

Moreover, several studies have shown that the lifespan-extending effect of resveratrol is mediated by sirtuin. Both in worms and flies, resveratrol failed to extend the lifespan due to the lack of a functional Sir2—a major member of sirtuin associated with longevity and anti-aging effects.^{17,18,22}

Although it has been attested that sirtuin activation is involved in the longevity benefit of resveratrol *in vivo*, whether resveratrol directly binds to and activates sirtuin is controversial since the allosteric interaction of resveratrol with SIRT1 has been challenged. In the first report that showed the longevity effect of resveratrol in yeast, authors used a fluorescence-conjugated peptide substrate, Fluor-de-Lys, to show the binding and activation of sirtuin by resveratrol.¹⁷ However, such an outcome was not reproducible in subsequent studies that used other peptides. Resveratrol failed to activate SIRT1 when *in vitro* full-length endogenous substrates or short, fluorescence-unconjugated peptide substrates were used.^{63,71,72} Therefore, a possibility of other potential pathways has been suggested, such as the activation of AMPK or other unknown pathways.^{8,73} Conversely, several studies confirmed the initial finding. In 2013, Hubbard *et al.* identified the allosteric binding site of SIRT1 with resveratrol.⁷⁴ They showed that when glutamate at position 230 was substituted for lysine or alanine, the activation of SIRT1 by resveratrol was attenuated regardless of the substrates used.⁷⁴ In addition, the substrate sequence was reported to affect the activation of SIRT1 by resveratrol.⁷⁵ In summary, although whether resveratrol directly binds to and activates SIRT1 remains to be clarified, the requirement of sirtuin activation for the lifespan-extending effects of resveratrol seems to be obvious.

13.2.4.3 Autophagy

Autophagy is an evolutionarily conserved lysosomal degradation process for old, supernumerary, and damaged cytoplasmic components, including proteins, lipids, organelles, and membranes, contributing eukaryotic cellular homeostasis. Moreover, several pieces of evidence show that autophagy is involved in organismal survival. The induction of autophagy has been reported to improve lifespan and reduce age-associated mortality,^{75–77} and the lifespan-extending effect of rapamycin and spermidine has been shown to be mediated by autophagy in *C. elegans* and *D. melanogaster*.^{76,78} In addition, the autophagy processes within the central nervous system have been shown to be particularly important for determining longevity, since brain-specific overexpression of *Atg8* extends the longevity of *Drosophila* by 50%.^{79,80}

Recently, autophagy and its related signaling pathways, including the mTOR pathway, were reported to contribute to the beneficial effects of resveratrol.⁸¹ Induction of autophagy by resveratrol has been well established thus far, although controversial evidence showed that autophagy induced by nutrient starvation or rapamycin treatment was suppressed by resveratrol.⁸² 100 μ M resveratrol induced autophagy indicated by the accumulation of LC3B-II and LC3 puncta through the inhibition of mTOR signaling in GFP-LC3-expressing HeLa Cells.⁸³ In addition, resveratrol can induce