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Review

Targeting mTOR signaling by polyphenols: A new therapeutic target for ageing

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ABSTRACT

Current ageing research is aimed not only at the promotion of longevity, but also at improving health span through the discovery and development of new therapeutic strategies by investigating molecular and cellular pathways involved in cellular senescence. Understanding the mechanism of action of polyphenolic compounds targeting mTOR (mechanistic target of rapamycin) and related pathways opens up new directions to revolutionize ways to slow down the onset and development of age-dependent degeneration. Herein, we will discuss the mechanisms by which polyphenols can delay the molecular pathogenesis of ageing via manipulation or more specifically inhibition of mTOR-signaling pathways. We will also discuss the implications of polyphenols in targeting mTOR and its related pathways on health life span extension and longevity.

Contents

1. Aging: a growing pandemic

The incidence of age-related degenerative diseases leading to morbidity and mortality has increased by more than 30 million individuals worldwide, and the number of individuals affected is expected to increase exponentially (Maiese 2014c). The increase in lifespan has been attributed to significant improvements in the current healthcare system. At present, the maximum life expectancy is 80 years for most individuals, and the number of individuals over the age of 65 years has doubled since the 1960s (Maiese 2014c). The elderly population in large developing countries such as India and China is expected to increase from 5% to 10% over the next several decades (Maiese 2015b). In line with an increase in the life span of the global population there has been a rise in non-communicable diseases (Gordon et al., 2014). The World Health Organization esti-

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mated that more than 60% of the 57 million global deaths are due to non-communicable diseases (Maiese 2015a). Of these disorders, the leading causes of death are cardiac disease, cancer, chronic lower respiratory disease, stroke and traumatic accidents (Minino and Murphy 2012). Cardiovascular disease in combination with hypercholesterolemia and hypertension can lead to neurodegenerative diseases including cognitive loss and stroke (Maiese 2014a).

There have been significant advances in the development of therapeutic agents for age-related disorders. For instance, owing to the success of preventative care, stroke is no longer the third leading cause of natural death which occurs due to cardiovascular disease. Reduction in smoking, greater control of hypertension and improved plasma lipid profiles, have most likely led to the lowered ranking for stroke (Pergola et al., 2014). Newer treatment strategies, such as recombinant tissue plasminogen activator have led to reduced mortality and morbidity in stroke patients (Chen et al., 2014). However, degenerative disorders continue to play a leading cause of death (Minino and Murphy 2012). Therefore, the development of treatments that can slow down or attenuate age-related degeneration is warranted.

2. Mechanisms of ageing

Ageing is a time-dependent deterioration of physiological functions of the cells that leads to an increased risk of cell death or the outbreak of diseases such as cancer (Zoncu et al., 2011). It is a natural phenomenon with a series of modifiable molecular events that confers disability of organisms to adapt in response to stress (Kyriakakis et al., 2015). The types of ageing processes include 'programmed ageing' and 'wear and tear ageing'. Programmed ageing is a specific controller program for entrance to other life-stages. Wear and tear ageing may occur in response to environmental conditions such as reactive oxygen species (ROS) and chemical toxins (Mondal et al., 2014; Munné-Bosch, 2015).

Cellular signaling pathways associated with ageing including Mammalian target of rapamycin (mTOR) kinase, the dynamic chromatin activities of Mi-2/Nucleosome Remodeling and Histone deacetylation (Mi- 2/NuRD) complexes (Zhang, 2012). Cellular processes such as autophagy, response to hypoxic insult, mitochondrial respiration, mRNA translation and ribosome biogenesis are regulated by proteins downstream of mTOR signaling, and are thought to play a pivotal role in the ageing process and age-related molecular pathologies (McCormick et al., 2011). The Mi-2/NuRD complex plays a significant role in the maintenance of pericentric heterochromatin assembly and normal progression in S phase. Declining Mi-2/NuRD complex function accelerates ageing through increasing level of DNA damage (Sims and Wade 2011; Zhang 2011).

Biomarkers of ageing may be associated with intercellular pathways. Interleukin 7 receptor (IL7R) is a novel biomarker for ageing and has been associated with longevity and age- related pathologies. PI3K activity, a protein upstream of mTOR, has been shown to affect the interleukin 7 receptor. IL7 R expression levels are reduced in older individuals. Reduced IL7 R gene expression levels is associated with increased risk of age- related pathologies (Kerdiles et al., 2009; Passtoors et al., 2015). Declining synaptic function in the outer retina in ageing may be associated with alterations in intercellular signaling pathway. Depletion of AMP-activated protein kinase (AMPK) can induce similar synaptic changes with ageing by upregulation of mTOR (Lewis, 2014).

Abnormal protein aggregation, inadequate anti-oxidant defense system, subcellular organelles and DNA damage plays an important role in the onset and development of ageing processes (He et al., 2014). During ageing, endoplasmic reticulum (ER) responds to stress by increasing the expression of proinflammatory cytokines that confer inflammation in adipose tissue (Ghosh et al., 2014). It is also recognized that during ageing, the ER stress response affects sleep homeostasis and increases age-associated sleep changes via up regulation of unfolded protein response (UPR) pathway (Brown et al., 2014).

Another important hallmark of ageing is metabolic dysfunction (Stout et al., 2015). Insulin/IGF-1 (IIS) metabolic pathway plays an essential role in the ageing process. It is well established that mitochondrial dysfunction can have a significant effect on age-related pathologies (Fischer et al., 2015). The concentration of ROS is increased in senescence and premature ageing (Kim et al., 2016; Li et al., 2016). During ageing, respiratory chain dysfunction leads to over-production of free radicals, and an imbalance in the endogenous antioxidant defense mechanisms. Ageing processes ultimately affect the efficiency of mitochondrial electron transport chain (ETC) to produce ATP by increasing proton leakage (Riera and Dillin 2015).

Cellular pathways that are involved in amino acid metabolism can affect lifespan and ageing (Zhang et al., 2014).The limitation of nutrient and calorie ingestion, without malnutrition, can decrease the onset of age-related deterioration and increases longevity (Ramos and Kaeberlein, 2012) Dysfunction in insulin sensitivity can lead to glucose intolerance and subsequently, cardiovascular and metabolic diseases (Riera and Dillin 2015; Wang et al., 2015). Lifespan extension is associated with metabolic health and respiratory exchange ratio (RER). Mitochondrial transcription factor A (TFAM) activity and nuclear respiratory factors (NRF1, NRF2) activity is regulated by PGC1α (proliferator-activated receptor γ coactivator α). Under conditions of low cellular energy, AMP-activated protein kinase (AMPK) inhibits anabolic processes such as protein synthesis by activating PGC1α, and thus enhancing senescence (de Lange et al., 2013; Slámová et al., 2015; Xu et al., 2015).

The integrity of damaged proteins (proteostasis) is also associated with the accumulation of ROS in ageing, and plays a significant role in age-related disorders such as osteoporosis, Huntington, amyotrophic lateral sclerosis (ALS), type2 diabetes mellitus, Alzheimer's disease, and Parkinson's disease (Chalil et al., 2015; Sharoar et al., 2015). During ageing, the deterioration of protein quality control ler mechanisms confer proteotoxicity. Misfolded or aggregated proteins confers dysfunction through the proteasome or autophagy related mechanism. Autophagy prevents the development of age- dependent pathologies by scavenging unnecessary proteins through an mTORC1 related pathway (Vilchez et al., 2014).

Chromatin defects are important in the molecular pathogenesis of ageing (White et al., 2015). RBBP4/7 knockdown leads to accumulation of heterochromatin structural damage, and subsequently the onset and development of ageing-related chromatin defects by impairing H4K20 histone methyltransferase PR-Set7. H4K20 histone methyltransferase PR-Set7s are master regulators of DNA replication, and their impairment can arrest cells in S phase and consequently confers the accumulation of DNA damage. During ageing, the components of NURD protein complex are downregulated, and reduction of their function is associated with chromatin defects (Pegoraro et al., 2009). It is also reported that telomere shortening is a natural "ageing clock" and exorbitance telomere shortening leads to DNA damage and chromatin instability (Badiola et al., 2015). Deterioration of circadian clock function is associated with senescence in the ageing brain. The circadian system, an internal time-keeping system that leads to coordination between physiological processes, behavior and environmental changes in 24 h rhythms, regulates rest activity cycle, hormone secretion, metabolism and other molecular and cellular processes (Kondratova and Kondratov 2012). During the circadian cycle, cAMP/MAPK/CREB transcriptional pathway is a master regulator of long-term memory (Eckel-Mahan et al., 2008). CREB is associated with the PI3K/Akt pathway (Chien et al., 2015). The

cAMP/MAPK/CREB transcriptional pathway is indirectly associated with the mTOR pathway.

The mTOR pathway also regulates cellular senescence. For instance, 'geroconversion' a form of futile growth that occurs during cell cycle arrest is suppressed by rapamycin, maintaining cellular quiescence (Demidenko et al., 2009). As well, low doses of dual mTORC1/mTORC2-selective inhibitors (PP242 and Torin1) can also restrict cellular senescence (Leontieva et al., 2015). Cellular senescence may mediate important roles in ageing and age-associated diseases, likely through the depletion of stem and progenitor cells, and the degenerative effects of senescence-associated secretory phenotype, such as proinflammatory cytokines and chemokines, highly volatile free radicals, matrix metalloproteinases and growth factors (Vicente et al., 2016; Yeh 2016). This is supported by evidence showing that interference with senescent cell accumulation may delay age-related degeneration. Moreover, recent studies have shown that senescent cells have beneficial effects in enhancing cellular repair following injury, and tissue remodeling (Baker et al., 2016; Cerella et al., 2016; Kaneda et al., 2016; Montesanto et al., 2016; Unruhe et al., 2016; Vicente et al., 2016; Yeh, 2016). Taken together, these studies suggest that senescence may drive age-dependent pathologies, and that mechanisms to facilitate the clearance of senescent cells may not distinguish between the removal of deleterious cells, and 'healthy' functional cells (Baker et al., 2011).

3. The mTOR signaling pathway

The mTOR signaling pathway is a key regulator of anabolic processes and a central controller of cell growth. It plays a significant role in cellular functions such as autophagy. It acts as a supplier of substrate in protein synthesis, and proteins quality controller through the promotion efficiency of protein repair and protein degradation (Zhang et al., 2014). The mTOR signaling pathways can also act as a central player in calorie restriction (Slack and Partridge 2013).

The mTOR pathway is a member of the phosphoinositide 3-kinase(PI3 K)-related protein kinases (PIKK) family (Yang et al., 2013). mTOR is encoded by the FRAP1gene and is expressed throughout different components of the body (Maiese 2014b). It is formed by two distinct multiprotein complexes including mTORC1and mTORC2 (Dalle Pezze et al., 2012; Thoreen et al., 2012). mTOR is a master regulator of cellular metabolism and cell growth (Mehrjerdi et al., 2013; Hukelmann et al., 2016). mTORC1 is formed by multiprotein including the regulatory-associated protein of mTOR (raptor), Deptor (DEP domain-containing mTOR interacting protein), proline rich Akt substrate 40 kDa (PRAS40) and mLST8/GâL (mammalian lethal with Sec13 protein 8) (Aylett et al., 2016). Akt maintain mTORC1 activity by inhibiting the phosphorylation of PRAS40 (Maiese, 2014b) . Phosphorylation of PRAS40 prevents the binding of mTORC1 to Raptor. In addition, the phosphorylation of Raptor by AMPK negatively controls mTORC1 activity. Ras homologue which is enriched in the brain (Rheb) is upstream of mTORC1 that phosphorylates residue serine^{863}in Raptor (Dibble and Cantley, 2015). Whereas mTORC2 contains mTOR kinase, Rictor (rapamycin-insensitive companion of mTOR), mLST8, DEPTOR, Protor, PRR5, PRR5 L and stress-activated protein kinase interacting protein (mSIN1).On the other hand, the mTORC2 activates Akt through mSIN1component. SGK1/protein kinase C (PKC)/Akt phosphorylation by mTORC2 at the hydrophobic motif sites regulates cytoskeletal organization, motility and subsequently spatial growth. Recent studies have reported that mTORC2 activation acts as a crucial component in tumorigenesis and fibrosis (Carr et al., 2015; Li et al., 2015).

4. Role of mTOR signaling pathways in ageing

Recently, the mTOR signaling pathway has been the focus of ageing research (Lamming et al., 2012). Evidence indicates that the mTOR signaling pathways acts as key controller of cellular ageing (Johnson et al., 2013). Previous studies suggest that mTORC1 plays a greater role than mTORC 2 in ageing. Inhibition of the mTOR signaling pathways extends lifespan through dietary restriction mimetic effects, whereas chronic activation of mTORC1 can extend age- dependent pathologies . The mutation of component or activation of upstream inhibitors of mTORC1 signaling pathways confers lifespan extension in model organisms such as *Drosophila melanogaster* and worms (Johnson et al., 2013). Rapamycin can promote glucose tolerance and insulin sensitivity and retard the ageing process in mammals by mTORC1 inhibition (Schreiber et al., 2015) . Several genetic and molecular studies suggest that calorie restriction and mTOR kinase can regulate ageing by overlapping mechanisms (Kaeberlein, 2013). Nutrient abundance plays an important role in the onset and development of the ageing process. Calorie restriction is a natural method that retards ageing via mTORC1 inhibition. AMPK can be activated by calorie restriction, and in turn, activates TSC1-TSC2 and consequently inhibits mTORC1 signaling and extends lifespan (Wrighton, 2011).

Rodents with nutrient restriction also display lower IGF-1 (Insulin-like growth factor) levels compared to control group (Kenyon 2010; Kanfi et al., 2012). The reduced levels of IGF-1 associated with calorie restriction plays a major role in slowing down the ageing process. Cellular amino acid and glucose levels increase Rag family of GTPases activity and subsequently mTORC1 activity. Nutrient restriction delays the ageing process by inhibiting of mTORC1 and provoking autophagy (Efeyan et al., 2015). The function of autophagy is one of key factors for determination of cellular ageing (Warr et al., 2013). Autophagy postpones ageing by eliminating damaged proteins and organelles. Abundance of phosphatidylethanolamine retards age- related pathologies by activating AMPK and subsequently autophagy via an mTORC1 related mechanism (Rockenfeller et al., 2015). Defective autophagy increases the onset of pathologies associated with ageing. mTORC1 signaling is a negative regulator of autophagy. Activation of mTORC1 signaling may enhance ageing and age-associated diseases by suppressing canonical autophagy (Levine et al., 2011). Indeed, mTORC1 activation suppresses the degradation of damaged proteins by inhibiting the formation of Atg1 complex and subsequently autophagosomal vesicle (Vilchez et al., 2014). It is well established that insulin/insulin-like growth factor 1 signaling pathway plays a key role in ageing. Insulin/IGF-1 signaling (IIS) suppression extends lifespan in worms, fruitflies and mammals (Bartke and Westbrook, 2012). Insulin/IGF-1 and other growth factors activate the PI3 K/phospho-AKT/mTOR pathway (Bitzer and Wiggins, 2016). Activated PI3 K pathway in turn stimulates Akt that inhibits tuberous sclerosis 2 (TSC2) and removes the inhibitory effects of TSC2 on Ras homologue enriched in brain (RHEB) and subsequently mTORC1 (Bender and Stewart, 2014). As well, low intracellular ATP content in response to AMP-activated protein kinase (AMPK) activation, facilitates TSC1-TSC2-mediated inhibition of Rheb, and consequently mTORC1.

It has been previously shown that metformin treatment activates AMPK, a negative regulator of mTOR, and then slows senescence and age-dependent centrosome amplification via repression of the AKT/TOR pathway (Miles et al., 2014; Na et al., 2015). Activation of the PI3K/phospho-AKT/mTOR pathway suppresses FOXO3A/autophagy and increases ribosomal synthesis. Inhibition of the ribosomal protein S6K1, promotes lifespan by improving insulin sensitivity and glucose tolerance through an mTORC1 related pathway (Mercado et al., 2015). Nonsteroidal anti-

inflammatory drug-activated gene (h NAG-1) or GDF15 suppresses the insulin/IGF-1 pathway and consequently, the PI3K/phospho-AKT/mTOR pathway, leading to the extension of longevity and lifespan in mice.

In addition, PI3K/phospho-AKT/mTOR pathway acts as a central player in Alzheimer's disease. IIS inhibition reduces mTOR activity followed by alterations in the phosphorylation and conformation of tau and Aβ protein. Altered protein homoeostasis in turn reduces the conformation of toxic proteins and subsequently confers lifespan extension and recovery of cognitive function (O'Neill et al., 2012).

ER stress accelerates ageing and age-associated deterioration (Brown and Naidoo 2012). During ER stress, mTORC1 activation results in the accumulation of unfolded proteins and damaged organelles by suppressing productive autophagy (Kyriakakis et al., 2015). ROS can also enhance ageing via the PI3K/phospho-AKT/mTOR. ROS initially activates PI3 K, and then inhibit FOXO3A/autophagy through PTEN and SIRT1 inactivation (Mercado et al., 2015). Ageing decreases ketogenesis by downregulating peroxisome-proliferator activated receptor (aPPARα) gene. During ageing, the activation of mTORC1 suppresses PPARα function and consequently, ketone production (Sengupta et al., 2010). Premature ageing can induce gene mutation or deletion of genes such as FOXO, Tsc1, ATM (ataxia telangiectasia mutated) and ATR (ataxia telangiectasia and Rad3 related). These genes have been associated with the mTORC1 pathway. For example, depletion of Tsc1, which is a negative central regulator of mTORC1, enhances the ageing process.

Moreover, the FOXO family of forkhead box protein is associated with the PI(3)K pathway (Sahin and DePinho 2010). Overexpression of FOXO genes can delay senescence and extend longevity in Drosophila (Proshkina et al., 2015). During ageing, mTOR activation enhances phosphorylation of the ULK1 protein that blocks AMPK-ULK1 interaction and consequently inhibits autophagy. Reduced Rictor expression or rictor mutants increases longevity by mTORC2 inhibition through an Akt- dependent mechanism. Inhibition of mTORC2 decreases Akt phosphorylation. Reduced Akt in turn stimulates Tsc1/Tsc2 complex, followed by suppression of mTORC1 activity (Soukas et al., 2009).

5. Modulation of mTOR signaling with polyphenol ic compounds

Polyphenols are organic chemical structures that can be found in natural and synthetic forms (Lu et al., 2016). Natural polyphenolic compounds, that are present in daily foods, spices, tea, oils, colourful fruits, and the skin and seed of grapes, can act as potent antioxidants. These compounds have been shown to induce the over expression of antioxidant enzymes such as manganese superoxide dismutase and catalase (Alañón et al., 2011; Schaffer et al., 2012; Moore 2015; Shen et al., 2016). Their antioxidant properties maybe associated with lifespan extension through mTOR related mechanisms. Polyphenols can also regulate mitochondrial activity and energy homeostasis (Lagouge et al., 2006). During ageing, polyphenolic compounds such as flavonoids play an important role in the reduction of oxidative stress levels and maintenance of mitochondrial homeostasis. Polyphenolic treatment confers over expression of genes for oxidative phosphorylation and mitochondrial biogenesis (Charles et al., 2013). Their anti-ageing properties have been associated with increasing collagen and ELN content, and decreasing the expression of metalloproteinase genes (Gopaul et al., 2012).

The polyphenol-rich extract of *Pimentadioica berries* (Allspice) displays potent anticancer activity by enhancing autophagy via the AKT/mTOR signaling pathway. These spices have been shown to lower pAkt and pmTOR levels, and stimulate the expression of autophagic genes in MCF7 cells (Zhang et al., 2015). *Hibiscus* leaf polyphenolics also causes similar effects on the Akt/mTOR signaling pathway in melanoma cells (Chiu et al., 2015). Polyphenols can induce both autophagy and apoptosis via similar mechanisms in human gastric cancer cells. The use of polyphenolic treatment together with autophagy inhibitors can increase apoptosis and cell death in human gastric cancer cells. Activated Akt can be linked to Ser184 of the BCL-2, an antiapoptic factor, which attenuates cell death (Nie et al., 2016).

Polyphenolic compounds such as can honokiol act as anticarcinogenic agents. Honokiol (3-,5-di-(2-propenyl)-1,1′-biphenyl-2,2′-diol), a naturally occurring dietary product isolated from an extract of seed cones from *Magnolia grandiflora*, can induce cellular autophagy by modulating the PI3K/Akt/mTOR signaling pathway in neuroblastoma cells (Yeh et al., 2016). Treatment with honokiol has been shown to reduce PI3K content, and in turn, inhibit Akt, which down regulates phosphorylation of mTOR. mTOR inhibition induces autophagy by upregulating the downstream protein kinases, such as ULK1and ATG13 (Yeh et al., 2016).

Polyphenolic compound such as curcumin, present in spices, also display anticancer activity through both the PI3K/phospho-AKT/mTOR and the AMPK-mTORC1 pathway. Activated AMPK stimulates Tsc1-Tsc2 activity and subsequently inhibits Rheb and mTORC1 signaling. In addition, these compounds can directly inhibit mTOR kinase (Wee et al., 2015). Curcumin blocks the association between Raptor and mTORC1 kinase. It also prevents the association of Rictor and mTORC2 (Cerella et al., 2015). Curcumin also suppresses the excessive activation of Insulin/IGF1 signaling and delays senescence in an experimental settings through the PI3K/AKT/mTOR pathway (Brown, 2015).

Apart from honokiol and curcumin, other polyphenols can target mTOR. Of particular interest are epigallocatechin 3-gallate (EGCG) from green tea, theaflavin digallate from black tea, the flavonoid quercetin, and the stilbenoid, resveratrol, extracted from grapes and red wine. Exposure of leukemia cells to polyphenols such as quercetin (3,3′,4′,5,7-pentahydroxy-flavone) led to cell cycle arrest through the Notch/AKT/mTOR signaling pathway. Quercetin inhibits Notch 1 expression, and subsequently downregulates phosphorylation of downstream protein kinases including Akt and mTOR, and promotes apoptosis (Chen et al., 2016). Recently, it has been shown that quercetin can effectively eliminate senescent human endothelial cells and mouse BM-SMCs by interfering with the expression of ephrin dependent receptor ligands EFNB1 or EFNB3 (Zhu et al., 2015). Silencing EFNB3 expression downregulates Akt in cancer cancers (Stahl et al., 2011). The combination of dasatinib (an oral Bcr-Abl tyrosine kinase inhibitor) and quercetin attenuated senescent cell burden in chronologically aged, radiation exposed, and progeroid Ercc1$^{-/\Delta}$ mice (Zhu et al., 2015). These results highlight the importance of quercetin and other polyphenols to ablate senescent cells and alleviate symptoms of frailty and promote lifespan extension.

Fisetin (3,7,3′,4′-tetrahydroxyflavone) belongs to the flavonol subgroup of flavonoids which includes quercetin, myricetin and kaempferol. Epidemiological and preclinical studies have shown that fisetin consumption affects various molecular targets which collectively slow the ageing process (George, 2016). Fisetin has been shown to inhibit the association between mTOR pathway signaling pathway constituents, Raptor, and Rictor (Syed et al., 2013, 2014; Chamcheu et al., 2015; Watanabe et al., 2015). Fisetin also decreases PI3-K content, and enhances phosphorylation of Akt. Phosphorylated Akt in turn inhibits mTOR activity. Fisetin can also directly activate negative regulators of mTOR such as Tsc complex and AMPK as shown in Fig. 1 (Adhami et al., 2012). Moreover, fisetin has been shown to enhance cytotoxic effects when used in

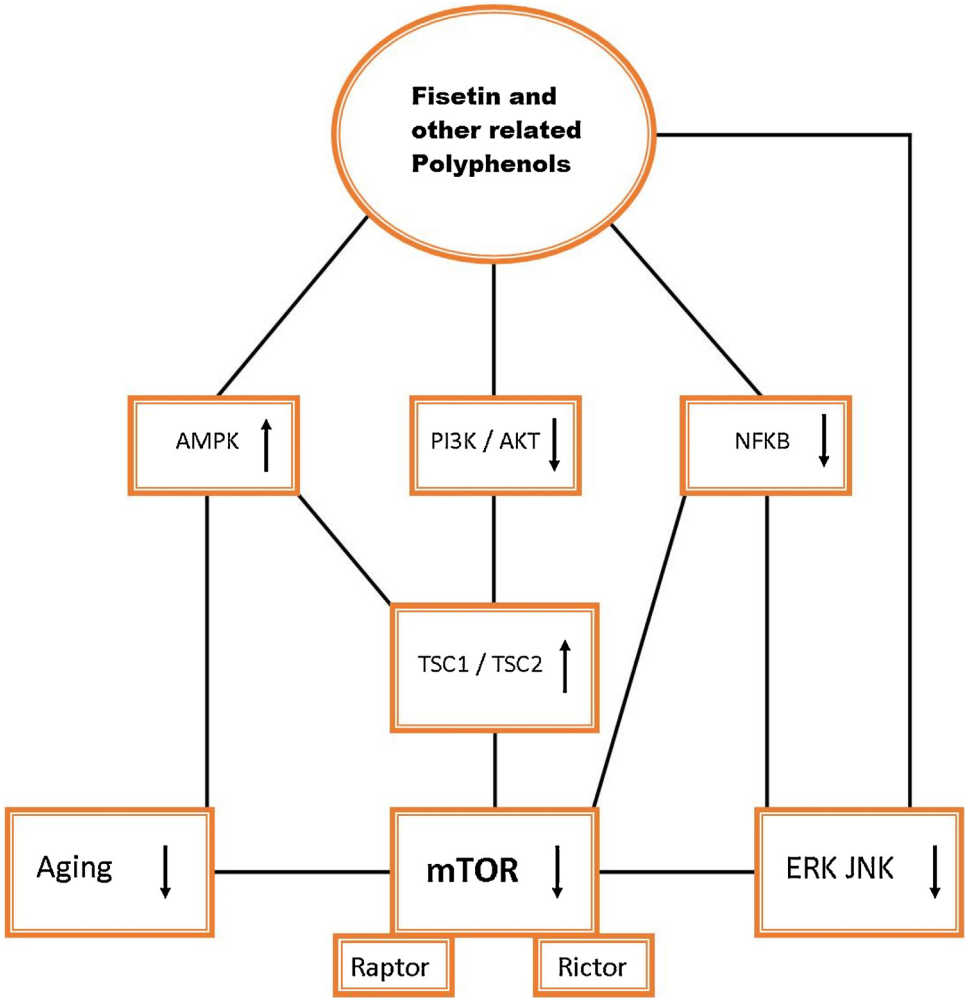

Fig. 1. Various molecular targets of fisetin and other related polyphenols.

combination with other chemotherapeutic drugs (Haddad et al., 2010; Klimaszewska-Wisniewska et al., 2016).

Polyphenolic compounds can also stimulate mTOR signaling to reduce cognitive defects in sleep deprivation. Sleep deprivation promotes cognitive impairment by downregulating CREB and mTOR signaling pathways. Combinational treatment with polyphenols ameliorates cognitive impairment.For instance, malvidin-3-*O*-glucoside activates mTOR/p70S6 K, whereas quercetin-3-*O*-glucuronide simulates CREB signaling activity. Punicalagin, a polyphenol present in pomegranate juice, can stimulate the expression of autophagic genes via mTOR inhibition (Banerjee et al., 2013). Overexpression of autophagic genes has been shown to limit apoptosis by decreasing phosphorylated ribosomal protein S6 expression in syncytiotrophoblasts (Wang et al., 2016a).

Several studies have identified the beneficial effects of oleuropein aglycone, the main polyphenol present in extra virgin olive oil, against senescence and ageing-related conditions in several models, with particular focus on autophagy activation (Hadrich et al., 2016). Similarly, oleuropein aglycone prevents the onset and development of age-related deterioration through Ca^{2+}/CaMKKβ/AMPK/mTOR axis (Rigacci et al., 2015). Oleuropein aglycone treatment leads to Ca^{2+} release from intracellular stores. Unbound Ca^{2+} activates CaMKKβ that is a protein present upstream of AMPK (Rigacci et al., 2015). CaMKKβ stimulates AMPK activity and induces autophagy by mTOR inhibition. This supports the notion that autophagy activation proceeds through mTOR activation (Rigacci et al., 2015).

Novel therapeutic strategies and new anti-ageing drugs based on caloric restriction mimetic properties, mTOR inhibition, insulin growth factor (IGF-I) inhibition, and activation of AMPK and sirtuins are currently in development. These therapeutic strategies are linked by the mTOR pathway (Stenvinkel et al., 2016). Polyphenols and caloric restriction have been shown to prevent ageing and age-associated diseases via similar mechanism. Polyphenolic compounds serve as important caloric restriction mimetics. They have similar effects on the function a new class of NAD-dependent histone deacetylases known as silent information regulators of gene transcription, or sirtuins (Pallauf et al., 2013). Recent studies have been reported that inhibition of mTOR inhibition confers lifespan extension (Mannick et al., 2014). Polyphenols, can act as anti-ageing agents by targeting mTOR and related pathways. Natural polyphenolic compounds can delay ageing by targeting AMPK through an mTOR dependent mechanism (Hwang et al., 2009).

Resveratrol, a polyphenolic compound isolated from the skin and seeds of grapes, can act as an anti-ageing agent via two distinctive mechanisms. Firstly, it suppresses IGF-1 and increases insulin sensitivity through PI3K/AKT/mTOR pathway. Secondly, it also promotes the activation of AMPK and Sirt1 protein (a nuclear sirtuin) in an Epac1-Dependent Manner (Park et al., 2012). Sirt1 is a positive regulator of AMPK that inhibits mTOR activity (Morselli et al., 2010). Activated AMPK can inhibit mTOR activity and delay the onset of age-related degeneration. In addition, resver-

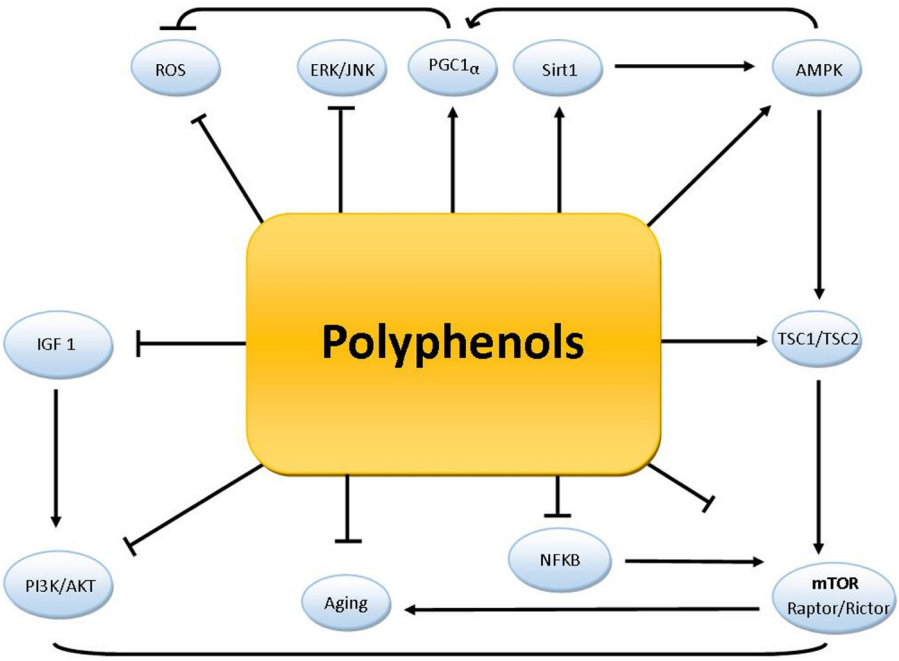

Fig. 2. Polyphenolic compounds delay the aging process and cognitive defects through activation or inhibition of various intracellular pathway.

atrol can increase peroxisome proliferator-activated receptor-c co- activator 1a (PPAR-co-1a) activity as shown in Fig. 2 (Barger et al., 2008).

6. Clinical implications of polyphenols as modulators of mTOR signaling

The beneficial effects of numerous naturally occurring polyphenols have been extensively documented in multiple *in vitro* and *in vivo* models (Table 1). Despite these findings, there are crucial imperative problems that need to be resolved prior to the use of these agents in human clinical trials. These problems include issues with absorption, bioavailability, and tissue distribution in animal models and humans for the most promising compounds.

Honokiol has demonstrated multiple biological properties relevant to antiaging including anti-arrhythmic, anti-inflammatory, anti-thrombocytic, anti-angiogenesis, anti-tumor, anxiolytic, antiviral, and antioxidative activities (Averett et al., 2016; Bunel et al., 2016; Lin et al., 2016; Sakaue et al., 2016; Suh et al., 2016; Tunc et al., 2016 Wang et al., 2016b Yu et al., 2016). It exhibited a favourable pharmacokinetic profile after intravenous (i.v.) administration in animal models (Jeong et al., 2016; Liu et al., 2016). One study showed that the maximal plasma concentration of honokiol following rectal administration of a houpo decoction equivalent to 13.5 mg/kg honokiol was approximately six times greater to that administered orally at an identical dose (Wu et al., 2003). Owing to concerns regarding poor aqueous solubility, liposomal formulations of honokiol have been developed. PEGylated (polyethylene glycol coated) liposomal honokiol showed greater serum concentrations, increased half-life (26 mins compared to 13 min following single i.v. administration of 20 mg/kg in balb/c mice), and reduced clearance (Gou et al., 2009; Gong et al., 2010; Gou et al., 2010). Another study demonstrated that honokiol can cross the blood brain barrier (BBB) after i.v. administration. The same study revealed the organ distribution of honokiol to be in the following order: lung > plasma > liver > brain > kidney > heart > spleen (Wang et al., 2011). Toxicity studies in animal models showed no significant macroscopic or microscopic hematological changes in Sprague Dawley rats after acute and sub-chronic oral dosage (Liu et al., 2007). Honokiol is clinically available for human testing and may be used as an adjunct to pre-existing cancer immunotherapies.

Curcumin is another polyphenol of major interest. Apart from its potent antioxidant and anti-inflammatory properties, it has been shown to antagonize pro-carcinogenic inflammatory mediators as well as cancer cell growth and viability (Guo et al., 2016; Seo et al., 2016; Zhao et al., 2016). However, the clinical administration of curcumin has been limited due to poor solubility and limited adsorption in the gastrointestinal tract. Other limiting factors include its rapid metabolism and clearance (Gupta et al., 2013). Plasma levels of curcumin are in the low micromolar concentrations after 1–2 h of administration. It has been suggested that the beneficial effects of curcumin can only be observed in compartments in full contact with pharmacological concentration, including the oropharyngeal and gastrointestinal tract following oral administration, and the skin or vaginal after topical application (Lao et al., 2006). Improved drug delivery systems have led to entrapping of curcumin and its derivatives in nanoparticles or liposomes, thus improving solubility and improving delivery to target tissue. Increased bioactivity has been reported in cancer cell models using nano-encapsulated curcumin. A human study using healthy volunteers reported increased plasma concentrations following oral consumption of nano-formulated curcumin (Kakkar et al., 2011). Curcumin has been shown to be well tolerated with little or no adverse effects when administered at doses between 0.5 and 12 g (Cheng et al., 2001). However, the safety and tolerability of encapsulated forms of curcumin are warranted.

There has been growing interest in the role of quercetin to ameliorate symptoms of degenerative and attenuate age-related degenerative diseases, including cancer, cardiovascular, inflammatory and neurodegenerative diseases (Hoek-van den Hil et al., 2015; Kobylinska and Janas 2015). Like honokiol, curcumin, and other related polyphenols, quercetin exhibits antioxidant, anti-inflammatory, and anti-amyloidogenic properties. Moreover, it can also regulate autophagic responses associated with protection against neurodegeneration, cancer and vascular disease (de Oliveira et al., 2015). To our knowledge, there is no published clinical trial on the anti-ageing effects of quercetin. However, there are few reports which have examined the effect of quercetin sup-

Table 1
Natural compounds implicated in the modulation of mTOR.

Compound	Contained in	Mechanism	Cellular studies	Concentrations used	References
Honokiol	Bark, seed cones, and leaves of trees belonging to the genus *Magnolia*	Inhibition of PI3k/Akt pathway	Glioma, breast and prostate cancer cells	25–100 μM	Yeh et al. (2016)
Curcumin	Turmeric *Curcuma longa*	Inhibition of PI3k/Akt pathway	Colorectal carcinoma cells	10–50 μM	Johnson et al. (2009)
		Blocks the association between Raptor and mTORC1 kinase	Rh1 cell lines	10–50 μM	Beevers et al. (2009)
		Suppresses the excessive activation of Insulin/IGF1 signaling	Streptozotocin-treated rat model	10–50 μM	Isik et al. (2009)
		Activation of AMPK via AMP/ATP balance	Macrophage-derived murine foam cells.	20–100 μM	Lin et al. (2015)
Epigallocatechin-3-gallate (EGCG)	Green tea	Activation of AMPK via AMP/ATP balance	Mouse Adipocytic 3T3-L1 cells	100 μM	Hsieh et al. (2010)
Theaflavin digallate	Black tea	Activation of AMPK via AMP/ATP balance	Human hepatoblastoma HepG2 cells	50 μM	Lin et al. (2007)
Quercetin	Capers, dill, onion, cranberry, apples, tomatoes	Inhibition of Notch 1 expression, and subsequently downregulates phosphorylation of Akt and mTOR	Human leukemia U937 cells	10–50 μM	Chen et al. (2016)
		Silencing of EFNB3 expression and downregulation of Akt	Human Preadipocytes, and *Ercc*$^{1-/\Delta}$ C57Bl/*6 mice*	50 μM	Zhu et al. (2015)
Punicalagin	Pomegranates	Inhibition of PI3k/Akt pathway	Sprague-Dawley rats	2 g/L	Banerjee et al. (2013)
Fisetin	Strawberries, apples, persimmons, kiwis, cucumbers, and onions	Inhibition of PI3k/Akt pathway	Human melanoma cells	10–50 μM	Suh et al. (2010), Adhami et al. (2012), Syed et al. (2013)
		Activation of AMPK via AMP/ATP balance	Mouse adipocytic 3T3-L1 cells	50 μM	Watanabe et al. (2015)
Oleuropein	Extra virgin olive oil	Activation of CaMKKβ stimulates AMPK activity and induces autophagy by mTOR inhibition	SH-SY5Y cells	50 μM	Rigacci et al. (2015)
Resveratrol	Grapes, red wine	Suppression of IGF-1 and increases insulin sensitivity through PI3K/AKT/mTOR pathway	C2C12 myoblast cells	50 μM	Park, et al. (2012)
		Activation of AMPK and Sirt1 protein in an Epac1-Dependent Manner	Mouse adipocytic 3T3-L1 cells	50 μM	Chen et al. (2015)

plementation on hypertension (Egert et al., 2010), and chronic lung disease sarcoidosis in healthy volunteers (Boots et al., 2011). Although the study found no significant changes on oxidative stress markers in healthy volunteers (Conquer et al., 1998), quercetin significantly reduced blood pressure in obese or hypertensive patients (Egert et al., 2010), and markedly reduced inflammation and oxidative stress present in sarcoidosis (Boots et al., 2011). This highlights the importance of quercetin to target physio-pathologic conditions.

Quercetin almost always undergoes glycation to yield the bioactive molecule, aglycon, which is responsible for the beneficial *in vitro* effects of quercetin (Russo et al., 2012). On the contrary to its natural glycosidic derivatives, the poor bioavailability of the aglycon form due to reduced absorption in the gastrointestinal tract raises a question of the clinical translation of quercetin (Russo et al., 2012). Quercetin has been shown to localize in several tissues following absorption through the gastrointestinal tract where it is metabolised. The greatest concentrations of quercetin have been reported in the colon, lungs, liver, and kidneys, the latter of which plays an important role in the clearance and excretion of quercetin and its derivatives (Wang et al., 2014). Another study in rats showed that quercetin can accumulate in the brain after oral administration, providing evidence for the ability of quercetin to cross the BBB (Ishisaka et al., 2011). It has been estimated that the plasma concentration of quercetin in a normal diet is typically <100 nM, which is below concentrations reported to produce favourable effects in experimental models (Russo et al., 2012). However, one study previously demonstrated that the plasma concentration may be increased to >10 μM when a quercetin rich diet composed of apples, onions or tomatoes is administered (Russo et al., 2012). Several strategies are currently under investigation to improve the bioavailability of the aglycon and enhance therapeutic outcomes in the clinic. These strategies include: (1) determining the mode of action of hemi-synthetic moieties of the aglycon product which demonstrate greater chemical stability (Kim et al., 2009); (2) investigating the biological properties of quercetin glycosides such as quercetin-3-*O*-rutinoside and quercetin-3-*O*-glucoside (Makino et al., 2009); and (3) development of encapsulation technique to entrap quercetin and its derivatives in nanoparticles or liposomes (Wang et al., 2014). Quercetin-containing liposomes have been shown to specifically accumulate in cancer cells, and arrest cancer cell growth when administered intravenously in lung tumor-bearing mice (Yuan et al., 2006).

Quercetin displays a favourable toxicity profile, and no adverse effects were reported after oral administration of quercetin at concentrations up to 4 g, or following supplementation with a maximal dose of 1 g fractioned into 500 mg given twice daily, in healthy human volunteers. However, renal toxicity was reported following i.v. infusion when administered over 3.5 g in a 70 kg human (Ferry et al., 1996). Therefore, while quercetin is generally considered to be safe, it is important to establish the toxicological profile prior to clinical use.

Fisetin, like quercetin, is another flavonol that is present in several fruits and vegetables, including strawberries, apples, persimmons, kiwis, cucumbers, and onions (Khan et al., 2013). Fisetin

has been shown to exhibit several biological properties relevant to ageing and age-related diseases (Khan et al., 2013). It has been identified as a potential antimicrobial agent in one study (Gabor and Eperjessy 1966). Several other studies have shown that fisetin can attenuate oxidative stress and inflammation following nerve damage (Inkielewicz-Stepniak et al., 2012 Piao et al., 2013; Hytti et al., 2015). Oral administration of fisetin improved long-term potentiation and memory in a rodent model (Prasath and Subramanian 2013; Prasath et al., 2013). The anti-angiogenic effects of fisetin have been well described and linked to the significantly reduced activity of Aurora B kinase (Gollapudi et al., 2014). The anti-inflammatory effects of fisetin have been attributed to modulation of NF-κB (Yao et al., 2008; Wu et al., 2011; Zhou et al., 2015; Sahu et al., 2016). NF-κB signaling may also activate mTOR through proinflammatory cytokines such as TNF-α. In particular, fisetin can suppress the activation of NF-κB which may be induced in response to several inflammatory agents, leading to reduced phosphorylation and degradation of IκBα, and suppression of the phosphorylation and nuclear translocation of NF-κB/p65 (Yao et al., 2008; Murtaza et al., 2009; Leotoing et al., 2013; Li et al., 2014). Another study showed that fisetin can attenuate cerebral damage in a murine stroke model and can accumulate in the brain following a single intraperitoneal (i.p.) dose, suggesting that it can cross the BBB (Zhou et al., 2015).

Fisetin appears to be well tolerated with no adverse effects previously reported. The bioavailability of fisetin has been previously examined following i.v. and oral administration (Shia et al., 2009). The serum levels of fisetin have been shown to decline rapidly within the first few hours. This occurs parallel to a significant increase in the levels of sulfated and glucuronidated derivatives. Following oral consumption, the serum concentration of bioactive fisetin sulfates or its glucuronide products has been shown to be maintained at 10 μM for at least 24 h. Therefore, the protective effects of fisetin against cancer cell proliferation, which have been reported at concentrations below 50 μM, are achievable following consumption of a fisetin-rich diet (Maher et al., 2007).

The beneficial effects of extra virgin olive oil have been associated with the content of polyphenols. These properties include antioxidant, anti-inflammatory, anti-cancer, anti-microbial, anti-viral, anti-atherogenic, hypoglycemic, hepatic-cardiac- and neuroprotective effects (Mourouti and Panagiotakos, 2016). Oleuropein, main molecule responsible for the favourable effects of olive oil, is the most prevalent phenolic compound in olives and accounts for up to 14% of the dry weight. Although the mode of absorption of oleuropein remains unclear, bioavailability studies have shown that the absorption of oleuropein derivatives (including ligistroside-aglycone, hydroxyltyrosol, tyrosol and oleuropein-aglycone) from virgin olive oil was 55–60% in humans (Garcia-Villalba et al., 2014). Moreover, another study showed that 15% of oleuropein glycosides were excreted in urine as hydroxyltyrosol and tyrosol. Oleuropein has been shown to be rapidly absorbed after oral administration, with maximal plasma concentration occurring 2 h after administration. Hydroxyltyrosol represents the most important metabolite and is excreted in urine mostly as glucoronides (de Bock et al., 2013).

Several studies have shown that consumption of olive oil can reduce the risk of coronary heart disease, which is a major facet of ageing. As well, several studies have shown that the phenol content of olive oil can reduce the oxidation of low-density lipoprotein (LDL) and biomarkers of oxidative stress in animal models (Vissers et al., 2004). However, the effect of these phenols in humans has not been confirmed. Moreover, there is no data on the phenol concentrations in plasma following dietary intake of olive oil. It has been estimated that 50 g of olive oil is required to provide 13 μM of hydroxytyrosol per day, with the plasma concentration of phenols in olive oil that demonstrate potent antioxidant properties is much lower (Vissers et al., 2004). Therefore, there is no available evidence to suggest that dietary olive oil will protect LDL against oxidative modification and delay the onset of vascular disease.

Resveratrol is an important stilbene that is present primarily in grapes and wine. The antioxidant and anti-inflammatory effects of resveratrol have been well described previously (Kelkel et al., 2010). Oral administration of resveratrol has been shown to reduce hypertension and dyslipidemia, and improve lung and endothelial function in humans (Bishayee 2009). Current clinical trials targeting cancer have used a daily oral dose of 500 mg (Bishayee 2009). Bioavailability studies have shown that resveratrol is rapidly absorbed in the gastrointestinal tract after oral consumption, with peak plasma concentrations recorded after 30 min (Soleas et al., 2001). Resveratrol appears to be excreted by the kidney and has been detected in the urine after 24 h (Soleas et al., 2001). Resveratrol is metabolised in its glucuronide and sulfate derivatives by rapid metabolism in enterocytes. Since concentrations of resveratrol are only detected in the blood in a nanomolar range, the possibility of reaching pharmacologically relevant doses in the clinic remains a challenge (Goldberg et al., 2003). The development of more active hemi-synthetic derivatives, or using encapsulation techniques are currently under development.

7. Conclusion

Our understanding of the molecular pathogenesis and cellular pathways involved in ageing has grown extensively in recent years. This is anticipated to lead to the slowing down or delaying the ageing and age- related diseases. Given the social and economic burden of the rapidly growing ageing population where the mTOR signaling pathway has already been implicated, it is likely that additional therapeutic uses for mTOR inhibitors will be identified in the near future.Polyphenolic compounds have been shown to delay the ageing process by targeting mTOR and related pathways, and provides a safe, non-invasive means to extend lifespan and reduce morbidity and mortality due to age-associated degeneration. Although the potential negative effects of mTOR inhibition remain unclear, the potential for naturally occurring polyphenolic compounds to interfere with the mTOR signaling as current drug targets appears effective and feasible *in vitro* cell culture setting and in experimental preclinical animal models.

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References

Adhami, V.M., Syed, D.N., Khan, N., Mukhtar, H., 2012. Dietary flavonoid fisetin: a novel dual inhibitor of PI3K/Akt and mTOR for prostate cancer management. Biochem. Pharmacol. 84 (10), 1277–1281.

Alañón, M., Castro-Vázquez, L., Díaz-Maroto, M., Gordon, M., Pérez-Coello, M., 2011. A study of the antioxidant capacity of oak wood used in wine ageing and the correlation with polyphenol composition. Food Chem. 128 (4), 997–1002.

Averett, C.A., Bhardwaj, A., Arora, S., Srivastava, S., Singh, S., Carter, J.E., Singh, A.P., 2016. Honokiol attenuates stemness, growth and metastasis of pancreatic tumor cells: a novel natural agent for therapy and prevention. Cancer Res. 76, http://dx.doi.org/10.1158/1538-7445.fbcr15-b02.

Aylett, C.H., Sauer, E., Imseng, S., Boehringer, D., Hall, M.N., Ban, N., Maier, T., 2016. Architecture of human mTOR complex 1. Science 351 (6268), 48–52.

Badiola, I., Santaolalla, F., Garcia-Gallastegui, P., Sánchez-del Rey, A., Unda, F., Ibarretxe, G., 2015. Biomolecular bases of the senescence process and cancer: a new approach to oncological treatment linked to ageing. Ageing Res. Rev. 23 (Pt B), 125–138.

Baker, D.J., Wijshake, T., Tchkonia, T., LeBrasseur, N.K., Childs, B.G., van de Sluis, B., Kirkland, J.L., van Deursen, J.M., 2011. Clearance of p16Ink4a-positive senescent cells delays ageing-associated disorders. Nature 479 (7372), 232–236.

Baker, D.J., Childs, B.G., Durik, M., Wijers, M.E., Sieben, C.J., Zhong, J., Saltness, R.A., Jeganathan, K.B., Verzosa, G.C., Pezeshki, A., Khazaie, K., Miller, J.D., van Deursen, J.M., 2016. Naturally occurring p16(Ink4a)-positive cells shorten healthy lifespan. Nature 530 (7589), 184–189.

Banerjee, N., Kim, H., Talcott, S., Mertens-Talcott, S., 2013. Pomegranate polyphenolics suppressed azoxymethane-induced colorectal aberrant crypt foci and inflammation: possible role of miR-126/VCAM-1 and miR-126/PI3K/AKT/mTOR. Carcinogenesis 34 (12), 2814–2822.

Barger, J.L., Kayo, T., Vann, J.M., Arias, E.B., Wang, J., Hacker, T.A., Wang, Y., Raederstorff, D., Morrow, J.D., Leeuwenburgh, C., 2008. A low dose of dietary resveratrol partially mimics caloric restriction and retards aging parameters in mice. PLoS One 3 (6), e2264.

Bartke, A., Westbrook, R., 2012. Metabolic characteristics of long-lived mice. Front. Genet. 3, 288.

Beevers, C.S., Chen, L., Liu, L., Luo, Y., Webster, N.J., Huang, S., 2009. Curcumin disrupts the Mammalian target of rapamycin-raptor complex. Cancer Res. 69 (3), 1000–1008.

Bender, A., Stewart, A.F., 2014. Good news for the ageing beta cell. Diabetologia 57 (2), 265–269.

Bishayee, A., 2009. Cancer prevention and treatment with resveratrol: from rodent studies to clinical trials. Cancer Prev. Res. 2 (5), 409–418.

Bitzer, M., Wiggins, J., 2016. Aging biology in the kidney. Adv. Chronic Kidney Dis. 23 (1), 12–18.

Boots, A.W., Drent, M., de Boer, V.C., Bast, A., Haenen, G.R., 2011. Quercetin reduces markers of oxidative stress and inflammation in sarcoidosis. Clin. Nutr. 30 (4), 506–512.

Brown, M.K., Naidoo, N., 2012. The endoplasmic reticulum stress response in aging and age-related diseases. Front. Physiol. 3, 263.

Brown, M.K., Chan, M.T., Zimmerman, J.E., Pack, A.I., Jackson, N.E., Naidoo, N., 2014. Aging induced endoplasmic reticulum stress alters sleep and sleep homeostasis. Neurobiol. Aging (Milano) 35 (6), 1431–1441.

Brown, D., 2015. Antibiotic resistance breakers: can repurposed drugs fill the antibiotic discovery void? Nature Rev. Drug Discovery 14 (12), 821–832.

Bunel, V., Antoine, M.H., Stevigny, C., Nortier, J., Duez, P., 2016. New in vitro insights on a cell death pathway induced by magnolol and honokiol in aristolochic acid tubulotoxicity. Food Chem. Toxicol. 87, 77–87.

Carr, T.D., Feehan, R.P., Hall, M.N., Rüegg, M.A., Shantz, L.M., 2015. Conditional disruption of rictor demonstrates a direct requirement for mTORC2 in skin tumor development and continued growth of established tumors. Carcinogenesis 36 (4), 487–497.

Cerella, C., Gaigneaux, A., Dicato, M., Diederich, M., 2015. Antagonistic role of natural compounds in mTOR-mediated metabolic reprogramming. Cancer Lett. 356 (2), 251–262.

Cerella, C., Grandjenette, C., Dicato, M., Diederich, M., 2016. Roles of apoptosis and cellular senescence in cancer and Aging. Curr. Drug Targets 17 (4), 405–415.

Chalil, S., Jaspers, R.T., Manders, R.J., Klein-Nulend, J., Bakker, A.D., Deldicque, L., 2015. Increased endoplasmic reticulum stress in mouse osteocytes with aging alters cox-2 response to mechanical stimuli. Calcif. Tissue Int. 96 (2), 123–128.

Chamcheu, J., Chaves-Rodriquez, M., Siddiqui, I., Adhami, V., Syed, D.N., Dodwad, S., Mukhtar, H., 2015. Fisetin, a small molecule and a natural inhibitor of mTOR for treating psoriasis. J. Invest. Dermatol. 135, S96.

Charles, A.L., Meyer, A., Dal-Ros, S., Auger, C., Keller, N., Ramamoorthy, T.G., Zoll, J., Metzger, D., Schini-Kerth, V., Geny, B., 2013. Polyphenols prevent ageing-related impairment in skeletal muscle mitochondrial function through decreased reactive oxygen species production. Exp. Physiol. 98 (2), 536–545.

Chen, H., Zhu, G.M., Liu, N., Zhang, W.W., 2014. Low-dose tissue plasminogen activator is as effective as standard tissue plasminogen activator administration for the treatment of acute ischemic stroke. Curr. Neurovasc. Res. 11 (1), 62–67.

Chen, S., Zhou, N., Zhang, Z., Li, W., Zhu, W., 2015. Resveratrol induces cell apoptosis in adipocytes via AMPK activation. Biochem. Biophys. Res. Commun. 457 (4), 608–613.

Chen, X., Dong, X.S., Gao, H.Y., Jiang, Y.F., Jin, Y.L., Chang, Y.Y., Chen, L.Y., Wang, J.H., 2016. Suppression of HSP27 increases the anti-tumor effects of quercetin in human leukemia U937 cells. Mol. Med. Rep 13 (1), 689–696.

Cheng, A.L., Hsu, C.H., Lin, J.K., Hsu, M.M., Ho, Y.F., Shen, T.S., Ko, J.Y., Lin, J.T., Lin, B.R., Ming-Shiang, W., Yu, H.S., Jee, S.H., Chen, G.S., Chen, T.M., Chen, C.A., Lai, M.K., Pu, Y.S., Pan, M.H., Wang, Y.J., Tsai, C.C., Hsieh, C.Y., 2001. Phase I clinical trial of curcumin, a chemopreventive agent, in patients with high-risk or pre-malignant lesions. Anticancer Res. 21 (4B), 2895–2900.

Chien, P.T.Y., Lin, C.C., Hsiao, L.D., Yang, C.M., 2015. c Src/Pyk2/EGFR/PI3 K/Akt/CREB-activated pathway contributes to human cardiomyocyte hypertrophy: role of COX-2 induction. Mol. Cell. Endocrinol. 409, 59–72.

Chiu, C.T., Hsuan, S.W., Lin, H.H., Hsu, C.C., Chou, F.P., Chen, J.H., 2015. Hibiscus sabdariffa leaf polyphenolic extract induces human melanoma cell death, apoptosis, and autophagy. J. Food Sci. 80 (3), H649–H658.

Conquer, J.A., Maiani, G., Azzini, E., Raguzzini, A., Holub, B.J., 1998. Supplementation with quercetin markedly increases plasma quercetin concentration without effect on selected risk factors for heart disease in healthy subjects. J. Nutr. 128 (3), 593–597.

Dalle Pezze, P., Sonntag, A.G., Thien, A., Prentzell, M.T., Gödel, M., Fischer, S., Neumann-Haefelin, E., Huber, T.B., Baumeister, R., Shanley, D.P., 2012. A dynamic network model of mTOR signaling reveals TSC-independent mTORC2 regulation. Sci. Signal 5 (217), ra25.

Demidenko, Z.N., Zubova, S.G., Bukreeva, E.I., Pospelov, V.A., Pospelova, T.V., Blagosklonny, M.V., 2009. Rapamycin decelerates cellular senescence. Cell Cycle 8 (12), 1888–1895.

Dibble, C.C., Cantley, L.C., 2015. Regulation of mTORC1 by PI3K signaling. Trends Cell Biol. 25 (9), 545–555.

Eckel-Mahan, K.L., Phan, T., Han, S., Wang, H., Chan, G.C., Scheiner, Z.S., Storm, D.R., 2008. Circadian oscillation of hippocampal MAPK activity and cAMP: implications for memory persistence. Nature Neurosci. 11 (9), 1074–1082.

Efeyan, A., Comb, W.C., Sabatini, D.M., 2015. Nutrient-sensing mechanisms and pathways. Nature 517 (7534), 302–310.

Egert, S., Boesch-Saadatmandi, C., Wolffram, S., Rimbach, G., Muller, M.J., 2010. Serum lipid and blood pressure responses to quercetin vary in overweight patients by apolipoprotein E genotype. J. Nutr. 140 (2), 278–284.

Ferry, D.R., Smith, A., Malkhandi, J., Fyfe, D.W., deTakats, P.G., Anderson, D., Baker, J., Kerr, D.J., 1996. Phase I clinical trial of the flavonoid quercetin: pharmacokinetics and evidence for *in vivo* tyrosine kinase inhibition. Clin. Cancer Res. 2 (4), 659–668.

Fischer, F., Filippis, C., Osiewacz, H.D., 2015. RCF1-dependent respiratory supercomplexes are integral for lifespan-maintenance in a fungal ageing model. Sci. Rep. 5, 12697.

Gabor, M., Eperjessy, E., 1966. Antibacterial effect of fisetin and fisetinidin. Nature 212 (5067), 1273.

Garcia-Villalba, R., Larrosa, M., Possemiers, S., Tomas-Barberan, F.A., Espin, J.C., 2014. Bioavailability of phenolics from an oleuropein-rich olive (*Olea europaea*) leaf extract and its acute effect on plasma antioxidant status: comparison between pre- and postmenopausal women. Eur. J. Nutr. 53 (4), 1015–1027.

George, V.C., 2016. Promising tumor inhibiting potentials of fisetin through PI3K/AKT/mTOR pathway. Am. J. Transl. Res. 8 (2), 1293–1294.

Ghosh, A.K., Garg, S.K., Mau, T., O'Brien, M., Liu, J., Yung, R., 2014. Elevated endoplasmic reticulum stress response contributes to adipose tissue inflammation in aging. J. Gerontol. Ser. A: Biol. Sci. Med. Sci. 70 (11), 1320–1329.

Goldberg, D.M., Yan, J., Soleas, G.J., 2003. Absorption of three wine-related polyphenols in three different matrices by healthy subjects. Clin. Biochem. 36 (1), 79–87.

Gollapudi, P., Hasegawa, L.S., Eastmond, D.A., 2014. A comparative study of the aneugenic and polyploidy-inducing effects of fisetin and two model Aurora kinase inhibitors. Mutat. Res. Genet. Toxicol. Environ. Mutagen 767, 37–43.

Gong, C.Y., Wei, X.W., Wang, X.H., Wang, Y.J., Guo, G., Mao, Y.Q., Luo, F., Qian, Z.Y., 2010. Biodegradable self-assembled PEG-PCL-PEG micelles for hydrophobic honokiol delivery: I: Preparation and characterization. Nanotechnology 21 (21), 215103.

Gopaul, R., Knaggs, H.E., Lephart, E.D., 2012. Biochemical investigation and gene analysis of equol: a plant and soy-derived isoflavonoid with antiaging and antioxidant properties with potential human skin applications. Biofactors 38 (1), 44–52.

Gordon, B.S., Kazi, A.A., Coleman, C.S., Dennis, M.D., Chau, V., Jefferson, L.S., Kimball, S.R., 2014. RhoA modulates signaling through the mechanistic target of rapamycin complex 1 (mTORC1) in mammalian cells. Cell. Signal. 26 (3), 461–467.

Gou, M.L., Zheng, L., Peng, X.Y., Men, K., Zheng, X.L., Zeng, S., Guo, G., Luo, F., Zhao, X., Chen, L.J., Wei, Y.Q., Qian, Z.Y., 2009. Poly(epsilon-caprolactone)-poly(ethylene glycol)–poly(epsilon-caprolactone) (PCL-PEG-PCL) nanoparticles for honokiol delivery *in vitro*. Int. J. Pharm. 375 (1-2), 170–176.

Gou, M.L., Gong, C.Y., Zhang, J., Wang, X.H., Wang, X.H., Gu, Y.C., Guo, G., Chen, L.J., Luo, F., Zhao, X., Wei, Y.Q., Qian, Z.Y., 2010. Polymeric matrix for drug delivery: honokiol-loaded PCL-PEG-PCL nanoparticles in PEG-PCL-PEG thermosensitive hydrogel. J. Biomed. Mater. Res. Part A 93A (1), 219–226.

Guo, S.Y., Long, M.Z., Li, X.Z., Zhu, S.S., Zhang, M., Yang, Z.J., 2016. Curcumin activates autophagy and attenuates oxidative damage in EA.hy926 cells via the Akt/mTOR pathway. Mol. Med. Rep. 13 (3), 2187–2193.

Gupta, S.C., Patchva, S., Aggarwal, B.B., 2013. Therapeutic roles of curcumin: lessons learned from clinical trials. AAPS J. 15 (1), 195–218.

Haddad, A.Q., Fleshner, N., Nelson, C., Saour, B., Musquera, M., Venkateswaran, V., Klotz, L., 2010. Antiproliferative mechanisms of the flavonoids 2,2'-Dihydroxychalcone and fisetin in human prostate cancer cells. Nutr. Cancer 62 (5), 668–681.

Hadrich, F., Garcia, M., Maalej, A., Moldes, M., Isoda, H., Feve, B., Sayadi, S., 2016. Oleuropein activated AMPK and induced insulin sensitivity in C2C12 muscle cells. Life Sci. 151, 167–173.

He, H., Liu, X., Lv, L., Liang, H., Leng, B., Zhao, D., Zhang, Y., Du, Z., Chen, X., Li, S., 2014. Calcineurin suppresses AMPK-dependent cytoprotective autophagy in cardiomyocytes under oxidative stress. Cell. Death. Dis. 5 (1), e997.

Hoek-van den Hil, E.F., van Schothorst, E.M., van der Stelt, I., Swarts, H.J., van Vliet, M., Amolo, T., Vervoort, J.J., Venema, D., Hollman, P.C., Rietjens, I.M., Keijer, J., 2015. Direct comparison of metabolic health effects of the flavonoids quercetin, hesperetin, epicatechin, apigenin and anthocyanins in high-fat-diet-fed mice. Genes Nutr. 4, 469.

Hsieh, C.F., Tsuei, Y.W., Liu, C.W., Kao, C.C., Shih, L.J., Ho, L.T., Wu, L.Y., Wu, C.P., Tsai, P.H., Chang, H.H., Ku, H.C., Kao, Y.H., 2010. Green tea epigallocatechin gallate inhibits insulin stimulation of adipocyte glucose uptake via the 67-kilodalton laminin receptor and AMP-activated protein kinase pathways. Planta Med. 76 (15), 1694–1698.

Hukelmann, J.L., Anderson, K.E., Sinclair, L.V., Grzes, K.M., Murillo, A.B., Hawkins, P.T., Stephens, L.R., Lamond, A.I., Cantrell, D.A., 2016. The cytotoxic T cell proteome and its shaping by the kinase mTOR. Nat. Immunol. 17 (1), 104–112.
Hwang, J.T., Kwon, D.Y., Yoon, S.H., 2009. AMP-activated protein kinase: a potential target for the diseases prevention by natural occurring polyphenols. New Biotechnol. 26 (1), 17–22.
Hytti, M., Piippo, N., Korhonen, E., Honkakoski, P., Kaarniranta, K., Kauppinen, A., 2015. Fisetin and luteolin protect human retinal pigment epithelial cells from oxidative stress-induced cell death and regulate inflammation. Sci. Rep. 5, 17645.
Inkielewicz-Stepniak, I., Radomski, M.W., Wozniak, M., 2012. Fisetin prevents fluoride- and dexamethasone-induced oxidative damage in osteoblast and hippocampal cells. Food Chem. Toxicol. 50 (3-4), 583–589.
Ishisaka, A., Ichikawa, S., Sakakibara, H., Piskula, M.K., Nakamura, T., Kato, Y., Ito, M., Miyamoto, K., Tsuji, A., Kawai, Y., Terao, J., 2011. Accumulation of orally administered quercetin in brain tissue and its antioxidative effects in rats. Free Radic. Biol. Med. 51 (7), 1329–1336.
Isik, A.T., Celik, T., Ulusoy, G., Ongoru, O., Elibol, B., Doruk, H., Bozoglu, E., Kayir, H., Mas, M.R., Akman, S., 2009. Curcumin ameliorates impaired insulin/IGF signalling and memory deficit in a streptozotocin-treated rat model. Age (Dordr) 31 (1), 39–49.
Jeong, H.U., Kim, J.H., Kong, T.Y., Choi, W.G., Lee, H.S., 2016. Comparative metabolism of honokiol in mouse, rat, dog, monkey, and human hepatocytes. Arch. Pharm. Res. 39 (4), 516–530.
Johnson, S.M., Gulhati, P., Arrieta, I., Wang, X., Uchida, T., Gao, T., Evers, B.M., 2009. Curcumin inhibits proliferation of colorectal carcinoma by modulating Akt/mTOR signaling. Anticancer Res. 29 (8), 3185–3190.
Johnson, S.C., Rabinovitch, P.S., Kaeberlein, M., 2013. mTOR is a key modulator of ageing and age-related disease. Nature 493 (7432), 338–345.
Kaeberlein, M., 2013. mTOR inhibition: from aging to autism and beyond. Scientifica 2013, 849186.
Kakkar, V., Singh, S., Singla, D., Kaur, I.P., 2011. Exploring solid lipid nanoparticles to enhance the oral bioavailability of curcumin. Mol. Nutr. Food Res. 55 (3), 495–503.
Kaneda, A., Nonaka, A., Fujita, T., Yamanaka, R., Fujimoto, M., Miyazono, K., Aburatani, H., 2016. Epigenomic regulation of smad1 signaling during cellular senescence induced by ras activation. Methods Mol. Biol. 1344, 341–353.
Kanfi, Y., Naiman, S., Amir, G., Peshti, V., Zinman, G., Nahum, L., Bar-Joseph, Z., Cohen, H.Y., 2012. The sirtuin SIRT6 regulates lifespan in male mice. Nature 483 (7388), 218–221.
Kelkel, M., Jacob, C., Dicato, M., Diederich, M., 2010. Potential of the dietary antioxidants resveratrol and curcumin in prevention and treatment of hematologic malignancies. Molecules 15 (10), 7035–7074.
Kenyon, C.J., 2010. The genetics of ageing. Nature 464 (7288), 504–512.
Kerdiles, Y.M., Beisner, D.R., Tinoco, R., Dejean, A.S., Castrillon, D.H., DePinho, R.A., Hedrick, S.M., 2009. Foxo1 links homing and survival of naive T cells by regulating L-selectin, CCR7 and interleukin 7 receptor. Nat. Immunol. 10 (2), 176–184.
Khan, N., Syed, D.N., Ahmad, N., Mukhtar, H., 2013. Fisetin: a dietary antioxidant for health promotion. Antioxid. Redox Signal. 19 (2), 151–162.
Kim, M.K., Park, K.S., Yeo, W.S., Choo, H., Chong, Y., 2009. In vitro solubility, stability and permeability of novel quercetin-amino acid conjugates. Bioorg. Med. Chem. 17 (3), 1164–1171.
Kim, D.B., Shin, G.H., Kim, J.M., Kim, Y.H., Lee, J.H., Lee, J.S., Song, H.J., Choe, S.Y., Park, I.J., Cho, J.H., 2016. Antioxidant and anti-ageing activities of citrus-based juice mixture. Food Chem. 194, 920–927.
Klimaszewska-Wisniewska, A., Halas-Wisniewska, M., Tadrowski, T., Gagat, M., Grzanka, D., Grzanka, A., 2016. Paclitaxel and the dietary flavonoid fisetin: a synergistic combination that induces mitotic catastrophe and autophagic cell death in A549 non-small cell lung cancer cells. Cancer Cell Int. 16, 10.
Kobylinska, A., Janas, K.M., 2015. Health–promoting effect of quercetin in human diet. Postepy Hig Med Dosw (Online) 69, 51–62.
Kondratova, A.A., Kondratov, R.V., 2012. The circadian clock and pathology of the ageing brain. Nat. Rev. Neurosci. 13 (5), 325–335.
Kyriakakis, E., Princz, A., Tavernarakis, N., 2015. Stress responses during ageing: molecular pathways regulating protein homeostasis. Stress Responses Methods Protoc., 215–234.
Lagouge, M., Argmann, C., Gerhart-Hines, Z., Meziane, H., Lerin, C., Daussin, F., Messadeq, N., Milne, J., Lambert, P., Elliott, P., 2006. Resveratrol improves mitochondrial function and protects against metabolic disease by activating SIRT1 and PGC-1α. Cell 127 (6), 1109–1122.
Lamming, D.W., Ye, L., Katajisto, P., Goncalves, M.D., Saitoh, M., Stevens, D.M., Davis, J.G., Salmon, A.B., Richardson, A., Ahima, R.S., 2012. Rapamycin-induced insulin resistance is mediated by mTORC2 loss and uncoupled from longevity. Science 335 (6076), 1638–1643.
Lao, C.D., Ruffin, M.T. t., Normolle, D., Heath, D.D., Murray, S.I., Bailey, J.M., Boggs, M.E., Crowell, J., Rock, C.L., Brenner, D.E., 2006. Dose escalation of a curcuminoid formulation. BMC Complement. Altern. Med. 6, 10.
Leontieva, O.V., Demidenko, Z.N., Blagosklonny, M.V., 2015. Dual mTORC1/C2 inhibitors suppress cellular geroconversion (a senescence program). Oncotarget 6 (27), 23238–23248.
Leotoing, L., Wauquier, F., Guicheux, J., Miot-Noirault, E., Wittrant, Y., Coxam, V., 2013. The polyphenol fisetin protects bone by repressing NF-kappaB and MKP-1-dependent signaling pathways in osteoclasts. PLoS One 8 (7), e68388.
Levine, B., Mizushima, N., Virgin, H.W., 2011. Autophagy in immunity and inflammation. Nature 469 (7330), 323–335.
Lewis, S., 2014. Ageing: synaptic slide into old age. Nat. Rev. Neurosci. 15 (9), 566.
Li, J., Qu, W., Cheng, Y., Sun, Y., Jiang, Y., Zou, T., Wang, Z., Xu, Y., Zhao, H., 2014. The inhibitory effect of intravesical fisetin against bladder cancer by induction of p53 and down-regulation of NF-kappa B pathways in a rat bladder carcinogenesis model. Basic Clin. Pharmacol. Toxicol. 115 (4), 321–329.
Li, J., Ren, J., Liu, X., Jiang, L., He, W., Yuan, W., Yang, J., Dai, C., 2015. Rictor/mTORC2 signaling mediates TGFβ1-induced fibroblast activation and kidney fibrosis. Kidney Int. 88 (3), 515–527.
Li, M., Hu, G., Chen, Y., 2016. Evaluation of the antioxidant capacity of natural polyphenolic compounds using a macrocyclic Ni-(II) complex-catalysed Briggs–Rauscher reaction. Food Chem. 197, 987–991.
Lin, C.L., Huang, H.C., Lin, J.K., 2007. Theaflavins attenuate hepatic lipid accumulation through activating AMPK in human HepG2 cells. J. Lipid Res. 48 (11), 2334–2343.
Lin, X.L., Liu, M.H., Hu, H.J., Feng, H.R., Fan, X.J., Zou, W.W., Pan, Y.Q., Hu, X.M., Wang, Z., 2015. Curcumin enhanced cholesterol efflux by upregulating ABCA1 expression through AMPK-SIRT1-LXRalpha signaling in THP-1 macrophage-derived foam cells. DNA Cell. Biol. 34 (9), 561–572.
Lin, C.J., Chang, Y.A., Lin, Y.L., Liu, S.H., Chang, C.K., Chen, R.M., 2016. Preclinical effects of honokiol on treating glioblastoma multiforme via G1 phase arrest and cell apoptosis. Phytomedicine 23 (5), 517–527.
Liu, Z.P., Zhang, X.P., Cui, W.M., Zhang, X., Li, N., Chen, S.J., Wong, A.W., Roberts, A., 2007. Evaluation of short-term and subchronic toxicity of magnolia bark extract in rats. Regul. Toxicol. Pharmacol. 49 (3), 160–171.
Liu, Y., Wang, D., Yang, G.J., Shi, Q.S., Feng, F., 2016. Comparative pharmacokinetics and brain distribution of magnolol and honokiol after oral administration of Magnolia officinalis cortex extract and its compatibility with other herbal medicines in Zhi-Zi-Hou-Po decoction to rats. Biomed. Chromatogr. 30 (3), 369–375.
Lu, W., Kelly, A.L., Miao, S., 2016. Emulsion-based encapsulation and delivery systems for polyphenols. Trends Food Sci. Technol. 47, 1–9.
Maher, P., Salgado, K.F., Zivin, J.A., Lapchak, P.A., 2007. A novel approach to screening for new neuroprotective compounds for the treatment of stroke. Brain Res. 1173, 117–125.
Maiese, K., 2014a. Cutting through the complexities of mTOR for the treatment of stroke. Curr. Neurovasc. Res. 11 (2), 177–186.
Maiese, K., 2014b. Driving neural regeneration through the mammalian target of rapamycin. Neural Regener. Res. 9 (15), 1413–1417.
Maiese, K., 2014c. Taking aim at Alzheimer's disease through the mammalian target of rapamycin. Ann. Med. 46 (8), 587–596.
Maiese, K., 2015a. SIRT1 and stem cells: in the forefront with cardiovascular disease, neurodegeneration and cancer. World J. Stem Cells 7 (2), 235–242.
Maiese, K., 2015b. Targeting molecules to medicine with mTOR, autophagy, and neurodegenerative disorders. Br. J. Clin. Pharmacol., http://dx.doi.org/10.1111/bcp.12804.
Makino, T., Shimizu, R., Kanemaru, M., Suzuki, Y., Moriwaki, M., Mizukami, H., 2009. Enzymatically modified isoquercitrin, alpha-oligoglucosyl quercetin 3-*O*-glucoside, is absorbed more easily than other quercetin glycosides or aglycone after oral administration in rats. Biol. Pharm. Bull. 32 (12), 2034–2040.
Mannick, J.B., Del Giudice, G., Lattanzi, M., Valiante, N.M., Praestgaard, J., Huang, B., Lonetto, M.A., Maecker, H.T., Kovarik, J., Carson, S., 2014. mTOR inhibition improves immune function in the elderly. Sci Transl. Med. 6 (268), 268ra179.
McCormick, M.A., Tsai, S.Y., Kennedy, B.K., 2011. TOR and ageing: a complex pathway for a complex process. Philos. Trans. R Soc. Lond. B Biol. Sci. 366 (1561), 17–27.
Mehrjerdi, F.Z., Aboutaleb, N., Habibey, R., Ajami, M., Soleimani, M., Arabian, M., Niknazar, S., Davoodi, S.H., Pazoki-Toroudi, H., 2013. Increased phosphorylation of mTOR is involved in remote ischemic preconditioning of hippocampus in mice. Brain Res. 1526, 94–101.
Mercado, N., Ito, K., Barnes, P.J., 2015. Accelerated ageing of the lung in COPD: new concepts. Thorax: Thoraxjnl-, 2014–206084.
Miles, J.M., Rule, A.D., Borlaug, B.A., 2014. Use of metformin in diseases of aging. Curr. Diabetes Rep. 14 (6), 1–9.
Minino, A.M., Murphy, S.L., 2012. Death in the United States, 2010. NCHS Data Brief 99, 1–8.
Mondal, S.C., Singh, P., Kumar, B., Singh, S.K., Gupta, S.K., Verma, A., 2014. Ageing and potential anti-aging phytochemicals: an overview. World J. Pharm. Pharm. Sci. 4, 426–454.
Montesanto, A., Geracitano, S., Garasto, S., Fusco, S., Lattanzio, F., Passarino, G., Corsonello, A., 2016. The impact of the emerging genomics data on the management of agerelated phenotypes in the context of cellular senescence. Curr. Drug Targets 17 (4), 428–438.
Moore, M.N., 2015. Do airborne biogenic chemicals interact with the PI3K/Akt/mTOR cell signalling pathway to benefit human health and wellbeing in rural and coastal environments? Environ. Res. 140, 65–75.
Morselli, E., Maiuri, M.C., Markaki, M., Megalou, E., Pasparaki, A., Palikaras, K., Criollo, A., Galluzzi, L., Malik, S.A., Vitale, I., 2010. The life span-prolonging effect of sirtuin-1 is mediated by autophagy. Autophagy 6 (1), 186–188.
Mourouti, N., Panagiotakos, D.B., 2016. The beneficial effect of a Mediterranean diet supplemented with extra virgin olive oil in the primary prevention of breast cancer among women at high cardiovascular risk in the PREDIMED Trial. Evid. Based Nurs., http://dx.doi.org/10.1136/ebnurs-2016-102303.
Munné-Bosch, S., 2015. Senescence: is it universal or not? Trends Plant Sci. 20 (11), 713–720.
Murtaza, I., Adhami, V.M., Hafeez, B.B., Saleem, M., Mukhtar, H., 2009. Fisetin, a natural flavonoid, targets chemoresistant human pancreatic cancer AsPC-1

cells through DR3-mediated inhibition of NF-kappaB. Int. J. Cancer 125 (10), 2465–2473.
Na, H.J., Park, J.S., Pyo, J.H., Jeon, H.J., Kim, Y.S., Arking, R., Yoo, M.A., 2015. Metformin inhibits age-related centrosome amplification in Drosophila midgut stem cells through AKT/TOR pathway. Mech. Ageing Dev. 149, 8–18.
Nie, C., Zhou, J., Qin, X., Shi, X., Zeng, Q., Liu, J., Yan, S., Zhang, L., 2016. Reduction of apoptosis by proanthocyanidin-induced autophagy in the human gastric cancer cell line MGC-803. Oncol. Rep. 35 (2), 649–658.
O'Neill, C., Kiely, A., Coakley, M., Manning, S., LongSmith, C., 2012. Insulin and IGF-1 signalling: longevity, protein homoeostasis and Alzheimer's disease. Biochem. Soc. Transact. 40 (4), 721.
Pallauf, K., Giller, K., Huebbe, P., Rimbach, G., 2013. Nutrition and healthy ageing: calorie restriction or polyphenol-rich MediterrAsian diet? Oxid. Med. Cell. Longevity 2013, 707421.
Park, S.J., Ahmad, F., Philp, A., Baar, K., Williams, T., Luo, H., Ke, H., Rehmann, H., Taussig, R., Brown, A.L., Kim, M.K., Beaven, M.A., Burgin, A.B., Manganiello, V., Chung, J.H., 2012. Resveratrol ameliorates aging-related metabolic phenotypes by inhibiting cAMP phosphodiesterases. Cell 148 (3), 421–433.
Passtoors, W.M., van den Akker, E.B., Deelen, J., Maier, A.B., van der Breggen, R., Jansen, R., Trompet, S., van Heemst, D., Derhovanessian, E., Pawelec, G., 2015. IL7R gene expression network associates with human healthy ageing. Immun. Ageing 12 (1), 1–9.
Pegoraro, G., Kubben, N., Wickert, U., Göhler, H., Hoffmann, K., Misteli, T., 2009. Ageing-related chromatin defects through loss of the NURD complex. Nat. Cell Biol. 11 (10), 1261–1267.
Pergola, P.E., White, C.L., Szychowski, J.M., Talbert, R., del Brutto, O., Castellanos, M., Graves, J.W., Matamala, G., Pretell, E.J., Yee, J., Rebello, R., Zhang, Y., Benavente, O.R., Investigators, S., 2014. Achieved blood pressures in the secondary prevention of small subcortical strokes (SPS3) study: challenges and lessons learned. Am. J. Hypertens. 27 (8), 1052–1060.
Piao, M.J., Kim, K.C., Chae, S., Keum, Y.S., Kim, H.S., Hyun, J.W., 2013. Protective effect of fisetin (3,7,3',4'-tetrahydroxyflavone) against gamma-irradiation-induced oxidative stress and cell damage. Biomol. Ther. (Seoul) 21 (3), 210–215.
Prasath, G.S., Subramanian, S.P., 2013. Fisetin, a tetra hydroxy flavone recuperates antioxidant status and protects hepatocellular ultrastructure from hyperglycemia mediated oxidative stress in streptozotocin induced experimental diabetes in rats. Food Chem. Toxicol. 59, 249–255.
Prasath, G.S., Sundaram, C.S., Subramanian, S.P., 2013. Fisetin averts oxidative stress in pancreatic tissues of streptozotocin-induced diabetic rats. Endocrine 44 (2), 359–368.
Proshkina, E.N., Shaposhnikov, M.V., Sadritdinova, A.F., Kudryavtseva, A.V., Moskalev, A.A., 2015. Basic mechanisms of longevity: a case study of Drosophila pro-longevity genes. Ageing Res. Rev. 24, 218–231.
Ramos, F.J., Kaeberlein, M., 2012. Ageing: a healthy diet for stem cells. Nature 486 (7404), 477–478.
Riera, C.E., Dillin, A., 2015. Tipping the metabolic scales towards increased longevity in mammals. Nat. Cell Biol. 17 (3), 196–203.
Rigacci, S., Miceli, C., Nediani, C., Berti, A., Cascella, R., Pantano, D., Nardiello, P., Luccarini, I., Casamenti, F., Stefani, M., 2015. Oleuropein aglycone induces autophagy via the AMPK/mTOR signalling pathway: a mechanistic insight. Oncotarget 6 (34), 35344–35357.
Rockenfeller, P., Koska, M., Pietrocola, F., Minois, N., Knittelfelder, O., Sica, V., Franz, J., Carmona-Gutierrez, D., Kroemer, G., Madeo, F., 2015. Phosphatidylethanolamine positively regulates autophagy and longevity. Cell Death Different. 22 (3), 499–508.
Russo, M., Spagnuolo, C., Tedesco, I., Bilotto, S., Russo, G.L., 2012. The flavonoid quercetin in disease prevention and therapy: facts and fancies. Biochem. Pharmacol. 83 (1), 6–15.
Sahin, E., DePinho, R.A., 2010. Linking functional decline of telomeres, mitochondria and stem cells during ageing. Nature 464 (7288), 520–528.
Sahu, B.D., Kumar, J.M., Sistla, R., 2016. Fisetin, a dietary flavonoid, ameliorates experimental colitis in mice: relevance of NF-kappaB signaling. J. Nutr. Biochem. 28, 171–182.
Sakaue, Y., Domon, H., Oda, M., Takenaka, S., Kubo, M., Fukuyama, Y., Okiji, T., Terao, Y., 2016. Anti-biofilm and bactericidal effects of magnolia bark-derived magnolol and honokiol on Streptococcus mutans. Microbiol. Immunol. 60 (1), 10–16.
Schaffer, S., Asseburg, H., Kuntz, S., Muller, W.E., Eckert, G.P., 2012. Effects of polyphenols on brain ageing and Alzheimer's disease: focus on mitochondria. Mol. Neurobiol. 46 (1), 161–178.
Schreiber, K.H., Ortiz, D., Academia, E.C., Anies, A.C., Liao, C.Y., Kennedy, B.K., 2015. Rapamycin-mediated mTORC2 inhibition is determined by the relative expression of FK506-binding proteins. Aging cell 14 (2), 265–273.
Sengupta, S., Peterson, T.R., Laplante, M., Oh, S., Sabatini, D.M., 2010. mTORC1 controls fasting-induced ketogenesis and its modulation by ageing. Nature 468 (7327), 1100–1104.
Seo, B.R., Min, K.J., Cho, I.J., Kim, S.C., Kwon, T.K., 2016. Curcumin significantly enhances dual PI3K/Akt and mTOR inhibitor NVP-BEZ235-induced apoptosis in human renal carcinoma caki cells through down-regulation of p53-dependent bcl-2 expression and inhibition of mcl-1 protein stability (vol 9, e95588, 2014). PLoS One 11 (3).
Sharoar, M., Shi, Q., Ge, Y., He, W., Hu, X., Perry, G., Zhu, X., Yan, R., 2015. Dysfunctional tubular endoplasmic reticulum constitutes a pathological feature of Alzheimer's disease. Mol. Psychiatry., http://dx.doi.org/10.1038/mp.2015.181.
Shen, Y., Zhang, H., Cheng, L., Wang, L., Qian, H., Qi, X., 2016. *In vitro* and *in vivo* antioxidant activity of polyphenols extracted from black highland barley. Food Chem. 194, 1003–1012.
Shia, C.S., Tsai, S.Y., Kuo, S.C., Hou, Y.C., Chao, P.D., 2009. Metabolism and pharmacokinetics of 3,3',4',7-tetrahydroxyflavone (fisetin), 5-hydroxyflavone, and 7-hydroxyflavone and antihemolysis effects of fisetin and its serum metabolites. J. Agric. Food Chem. 57 (1), 83–89.
Sims, J.K., Wade, P.A., 2011. Mi-2/NuRD complex function is required for normal S phase progression and assembly of pericentric heterochromatin. Mol. Biol. Cell 22 (17), 3094–3102.
Slámová, K., Papoušek, F., Janovská, P., Kopecký, J., Kolář, F., 2015. Adverse effects of AMP-activated protein kinase alpha2-subunit deletion and high-fat diet on heart function and ischemic tolerance in aged female mice. Physiol. Res. 65 (1), 33–42.
Slack, C., Partridge, L., 2013. Genes, pathways and metabolism in ageing. Drug Discov. Today 10 (2), e87–e93.
Soleas, G.J., Yan, J., Goldberg, D.M., 2001. Ultrasensitive assay for three polyphenols (catechin, quercetin and resveratrol) and their conjugates in biological fluids utilizing gas chromatography with mass selective detection. J. Chromatogr. B 757 (1), 161–172.
Soukas, A.A., Kane, E.A., Carr, C.E., Melo, J.A., Ruvkun, G., 2009. Rictor/TORC2 regulates fat metabolism, feeding, growth, and life span in Caenorhabditis elegans. Genes Dev. 23 (4), 496–511.
Stahl, S., Branca, R.M., Efazat, G., Ruzzene, M., Zhivotovsky, B., Lewensohn, R., Viktorsson, K., Lehtio, J., 2011. Phosphoproteomic profiling of NSCLC cells reveals that ephrin B3 regulates pro-survival signaling through akt1-mediated phosphorylation of the EphA2 receptor. J. Proteome Res. 10 (5), 2566–2578.
Stenvinkel, P., Kooman, J.P., Shiels, P.G., 2016. Nutrients and ageing: what can we learn about ageing interactions from animal biology? Curr. Opin. Clin. Nutr. Metab. Care 19 (1), 19–25.
Stout, M.B., Evans, G.L., Jurczak, M.J., Zhu, Y., Xu, M., Palmer, A.K., Pirtskhalava, T., White, T.A., Singh, R.J., LeBrasseur, N.K., 2015. LB-OR03-3: alleviation of age-related metabolic dysfunction by 17α-estradiol: a novel therapeutic strategy. Endocr. Rev. 36 (2).
Suh, Y., Afaq, F., Khan, N., Johnson, J.J., Khusro, F.H., Mukhtar, H., 2010. Fisetin induces autophagic cell death through suppression of mTOR signaling pathway in prostate cancer cells. Carcinogenesis 31 (8), 1424–1433.
Suh, K.S., Chon, S., Choi, E.M., 2016. Protective effects of honokiol against methylglyoxal-induced osteoblast damage. Chem. Biol. Interact. 244, 169–177.
Syed, D.N., Adhami, V.M., Khan, M.I., Mukhtar, H., 2013. Inhibition of Akt/mTOR signaling by the dietary flavonoid fisetin. Anti-Cancer Agents Med. Chem. 13 (7), 995–1001.
Syed, D.N., Chamcheu, J.C., Khan, M.I., Sechi, M., Lall, R.K., Adhami, V.M., Mukhtar, H., 2014. Fisetin inhibits human melanoma cell growth through direct binding to p70S6K and mTOR: findings from 3-D melanoma skin equivalents and computational modeling. Biochem. Pharmacol. 89 (3), 349–360.
Thoreen, C.C., Chantranupong, L., Keys, H.R., Wang, T., Gray, N.S., Sabatini, D.M., 2012. A unifying model for mTORC1-mediated regulation of mRNA translation. Nature 485 (7396), 109–113.
Tunc, S.Y., Agacayak, E., Goruk, N.Y., Icen, M.S., Turgut, A., Alabalik, U., Togrul, C., Ekinci, C., Ekinci, A., Gul, T., 2016. Protective effects of honokiol on ischemia/reperfusion injury of rat ovary: an experimental study. Drug Des. Dev. Ther. 10, 1077–1083.
Unruhe, B., Schroder, E., Wunsch, D., Knauer, S.K., 2016. An old flame never dies: survivin in cancer and cellular senescence. Gerontology 62 (2), 173–181.
Vicente, R., Mausset-Bonnefont, A.L., Jorgensen, C., Louis-Plence, P., Brondello, J.M., 2016. Cellular senescence impact on immune cell fate and function. Aging Cell 15 (3), 400–406.
Vilchez, D., Saez, I., Dillin, A., 2014. The role of protein clearance mechanisms in organismal ageing and age-related diseases. Nat. Commun. 5, 5659.
Vissers, M.N., Zock, P.L., Katan, M.B., 2004. Bioavailability and antioxidant effects of olive oil phenols in humans: a review. Eur. J. Clin Nutr. 58 (6), 955–965.
Wang, X.H., Duan, X.M., Yang, G.L., Zhang, X.Y., Deng, L.Y., Zheng, H., Deng, C.Y., Wen, J.L., Wang, N., Peng, C., Zhao, X., Wei, Y.Q., Chen, L.J., 2011. Honokiol crosses BBB and BCSFB, and inhibits brain tumor growth in rat 9L intracerebral gliosarcoma model and human U251 xenograft glioma model. PLoS One 6 (4), e18490.
Wang, S., Su, R., Nie, S., Sun, M., Zhang, J., Wu, D., Moustaid-Moussa, N., 2014. Application of nanotechnology in improving bioavailability and bioactivity of diet-derived phytochemicals. J. Nutr. Biochem. 25 (4), 363–376.
Wang, S., Liu, X., Sun, Z., 2015. MicroRNA-150 knockout (miR150-KO) prevents age-Related glucose intolerance and extends life-span. FASEB J. 29 (1 Supplement), 996–999.
Wang, Y., Chen, B., Longtine, M.S., Nelson, D.M., 2016a. Punicalagin promotes autophagy to protect primary human syncytiotrophoblasts from apoptosis. Reproduction 151 (2), 97–104.
Wang, Y., Jiang, L.L., Wu, J.F., Liu, Z., 2016b. Protective effect of Honokiol against endometriosis in rats via attenuating survivin and Bcl-2: a mechanistic study. Cell. Mol. Biol. 62 (1), 1–5.
Warr, M.R., Binnewies, M., Flach, J., Reynaud, D., Garg, T., Malhotra, R., Debnath, J., Passegué, E., 2013. FOXO3A directs a protective autophagy program in haematopoietic stem cells. Nature 494 (7437), 323–327.
Watanabe, M., Hisatake, M., Fujimori, K., 2015. Fisetin suppresses lipid accumulation in mouse adipocytic 3T3-L1Cells by repressing GLUT4-mediated glucose uptake through inhibition of mTOR-C/EBP alpha signaling. J. Agric. Food Chem. 63 (20), 4979–4987.

Wee, L.H., Morad, N.A., Aan, G.J., Makpol, S., Ngah, W.Z.W., Yusof, Y.A.M., 2015. Mechanism of chemoprevention against colon cancer cells using combined gelam honey and ginger extract via mTOR and Wnt/β-catenin pathways. Asian Pacific J. Cancer Prev. 16 (15), 6549–6556.

White, R.R., Milholland, B., de Bruin, A., Curran, S., Laberge, R.M., van Steeg, H., Campisi, J., Maslov, A.Y., Vijg, J., 2015. Controlled induction of DNA double-strand breaks in the mouse liver induces features of tissue ageing. Nat. Commun. 6, 6790.

Wrighton, K.H., 2011. Ageing: staying alive without CRTC-1. Nat. Rev. Mol. Cell Biol 12 (4), 206–207.

Wu, X.N., Chen, X.G., Hu, Z.D., 2003. High-performance liquid simultaneous determination chromatographic method for of honokiol and magnolol in rat plasma. Talanta 59 (1), 115–121.

Wu, M.Y., Hung, S.K., Fu, S.L., 2011. Immunosuppressive effects of fisetin in ovalbumin-induced asthma through inhibition of NF-kappaB activity. J. Agric. Food Chem. 59 (19), 10496–10504.

Xu, L., Wang, S., Li, B., Sun, A., Zou, Y., Ge, J., 2015. A protective role of ciglitazone in ox-LDL-induced rat microvascular endothelial cells via modulating PPAR γ-dependent AMPK/eNOS pathway. J. Cell. Mol. Med. 19 (1), 92–102.

Yang, H., Rudge, D.G., Koos, J.D., Vaidialingam, B., Yang, H.J., Pavletich, N.P., 2013. mTOR kinase structure, mechanism and regulation. Nature 497 (7448), 217–223.

Yao, K., Zhang, L., Zhang, Y., Ye, P., Zhu, N., 2008. The flavonoid, fisetin, inhibits UV radiation-induced oxidative stress and the activation of NF-kappaB and MAPK signaling in human lens epithelial cells. Mol. Vis. 14, 1865–1871.

Yeh, P.S., Wang, W., Chang, Y.A., Lin, C.J., Wang, J.J., Chen, R.M., 2016. Honokiol induces autophagy of neuroblastoma cells through activating the PI3K/Akt/mTOR and endoplasmic reticular stress/ERK1/2 signaling pathways and suppressing cell migration. Cancer Lett. 370 (1), 66–77.

Yeh, C.K., 2016. Cellular senescence and aging. Oral Dis., http://dx.doi.org/10.1111/odi.12483.

Yu, Y.W., Li, M.X., Su, N., Zhang, Z.Y., Zhao, H.D., Yu, H., Xu, Y.L., 2016. Honokiol protects against renal ischemia/reperfusion injury via the suppression of oxidative stress, iNOS, inflammation and STAT3 in rats. Mol. Med. Rep. 13 (2), 1353–1360.

Yuan, Z.P., Chen, L.J., Fan, L.Y., Tang, M.H., Yang, G.L., Yang, H.S., Du, X.B., Wang, G.Q., Yao, W.X., Zhao, Q.M., Ye, B., Wang, R., Diao, P., Zhang, W., Wu, H.B., Zhao, X., Wei, Y.Q., 2006. Liposomal quercetin efficiently suppresses growth of solid tumors in murine models. Clin. Cancer Res. 12 (10), 3193–3199.

Zhang, Y., Nicholatos, J., Dreier, J.R., Ricoult, S.J., Widenmaier, S.B., Hotamisligil, G.S., Kwiatkowski, D.J., Manning, B.D., 2014. Coordinated regulation of protein synthesis and degradation by mTORC1. Nature 513 (7518), 440–443.

Zhang, L., Shamaladevi, N., Jayaprakasha, G., Patil, B., Lokeshwar, B., 2015. Polyphenol-rich extract of *Pimenta dioica* berries (Allspice) kills breast cancer cells by autophagy and delays growth of triple negative breast cancer in athymic mice. Oncotarget 6 (18), 16379–16395.

Zhang, Y., 2011. Biology of the Mi-2/NuRD complex in SLAC (stemness, longevity/ageing, and cancer). Gene Regul. Syst. Biol. 5, 1–26.

Zhang, Y., 2012. Ageing and cancer: breaking the don't put all eggs in one basket and natural self-organisation, and their potential reprogramming via modulation of mi-2/NuRD, mTOR kinase and metabolism. Enz. Eng. 2, 109.

Zhao, G.M., Han, X.D., Zheng, S.W., Li, Z., Sha, Y., Ni, J., Sun, Z., Qiao, S., Song, Z.Q., 2016. Curcumin induces autophagy, inhibits proliferation and invasion by downregulating AKT/mTOR signaling pathway in human melanoma cells. Oncol. Rep. 35 (2), 1065–1074.

Zhou, C.H., Wang, C.X., Xie, G.B., Wu, L.Y., Wei, Y.X., Wang, Q., Zhang, H.S., Hang, C.H., Zhou, M.L., Shi, J.X., 2015. Fisetin alleviates early brain injury following experimental subarachnoid hemorrhage in rats possibly by suppressing TLR 4/NF-kappaB signaling pathway. Brain Res. 1629, 250–259.

Zhu, Y., Tchkonia, T., Pirtskhalava, T., Gower, A.C., Ding, H., Giorgadze, N., Palmer, A.K., Ikeno, Y., Hubbard, G.B., Lenburg, M., O'Hara, S.P., LaRusso, N.F., Miller, J.D., Roos, C.M., Verzosa, G.C., LeBrasseur, N.K., Wren, J.D., Farr, J.N., Khosla, S., Stout, M.B., McGowan, S.J., Fuhrmann-Stroissnigg, H., Gurkar, A.U., Zhao, J., Colangelo, D., Dorronsoro, A., Ling, Y.Y., Barghouthy, A.S., Navarro, D.C., Sano, T., Robbins, P.D., Niedernhofer, L.J., Kirkland, J.L., 2015. The Achilles' heel of senescent cells: from transcriptome to senolytic drugs. Aging Cell 14 (4), 644–658.

Zoncu, R., Efeyan, A., Sabatini, D.M., 2011. mTOR: from growth signal integration to cancer, diabetes and ageing. Nat. Rev. Mol. Cell Biol. 12 (1), 21–35.

de Bock, M., Thorstensen, E.B., Derraik, J.G.B., Henderson, H.V., Hofman, P.L., Cutfield, W.S., 2013. Human absorption and metabolism of oleuropein and hydroxytyrosol ingested as olive (*Olea europaea* L.) leaf extract. Mol. Nutr. Food Res. 57 (11), 2079–2085.

de Lange, P., Cioffi, F., Silvestri, E., Moreno, M., Goglia, F., Lanni, A., 2013. (Healthy) ageing: focus on iodothyronines. Int. J. Mol. Sci. 14 (7), 13873–13892.

de Oliveira, M.R., Nabavi, S.M., Braidy, N., Setzer, W.N., Ahmed, T., Nabavi, S.F., 2015. Quercetin and the mitochondria: a mechanistic view. Biotechnol. Adv., http://dx.doi.org/10.1016/j.biotechadv.2015.12.014.