REVIEW

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mTOR is a key modulator of ageing and age-related disease

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Many experts in the biology of ageing believe that pharmacological interventions to slow ageing are a matter of 'when' rather than 'if'. A leading target for such interventions is the nutrient response pathway defined by the mechanistic target of rapamycin (mTOR). Inhibition of this pathway extends lifespan in model organisms and confers protection against a growing list of age-related pathologies. Characterized inhibitors of this pathway are already clinically approved, and others are under development. Although adverse side effects currently preclude use in otherwise healthy individuals, drugs that target the mTOR pathway could one day become widely used to slow ageing and reduce age-related pathologies in humans.

The mechanistic target of rapamycin (mTOR) story began in the 1970s when new antifungal activity was discovered in soil samples from the Polynesian island of Rapa Nui (Box 1). Thus, the compound, which was isolated from *Streptomyces hygroscopicus*, was named rapamycin. Also known as sirolimus, rapamycin was widely studied as an immunosuppressant before its mechanism of action was well understood, and in 1999 it was approved for use in post-transplantation therapy. Since then, rapamycin and several derivative compounds (including everolimus, temsirolimus, ridaforolimus, umirolimus and zotarolimus, collectively referred to as 'rapamycins' in this Review) have been approved for a variety of uses, including prevention of restenosis following angioplasty and as a treatment for certain forms of cancer.

Studies in the budding yeast *Saccharomyces cerevisiae* first identified the target of rapamycin genes *TOR1* and *TOR2* as genetic mediators of rapamycin's growth inhibitory effects, and soon afterwards the mTOR protein was purified from mammalian cells and demonstrated to be the physical target of rapamycin[1]. mTOR is a serine/threonine protein kinase of the phosphatidylinositol-3-OH kinase (PI(3)K)-related family that functions as a master regulator of cellular growth and metabolism in response to nutrient and hormonal cues[2]. mTOR functions in two distinct complexes: mTOR complex 1 (mTORC1) and mTOR complex 2 (mTORC2) (Fig. 1). Rapamycins inhibit mTORC1 by binding the FK506-binding protein FKBP12, which then interacts physically with the complex and decreases activity. Although mTORC2 is not directly affected by rapamycin, chronic exposure can sequester mTOR from mTORC2, inhibiting mTORC2 assembly. This effect on mTORC2 is thought to contribute to metabolic complications associated with chronic rapamycin treatment, including glucose intolerance and abnormal lipid profiles[3] (described further later).

Much more is known about both the upstream regulation and downstream outputs of mTORC1 compared with mTORC2. mTORC1 is activated by insulin and other growth factors through PI(3)K and AKT kinase signalling[1]. mTORC1 is also activated by environmental nutrients (for example, amino acids) and repressed by AMP-activated protein kinase (AMPK), a key sensor of cellular energy status (discussed further later). In response to these growth signals, mTORC1 is thought to promote messenger RNA translation and protein synthesis through at least two mTORC1 substrates, ribosomal protein S6 kinases (S6Ks) and eukaryotic translation initiation factor 4E-binding protein 1 (4E-BP1). mTORC1 also promotes lipid biosynthesis, represses degradation through the autophagy pathway, and regulates glucose metabolism and mitochondrial function through the hypoxic response transcription factor HIF-1α (Fig. 1) as well as the peroxisome-proliferator-activated receptor coactivator PGC-1α. These multiple inputs and outputs place mTORC1 as a key regulatory nexus, modulating anabolic processes versus catabolic processes in response to nutrients, growth cues and cellular energy status.

mTOR and known longevity factors

The first indication that mTOR regulates ageing came from studies in *S. cerevisiae*, in which it was found that deletion of the gene encoding the yeast orthologue of S6K — *SCH9* — resulted in a doubling of yeast chronological lifespan (defined as the duration of time that cells in stationary phase remain viable)[4]. Soon after, mTORC1 was shown to affect longevity with the finding that mutation or RNA interference (RNAi) knockdown of mTOR (*let-363*) or the mTORC1 component raptor (*daf-15*) could extend lifespan in the nematode *Caenorhabditis elegans*[5,6]. This was followed by studies in the fruitfly *Drosophila melanogaster* and using the yeast replicative ageing model, showing that mutations in mTOR and several other components of the mTORC1 pathway also increase lifespan in these systems[7,8]. In addition, a series of studies showed that rapamycin extended lifespan in yeast[9,10], nematodes[11], fruitflies[12] and mice[13–15], firmly establishing mTORC1 as a central, evolutionarily conserved regulator of longevity (Table 1).

Dietary restriction

Reduction in nutrient intake in the absence of malnutrition, or dietary restriction (also referred to as calorie restriction), extends lifespan in many different species[16]. In fact, other than mTORC1 inhibition, dietary restriction is currently the only intervention known to extend lifespan in yeast ageing models and in worms, flies and mice. mTORC1 is thought to play a part in mediating longevity and health benefits as a result of dietary restriction, which is intuitive given its function in responding to nutrient and growth cues. Dietary restriction reduces mTORC1 activity in invertebrate organisms and in some mammalian tissues, and pharmacological or genetic disruption of mTORC1 is sufficient to extend lifespan in both invertebrates and mice under non-dietary restriction conditions[2].

Genetic evidence that suggests mTORC1 acts downstream of dietary restriction comes mainly from epistasis experiments in which

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dietary restriction is combined with mutations in the mTORC1 pathway. In yeast, for example, dietary restriction fails to further extend replicative lifespan when combined with deletion of the genes encoding the mTOR and S6K homologues[8]. A similar relationship is observed when RNAi knockdown of mTOR is combined with dietary restriction in *C. elegans*[17]. In fruitflies, 4E-BP1 is required for maximal lifespan extension from at least one method of dietary restriction[18], and lifespan extension from dominant-negative alleles of mTOR and S6K extend lifespan in fruitflies in a nutrient-dependent manner[7]. However, the interaction between dietary restriction and mTORC1 signalling is complex, as evidenced in reports that RNAi knockdown of S6K and translation initiation factors yield an additive lifespan extension when combined with dietary restriction in *C. elegans*[17]. Interpretation of these types of experiments is complicated by the fact that several different dietary restriction protocols are used, and quantitative assessments of activity are often missing. Despite these challenges, numerous studies have linked mTORC1-regulated processes, including both reduced mRNA translation and induction of autophagy, to lifespan extension from dietary restriction in different organisms (discussed further later). Thus, there is broad consensus that reduced mTORC1 signalling probably contributes to longevity and health benefits as a result of dietary restriction[19].

Insulin/IGF-1-like signalling.

Reduced insulin/IGF-1-like signalling (IIS) increases longevity in nematodes, fruitflies and mice[16,19]. Lifespan extension from reduced IIS is mediated by the FOXO family of transcription factors, and several studies have associated polymorphisms in FOXO3 with longevity in humans[19]. mTOR activity is linked to IIS through multiple connections (Fig. 2). As already mentioned, mTORC1 is activated by IIS through AKT, and mTORC1 can negatively regulate IIS through S6K, which inhibits insulin receptor substrate 1 (IRS-1)[20]. Additional studies have indicated that FOXO3A can transcriptionally regulate tuberous sclerosis protein 1 (TSC1), an upstream inhibitor of mTORC1, in mammalian cells[21] and that the mTORC1 target 4E-BP1 is also a transcriptional target of FOXO in fruitflies[22]. mTOR and IIS also interact through mTORC2, which activates AKT to repress FOXO1 and FOXO3 in mammalian cells[23].

Genetic studies that examine the relationship between mTORC1 and IIS with respect to longevity reflect this complexity. In nematodes, the FOXO orthologue *daf-16* is proposed to repress raptor expression, and lifespan extension from mutation of raptor requires DAF-16 — a defining characteristic of components of the IIS pathway[5]. Paradoxically, mutation of mTOR itself[6] or treatment with rapamycin[11] does not require DAF-16 for lifespan extension. Similarly, lifespan extension that results from mutations or RNAi knockdown of mTOR-pathway downstream targets such as S6K and the translation initiation factors eIF4E and eIF4G does not require DAF-16 (refs 17, 24). Thus, current evidence supports the idea that mTORC1 modulates ageing by mechanisms that overlap but

BOX 1

Key events for rapamycins in ageing

- **1970** Soil samples containing rapamycin-producing *Streptomyces hygroscopicus* taken from the Polynesian island of Rapa Nui
- **1975** Purification of rapamycin and identification of fungicidal activity
- **1977** Immunosuppressive activity discovered
- **1984** Antitumour activity discovered
- **1991** *TOR1* and *TOR2* genes identified in yeast
- **1994** mTOR gene *FRAP1*, now *MTOR*, identified in mammals
- **1995** Mechanism of action of rapamycin discovered
- **1999** Approved by US Food and Drug Administration (FDA) for use in preventing host-rejection in patients undergoing kidney transplantation
- **2003** Approved by FDA for use in drug-eluting stents
- **2006** Shown to extend lifespan in budding yeast
- **2007** Approved by FDA for treatment of renal-cell carcinoma
- **2008** Approved by FDA for treatment of mantle cell lymphoma
- **2009** Shown to extend lifespan in mice
- **2010** Shown to extend lifespan in fruitflies
- **2010** Approved by FDA for treatment of tuberous sclerosis
- **2011** Shown to improve outcome in mouse models of Alzheimer's disease
- **2011** Approved by FDA for treatment of pancreatic cancer
- **2012** Shown to extend lifespan in nematodes
- **2012** More than 1,300 clinical trials under way or completed

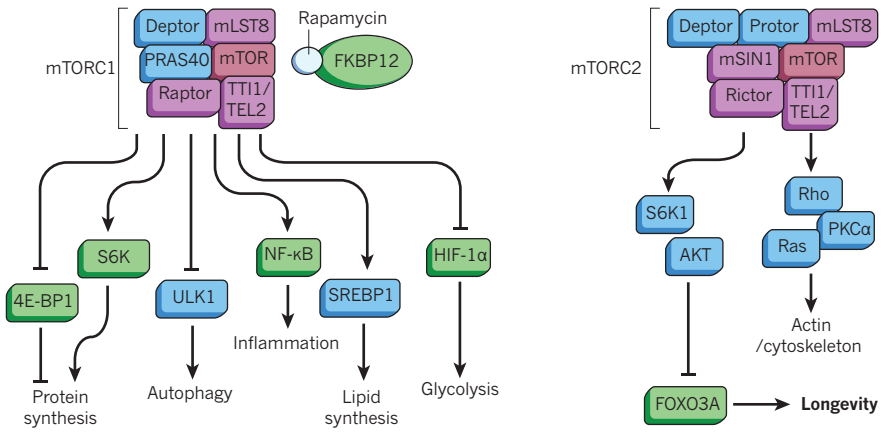

Figure 1 | The two mTOR complexes have distinct constituent proteins and regulate different downstream processes. Here (figure represents data from studies in mice) mTORC1 comprises deptor, PRAS40, raptor, mLST8, mTOR and TTI1–TEL2. mTORC2 is comprised of deptor, mLST8, protor, rictor, mSIN1, mTOR and TTI1–TEL2. Rapamycin binds to FKBP12 and inhibits mTORC1 by disrupting the interaction between mTOR and raptor. Regulation of lipid synthesis by mTORC1 is thought to occur mainly through sterol-regulatory-element-binding protein transcription factors (shown here as SREBP1) by a mechanism that is not completely understood. mTORC1 negatively regulates autophagy through multiple inputs, including inhibitory phosphorylation of ULK1, preventing formation of the ULK1–ATG13–FIP200 complex (which is required for initiation of autophagy). mTORC1 promotes protein synthesis through activation of the translation initiation promoter S6K and through inhibition of the inhibitory mRNA cap binding 4E-BP1, and regulates glycolysis through HIF-1α. mTORC2 inhibits FOXO3a through S6K1 and AKT, which can lead to increased longevity. The complex also regulates actin cytoskeleton assembly through protein kinase C α (PKCα), Rho GTPases and Ras proteins.

Table 1 | Comparison of species in which genetic or pharmacological inhibition of an mTORC1-pathway component extends lifespan

	*Saccharomyces cerevisiae**	*Caenorhabditis elegans*	*Drosophila melanogaster*	*Mus musculus*
Rapamycin	Yes[9,10]	Yes[11]	Yes[12]	Yes[13–15]
mTOR gene mutation and knockdown	Yes[8,9]	Yes[6]	Yes[7]	Yes[3]
Raptor gene mutation and knockdown	Not reported	Yes[5]	Not reported	Not reported
Tsc1 and *Tsc2* activation	Not applicable†	Not applicable	Yes[7]	Not reported
S6K gene mutation and knockdown	Yes[4,8]	Yes[24]	Yes[7]	Yes[37]
4E-BP activation	Not applicable	Not applicable	Yes[18]	Not reported
Translation initiation factor mutation and knockdown	Yes[44]	Yes[17,24,74,75]	Not reported	Not reported
Ribosomal protein mutation and knockdown	Yes[44,47,76]	Yes[17,75]	Not reported	Not reported

*Includes both replicative and chronological lifespan; †Not applicable is used in cases for which homologues have not yet been identified.

are distinct from IIS. This is consistent with the model that mTORC1 acts mainly downstream of dietary restriction, which, based on similar epistasis experiments, also seems to increase lifespan by mechanisms that are overlapping but distinct from IIS.

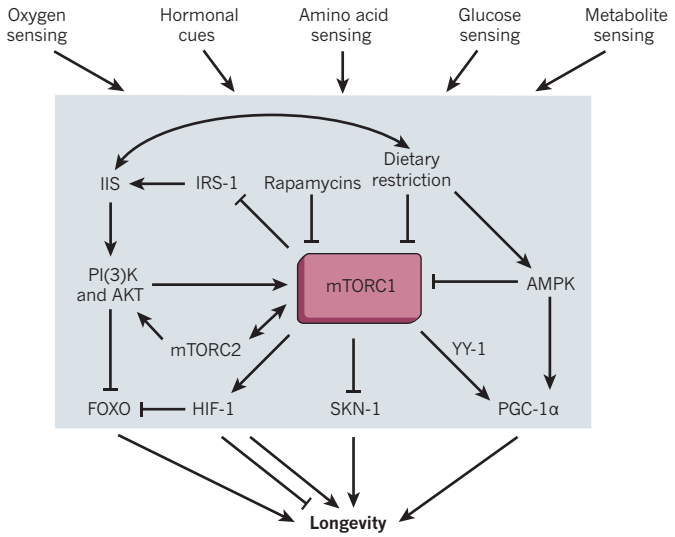

Figure 2 | Interactions between mTOR and other longevity pathways. mTORC1 responds to a variety of environmental cues and communicates with several known longevity factors in a complex network of interactions. mTORC1 activity limits longevity, and several pathways related to mTORC1 can result in the extension of lifespan. Rapamycins inhibit mTORC1 and decrease its activity. mTORC1 can be activated by environmental nutrients such as amino acids. Hormonal cues can stimulate IIS, which can increase longevity through PI(3)K and AKT mediation of the FOXO family of transcription factors. mTORC1 can also negatively regulate IIS through inhibition of IRS-1. Sensing of low oxygen levels stimulates mTORC1 to activate the hypoxic response by enhancing translation of HIF-1, which inhibits FOXO family members and increases longevity. HIF-1 is also thought to extend longevity at high and low temperatures, or inhibit it at low temperatures. Inhibition of mTORC1 signalling has been linked to stress resistance, and inhibition of the stress response transcription factor SKN-1 (an orthologue of Nrf2) by mTORC1 has been implicated in lifespan extension. Glucose and metabolite sensing as a result of dietary restriction can lower mTORC1 signalling partly through activation of AMPK, which can inhibit mTORC1 and promote longevity. The complex actively promotes mitochondrial biogenesis and metabolism through PGC-1α and YY-1. Inhibition of this results in greater mitochondrial respiration, leading to increased longevity. Dietary restriction and IIS can also influence each other and their related pathways. The relationship between mTORC2 and mTORC1 is still unclear. The interactions shown have been described in studies of yeast, nematodes, fruitflies and mice.

AMP-activated protein kinase

AMPK is a conserved sensor of energy status that is activated in response to low ATP levels and negatively regulates mTORC1 (Fig. 2). Overexpression of AMPK is sufficient to extend lifespan in *C. elegans*, and the AMPK activator metformin (a widely used antidiabetic drug) is reported to extend lifespan in *C. elegans* and in short-lived, cancer-prone strains of mice[25–27]. AMPK inhibits mTORC1 through at least two distinct mechanisms: AMPK phosphorylates TSC2 on conserved serine sites, resulting in activation of TSC2 and downregulation of mTORC1 activity[28], and AMPK directly phosphorylates raptor to impair mTORC1 signalling[29]. These data suggest that dietary restriction could reduce mTORC1 signalling partly through activation of AMPK. The actual situation is likely to be more complex, however, as both AMPK and mTOR interact with multiple additional factors, and it has been reported that activation of AMPK extends the lifespan of *C. elegans* through the CREB-regulated transcriptional coactivator 1 (CRTC-1) rather than through mTORC1 (ref. 30). Importantly, so far, there is no direct evidence for regulation of mTORC1 by AMPK in *C. elegans*, which does not have a TSC2 orthologue.

The hypoxic response

HIF-1α is a highly conserved transcription factor that is activated in response to low oxygen availability (hypoxia) and regulates the expression of a suite of genes involved in maintaining cellular homeostasis under these conditions[31]. Under normoxic conditions, HIF-1α is targeted for proteasomal degradation, and ectopic stabilization of HIF-1α is a hallmark of many cancers. A major feature of the hypoxic response is to drive ATP production under oxygen-limiting conditions by promoting a metabolic shift from oxidative phosphorylation towards glycolysis and lactic acid fermentation. mTORC1 activates the early hypoxic response in mammals by enhancing translation and stabilization of HIF-1 itself, as well as by enhancing translation of mRNAs encoding a subset of HIF-1 target genes that include vascular endothelial growth factor (VEGF)[32]. Prolonged hypoxia, however, results in downregulation of mTORC1.

The hypoxic response has been implicated in ageing by studies in *C. elegans* in which HIF-1 acts as both a positive and negative regulator of longevity[31]. Stabilization of HIF-1 results in lifespan extension by a mechanism that is distinct from both IIS and dietary restriction[33,34]. Interestingly, deletion of *hif-1* also extends lifespan of animals grown at a high temperature (25 °C), but not at a low temperature (15 °C)[35]. In this context, HIF-1 has been proposed to act downstream of dietary restriction and mTORC1 (ref. 36) (Fig. 2). However, direct evidence for regulation of HIF-1 by mTORC1 in *C. elegans* has yet to be obtained.

mTOR in mammalian ageing and healthspan

The first indication that mTOR modulates mammalian ageing came from longevity studies as part of the National Institute on Aging's Interventions Testing Program (ITP). Rapamycin was found to significantly

extend lifespan in a genetically heterogeneous strain background at three independent test locations[13]. A particularly interesting aspect of this study was that the animals did not begin receiving the drug until they had reached 600 days of age, roughly equivalent to 60 years of age in a person. Follow-up studies that began rapamycin treatment at an earlier age replicated the initial result but failed to show substantially larger effects on longevity[14]. In mice treated with rapamycin from 6 months of age, mean lifespan extension was about 18% in females, but only 10% in males[14]. Both higher and lower doses of rapamycin are currently being tested, and it will be of particular interest to see whether a greater longevity benefit can be achieved. Soon after the ITP rapamycin study was published, knockout of the mouse gene encoding S6K1 (*Rps6kb1*) was reported to increase lifespan in females[37]. In this case, the lifespan extension was sex-specific, with males receiving no longevity benefit.

A key prediction of the hypothesis that ageing is caused by specific molecular changes is that it should be possible to slow the ageing process and, thereby, delay the onset and progression of multiple age-related diseases (Fig. 3). Thus, slowing ageing should increase both lifespan and healthspan — the period of life spent in relatively good health, free from chronic disease or disability. This seems to be the case for dietary restriction, which not only extends lifespan but also delays the incidence of age-related decline and disease in rodents, including cancer, cognitive decline and neurodegeneration[16]. Similarly, dietary restriction reduces the incidence of cancer, cardiovascular disease, brain atrophy and diabetes in rhesus monkeys[38]. Emerging evidence suggests that, like dietary restriction, mTORC1 inhibition may have similar positive effects on multiple age-related pathologies in rodents and, in some cases, humans (Box 2).

From a public-health perspective, an intervention that results in compression of morbidity, in which most of a lifetime's illness is compressed into a shorter period of time near the end of life[39], is particularly desirable. Although it is too early to know whether rapamycins could represent such an intervention, end-of-life analysis of the long-lived rapamycin-treated mice in the ITP studies indicates that the spectrum of causes of death was not significantly altered[14]. However, age-related alterations in heart, liver, adrenal glands, endometrium and tendon, as well as the decline in spontaneous activity, all occur more slowly in rapamycin-treated mice[40]. Another study found that, in addition to lifespan extension, spontaneous tumorigenesis and deaths owing to cancer in the inbred C57BL/6 mouse strain were significantly reduced by rapamycin[15]. These data suggest that rapamycin slows the ageing process in mice such that many normal causes of morbidity and death are delayed. The evidence that rapamycin can protect against multiple age-related diseases (Box 2) — including reduced cognitive function[41,42] — is consistent with a compression of morbidity, although further studies will be needed to definitively test this. As previously alluded to, there has not yet been a careful analysis of different rapamycin regimens, and the observed effects on longevity and healthspan may be quite different at higher or lower doses of the drug.

Mechanisms of longevity regulation by mTOR

In recent years, much effort has been applied to defining the mechanisms by which inhibition of mTORC1 enhances longevity. Not surprisingly, the picture that has emerged is complex. Multiple mTORC1-regulated processes seem to contribute to the pro-longevity effects of mTORC1 inhibition in a coordinated and overlapping manner.

mRNA translation

Among the most crucial of mTORC1 functions is the promotion of mRNA translation and protein synthesis under conditions that are favourable for growth, and there is evidence that regulation of mRNA translation can modulate longevity in yeast, nematodes, fruitflies and mice[43]. For example, mutations in S6K have been found to extend lifespan in all four of these model organisms (Table 1). Likewise, mutation or knockdown of multiple translation initiation factors and ribosomal proteins has also been shown to extend lifespan in yeast and nematodes[44], and overexpression of an activated allele of 4E-BP extends lifespan in flies[18].

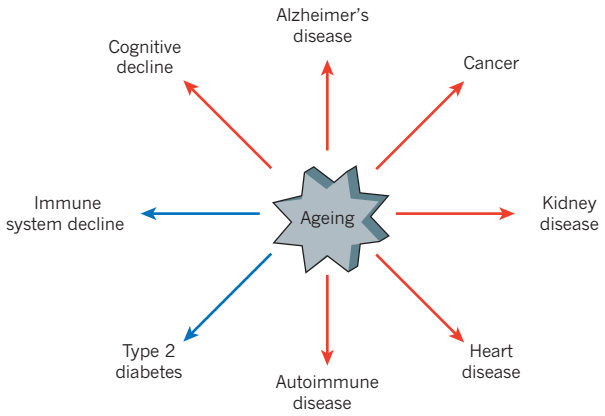

Figure 3 | The impact of mTORC1 on diseases of ageing. Ageing drives the onset and progression of multiple disorders that are modulated by mTORC1 signalling. Data from animal and human studies indicate that some disorders (red arrows) are improved by rapamycins. However, for others (blue arrows), although influenced by rapamycins, evidence suggests there are both beneficial and detrimental consequences of mTORC1 inhibition.

It has been proposed that a global reduction in mRNA translation could be beneficial during ageing by allowing endogenous protein repair and degradation machinery to better maintain protein homeostasis in the face of protein aggregation and oxidative damage[45]. Alternatively, a general reduction in mRNA translation could attenuate age-associated pathologies that result from 'hyperfunctional' overactive biosynthetic and proliferative processes that are important during development but detrimental during adulthood[46]. Instead of a global reduction in mRNA translation, however, molecular evidence suggests that differential translation of specific mRNAs is more important for effects on lifespan. The evidence for this came from yeast, in which lifespan extension in ribosomal protein mutants is associated with increased translation of the transcriptional activator Gcn4, which regulates the expression of many target genes, including several that are involved in enhanced stress resistance and response to amino acid starvation[47]. This regulation of Gcn4 is mediated by structural features of the 5′ region of the *GCN4* mRNA that cause it to be preferentially translated when mTORC1 is inhibited. Mechanistically similar observations have been made in both nematodes and fruitflies, in which a subset of mRNAs are translated more efficiently when global mRNA translation is reduced through inhibition of mTORC1-pathway components. In worms, these differentially translated mRNAs are enriched for stress response genes, and in flies they are enriched for mRNAs encoding components of the mitochondrial electron transport chain[18,48]. Although the specific mRNAs seem to differ across species, a common theme of enhanced translation efficiency of metabolic and stress genes in response to mTORC1 inhibition has emerged.

The mechanism by which mTORC1 regulates mRNA translation in mammals continues to be an active area of research. Although *S6k1* knockout mice are long-lived and have a small body size[37], there is no direct evidence that these animals have reduced rates of global mRNA translation. In both yeast and worms, a deficiency of S6K causes a substantial reduction in global mRNA translation and protein synthesis[24,47], so it would be somewhat surprising if mice that lack S6K1 do not. However, one recent study has questioned whether S6K1 is a major component of mTORC1 regulation of mRNA translation in mammalian cells[49]. In experiments using the mTOR catalytic inhibitor Torin 1, acute control of translation by mTORC1 in $p53^{-/-}$ mouse embryonic fibroblasts was found to be mediated mainly by 4E-BPs, and was largely limited to mRNAs with 5′ terminal oligopyrimidine (TOP) motifs[49]. This study also found no evidence that complexity of the 5′ untranslated region is a determinant of mTORC1-dependent differential mRNA translation[49],

BOX 2

Effects of mTORC1 inhibition on age-related disease

Inhibition of mTORC1 has positive, and sometimes negative, effects on the age-related pathologies in model organisms and, in some cases, humans.

- **Neurodegenerative diseases** Protection against dopaminergic neuron death in fruitfly and mouse models of Parkinson's disease[77,78], delayed disease progression in mouse models of Alzheimer's disease[79,80] and frontotemporal lobar dementia[81], and improved spatial learning and memory in old-aged mice[41,42].
- **Cancer** Reduced growth of solid-tumour cell lines, but disappointing efficacy in clinical trials, with the exception of renal-cell carcinoma, breast cancer and several rare forms of cancer[82].
- **Heart disease** Widespread use to prevent restenosis following angioplasty. Reduced hypertrophy[83] and regression of failure[84] in mouse aortic-constriction model. Reduced cardiomyopathy in models of the zebrafish *Danio rerio*[85] and in mice[86].
- **Metabolic disease and obesity** Resistance to diet-induced[60,87] and age-associated[88] obesity in mice. However, inhibition is also associated with glucose intolerance and insulin resistance in mice[3], and dyslipidaemia in humans.
- **Immune function** Rapamycins are used in combined immunosuppression therapies in humans. Rapamycin pre-treatment enhances immune function in tuberculosis[89], influenza[68] and antitumour vaccine responses in mice[90] and in vaccinia vaccine responses in non-human primates[91].
- **Kidney disease** Decreased rejection and nephrotoxicity in allograft and renal-cancer therapies in patients[92]. Improved outcome in animal models of polycystic kidney disease and diabetic nephropathy.
- **Other health effects** Beneficial effects in animal models of age-associated retinopathy[93], and in a variety of disorders that are not obviously age-related, including cerebral ischaemia and reperfusion injury[94], depression[41], autism spectrum disorder[95], muscular dystrophy[86], epilepsy[96], food allergies[97] and autoimmune disorders[98]. Decreased need for anti-VEGF intravitreal injections in humans with age-related macular degeneration[99]. Reduced toxicity and enhanced clearance of progerin in cells from patients with Hutchinson–Gilford progeria syndrome[100]. Negative effects include testicular degeneration and cataracts in mice[58], oral mucositis and stomatitis in humans, and impaired wound healing in humans and animal models.

which is in contrast with the yeast and invertebrate models already described. It may be that the precise mechanisms by which mTORC1 regulates mRNA translation, and the impact of this regulation on longevity, is different in mammals and invertebrate species. Alternatively, these differences could arise from effects of the catalytic inhibition of mTOR, as opposed to genetic inhibition or inhibition with rapamycin, or from differences in mTOR function in cells grown between culture and *in vivo*.

Autophagy

Activation of autophagy is another key mTORC1-regulated process that probably has a central role in promoting longevity. Autophagy is a major degradation pathway in eukaryotic cells that is essential for removing damaged organelles and macromolecules from the cytoplasm and recycling amino acids during periods of starvation[50]. Evidence suggests that autophagic degradation declines with age, and it has been proposed that this leads to an accumulation of damage, such as protein aggregates and degenerate mitochondria, that contribute to age-related cellular dysfunction[51]. Activation of autophagy by inhibition of mTORC1 presumably maintains cellular function during ageing by allowing enhanced degradation of aged cellular components. However, this hypothesis has been difficult to test directly, partly because the tools for quantifying autophagic flux are inadequate and rely mainly on secondary assays — such as the abundance of proteins involved in various steps of autophagy. Evidence from studies in yeast and invertebrates supports the model that mTORC1-mediated induction of autophagy is required for lifespan extension from dietary restriction or from rapamycin[12,52,53], although it remains to be determined whether induction of autophagy is sufficient to promote longevity in the absence of mTORC1 inhibition. In addition to longevity, aberrant regulation of autophagy has been linked to several diseases of ageing, including cancer, diabetes, cardiovascular disease and neurodegenerative diseases[50], and it seems likely that enhanced autophagy underlies many of the beneficial effects of mTORC1 inhibition in these disease models.

Stress resistance and xenobiotic metabolism

Inhibition of mTORC1 signalling has been linked most clearly to enhanced stress resistance in yeast and nematodes. In yeast, for example, reduced activity of Tor1 and Sch9 activates the protein kinase Rim15, which in turn activates the stress response transcription factors Msn2, Msn4 and Gis1 (ref. 54). Induction of these stress response pathways is required for yeast chronological lifespan extension from dietary restriction, or in response to reduced mTORC1 signalling through deletion of *SCH9* (ref. 54). In nematodes, treatment with rapamycin has been shown to activate the stress response transcription factor SKN-1, which is encoded by the orthologue of mammalian *NRF2* (ref. 11). SKN-1 is required for lifespan extension from rapamycin[11], and has also been implicated in lifespan extension from dietary restriction[55]. Interestingly, NRF2 is activated in mice treated with rapamycin and in those subjected to dietary restriction[56]. In both cases, NRF2 activation results in the induction of several enzymes involved in xenobiotic defence, suggesting that enhanced resistance to such environmental insults may be important for lifespan extension.

Mitochondrial function

Regulation of mitochondrial function by mTORC1 is complex and seems to involve multiple mechanisms. As mentioned previously, mTORC1 promotes activation of HIF-1, which in turn enhances glycolytic flux while simultaneously downregulating mitochondrial oxygen consumption[32]. Another report suggests that mTORC1 actively promotes mitochondrial biogenesis and metabolism through PGC-1α and the transcription factor YY-1 (ref. 57). The best evidence for a direct mitochondrial role in longevity downstream of mTOR signalling comes from yeast, in which inhibition of mTORC1 results in a metabolic shift towards greater mitochondrial respiration, thereby increasing chronological lifespan[58]. This effect has been proposed to involve an adaptive signalling response owing to elevated levels of mitochondrial superoxide[59]. Mice that lack mTORC1 activity in adipose tissue also show enhanced mitochondrial respiration[60], suggesting a similar relationship between mTORC1 and mitochondrial function in mammals, at least in some tissues.

Inflammation

Inflammation is associated with several age-related disorders[61], and a reduction in inflammation is proposed to be a primary mechanism by which dietary restriction promotes longevity and healthspan[62]. Hyperactivation of mTOR is often associated with inflammation, and rapamycin has been shown to have anti-inflammatory effects in multiple

settings, including chronic kidney disease[63], vascular inflammation after angioplasty[64], atherosclerotic plaques[65] and lung infection[66]. Thus, a reduction in chronic, age-associated inflammation is another attractive mechanism by which mTORC1 inhibition could slow multiple age-related pathologies in mammals.

Stem cells

In mammals, a decline in stem-cell function is likely to be an important cause of age-related pathology[67]. There is increasing evidence that mTORC1 has a central role in this process, and that inhibition of mTORC1 can preserve, and perhaps even rejuvenate, stem-cell function in a variety of tissues. For example, rapamycin protects old mice from an immune challenge with influenza virus, which has been attributed to a rejuvenation of haematopoietic stem-cell function in the treated animals[68]. Furthermore, treating old mice with rapamycin has been found to enhance intestinal stem-cell function[69]. Interestingly, the mechanism accounting for this improvement is linked to inhibition of mTORC1 in the surrounding Paneth cells, resulting in a more favourable niche, rather than a direct effect of mTORC1 inhibition in the intestinal stem cells. In another study, dietary restriction was shown to enhance the function of muscle stem cells in both young and old animals through both cell-intrinsic and cell-extrinsic mechanisms[70]. Although the authors did not directly examine a role for mTORC1, these effects have been proposed to be partly mediated by reduced mTORC1 activity[71].

Future directions and perspectives

mTOR is a key modulator of ageing in evolutionarily divergent organisms, ranging from yeast to rodents, and it is likely that this function has been conserved to some extent in humans. The complexity of the mTOR network presents a hurdle in defining the mechanistic details of how mTOR influences longevity and healthspan. Several mTOR-regulated processes are likely to contribute to the effects of mTORC1 inhibition on ageing and disease, and it will be challenging to untangle the context-specific and tissue-specific relationships. Nonetheless, impressive progress has been made in this area over the past few years, and we anticipate that this trend will continue.

As the list of beneficial effects of rapamycin in invertebrate and mouse models continues to grow longer (Box 2), it becomes increasingly tempting to speculate on similar benefits in people. As mentioned previously, rapamycins have been approved clinically for a variety of uses but have yet to be tested against a broad spectrum of age-related diseases. At the time of writing, the search term 'rapamycin' generates 1,343 results in the National Institutes of Health clinical trials database (http://clinicaltrials.gov). We therefore anticipate a bounty of additional data on the effects of rapamycin in humans over the next few years.

All of this then raises the question, at what point should we consider giving these drugs to otherwise healthy people? Perhaps the greatest upside of mTORC1 inhibitors is their potential to delay cognitive decline during ageing. The evidence for improved cognitive function in old mice treated with rapamycin is striking[41]. Current estimates suggest cognitive decline can be detected as early as 45 years of age in otherwise healthy people[72]. Loss of cognitive function is a leading concern among geriatricians and their elderly patients and is a significant and growing public-health burden. If mTORC1 inhibition has even a modest positive effect on cognitive function, it could improve the quality of life for millions of middle-aged and older adults. Add to this the likelihood that risks of developing some forms of cancer, cardiovascular disease, and neurodegenerative disease would be reduced, then mTOR inhibitors may offer an attractive opportunity to have a significant impact on preventive health care.

Before this can happen, however, important questions must be answered. Rapamycin is not without side effects, including hyperlipidaemia and hyperglycaemia, anaemia and stomatitis, in patients. A recent study of long-term rapamycin treatment in mice reported increased incidence of cataracts and testicular degeneration[40]. The effects of mTORC1 inhibition on immune function and wound healing are also of particular concern. Although it is unclear whether rapamycin alone has substantial immunosuppressive effects in healthy individuals, it undoubtedly is immunomodulatory. It would be unfortunate to take a drug that slows the rate of ageing, only to succumb to infection from an otherwise innocuous bacterium or virus at an early age. Optimal dosage and duration of treatment are also unknown factors. Almost no information exists on the most effective dose of rapamycin for longevity or healthspan in mice, and the current data suggest that rapamycin therapy that begins late in life is nearly as effective as therapy that begins early in life, at least for longevity. Understanding whether this is also true for a variety of age-related pathologies will be important. Assessing risks and side effects from studies of patients with disorders such as the rare lung disease lymphangioleiomyomatosis, for which rapamycin is indicated for chronic use, may be informative in determining the clinical potential of this drug and other mTOR inhibitors in the setting of age-related disease.

The development of chemical inhibitors of mTOR, as well as drugs that target other components of the mTOR pathway, promises to greatly aid research, while also potentially providing drugs of therapeutic value. Until recently, pharmacological inhibition of mTOR has been largely limited to rapamycins, but newer ATP-competitive inhibitors of mTOR kinase activity have been developed that block the phosphorylation of all known substrates of both mTORC1 and mTORC2 (ref. 73). These drugs show early promise as anticancer therapies, but as yet there are no data regarding their effects on longevity and other age-related diseases. S6K inhibitors, 4E-BP1 activators, mTORC1-specific inhibitors and mTOR-independent activators of autophagy are also attractive approaches for potentially modulating longevity and healthspan without incurring the side effects that are associated with rapamycins.

The search for a way to delay human ageing has proven long and elusive. Although still far from certain, there is reason to be optimistic that mTOR inhibitors may accomplish this goal. mTORC1 inhibition slows ageing in yeast and invertebrates, extends lifespan in mice, and has an impact on a diverse array of age-related diseases. Are we finally on the threshold of being able to fundamentally alter human ageing? Only time will tell, but if the pace and direction of recent progress are any indication, the next few chapters in the mTOR story should prove very interesting indeed. ■

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1. Laplante, M. & Sabatini, D. M. mTOR signaling in growth control and disease. *Cell* **149,** 274–293 (2012).
2. Stanfel, M. N., Shamieh, L. S., Kaeberlein, M. & Kennedy, B. K. The TOR pathway comes of age. *Biochim. Biophys. Acta* **1790,** 1067–1074 (2009).
3. Lamming, D. W. *et al.* Rapamycin-induced insulin resistance is mediated by mTORC2 loss and uncoupled from longevity. *Science* **335,** 1638–1643 (2012). **This study uses genetic models to confirm that reduced mTORC1 activity extends lifespan in mammals, and reports that the insulin resistance associated with chronic mTORC1 inhibition is caused by altered mTORC2 signalling and is not causally involved in lifespan extension.**
4. Fabrizio, P., Pozza, F., Pletcher, S. D., Gendron, C. M. & Longo, V. D. Regulation of longevity and stress resistance by Sch9 in yeast. *Science* **292,** 288–290 (2001).
5. Jia, K., Chen, D. & Riddle, D. L. The TOR pathway interacts with the insulin signaling pathway to regulate *C. elegans* larval development, metabolism and life span. *Development* **131,** 3897–3906 (2004).
6. Vellai, T. *et al.* Influence of TOR kinase on lifespan in *C. elegans*. *Nature* **426,** 620 (2003).
7. Kapahi, P. *et al.* Regulation of lifespan in *Drosophila* by modulation of genes in the TOR signaling pathway. *Curr. Biol.* **14,** 885–890 (2004).
8. Kaeberlein, M. *et al.* Regulation of yeast replicative life span by TOR and Sch9 in response to nutrients. *Science* **310,** 1193–1196 (2005).
9. Powers, R. W. III, Kaeberlein, M., Caldwell, S. D., Kennedy, B. K. & Fields, S. Extension of chronological life span in yeast by decreased TOR pathway signaling. *Genes Dev.* **20,** 174–184 (2006).
10. Medvedik, O., Lamming, D. W., Kim, K. D. & Sinclair, D. A. MSN2 and MSN4 link calorie restriction and TOR to sirtuin-mediated lifespan extension in *Saccharomyces cerevisiae*. *PLoS Biol.* **5,** e261 (2007).
11. Robida-Stubbs, S. *et al.* TOR signaling and rapamycin influence longevity by regulating SKN-1/Nrf and DAF-16/FoxO. *Cell Metab.* **15,** 713–724 (2012).
12. Bjedov, I. *et al.* Mechanisms of life span extension by rapamycin in the fruit fly *Drosophila melanogaster*. *Cell Metab.* **11,** 35–46 (2010).
13. Harrison, D. E. *et al.* Rapamycin fed late in life extends lifespan in genetically heterogeneous mice. *Nature* **460,** 392–395 (2009). **This study reports lifespan extension from mTORC1 inhibition in a mammal.**

14. Miller, R. A. *et al.* Rapamycin, but not resveratrol or simvastatin, extends life span of genetically heterogeneous mice. *J. Gerontol. A Biol. Sci. Med. Sci.* **66,** 191–201 (2011).
15. Anisimov, V. N. *et al.* Rapamycin increases lifespan and inhibits spontaneous tumorigenesis in inbred female mice. *Cell Cycle* **10,** 4230–4236 (2011).
16. Fontana, L., Partridge, L. & Longo, V. D. Extending healthy lifespan — from yeast to humans. *Science* **328,** 321–326 (2010).
17. Hansen, M. *et al.* Lifespan extension by conditions that inhibit translation in *Caenorhabditis elegans*. *Aging Cell* **6,** 95–110 (2007).
18. Zid, B. M. *et al.* 4E-BP extends lifespan upon dietary restriction by enhancing mitochondrial activity in *Drosophila*. *Cell* **139,** 149–160 (2009).
19. Kenyon, C. J. The genetics of ageing. *Nature* **464,** 504–512 (2010).
20. Takano, A. *et al.* Mammalian target of rapamycin pathway regulates insulin signaling via subcellular redistribution of insulin receptor substrate 1 and integrates nutritional signals and metabolic signals of insulin. *Mol. Cell. Biol.* **21,** 5050–5062 (2001).
21. Khatri, S., Yepiskoposyan, H., Gallo, C. A., Tandon, P. & Plas, D. R. FOXO3a regulates glycolysis via transcriptional control of tumor suppressor TSC1. *J. Biol. Chem.* **285,** 15960–15965 (2010).
22. Tettweiler, G., Miron, M., Jenkins, M., Sonenberg, N. & Lasko, P. F. Starvation and oxidative stress resistance in *Drosophila* are mediated through the eIF4E-binding protein, d4E-BP. *Genes Dev.* **19,** 1840–1843 (2005).
23. Guertin, D. A. *et al.* Ablation in mice of the mTORC components raptor, rictor, or mLST8 reveals that mTORC2 is required for signaling to Akt-FOXO and PKCα, but not S6K1. *Dev. Cell* **11,** 859–871 (2006).
24. Pan, K. Z. *et al.* Inhibition of mRNA translation extends lifespan in *Caenorhabditis elegans*. *Aging Cell* **6,** 111–119 (2007).
25. Apfeld, J., O'Connor, G., McDonagh, T., DiStefano, P. S. & Curtis, R. The AMP-activated protein kinase AAK-2 links energy levels and insulin-like signals to lifespan in *C. elegans*. *Genes Dev.* **18,** 3004–3009 (2004).
26. Onken, B. & Driscoll, M. Metformin induces a dietary restriction-like state and the oxidative stress response to extend *C. elegans* healthspan via AMPK, LKB1, and SKN-1. *PLoS ONE* **5,** e8758 (2010).
27. Anisimov, V. N. Metformin for aging and cancer prevention. *Aging (Albany NY)* **2,** 760–774 (2010).
28. Inoki, K., Zhu, T. & Guan, K. L. TSC2 mediates cellular energy response to control cell growth and survival. *Cell* **115,** 577–590 (2003).
29. Gwinn, D. M. *et al.* AMPK phosphorylation of raptor mediates a metabolic checkpoint. *Mol. Cell* **30,** 214–226 (2008).
30. Mair, W. *et al.* Lifespan extension induced by AMPK and calcineurin is mediated by CRTC-1 and CREB. *Nature* **470,** 404–408 (2011).
31. Leiser, S. F. & Kaeberlein, M. The hypoxia-inducible factor HIF-1 functions as both a positive and negative modulator of aging. *Biol. Chem.* **391,** 1131–1137 (2010).
32. Hudson, C. C. *et al.* Regulation of hypoxia-inducible factor 1α expression and function by the mammalian target of rapamycin. *Mol. Cell. Biol.* **22,** 7004–7014 (2002).
33. Mehta, R. *et al.* Proteasomal regulation of the hypoxic response modulates aging in *C. elegans*. *Science* **324,** 1196–1198 (2009).
34. Zhang, Y., Shao, Z., Zhai, Z., Shen, C. & Powell-Coffman, J. A. The HIF-1 hypoxia-inducible factor modulates lifespan in *C. elegans*. *PLoS ONE* **4,** e6348 (2009).
35. Leiser, S. F., Begun, A. & Kaeberlein, M. HIF-1 modulates longevity and healthspan in a temperature-dependent manner. *Aging Cell* **10,** 318–326 (2011).
36. Chen, D., Thomas, E. L. & Kapahi, P. HIF-1 modulates dietary restriction-mediated lifespan extension via IRE-1 in *Caenorhabditis elegans*. *PLoS Genet.* **5,** e1000486 (2009).
37. Selman, C. *et al.* Ribosomal protein S6 kinase 1 signaling regulates mammalian lifespan. *Science* **326,** 140–144 (2009).
This study shows that knockout of the mTORC1 substrate S6K1 extends lifespan in mice.
38. Colman, R. J. *et al.* Caloric restriction delays disease onset and mortality in rhesus monkeys. *Science* **325,** 201–204 (2009).
39. Fries, J. F. Aging, natural death, and the compression of morbidity. *N. Engl. J. Med.* **303,** 130–135 (1980).
40. Wilkinson, J. E. *et al.* Rapamycin slows aging in mice. *Aging Cell* **11,** 675–682 (2012).
41. Halloran, J. *et al.* Chronic inhibition of mammalian target of rapamycin by rapamycin modulates cognitive and non-cognitive components of behavior throughout lifespan in mice. *Neuroscience* **223,** 102–113 (2012).
42. Majumder, S. *et al.* Lifelong rapamycin administration ameliorates age-dependent cognitive deficits by reducing IL-1β and enhancing NMDA signaling. *Aging Cell* **11,** 326–335 (2012).
References 41 and 42 report that rapamycin improves cognitive function and protects against age-related cognitive decline in mice.
43. Kaeberlein, M. & Kennedy, B. K. Hot topics in aging research: protein translation and TOR signaling. *Aging Cell* **10,** 185–190 (2011).
44. Smith, E. D. *et al.* Quantitative evidence for conserved longevity pathways between divergent eukaryotic species. *Genome Res.* **18,** 564–570 (2008).
45. Kaeberlein, M. & Kennedy, B. K. Protein translation. *Aging Cell* **6,** 731–734 (2007).
46. Blagosklonny, M. V. Aging and immortality: quasi-programmed senescence and its pharmacologic inhibition. *Cell Cycle* **5,** 2087–2102 (2006).
47. Steffen, K. K. *et al.* Yeast lifespan extension by depletion of 60S ribosomal subunits is mediated by Gcn4. *Cell* **133,** 292–302 (2008).
48. Rogers, A. N. *et al.* Lifespan extension via eIF4G inhibition is mediated by post-transcriptional remodeling of stress response gene expression in *C. elegans*. *Cell Metab.* **14,** 55–66 (2011).
49. Thoreen, C. C. *et al.* A unifying model for mTORC1-mediated regulation of mRNA translation. *Nature* **485,** 109–113 (2012).
50. Mizushima, N., Levine, B., Cuervo, A. M. & Klionsky, D. J. Autophagy fights disease through cellular self-digestion. *Nature* **451,** 1069–1075 (2008).
51. Cuervo, A. M. Autophagy and aging: keeping that old broom working. *Trends Genet.* **24,** 604–612 (2008).
52. Hansen, M. *et al.* A role for autophagy in the extension of lifespan by dietary restriction in *C. elegans*. *PLoS Genet.* **4,** e24 (2008).
53. Alvers, A. L. *et al.* Autophagy is required for extension of yeast chronological life span by rapamycin. *Autophagy* **5,** 847–849 (2009).
54. Wei, M. *et al.* Life span extension by calorie restriction depends on Rim15 and transcription factors downstream of Ras/PKA, Tor, and Sch9. *PLoS Genet.* **4,** e13 (2008).
55. Bishop, N. A. & Guarente, L. Two neurons mediate diet-restriction-induced longevity in *C. elegans*. *Nature* **447,** 545–549 (2007).
56. Steinbaugh, M. J., Sun, L. Y., Bartke, A. & Miller, R. A. Activation of genes involved in xenobiotic metabolism is a shared signature of mouse models with extended lifespan. *Am. J. Physiol. Endocrinol. Metab.* **303,** E488–E495 (2012).
57. Cunningham, J. T. *et al.* mTOR controls mitochondrial oxidative function through a YY1–PGC-1α transcriptional complex. *Nature* **450,** 736–740 (2007).
58. Bonawitz, N. D., Chatenay-Lapointe, M., Pan, Y. & Shadel, G. S. Reduced TOR signaling extends chronological lifespan via increased respiration and upregulation of mitochondrial gene expression. *Cell Metab.* **5,** 265–277 (2007).
59. Pan, Y., Schroeder, E. A., Ocampo, A., Barrientos, A. & Shadel, G. S. Regulation of yeast chronological lifespan by TORC1 via adaptive mitochondrial ROS signaling. *Cell Metab.* **13,** 668–678 (2011).
60. Polak, P. *et al.* Adipose-specific knockout of raptor results in lean mice with enhanced mitochondrial respiration. *Cell Metab.* **8,** 399–410 (2008).
61. Chung, H. Y. *et al.* Molecular inflammation: underpinnings of aging and age-related diseases. *Ageing Res. Rev.* **8,** 18–30 (2009).
62. Morgan, T. E., Wong, A. M. & Finch, C. E. Anti-inflammatory mechanisms of dietary restriction in slowing aging processes. *Interdiscip. Top. Gerontol.* **35,** 83–97 (2007).
63. Liu, Y. Rapamycin and chronic kidney disease: beyond the inhibition of inflammation. *Kidney Int.* **69,** 1925–1927 (2006).
64. Nuhrenberg, T. G. *et al.* Rapamycin attenuates vascular wall inflammation and progenitor cell promoters after angioplasty. *FASEB J.* **19,** 246–248 (2005).
65. Chen, W. Q. *et al.* Oral rapamycin attenuates inflammation and enhances stability of atherosclerotic plaques in rabbits independent of serum lipid levels. *Br. J. Pharmacol.* **156,** 941–951 (2009).
66. Abdulrahman, B. A. *et al.* Autophagy stimulation by rapamycin suppresses lung inflammation and infection by *Burkholderia cenocepacia* in a model of cystic fibrosis. *Autophagy* **7,** 1359–1370 (2011).
67. Sharpless, N. E. & DePinho, R. A. How stem cells age and why this makes us grow old. *Nature Rev. Mol. Cell Biol.* **8,** 703–713 (2007).
68. Chen, C., Liu, Y. & Zheng, P. mTOR regulation and therapeutic rejuvenation of aging hematopoietic stem cells. *Sci. Signal.* **2,** ra75 (2009).
69. Yilmaz, O. H. *et al.* mTORC1 in the Paneth cell niche couples intestinal stem-cell function to calorie intake. *Nature* **486,** 490–495 (2012).
This study shows that rapamycin protects against loss of intestinal stem-cell function during ageing by preserving the stem-cell niche.
70. Cerletti, M., Jang, Y. C., Finley, L. W., Haigis, M. C. & Wagers, A. J. Short-term calorie restriction enhances skeletal muscle stem cell function. *Cell Stem Cell* **10,** 515–519 (2012).
71. Ramos, F. J. & Kaeberlein, M. A healthy diet for stem cells. *Nature* **486,** 477–478 (2012).
72. Singh-Manoux, A. *et al.* Timing of onset of cognitive decline: results from Whitehall II prospective cohort study. *Br. Med. J.* **344,** d7622 (2012).
73. Benjamin, D., Colombi, M., Moroni, C. & Hall, M. N. Rapamycin passes the torch: a new generation of mTOR inhibitors. *Nature Rev. Drug Discov.* **10,** 868–880 (2011).
74. Syntichaki, P., Troulinaki, K. & Tavernarakis, N. eIF4E function in somatic cells modulates ageing in *Caenorhabditis elegans*. *Nature* **445,** 922–926 (2007).
75. Curran, S. P. & Ruvkun, G. Lifespan regulation by evolutionarily conserved genes essential for viability. *PLoS Genet.* **3,** e56 (2007).
76. Chiocchetti, A. *et al.* Ribosomal proteins Rpl10 and Rps6 are potent regulators of yeast replicative life span. *Exp. Gerontol.* **42,** 275–286 (2007).
77. Malagelada, C., Jin, Z. H., Jackson-Lewis, V., Przedborski, S. & Greene, L. A. Rapamycin protects against neuron death in *in vitro* and *in vivo* models of Parkinson's disease. *J. Neurosci.* **30,** 1166–1175 (2010).
78. Tain, L. S. *et al.* Rapamycin activation of 4E-BP prevents parkinsonian dopaminergic neuron loss. *Nature Neurosci.* **12,** 1129–1135 (2009).
79. Majumder, S., Richardson, A., Strong, R. & Oddo, S. Inducing autophagy by rapamycin before, but not after, the formation of plaques and tangles ameliorates cognitive deficits. *PLoS ONE* **6,** e25416 (2011).
80. Spilman, P. *et al.* Inhibition of mTOR by rapamycin abolishes cognitive deficits and reduces amyloid-β levels in a mouse model of Alzheimer's disease. *PLoS ONE* **5,** e9979 (2010).
81. Wang, I. F. *et al.* Autophagy activators rescue and alleviate pathogenesis of a mouse model with proteinopathies of the TAR DNA-binding protein 43. *Proc. Natl Acad. Sci. USA* **109,** 15024–15029 (2012).
82. Garber, K. Targeting mTOR: something old, something new. *J. Natl Cancer Inst.* **101,** 288–290 (2009).
83. Shioi, T. *et al.* Rapamycin attenuates load-induced cardiac hypertrophy in mice. *Circulation* **107,** 1664–1670 (2003).
84. McMullen, J. R. *et al.* Inhibition of mTOR signaling with rapamycin regresses established cardiac hypertrophy induced by pressure overload. *Circulation* **109,** 3050–3055 (2004).

85. Ding, Y. *et al.* Haploinsufficiency of target of rapamycin attenuates cardiomyopathies in adult zebrafish. *Circ. Res.* **109,** 658–669 (2011).
86. Ramos, F. J. *et al.* Rapamycin reverses elevated mTORC1 signaling in lamin A/C-deficient mice, rescues cardiac and skeletal muscle function, and extends survival. *Sci. Transl. Med.* **4,** 144ra103 (2012).
87. Um, S. H. *et al.* Absence of S6K1 protects against age- and diet-induced obesity while enhancing insulin sensitivity. *Nature* **431,** 200–205 (2004).
88. Yang, S. B. *et al.* Rapamycin ameliorates age-dependent obesity associated with increased mTOR signaling in hypothalamic POMC neurons. *Neuron* **75,** 425–436 (2012).
89. Jagannath, C. & Bakhru, P. Rapamycin-induced enhancement of vaccine efficacy in mice. *Methods Mol. Biol.* **821,** 295–303 (2012).
90. Amiel, E. *et al.* Inhibition of mechanistic target of rapamycin promotes dendritic cell activation and enhances therapeutic autologous vaccination in mice. *J. Immunol.* **189,** 2151–2158 (2012).
91. Araki, K. *et al.* mTOR regulates memory CD8 T-cell differentiation. *Nature* **460,** 108–112 (2009).
92. Lieberthal, W. & Levine, J. S. Mammalian target of rapamycin and the kidney. II. Pathophysiology and therapeutic implications. *Am. J. Physiol. Renal Physiol.* **303,** F180–F191 (2012).
93. Kolosova, N. G. *et al.* Prevention of age-related macular degeneration-like retinopathy by rapamycin in rats. *Am. J. Pathol.* **181,** 472–477 (2012).
94. Yin, L., Ye, S., Chen, Z. & Zeng, Y. Rapamycin preconditioning attenuates transient focal cerebral ischemia/reperfusion injury in mice. *Int. J. Neurosci.* **122,** 748–756 (2012).
95. Tsai, P. T. *et al.* Autistic-like behaviour and cerebellar dysfunction in Purkinje cell *Tsc1* mutant mice. *Nature* **488,** 647–651(2012).
96. Ryther, R. C. & Wong, M. Mammalian target of rapamycin (mTOR) inhibition: potential for antiseizure, antiepileptogenic, and epileptostatic therapy. *Curr. Neurol. Neurosci. Rep.* **12,** 410–418 (2012).
97. Yamaki, K. & Yoshino, S. Preventive and therapeutic effects of rapamycin, a mammalian target of rapamycin inhibitor, on food allergy in mice. *Allergy* **67,** 1259–1270 (2012).
98. Pierdominici, M., Vacirca, D., Delunardo, F. & Ortona, E. mTOR signaling and metabolic regulation of T cells: new potential therapeutic targets in autoimmune diseases. *Curr. Pharm. Des.* **17,** 3888–3897 (2011).
99. Nussenblatt, R. B. *et al.* A randomized pilot study of systemic immunosuppression in the treatment of age-related macular degeneration with choroidal neovascularization. *Retina* **30,** 1579–1587 (2010).
100. Cao, K. *et al.* Rapamycin reverses cellular phenotypes and enhances mutant protein clearance in Hutchinson–Gilford progeria syndrome cells. *Sci. Transl. Med.* **3,** 89ra58 (2011).

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