review and a working model. FEBS Lett 2011; 585: 986–94.

62 Maiese K, Chong ZZ, Shang YC, Hou J. FoxO proteins: cunning concepts and considerations for the cardiovascular system. Clin Sci (Lond) 2009; 116: 191–203.

63 Canto C, Auwerx J. Caloric restriction, SIRT1 and longevity. Trends Endocrinol Metab 2009; 20: 325–31.

64 Maiese K, Chong ZZ, Shang YC, Wang S. Novel directions for diabetes mellitus drug discovery. Expet Opin Drug Discov 2013; 8: 35–48.

65 Jin X, Chen M, Yi L, Chang H, Zhang T, Wang L, Ma W, Peng X, Zhou Y, Mi M. Delphinidin-3-glucoside protects human umbilical vein endothelial cells against oxidized low-density lipoprotein-induced injury by autophagy upregulation via the AMPK/SIRT1 signaling pathway. Mol Nutr Food Res 2014; 58: 1941–51.

66 Chong ZZ, Shang YC, Wang S, Maiese K. A Critical Kinase Cascade in Neurological Disorders: PI 3-K, Akt, and mTOR. Future Neurol 2012; 7: 733–48.

67 Janku F, Wheler JJ, Westin SN, Moulder SL, Naing A, Tsimberidou AM, Fu S, Falchook GS, Hong DS, Garrido-Laguna I, Luthra R, Lee JJ, Lu KH, Kurzrock R. PI3K/AKT/mTOR inhibitors in patients with breast and gynecologic malignancies harboring PIK3CA mutations. J Clin Oncol 2012; 30: 777–82.

68 Maiese K. Therapeutic targets for cancer: current concepts with PI 3-K, Akt and mTOR. Indian J Med Res 2013; 137: 243–6.

69 Morgan-Warren PJ, Berry M, Ahmed Z, Scott RA, Logan A. Exploiting mTOR signaling: a novel translatable treatment strategy for traumatic optic neuropathy? Invest Ophthalmol Vis Sci 2013; 54: 6903–16.

70 Cai SL, Tee AR, Short JD, Bergeron JM, Kim J, Shen J, Guo R, Johnson CL, Kiguchi K, Walker CL. Activity of TSC2 is inhibited by AKT-mediated phosphorylation and membrane partitioning. J Cell Biol 2006; 173: 279–89.

71 Hahn-Windgassen A, Nogueira V, Chen CC, Skeen JE, Sonenberg N, Hay N. Akt activates the mammalian target of rapamycin by regulating cellular ATP level and AMPK activity. J Biol Chem 2005; 280: 32081–9.

72 Shang YC, Chong ZZ, Wang S, Maiese K. Tuberous sclerosis protein two (TSC2) modulates CCN4 cytoprotection during apoptotic amyloid toxicity in microglia. Curr Neurovasc Res 2013; 10: 29–38.

73 Shang YC, Chong ZZ, Hou J, Maiese K. Wnt1, FoxO3a, and NF-kappaB oversee microglial integrity and activation during oxidant stress. Cell Signal 2010; 22: 1317–29.

74 Viola G, Bortolozzi R, Hamel E, Moro S, Brun P, Castagliuolo I, Ferlin MG, Basso G. MG-2477, a new tubulin inhibitor, induces autophagy through inhibition of the Akt/mTOR pathway and delayed apoptosis in A549 cells. Biochem Pharmacol 2012; 83: 16–26.

75 Wong DZ, Kadir HA, Lee CL, Goh BH. Neuroprotective properties of Loranthus parasiticus aqueous fraction against oxidative stress-induced damage in NG108-15 cells. J Nat Med 2012; 66: 544–51.

76 Troy CM, Akpan N, Jean YY. Regulation of caspases in the nervous system implications for functions in health and disease. Prog Mol Biol Transl Sci 2011; 99: 265–305.

77 Bailey TJ, Fossum SL, Fimbel SM, Montgomery JE, Hyde DR. The inhibitor of phagocytosis, O-phospho-L-serine, suppresses Muller glia proliferation and cone cell regeneration in the light-damaged zebrafish retina. Exp Eye Res 2010; 91: 601–12.

78 Hou J, Chong ZZ, Shang YC, Maiese K. Early apoptotic vascular signaling is determined by Sirt1 through nuclear shuttling, forkhead trafficking, bad, and mitochondrial caspase activation. Curr Neurovasc Res 2010; 7: 95–112.

79 Shang YC, Chong ZZ, Hou J, Maiese K. FoxO3a governs early microglial proliferation and employs mitochondrial depolarization with caspase three, eight, and nine cleavage during oxidant induced apoptosis. Curr Neurovasc Res 2009; 6: 223–38.

80 Wei L, Sun C, Lei M, Li G, Yi L, Luo F, Li Y, Ding L, Liu Z, Li S, Xu P. Activation of Wnt/beta-catenin Pathway by Exogenous Wnt1 Protects SH-SY5Y Cells Against 6-Hydroxydopamine Toxicity. J Mol Neurosci 2013; 49: 105–15.

81 Weinberg E, Maymon T, Weinreb M. AGEs induce caspase-mediated apoptosis of rat BMSCs via TNFalpha production and oxidative stress. J Mol Endocrinol 2014; 52: 67–76.

82 Yang Y, Li H, Hou S, Hu B, Liu J, Wang J. The noncoding RNA expression profile and the effect of lncRNA AK126698 on cisplatin resistance in non-small-cell lung cancer cell. PLoS One 2013; 8: e65309.

83 Chong ZZ, Li F, Maiese K. Cellular demise and inflammatory microglial activation during beta-amyloid toxicity are governed by Wnt1 and canonical signaling pathways. Cell Signal 2007; 19: 1150–62.

84 Lin SH, Maiese K. The metabotropic glutamate receptor system protects against ischemic free radical programmed cell death in rat brain endothelial cells. J Cereb Blood Flow Metab 2001; 21: 262–75.

85 Popescu NI, Lupu C, Lupu F. Extracellular protein disulfide isomerase regulates coagulation on endothelial cells through modulation of phosphatidylserine exposure. Blood 2010; 116: 993–1001.

86 Kim S, Kang IH, Nam JB, Cho Y, Chung DY, Kim SH, Kim JS, Cho YD, Hong EK, Sohn NW, Shin JW. Ameliorating the effect of astragaloside IV on learning and memory deficit after chronic cerebral hypoperfusion in rats. Molecules 2015; 20: 1904–21.

87 Maiese K. Novel applications of trophic factors, Wnt and WISP for neuronal repair and regeneration in metabolic disease. Neural Regen Res 2015; 10: 518–28.

88 Xin YJ, Yuan B, Yu B, Wang YQ, Wu JJ, Zhou WH, Qiu Z. Tet1-mediated DNA demethylation regulates neuronal cell death induced by oxidative stress. Sci Rep 2015; 5: 7645.

89 Yu T, Li L, Chen T, Liu Z, Liu H, Li Z. Erythropoietin attenuates advanced glycation endproducts-induced toxicity of schwann cells *in vitro*. Neurochem Res 2015; 40: 698–712.

90 Chen L, Xu B, Liu L, Luo Y, Yin J, Zhou H, Chen W, Shen T, Han X, Huang S. Hydrogen peroxide inhibits mTOR signaling by activation of AMPKalpha leading to apoptosis of neuronal cells. Lab Invest 2010; 90: 762–73.

91 Choi KC, Kim SH, Ha JY, Kim ST, Son JH. A novel mTOR activating protein protects dopamine neurons against

oxidative stress by repressing autophagy related cell death. J Neurochem 2010; 112: 366–76.

92 Zare Mehrjerdi F, Aboutaleb N, Habibey R, Ajami M, Soleimani M, Arabian M, Niknazar S, Hossein Davoodi S, Pazoki-Toroudi H. Increased phosphorylation of mTOR is involved in remote ischemic preconditioning of hippocampus in mice. Brain Res 2013; 1526: 94–101.

93 Shi GD, OuYang YP, Shi JG, Liu Y, Yuan W, Jia LS. PTEN deletion prevents ischemic brain injury by activating the mTOR signaling pathway. Biochem Biophys Res Commun 2011; 404: 941–5.

94 Salto R, Vilchez JD, Giron MD, Cabrera E, Campos N, Manzano M, Rueda R, Lopez-Pedrosa JM. Beta-hydroxy-beta-methylbutyrate (HMB) promotes neurite Outgrowth in Neuro2a Cells. PLoS One 2015; 10: e0135614.

95 Walker CL, Walker MJ, Liu NK, Risberg EC, Gao X, Chen J, Xu XM. Systemic bisperoxovanadium activates Akt/mTOR, reduces autophagy, and enhances recovery following cervical spinal cord injury. PLoS One 2012; 7: e30012.

96 Yang H, Shi O, Jin Y, Henrich-Noack P, Qiao H, Cai C, Tao H, Tian X. Functional protection of learning and memory abilities in rats with vascular dementia. Restor Neurol Neurosci 2014; 32: 689–700.

97 Shang YC, Chong ZZ, Wang S, Maiese K. Prevention of beta-amyloid degeneration of microglia by erythropoietin depends on Wnt1, the PI 3-K/mTOR pathway, Bad, and Bcl-xL. Aging (Albany NY) 2012; 4: 187–201.

98 Wang Y, Wang YX, Liu T, Law PY, Loh HH, Qiu Y, Chen HZ. mu-Opioid Receptor Attenuates Abeta Oligomers-Induced Neurotoxicity Through mTOR Signaling. CNS Neurosci Ther 2015; 21: 8–14.

99 Xue Z, Guo Y, Zhang S, Huang L, He Y, Fang R, Fang Y. Beta-asarone attenuates amyloid beta-induced autophagy via Akt/mTOR pathway in PC12 cells. Eur J Pharmacol 2014; 741: 195–204.

100 Huang H, Huang F, Huang JP. Transplantation of bone marrow derived endothelial progenitor cells overexpressing Deltalike4 enhances functional neovascularization in ischemic myocardium. Mol Med Rep 2013; 8: 1556–62.

101 Gubern C, Camos S, Hurtado O, Rodriguez R, Romera VG, Sobrado M, Canadas R, Moro MA, Lizasoain I, Serena J, Mallolas J, Castellanos M. Characterization of Gcf2/Lrrfip1 in experimental cerebral ischemia and its role as a modulator of Akt, mTOR and beta-catenin signaling pathways. Neuroscience 2014; 268C: 48–65.

102 Koh PO. Ferulic acid attenuates focal cerebral ischemia-induced decreases in p70S6 kinase and S6 phosphorylation. Neurosci Lett 2013; 555: 7–11.

103 Zhong X, Lin R, Li Z, Mao J, Chen L. Effects of salidroside on cobalt chloride-induced hypoxia damage and mTOR signaling repression in PC12 cells. Biol Pharm Bull 2014; 37: 1199–206.

104 Zhou J, Wu J, Zheng F, Jin M, Li H. Glucagon-like peptide-1 analog-mediated protection against cholesterol-induced apoptosis via mammalian target of rapamycin activation in pancreatic betaTC-6 cells -1mTORbetaTC-6. J Diabetes 2015; 7: 231–9.

105 Maiese K. mTOR: Driving apoptosis and autophagy for neurocardiac complications of diabetes mellitus. World J Diabetes 2015; 6: 217–24.

106 Chang ZY, Yeh MK, Chiang CH, Chen YH, Lu DW. Erythropoietin protects adult retinal ganglion cells against NMDA-, trophic factor withdrawal-, and TNF-alpha-induced damage. PLoS One 2013; 8: e55291.

107 Czubak P, Bojarska-Junak A, Tabarkiewicz J, Putowski L. A modified method of insulin producing cells' generation from bone marrow-derived mesenchymal stem cells. J Diabetes Res 2014; 2014: 628591.

108 Maiese K. Protein kinase C modulates the protective ability of peptide growth factors during anoxia. J Auton Nerv Syst 1994; 49 (Suppl): S187–93.

109 Maiese K, Boccone L. Neuroprotection by peptide growth factors against anoxia and nitric oxide toxicity requires modulation of protein kinase C. J Cereb Blood Flow Metab 1995; 15: 440–9.

110 Maiese K, Boniece I, DeMeo D, Wagner JA. Peptide growth factors protect against ischemia in culture by preventing nitric oxide toxicity. J Neurosci 1993; 13: 3034–40.

111 Sato A, Sunayama J, Matsuda K, Tachibana K, Sakurada K, Tomiyama A, Kayama T, Kitanaka C. Regulation of neural stem/progenitor cell maintenance by PI3K and mTOR. Neurosci Lett 2010; 470: 115–20.

112 Kimura R, Okouchi M, Kato T, Imaeda K, Okayama N, Asai K, Joh T. Epidermal growth factor receptor transactivation is necessary for glucagon-like peptide-1 to protect PC12 cells from apoptosis. Neuroendocrinology 2013; 97: 300–8.

113 Ramanan VK, Nho K, Shen L, Risacher SL, Kim S, McDonald BC, Farlow MR, Foroud TM, Gao S, Soininen H, Kloszewska I, Mecocci P, Tsolaki M, Vellas B, Lovestone S, Aisen PS, Petersen RC, Jack CR Jr, Shaw LM, Trojanowski JQ, Weiner MW, Green RC, Toga AW, De Jager PL, Yu L, Bennett DA, Saykin AJ. FASTKD2 is associated with memory and hippocampal structure in older adults. Mol Psychiatry 2015 Oct; 20: 1197–204.

114 Slipczuk L, Bekinschtein P, Katche C, Cammarota M, Izquierdo I, Medina JH. BDNF activates mTOR to regulate GluR1 expression required for memory formation. PLoS One 2009; 4: e6007.

115 Maiese K, Chong ZZ, Hou J, Shang YC. Oxidative stress: Biomarkers and novel therapeutic pathways. Exp Gerontol 2010; Mar;45: 217–34.

116 Maiese K, Li F, Chong ZZ. New avenues of exploration for erythropoietin. JAMA 2005; 293: 90–5.

117 Shang YC, Chong ZZ, Wang S, Maiese K. Erythropoietin and Wnt1 Govern Pathways of mTOR, Apaf-1, and XIAP in Inflammatory Microglia. Curr Neurovasc Res 2011; 8: 270–85.

118 Wang GB, Ni YL, Zhou XP, Zhang WF. The AKT/mTOR pathway mediates neuronal protective effects of erythropoietin in sepsis. Mol Cell Biochem 2014; 385: 125–32.

119 Marfia G, Madaschi L, Marra F, Menarini M, Bottai D, Formenti A, Bellardita C, Di Giulio AM, Carelli S, Gorio A. Adult neural precursors isolated from post mortem brain yield mostly neurons: an erythropoietin-dependent process. Neurobiol Dis 2011; 43: 86–98.

120 Ryou MG, Choudhury GR, Li W, Winters A, Yuan F, Liu R, Yang SH. Methylene blue-induced neuronal protective mechanism against hypoxia-reoxygenation stress. Neuroscience 2015; 301: 193–203.

121 Sanghera KP, Mathalone N, Baigi R, Panov E, Wang D, Zhao X, Hsu H, Wang H, Tropepe V, Ward M, Boyd SR. The PI3K/Akt/mTOR pathway mediates retinal progenitor cell survival under hypoxic and superoxide stress. Mol Cell Neurosci 2011; 47: 145–53.

122 Berwick DC, Harvey K. The regulation and deregulation of Wnt signaling by PARK genes in health and disease. J Mol Cell Biol 2014; 6: 3–12.

123 Maiese K, Li F, Chong ZZ, Shang YC. The Wnt signaling pathway: Aging gracefully as a protectionist? Pharmacol Ther 2008; 118: 58–81.

124 Chong ZZ, Maiese K. Erythropoietin involves the phosphatidylinositol 3-kinase pathway, 14-3-3 protein and FOXO3a nuclear trafficking to preserve endothelial cell integrity. Br J Pharmacol 2007; 150: 839–50.

125 Maiese K. FoxO proteins in the nervous system. Anal Cell Pathol (Amst) 2015; 2015: 569392.

126 Krupska I, Bruford EA, Chaqour B. Eyeing the Cyr61/CTGF/NOV (CCN) group of genes in development and diseases: highlights of their structural likenesses and functional dissimilarities. Hum Genomics 2015; 9: 24.

127 Maiese K. WISP1: Clinical Insights for a Proliferative and Restorative Member of the CCN Family. Curr Neurovasc Res 2014; 11: 378–89.

128 Wang S, Chong ZZ, Shang YC, Maiese K. WISP1 neuroprotection requires FoxO3a post-translational modulation with autoregulatory control of SIRT1. Curr Neurovasc Res 2013; 10: 54–60.

129 Hou J, Wang S, Shang YC, Chong ZZ, Maiese K. Erythropoietin employs cell longevity pathways of SIRT1 to foster endothelial vascular integrity during oxidant stress. Curr Neurovasc Res 2011; 8: 220–35.

130 Tanno M, Kuno A, Yano T, Miura T, Hisahara S, Ishikawa S, Shimamoto K, Horio Y. Induction of manganese superoxide dismutase by nuclear translocation and activation of SIRT1 promotes cell survival in chronic heart failure. J Biol Chem 2010; 285: 8375–82.

131 Chong ZZ, Maiese K. Mammalian target of rapamycin signaling in diabetic cardiovascular disease. Cardiovasc Diabetol 2012; 11: 45.

132 Kopp C, Hosseini A, Singh SP, Regenhard P, Khalilvandi-Behroozyar H, Sauerwein H, Mielenz M. Nicotinic acid increases adiponectin secretion from differentiated bovine preadipocytes through G-protein coupled receptor signaling. Int J Mol Sci 2014; 15: 21401–18.

133 Martinez de Morentin PB, Martinez-Sanchez N, Roa J, Ferno J, Nogueiras R, Tena-Sempere M, Dieguez C, Lopez M. Hypothalamic mTOR: the rookie energy sensor. Curr Mol Med 2014; 14: 3–21.

134 Tsai CF, Kuo YH, Yeh WL, Wu CY, Lin HY, Lai SW, Liu YS, Wu LH, Lu JK, Lu DY. Regulatory effects of caffeic acid phenethyl ester on neuroinflammation in microglial cells. Int J Mol Sci 2015; 16: 5572–89.

135 Wang L, Di L, Noguchi CT. AMPK is involved in mediation of erythropoietin influence on metabolic activity and reactive oxygen species production in white adipocytes. Int J Biochem Cell Biol 2014; 54: 1–9.

136 Chen A, Xiong LJ, Tong Y, Mao M. Neuroprotective effect of brain-derived neurotrophic factor mediated by autophagy through the PI3K/Akt/mTOR pathway. Mol Med Rep 2013; 8: 1011–6.

137 Uzdensky A, Berezhnaya E, Khaitin A, Kovaleva V, Komandirov M, Neginskaya M, Rudkovskii M, Sharifulina S. Protection of the crayfish mechanoreceptor neuron and glial cells from photooxidative injury by modulators of diverse signal transduction pathways. Mol Neurobiol 2015; 52: 811–25.

138 Andreucci M, Fuiano G, Presta P, Lucisano G, Leone F, Fuiano L, Bisesti V, Esposito P, Russo D, Memoli B, Faga T, Michael A. Downregulation of cell survival signalling pathways and increased cell damage in hydrogen peroxide-treated human renal proximal tubular cells by alpha-erythropoietin. Cell Prolif 2009; 42: 554–61.

139 Francois A, Terro F, Quellard N, Fernandez B, Chassaing D, Janet T, Rioux-Bilan A, Paccalin M, Page G. Impairment of autophagy in the central nervous system during lipopolysaccharide-induced inflammatory stress in mice. Mol Brain 2014; 7: 56.

140 Maiese K, Chong ZZ, Shang YC, Wang S. Targeting disease through novel pathways of apoptosis and autophagy. Expert Opin Ther Targets 2012; 16: 1203–14.

141 Vakifahmetoglu-Norberg H, Xia HG, Yuan J. Pharmacologic agents targeting autophagy. J Clin Invest 2015; 125: 5–13.

142 Peng N, Meng N, Wang S, Zhao F, Zhao J, Su L, Zhang S, Zhang Y, Zhao B, Miao J. An activator of mTOR inhibits oxLDL-induced autophagy and apoptosis in vascular endothelial cells and restricts atherosclerosis in apolipoprotein E(-/-) mice. Sci Rep 2014; 4: 5519.

143 He C, Zhu H, Li H, Zou MH, Xie Z. Dissociation of Bcl-2-Beclin1 complex by activated AMPK enhances cardiac autophagy and protects against cardiomyocyte apoptosis in diabetes. Diabetes 2013; 62: 1270–81.

144 Kim KA, Shin YJ, Akram M, Kim ES, Choi KW, Suh H, Lee CH, Bae ON. High glucose condition induces autophagy in endothelial progenitor cells contributing to angiogenic impairment. Biol Pharm Bull 2014; 37: 1248–52.

145 Lim YM, Lim H, Hur KY, Quan W, Lee HY, Cheon H, Ryu D, Koo SH, Kim HL, Kim J, Komatsu M, Lee MS. Systemic autophagy insufficiency compromises adaptation to metabolic stress and facilitates progression from obesity to diabetes. Nat Commun 2014; 5: 4934.

146 Chen W, Sun Y, Liu K, Sun X. Autophagy: a double-edged sword for neuronal survival after cerebral ischemia. Neural Regen Res 2014; 9: 1210–6.

147 Chen Y, Liu XR, Yin YQ, Lee CJ, Wang FT, Liu HQ, Wu XT, Liu J. Unravelling the multifaceted roles of Atg proteins to improve cancer therapy. Cell Prolif 2014; 47: 105–12.

148 Francois A, Rioux-Bilan A, Quellard N, Fernandez B, Janet T, Chassaing D, Paccalin M, Terro F, Page G. Longitudinal follow-up of autophagy and inflammation in brain of APPswePS1dE9 transgenic mice. J Neuroinflammation 2014; 11: 139.

149 Fu Y, Chang H, Peng X, Bai Q, Yi L, Zhou Y, Zhu J, Mi M. Resveratrol inhibits breast cancer stem-like cells and induces autophagy via suppressing Wnt/beta-catenin signaling pathway. PLoS One 2014; 9: e102535.

150 Geng Y, Ju Y, Ren F, Qiu Y, Tomita Y, Tomoeda M, Kishida M, Wang Y, Jin L, Su F, Wei C, Jia B, Li Y, Chang Z. Insulin receptor substrate 1/2 (IRS1/2) regulates Wnt/beta-catenin signaling through blocking autophagic degradation of dishevelled2. J Biol Chem 2014; 289: 11230–41.

151 Wu H, Lu MH, Wang W, Zhang MY, Zhu QQ, Xia YY, Xu RX, Yang Y, Chen LH, Ma QH. Lamotrigine reduces beta-site abetaPP-cleaving enzyme one protein levels through induction of autophagy. J Alzheimers Dis 2015; 46: 863–876.

152 Kamada Y, Funakoshi T, Shintani T, Nagano K, Ohsumi M, Ohsumi Y. Tor-mediated induction of autophagy via an Apg1 protein kinase complex. J Cell Biol 2000; 150: 1507–13.

153 Kabeya Y, Kamada Y, Baba M, Takikawa H, Sasaki M, Ohsumi Y. Atg17 functions in cooperation with Atg1 and Atg13 in yeast autophagy. Mol Biol Cell 2005; 16: 2544–53.

154 Hosokawa N, Hara T, Kaizuka T, Kishi C, Takamura A, Miura Y, Iemura S, Natsume T, Takehana K, Yamada N, Guan JL, Oshiro N, Mizushima N. Nutrient-dependent mTORC1 association with the ULK1-Atg13-FIP200 complex required for autophagy. Mol Biol Cell 2009; 20: 1981–91.

155 Jung CH, Jun CB, Ro SH, Kim YM, Otto NM, Cao J, Kundu M, Kim DH. ULK-Atg13-FIP200 complexes mediate mTOR signaling to the autophagy machinery. Mol Biol Cell 2009; 20: 1992–2003.

156 Cai Z, Yan LJ. Rapamycin, autophagy, and Alzheimer's disease. J Biochem Pharmacol Res 2013; 1: 84–90.

157 Balduini W, Carloni S, Buonocore G. Autophagy in hypoxia-ischemia induced brain injury. J Matern Fetal Neonatal Med 2012; 25 (Suppl 1): 30–4.

158 Kulbe JR, Mulcahy Levy JM, Coultrap SJ, Thorburn A, Bayern KU. Excitotoxic glutamate insults block autophagic flux in hippocampal neurons. Brain Res 2014; 1542: 12–9.

159 Sekiguchi A, Kanno H, Ozawa H, Yamaya S, Itoi E. Rapamycin promotes autophagy and reduces neural tissue damage and locomotor impairment after spinal cord injury in mice. J Neurotrauma 2012; 29: 946–56.

160 Jeong JK, Moon MH, Bae BC, Lee YJ, Seol JW, Kang HS, Kim JS, Kang SJ, Park SY. Autophagy induced by resveratrol prevents human prion protein-mediated neurotoxicity. Neurosci Res 2012; 73: 99–105.

161 Chan TM, Chen JY, Ho LI, Lin HP, Hsueh KW, Liu DD, Chen YH, Hsieh AC, Tsai NM, Hueng DY, Tsai ST, Chou PW, Lin SZ, Harn HJ. ADSC therapy in neurodegenerative disorders. Cell Transplant 2014; 23: 549–57.

162 Dong W, Wang R, Ma LN, Xu BL, Zhang JS, Zhao ZW, Wang YL, Zhang X. Influence of age-related learning and memory capacity of mice: different effects of a high and low caloric diet. Aging Clin Exp Res 2015; 3.

163 Zhu Z, Yan J, Jiang W, Yao XG, Chen J, Chen L, Li C, Hu L, Jiang H, Shen X. Arctigenin effectively ameliorates memory impairment in Alzheimer's disease model mice targeting both beta-amyloid production and clearance. J Neurosci 2013; 33: 13138–49.

164 Urbanek T, Kuczmik W, Basta-Kaim A, Gabryel B. Rapamycin induces of protective autophagy in vascular endothelial cells exposed to oxygen-glucose deprivation. Brain Res 2014; 1553: 1–11.

165 Balan V, Miller GS, Kaplun L, Balan K, Chong ZZ, Li F, Kaplun A, VanBerkum MF, Arking R, Freeman DC, Maiese K, Tzivion G. Life span extension and neuronal cell protection by Drosophila nicotinamidase. J Biol Chem 2008; 283: 27810–9.

166 Chong ZZ, Wang S, Shang YC, Maiese K. Targeting cardiovascular disease with novel SIRT1 pathways. Future Cardiol 2012; 8: 89–100.

167 Maiese K, Chong ZZ, Shang YC, Wang S. Translating cell survival and cell longevity into treatment strategies with SIRT1. Rom J Morphol Embryol 2011; 52: 1173–85.

168 Paraiso AF, Mendes KL, Santos SH. Brain activation of SIRT1: role in neuropathology. Mol Neurobiol 2013; 48: 681–9.

169 Wang W, Yan C, Zhang J, Lin R, Lin Q, Yang L, Ren F, Zhang J, Ji M, Li Y. SIRT1 inhibits TNF-alpha-induced apoptosis of vascular adventitial fibroblasts partly through the deacetylation of FoxO1. Apoptosis 2013; 18: 689–701.

170 Xiong S, Salazar G, Patrushev N, Alexander RW. FoxO1 Mediates an Autofeedback Loop Regulating SIRT1 Expression. J Biol Chem 2011; 286: 5289–99.

171 Vidal RL, Figueroa A, Court FA, Thielen P, Molina C, Wirth C, Caballero B, Kiffin R, Segura-Aguilar J, Cuervo AM, Glimcher LH, Hetz C. Targeting the UPR transcription factor XBP1 protects against Huntington's disease through the regulation of FoxO1 and autophagy. Hum Mol Genet 2012; 21: 2245–62.

172 Chen J, Xavier S, Moskowitz-Kassai E, Chen R, Lu CY, Sanduski K, Spes A, Turk B, Goligorsky MS. Cathepsin cleavage of sirtuin one in endothelial progenitor cells mediates stress-induced premature senescence. Am J Pathol 2012; 180: 973–83.

173 Ou X, Lee MR, Huang X, Messina-Graham S, Broxmeyer HE. SIRT1 positively regulates autophagy and mitochondria function in embryonic stem cells under oxidative stress. Stem Cells 2014; 32: 1183–94.

174 Guo W, Qian L, Zhang J, Zhang W, Morrison A, Hayes P, Wilson S, Chen T, Zhao J. Sirt1 overexpression in neurons promotes neurite outgrowth and cell survival through inhibition of the mTOR signaling. J Neurosci Res 2011; 89: 1723–36.

175 Ma L, Dong W, Wang R, Li Y, Xu B, Zhang J, Zhao Z, Wang Y. Effect of caloric restriction on the SIRT1/mTOR signaling pathways in senile mice. Brain Res Bull 2015; 116: 67–72.

176 Yin B, Liang H, Chen Y, Chu K, Huang L, Fang L, Matro E, Jiang W, Luo B. EGB1212 post-treatment ameliorates hippocampal CA1 neuronal death and memory impairment induced by transient global cerebral ischemia/reperfusion. Am J Chin Med 2013; 41: 1329–41.

177 Francois A, Terro F, Janet T, Bilan AR, Paccalin M, Page G. Involvement of interleukin-1beta in the autophagic process of microglia: relevance to Alzheimer's disease. J Neuroinflammation 2013; 10: 151.

178 Yu Y, Shiou SR, Guo Y, Lu L, Westerhoff M, Sun J, Petrof EO, Claud EC. Erythropoietin protects epithelial cells from excessive autophagy and apoptosis in experimental neonatal necrotizing enterocolitis. PLoS One 2013; 8: e69620.

179 Bendix I, Schulze C, Haefen C, Gellhaus A, Endesfelder S, Heumann R, Felderhoff-Mueser U, Sifringer M. Erythropoietin modulates autophagy signaling in the developing rat brain in an *in vivo* model of oxygen-toxicity. Int J Mol Sci 2012; 13: 12939–51.

180 Bains M, Zaegel V, Mize-Berge J, Heidenreich KA. IGF-I stimulates Rab7-RILP interaction during neuronal autophagy. Neurosci Lett 2011 Jan 20; 488: 112–17.

181 Maiese K. Regeneration in the Nervous System with Erythropoietin. Frontiers Biosci (Landmark edition) 2016; 21: 561–96.

182 Wang S, Chong ZZ, Shang YC, Maiese K. WISP1 (CCN4) autoregulates its expression and nuclear trafficking of beta-catenin during oxidant stress with limited effects upon neuronal autophagy. Curr Neurovasc Res 2012; 9: 89–99.

183 Danielyan L, Schafer R, Schulz A, Ladewig T, Lourhmati A, Buadze M, Schmitt AL, Verleysdonk S, Kabisch D, Koeppen K, Siegel G, Proksch B, Kluba T, Eckert A, Kohle C, Schoneberg T, Northoff H, Schwab M, Gleiter CH. Survival, neuron-like differentiation and functionality of mesenchymal stem cells in neurotoxic environment: the critical role of erythropoietin. Cell Death Differ 2009; 16: 1599–614.

184 Kim J, Jung Y, Sun H, Joseph J, Mishra A, Shiozawa Y, Wang J, Krebsbach PH, Taichman RS. Erythropoietin mediated bone formation is regulated by mTOR signaling. J Cell Biochem 2012; 113: 220–8.

185 Han J, Wang B, Xiao Z, Gao Y, Zhao Y, Zhang J, Chen B, Wang X, Dai J. Mammalian target of rapamycin (mTOR) is involved in the neuronal differentiation of neural progenitors induced by insulin. Mol Cell Neurosci 2008; 39: 118–24.

186 Hartman NW, Lin TV, Zhang L, Paquelet GE, Feliciano DM, Bordey A. mTORC1 targets the translational repressor 4E-BP2, but not S6 kinase 1/2, to regulate neural stem cell self-renewal *in vivo*. Cell Reprogram 2013; 5: 433–44.

187 Magri L, Cambiaghi M, Cominelli M, Alfaro-Cervello C, Cursi M, Pala M, Bulfone A, Garcia-Verdugo JM, Leocani L, Minicucci F, Poliani PL, Galli R. Sustained activation of mTOR pathway in embryonic neural stem cells leads to development of tuberous sclerosis complex-associated lesions. Cell Stem Cell 2011; 9: 447–62.

188 Malagelada C, Lopez-Toledano MA, Willett RT, Jin ZH, Shelanski ML, Greene LA. RTP801/REDD1 regulates the timing of cortical neurogenesis and neuron migration. J Neurosci 2011; 31: 3186–96.

189 Romine J, Gao X, Xu XM, So KF, Chen J. The proliferation of amplifying neural progenitor cells is impaired in the aging brain and restored by the mTOR pathway activation. Neurobiol Aging 2015; 36: 1716–26.

190 Zhang D, Yan B, Yu S, Zhang C, Wang B, Wang Y, Wang J, Yuan Z, Zhang L, Pan J. Coenzyme Q10 inhibits the aging of mesenchymal stem cells induced by D-galactose through Akt/mTOR signaling. Oxid Med Cell Longev 2015; 2015: 867293.

191 Galan-Moya EM, Le Guelte A, Lima Fernandes E, Thirant C, Dwyer J, Bidere N, Couraud PO, Scott MG, Junier MP, Chneiweiss H, Gavard J. Secreted factors from brain endothelial cells maintain glioblastoma stem-like cell expansion through the mTOR pathway. EMBO Rep 2011; 12: 470–6.

192 Sun JJ, Ren QG, Xu L, Zhang ZJ. LINGO-1 antibody ameliorates myelin impairment and spatial memory deficits in experimental autoimmune encephalomyelitis mice. Sci Rep 2015; 5: 14235.

193 Maiese K, Chong ZZ, Shang YC, Hou J. Therapeutic promise and principles: Metabotropic glutamate receptors. Oxid Med Cell Longev. 2008; 1: 1–14.

194 Hullinger R, O'Riordan K, Burger C. Environmental enrichment improves learning and memory and long-term potentiation in young adult rats through a mechanism requiring mGluR5 signaling and sustained activation of p70s6k. Neurobiol Learn Mem 2015; 125: 126–34.

195 Ma T, Hoeffer CA, Capetillo-Zarate E, Yu F, Wong H, Lin MT, Tampellini D, Klann E, Blitzer RD, Gouras GK. Dysregulation of the mTOR pathway mediates impairment of synaptic plasticity in a mouse model of Alzheimer's disease. PLoS One 2010 Sep 20; 5: e12845.

196 Li L, Xu B, Zhu Y, Chen L, Sokabe M. DHEA prevents Abeta25-35-impaired survival of newborn neurons in the dentate gyrus through a modulation of PI3K-Akt-mTOR signaling. Neuropharmacology 2010; 59: 323–33.

197 Gong R, Park CS, Abbassi NR, Tang SJ. Roles of glutamate receptors and the mammalian target of rapamycin (mTOR) signaling pathway in activity-dependent dendritic protein synthesis in hippocampal neurons. J Biol Chem 2006; 281: 18802–15.

198 Parsons RG, Gafford GM, Helmstetter FJ. Translational control via the mammalian target of rapamycin pathway is critical for the formation and stability of long-term fear memory in amygdala neurons. J Neurosci 2006; 26: 12977–83.

199 Zhao H, Wang ZC, Wang KF, Chen XY. Abeta peptide secretion is reduced by Radix Polygalaeinduced autophagy via activation of the AMPK/mTOR pathway. Mol Med Rep 2015 Aug; 12: 2771–76.

200 Paccalin M, Pain-Barc S, Pluchon C, Paul C, Besson MN, Carret-Rebillat AS, Rioux-Bilan A, Gil R, Hugon J. Activated mTOR and PKR kinases in lymphocytes correlate with memory and cognitive decline in Alzheimer's disease. Dement Geriatr Cogn Disord 2006; 22: 320–6.

201 Chano T, Okabe H, Hulette CM. RB1CC1 insufficiency causes neuronal atrophy through mTOR signaling alteration and involved in the pathology of Alzheimer's diseases. Brain Res 2007; 1168: 97–105.

202 Shahani N, Pryor W, Swarnkar S, Kholodilov N, Thinakaran G, Burke RE, Subramaniam S. Rheb GTPase regulates beta-secretase levels and amyloid beta generation. J Biol Chem 2014; 289: 5799–808.

203 Chen TJ, Wang DC, Chen SS. Amyloid-beta interrupts the PI3K-Akt-mTOR signaling pathway that could be involved in brain-derived neurotrophic factor-induced Arc expression in rat cortical neurons. J Neurosci Res 2009; 87: 2297–307.

204 Cai Z, Chen G, He W, Xiao M, Yan LJ. Activation of mTOR: a culprit of Alzheimer's disease? Neuropsych Dis Treat 2015; 11: 1015–30.

205 An WL, Cowburn RF, Li L, Braak H, Alafuzoff I, Iqbal K, Iqbal IG, Winblad B, Pei JJ. Up-regulation of phosphorylated/activated p70 S6 kinase and its relationship to neurofibrillary pathology in Alzheimer's disease. Am J Pathol 2003; 163: 591–607.

206 Caccamo A, De Pinto V, Messina A, Branca C, Oddo S. Genetic reduction of mammalian target of rapamycin ameliorates Alzheimer's disease-like cognitive and pathological deficits by restoring hippocampal gene expression signature. J Neurosci 2014; 34: 7988–98.

207 Liu YW, Zhang L, Li Y, Cheng YQ, Zhu X, Zhang F, Yin XX. Activation of mTOR signaling mediates the increased expression of AChE in high glucose condition: in vitro and *in vivo* evidences. Mol Neurobiol 2015; 15.

208 Jiang T, Yu JT, Zhu XC, Tan MS, Wang HF, Cao L, Zhang QQ, Shi JQ, Gao L, Qin H, Zhang YD, Tan L. Temsirolimus promotes autophagic clearance of amyloid-beta and provides protective effects in cellular and animal models of Alzheimer's disease. Pharmacol Res 2014; 81C: 54–63.

209 Frederick C, Ando K, Leroy K, Heraud C, Suain V, Buee L, Brion JP. Rapamycin ester analog CCI-779/Temsirolimus alleviates tau pathology and improves motor deficit in mutant tau transgenic mice. J Alzheimers Dis 2015; 44: 1145–56.

210 Shibuya Y, Chang CC, Huang LH, Bryleva EY, Chang TY. Inhibiting ACAT1/SOAT1 in microglia stimulates autophagy-mediated lysosomal proteolysis and increases Abeta1-42 clearance. J Neurosci 2014; 34: 14484–501.

211 Dijkstra AA, Ingrassia A, de Menezes RX, van Kesteren RE, Rozemuller AJ, Heutink P, van de Berg WD. Evidence for immune response, axonal dysfunction and reduced endocytosis in the substantia nigra in early stage Parkinson's disease. PLoS One 2015; 10: e0128651.

212 Zhou Q, Liu C, Liu W, Zhang H, Zhang R, Liu J, Zhang J, Xu C, Liu L, Huang S, Chen L. Rotenone induction of hydrogen peroxide inhibits mTOR-mediated S6K1 and 4E-BP1/eIF4E pathways, leading to neuronal apoptosis. Toxicol Sci 2015; 143: 81–96.

213 Imai Y, Gehrke S, Wang HQ, Takahashi R, Hasegawa K, Oota E, Lu B. Phosphorylation of 4E-BP by LRRK2 affects the maintenance of dopaminergic neurons in Drosophila. EMBO J 2008; 27: 2432–43.

214 Tain LS, Mortiboys H, Tao RN, Ziviani E, Bandmann O, Whitworth AJ. Rapamycin activation of 4E-BP prevents parkinsonian dopaminergic neuron loss. Nat Neurosci 2009; 12: 1129–35.

215 Wu J, Zhu D, Zhang J, Li G, Liu Z, Sun J. Lithium protects against methamphetamine-induced neurotoxicity in PC12 cells via Akt/GSK3beta/mTOR pathway. Biochem Biophys Res Commun 2015; 465: 368–73.

216 Xu Y, Liu C, Chen S, Ye Y, Guo M, Ren Q, Liu L, Zhang H, Xu C, Zhou Q, Huang S, Chen L. Activation of AMPK and inactivation of Akt result in suppression of mTOR-mediated S6K1 and 4E-BP1 pathways leading to neuronal cell death in *in vitro* models of Parkinson's disease. Cell Signal 2014; 26: 1680–9.

217 Zhang H, Duan C, Yang H. Defective Autophagy in Parkinson's Disease: Lessons from Genetics. Mol Neurobiol 2015; 51: 89–104.

218 Crews L, Spencer B, Desplats P, Patrick C, Paulino A, Rockenstein E, Hansen L, Adame A, Galasko D, Masliah E. Selective molecular alterations in the autophagy pathway in patients with Lewy body disease and in models of alpha-synucleinopathy. PLoS One 2010; 5: e9313.

219 Maiese K, Chong ZZ, Wang S, Shang YC. Oxidant stress and signal transduction in the nervous system with the PI 3-K, Akt and mTOR cascade. Int J Mol Sci 2013; 13: 13830–66.

220 Pino E, Amamoto R, Zheng L, Cacquevel M, Sarria JC, Knott GW, Schneider BL. FOXO3 determines the accumulation of alpha-synuclein and controls the fate of dopaminergic neurons in the substantia nigra. Hum Mol Genet 2014; 23: 1435–52.

221 Williams AC, Hill LJ, Ramsden DB. Nicotinamide, NAD(P)(H), and Methyl-Group Homeostasis Evolved and Became a Determinant of Ageing Diseases: Hypotheses and Lessons from Pellagra. Curr Gerontol Geriatr Res 2012; 2012: 302875.

222 Sasazawa Y, Sato N, Umezawa K, Simizu S. Conophylline protects cells in cellular models of neurodegenerative diseases by inducing mammalian target of rapamycin (mTOR)-independent autophagy. J Biol Chem 2015; 290: 6168–78.

223 Tourette C, Li B, Bell R, O'Hare S, Kaltenbach LS, Mooney SD, Hughes RE. A Large-scale huntingtin protein interaction network implicates rho GTPase signaling pathways in Huntington's disease. J Biol Chem 2014 Mar 7; 289: 6709–26.

224 Berger Z, Ravikumar B, Menzies FM, Oroz LG, Underwood BR, Pangalos MN, Schmitt I, Wullner U, Evert BO, O'Kane CJ, Rubinsztein DC. Rapamycin alleviates toxicity of different aggregate-prone proteins. Hum Mol Genet 2006; 15: 433–42.

225 Pryor WM, Biagioli M, Shahani N, Swarnkar S, Huang WC, Page DT, MacDonald ME, Subramaniam S. Huntingtin promotes mTORC1 signaling in the pathogenesis of Huntington's disease. Sci Signal 2014; 7: ra103.

226 Roscic A, Baldo B, Crochemore C, Marcellin D, Paganetti P. Induction of autophagy with catalytic mTOR inhibitors reduces huntingtin aggregates in a neuronal cell model. J Neurochem 2011; 119: 398–407.

227 Fox JH, Connor T, Chopra V, Dorsey K, Kama JA, Bleckmann D, Betschart C, Hoyer D, Frentzel S, Difiglia M, Paganetti P, Hersch SM. The mTOR kinase inhibitor Everolimus decreases S6 kinase phosphorylation but fails to reduce mutant huntingtin levels in brain and is not neuroprotective in the R6/2 mouse model of Huntington's disease. Mol Neurobiol 2010; 5: 26.

228 Lee JH, Tecedor L, Chen YH, Monteys AM, Sowada MJ, Thompson LM, Davidson BL. Reinstating aberrant mTORC1 activity in Huntington's disease mice improves disease phenotypes. Neuron 2015; 85: 303–15.

229 Holmes GL, Stafstrom CE. Tuberous sclerosis complex and epilepsy: recent developments and future challenges. Epilepsia 2007; 48: 617–30.

230 Liu J, Reeves C, Michalak Z, Coppola A, Diehl B, Sisodiya SM, Thom M. Evidence for mTOR pathway activation in a spectrum of epilepsy-associated pathologies. Acta Neuropathol Commun 2014; 2: 71.

231 Curatolo P, Moavero R. mTOR inhibitors as a new therapeutic option for epilepsy. Expert Rev Neurother 2013; 13: 627–38.

232 Meng XF, Yu JT, Song JH, Chi S, Tan L. Role of the mTOR signaling pathway in epilepsy. J Neurol Sci 2013; 332: 4–15.

233 Huang X, McMahon J, Huang Y. Rapamycin attenuates aggressive behavior in a rat model of pilocarpine-induced epilepsy. Neuroscience 2012 Jul 26; 215: 90–7.

234 Russo E, Andreozzi F, Iuliano R, Dattilo V, Procopio T, Fiume G, Mimmi S, Perrotti N, Citraro R, Sesti G, Constanti A, De Sarro G. Early molecular and behavioral response to lipopolysaccharide in the WAG/Rij rat model of absence epilepsy and depressive-like behavior, involves interplay between AMPK, AKT/mTOR pathways and neuroinflammatory cytokine release. Brain Behav Immun 2014; 42: 157–68.

235 Talos DM, Sun H, Zhou X, Fitzgerald EC, Jackson MC, Klein PM, Lan VJ, Joseph A, Jensen FE. The interaction between early life epilepsy and autistic-like behavioral consequences: a role for the mammalian target of rapamycin (mTOR) pathway. PLoS One 2012; 7: e35885.

236 Benz AP, Niquet J, Wasterlain CG, Rami A. Status epilepticus in the immature rodent brain alters the dynamics of autophagy. Curr Neurovasc Res 2014; 11: 125–35.

237 Reith RM, Way S, McKenna J 3rd, Haines K, Gambello MJ. Loss of the tuberous sclerosis complex protein tuberin causes Purkinje cell degeneration. Neurobiol Dis 2011; 43: 113–22.

238 Sosunov AA, Wu X, McGovern RA, Coughlin DG, Mikell CB, Goodman RR, McKhann GM 2nd. The mTOR pathway is activated in glial cells in mesial temporal sclerosis. Epilepsia 2012; 53 (Suppl 1): 78–86.

239 Rozas NS, Redell JB, McKenna J 3rd, Moore AN, Gambello MJ, Dash PK. Prolonging the survival of Tsc2 conditional knockout mice by glutamine supplementation. Biochem Biophys Res Commun 2015; 457: 635–9.

240 Curran MP. Everolimus: in patients with subependymal giant cell astrocytoma associated with tuberous sclerosis complex. Paediatr Drugs 2012; 14: 51–60.

241 Krueger DA, Care MM, Holland K, Agricola K, Tudor C, Mangeshkar P, Wilson KA, Byars A, Sahmoud T, Franz DN. Everolimus for subependymal giant-cell astrocytomas in tuberous sclerosis. N Engl J Med 2010; 363: 1801–11.

242 Krueger DA, Wilfong AA, Holland-Bouley K, Anderson AE, Agricola K, Tudor C, Mays M, Lopez CM, Kim MO, Franz DN. Everolimus treatment of refractory epilepsy in tuberous sclerosis complex. Ann Neurol 2013; 74: 679–87.

243 Abe N, Borson SH, Gambello MJ, Wang F, Cavalli V. Mammalian target of rapamycin (mTOR) activation increases axonal growth capacity of injured peripheral nerves. J Biol Chem 2010; 285: 28034–43.

244 Xie R, Wang P, Ji X, Zhao H. Ischemic post-conditioning facilitates brain recovery after stroke by promoting Akt/mTOR activity in nude rats. J Neurochem 2013; 127: 723–32.

245 You H, Li T, Zhang J, Lei Q, Tao X, Xie P, Lu W. Reduction in ischemic cerebral infarction is mediated through Golgi phosphoprotein three and Akt/mTOR signaling following salvianolate administration. Curr Neurovasc Res 2014 May; 11: 107–13.

246 Nguyen AQ, Cherry BH, Scott GF, Ryou MG, Mallet RT. Erythropoietin: Powerful protection of ischemic and post-ischemic brain. Exp Biol Med (Maywood) 2014; 4.

247 Maiese K. MicroRNAs and stem cells to the rescue. Curr Neurovasc Res 2015.

248 Selvamani A, Williams MH, Miranda RC, Sohrabji F. Circulating miRNA profiles provide a biomarker for severity of stroke outcomes associated with age and sex in a rat model. Clin Sci (Lond) 2014.

249 Liu G, Detloff MR, Miller KN, Santi L, Houle JD. Exercise modulates microRNAs that affect the PTEN/mTOR pathway in rats after spinal cord injury. Exp Neurol 2012; 233: 447–56.

250 Chong ZZ, Li F, Maiese K. The pro-survival pathways of mTOR and protein kinase B target glycogen synthase kinase-3beta and nuclear factor-kappaB to foster endogenous microglial cell protection. Int J Mol Med 2007; 19: 263–72.

251 Dello Russo C, Lisi L, Tringali G, Navarra P. Involvement of mTOR kinase in cytokine-dependent microglial activation and cell proliferation. Biochem Pharmacol 2009; 78: 1242–51.

252 Ji YF, Zhou L, Xie YJ, Xu SM, Zhu J, Teng P, Shao CY, Wang Y, Luo JH, Shen Y. Upregulation of glutamate transporter GLT-1 by mTOR-Akt-NF-small ka, CyrillicB cascade in astrocytic oxygen-glucose deprivation. Glia 2013; 61: 1959–75.

253 Yan H, Zhang X, Hu W, Ma J, Hou W, Zhang X, Wang X, Gao J, Shen Y, Lv J, Ohtsu H, Han F, Wang G, Chen Z. Histamine H3 receptors aggravate cerebral ischaemic injury by histamine-independent mechanisms. Nat Commun 2014; 5: 3334.

254 Fletcher L, Evans TM, Watts LT, Jimenez DF, Digicaylioglu M. Rapamycin treatment improves neuron viability in an *in vitro* model of stroke. PLoS One 2013; 8: e68281.

255 Xia DY, Li W, Qian HR, Yao S, Liu JG, Qi XK. Ischemia preconditioning is neuroprotective in a rat cerebral ischemic injury model through autophagy activation and apoptosis inhibition. Braz J Med Biol Res 2013; 46: 580–8.

256 Maiese K. Neuronal activity, mitogens, and mTOR: Overcoming the hurdles for the treatment of glioblastoma multiforme. J Transl Sci 2015; 1: 2.

257 Venkatesh HS, Johung TB, Caretti V, Noll A, Tang Y, Nagaraja S, Gibson EM, Mount CW, Polepalli J, Mitra SS, Woo PJ, Malenka RC, Vogel H, Bredel M, Mallick P, Monje M. Neuronal activity promotes glioma growth through neuroligin-3 secretion. Cell 2015; 161: 803–16.

258 Kolev VN, Wright QG, Vidal CM, Ring JE, Shapiro IM, Ricono J, Weaver DT, Padval MV, Pachter JA, Xu Q. PI3K/mTOR dual inhibitor VS-5584 preferentially targets cancer stem cells. Cancer Res 2015; 75: 446–55.

259 Karthik GM, Ma R, Lovrot J, Kis LL, Lindh C, Blomquist L, Fredriksson I, Bergh J, Hartman J. mTOR inhibitors counteract tamoxifen-induced activation of breast cancer stem cells. Cancer Lett 2015; 367: 76–87.

260 Hedley BD, Allan AL, Xenocostas A. The role of erythropoietin and erythropoiesis-stimulating agents in tumor progression. Clin Cancer Res 2011; 17: 6373–80.

261 Maiese K, Li F, Chong ZZ. Erythropoietin and cancer. JAMA 2005; 293: 1858–9.

262 Zhang C, Li Z, Cao Q, Qin C, Cai H, Zhou H, Qian J, Tao L, Ju X, Yin C. Association of erythropoietin gene rs576236 polymorphism and risk of adrenal tumors in a Chinese population. J Biomed Res 2014; 28: 456–61.

263 Ogunshola OO, Moransard M, Gassmann M. Constitutive excessive erythrocytosis causes inflammation and increased vascular permeability in aged mouse brain. Brain Res 2013; 1531: 48–57.

264 Liu Y, Yan W, Zhang W, Chen L, You G, Bao Z, Wang Y, Wang H, Kang C, Jiang T. MiR-218 reverses high invasiveness of glioblastoma cells by targeting the oncogenic transcription factor LEF1. Oncol Rep 2012; 28: 1013–21.

265 Tu Y, Gao X, Li G, Fu H, Cui D, Liu H, Jin W, Zhang Y. MicroRNA-218 inhibits glioma invasion, migration, proliferation, and cancer stem-like cell self-renewal by targeting the polycomb group gene Bmi1. Cancer Res 2013; 73: 6046–55.

266 James RG, Davidson KC, Bosch KA, Biechele TL, Robin NC, Taylor RJ, Major MB, Camp ND, Fowler K, Martins TJ, Moon RT. WIKI4, a novel inhibitor of tankyrase and Wnt/ss-catenin signaling. PLoS One 2012; 7: e50457.

267 Kafka A, Basic-Kinda S, Pecina-Slaus N. The cellular story of dishevelleds. Croat Med J 2014; 55: 459–67.

268 Klinke DJ 2nd. Induction of Wnt-inducible signaling protein-1 correlates with invasive breast cancer oncogenesis and reduced type one cell-mediated cytotoxic immunity: a retrospective study. PLoS Comput Biol 2014; 10: e1003409.

269 Knoblich K, Wang HX, Sharma C, Fletcher AL, Turley SJ, Hemler ME. Tetraspanin TSPAN12 regulates tumor growth and metastasis and inhibits beta-catenin degradation. Cell Mol Life Sci 2014; 71: 1305–14.

270 Uzdensky AB, Demyanenko SV, Bibov MY. Signal transduction in human cutaneous melanoma and target drugs. Curr Cancer Drug Targets 2013; 13: 843–66.

271 Ono M, Inkson CA, Sonn R, Kilts TM, de Castro LF, Maeda A, Fisher LW, Robey PG, Berendsen AD, Li L, McCartney-Francis N, Brown AC, Crawford NP, Molinolo A, Jain A, Fedarko NS, Young MF. WISP1/CCN4: a potential target for inhibiting prostate cancer growth and spread to bone. PLoS One 2013; 8: e71709.

272 Soon LL, Yie TA, Shvarts A, Levine AJ, Su F, Tchou-Wong KM. Overexpression of WISP-1 down-regulated motility and invasion of lung cancer cells through inhibition of Rac activation. J Biol Chem 2003; 278: 11465–70.

273 Castilho RM, Squarize CH, Chodosh LA, Williams BO, Gutkind JS. mTOR mediates Wnt-induced epidermal stem cell exhaustion and aging. Cell Stem Cell 2009; 5: 279–89.

274 Dello Russo C, Lisi L, Feinstein DL, Navarra P. mTOR kinase, a key player in the regulation of glial functions: Relevance for the therapy of multiple sclerosis. Glia 2013; 61: 301–11.

275 Pilar-Cuellar F, Vidal R, Diaz A, Castro E, dos Anjos S, Pascual-Brazo J, Linge R, Vargas V, Blanco H, Martinez-Villayandre B, Pazos A, Valdizan EM. Neural plasticity and proliferation in the generation of antidepressant effects: hippocampal implication. Neural Plast 2013; 2013: 537265.

276 Chiarini F, Evangelisti C, McCubrey JA, Martelli AM. Current treatment strategies for inhibiting mTOR in cancer. Trends Pharmacol Sci 2015; 36: 124–35.

277 Jin N, Jiang T, Rosen DM, Nelkin BD, Ball DW. Dual inhibition of mitogen-activated protein kinase kinase and mammalian target of rapamycin in differentiated and anaplastic thyroid cancer. J Clin Endocrinol Metab 2009; 94: 4107–12.

278 Gulhati P, Zaytseva YY, Valentino JD, Stevens PD, Kim JT, Sasazuki T, Shirasawa S, Lee EY, Weiss HL, Dong J, Gao T, Evers BM. Sorafenib enhances the therapeutic efficacy of rapamycin in colorectal cancers harboring oncogenic KRAS and PIK3CA. Carcinogenesis 2012 Sep; 33: 1782–90.

279 Mi R, Ma J, Zhang D, Li L, Zhang H. Efficacy of combined inhibition of mTOR and ERK/MAPK pathways in treating a tuberous sclerosis complex cell model. J Genet Genomics (Yi Chuan Xue Bao) 2009; 36: 355–61.

280 Pavel ME, Hainsworth JD, Baudin E, Peeters M, Horsch D, Winkler RE, Klimovsky J, Lebwohl D, Jehl V, Wolin EM, Oberg K, Van Cutsem E, Yao JC. Everolimus plus octreotide long-acting repeatable for the treatment of advanced neuroendocrine tumours associated with carcinoid

syndrome (RADIANT-2): a randomised, placebo-controlled, phase 3 study. Lancet 2011; 378: 2005–12.

281 Sanchez-Fructuoso AI, Ruiz JC, Calvo N, Rodrigo E, Perez-Flores I, Gomez-Alamillo C, Fernandez-Perez C, Arias M, Barrientos A. Everolimus as primary immunosuppression in kidney transplantation: experience in conversion from calcineurin inhibitors. Transplantation 2012; 93: 398–405.