



# SGLT2 Inhibitors Produce Cardiorenal Benefits by Promoting Adaptive Cellular Reprogramming to Induce a State of Fasting Mimicry: A Paradigm Shift in Understanding Their Mechanism of Action

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There is compelling evidence that sodium–glucose cotransporter 2 (SGLT2) inhibitors exert cardioprotective and renoprotective effects that are far greater than expected based on their effects on glycemia or glycosuria. In large-scale randomized controlled trials, SGLT2 inhibitors reduce the risk of hospitalizations for heart failure by ~30% and often decrease the risk of cardiovascular death (1). This benefit is particularly striking in patients who have the most marked impairment of systolic function prior to treatment. In parallel, SGLT2 inhibitors also reduce the risk of end-stage renal events, including the occurrence of renal death and the need for dialysis or renal transplantation by ~30% (2). This benefit is seen even when glomerular filtration rates are sufficiently low to abolish the glycosuric effect of these drugs.

These cardiorenal benefits cannot be explained by an action of SGLT2 inhibitors to lower blood glucose, since similar effects are not seen with antidiabetes drugs that have greater antihyperglycemic actions. Additionally, they cannot be ascribed to a natriuretic action, since these drugs exert only a modest effect on plasma volume or on circulating natriuretic peptides (3).

In 2016, two articles published in *Diabetes Care* proposed the “thrifty fuel hypothesis” to explain the cardiorenal benefits of SGLT2 inhibitors. Ferrannini et al. (4) and Mudaliar et al. (5) postulated that the action of SGLT2 inhibitors to promote ketogenesis might account for their favorable effects on the heart and kidney because enhanced ketone bodies formation might possibly provide an efficient fuel that could boost the energy state of organs under stress. However, type 2 diabetes represents a state of nutrient overabundance and not energy deprivation, as is evidenced by the enhancement of molecular pathways that promote both energy retention (e.g., through SGLT2 upregulation) and energy storage (6,7). In fact, the stressed heart already preferentially utilizes ketone bodies, and the diabetic kidney is a ketogenic organ (8,9). Therefore, it is not clear that SGLT2 inhibitors can augment ketone body utilization in a manner that improves ATP generation (10). More concerning, ketonemia leads to glomerular hyperfiltration (11), and ketogenesis has been proposed as a contributor to the development of diabetic nephropathy and renal fibrosis (12).

In light of these inconsistencies, it is noteworthy that, in this issue of *Diabetes Care*, Avogaro et al. (13) propose precisely the opposite hypothesis. Instead of postulating a drug-induced enhancement of fuel supply, the authors suggest that SGLT2 inhibitors induce a “dormancy state” that mimics starvation, i.e., these drugs recapitulate the conditions seen in hibernating animals. How is it possible that our conceptual framework of SGLT2 inhibitors has been turned upside down in only a few years? To comprehend the basis for this paradigm shift, it is important to understand the molecular mechanisms by which the cellular response to nutrient availability determines the health of vital organs.

## Activation of SIRT1 and AMPK Signaling as an Adaptive Response to Starvation and Cellular Stress

When cells are stressed by starvation, they activate a transcriptional program that facilitates adaptation to low-nutrient conditions. The enzymes that play a critical role as low-energy sensors include sirtuin 1 (SIRT1) and adenosine monophosphate–activated protein kinase (AMPK). SIRT1 responds to levels of nicotinamide adenine dinucleotide and serves as a redox

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rheostat, whereas AMPK discerns the balance between ATP and AMP in the cytosol.

Importantly, SIRT1 and AMPK are activated not only by fasting but also by a broad range of cellular stresses, including hypoxia, reactive oxygen species, injured organelles, and misfolded proteins. In response, SIRT1 mutes oxidative stress by enhancing antioxidant activity, directly reduces the inflammatory response to oxygen free radicals, and decreases the lethality of oxidative stress (14). AMPK preserves mitochondrial function, thereby reducing the formation of reactive oxygen species, and it attenuates the resulting proinflammatory and proapoptotic response (15). These enzymes mediate and reinforce each other's actions (16), and they coordinately regulate the expression of hundreds of genes that play a crucial role in maintaining cellular homeostasis and survival (14,15). In addition, both enzymes facilitate organismal adaption to a low-energy state, inhibiting energy storage (glycogen synthesis and lipogenesis) while promoting energy utilization (fatty acid oxidation leading to ketonemia).

Perhaps more importantly, coordinated activation of SIRT1/AMPK signaling stimulates autophagy (17,18). Autophagy is a lysosome-mediated degradative pathway that removes potentially dangerous constituents and recycles cellular components (19). The recycling process degrades both glycogen and lipids, thereby replenishing the supply of ATP to energy-starved cells, but in addition, enhanced autophagic flux performs a critical housekeeping function. By clearing the cytosol of damaged mitochondria and other dysfunctional organelles, autophagy directly reduces oxidative and endoplasmic reticular stress and mutes the resulting inflammatory response that leads to cellular dysfunction and death, thereby preserving cellular homeostasis. As is the case for SIRT1/AMPK signaling, autophagy is induced by nutrient, oxidative, and other forms of cellular stress. Excessive autophagy is prevented by stimulation of the PI3K/Akt/mTOR pathway (19), whose actions oppose those of SIRT1/AMPK.

**Suppression of SIRT1/AMPK Signaling and Autophagy in Type 2 Diabetes**

Since SIRT1/AMPK signaling is activated in states of nutrient deprivation, it is not surprising that these pathways are suppressed in states of energy overabundance.

Since type 2 diabetes is perceived by cells as a state of nutrient excess, it is accompanied by suppression of SIRT1 and AMPK as well as of autophagy (20–22). In striking contrast, autophagic flux is increased in type 1 diabetes, potentially because it is the hyperinsulinemia of type 2 diabetes—rather than the hyperglycemia—that causes downregulation of autophagy under conditions of glucose intolerance (20). In fact, insulin itself suppresses autophagic flux, potentially by activating Akt/mTOR signaling (23).

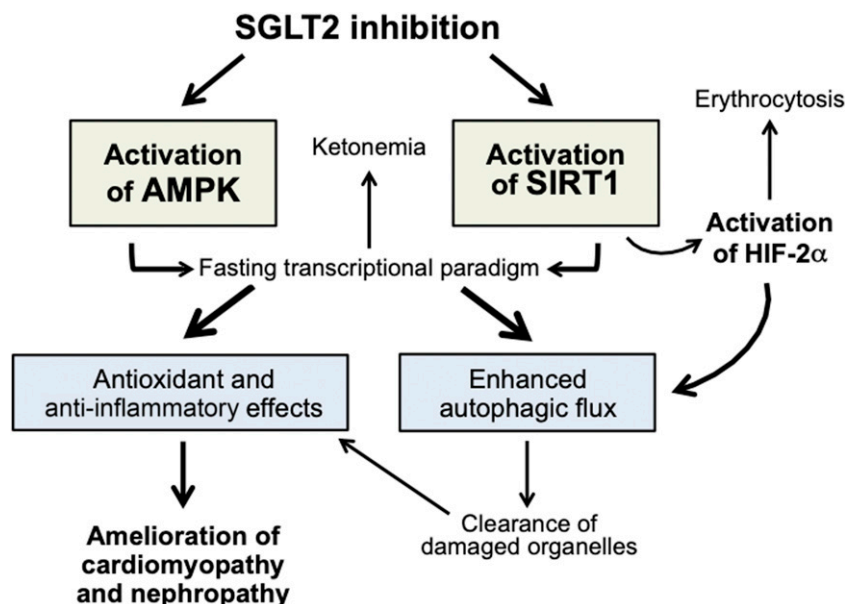
Impairment of SIRT1/AMPK signaling and autophagic flux has been implicated in the pathogenesis of cardiac and renal disease in type 2 diabetes. Specifically, a deficiency of SIRT1/AMPK activation and impaired autophagy appears to promote the development of cardiomyocyte dysfunction as well as inflammatory processes in diabetic hearts (20,24). In parallel, suppression of SIRT1/AMPK signaling and autophagy has been evoked to explain the pathogenesis of the glomerular and tubular lesions in diabetic nephropathy, particularly the podocyte injury that can lead to proteinuria and the derangements of proximal and distal tubular function that have been linked to advanced glycation end products (21,22,25).

Intriguingly, the sodium–glucose transporter (SGLT2) is activated by environmental glucose, and thus it acts as a sensor of energy overabundance.

Cellular homeostasis favors a counterbalance between SGLT2 and the activation of enzymes that are sensitive to nutrient deprivation. Accordingly, there exists an inverse relationship between the activity of SGLT2 and the intensity of proautophagic signaling in the kidney. High levels of renal tubular glucose promote the expression of SGLT2 but reduce the activation of SIRT1 (26). This intriguing inverse relationship supports the possibility that pharmacological modulation of SGLT2 may represent an effective entry point for regulating the activity of energy/redox sensing molecules that can stimulate autophagic flux and mute cellular stress.

**SGLT2 Inhibitors Produce Cardioprotective and Renoprotective Benefits by Inducing a State of Fasting Mimicry**

It is therefore noteworthy that SGLT2 inhibitors induce a transcriptional paradigm that closely mimics the cellular response to starvation (Fig. 1) (27). These drugs activate SIRT1/AMPK and suppress Akt/mTOR signaling and, consequently, they can promote autophagy, independent of their effects on glucose or insulin (28–31). Importantly, the effect of SGLT2 to stimulate the activity of low-energy sensors is not mediated by interference with SGLT2 protein on an individual cellular level, since it is seen in organs that do not express SGLT2 (30,32).



**Figure 1**—Induction of fasting transcriptional paradigm by SGLT2 inhibitors underlies their action to reduce heart failure and serious adverse renal events.

The actions of SGLT2 inhibitors to activate SIRT1/AMPK and suppress Akt/mTOR signaling lead to a reduction in oxidative stress, normalized mitochondrial structure and function, suppression of inflammation, minimization of coronary microvascular injury, enhanced contractile performance, and attenuation of the development of cardiomyopathy (33). The enhanced interplay of AMPK and SIRT1 also underlies the effect of SGLT2 inhibitors to ameliorate glomerular and tubular injury and inflammation and to mitigate the development of nephropathy (34). An increase in AMPK and SIRT1 may also directly enhance tubuloglomerular feedback, thereby linking changes in the activity of these energy sensors to the action of SGLT2 inhibitors to ameliorate glomerular hyperfiltration (35,36). Finally, activation of AMPK/SIRT1 and autophagic flux has been linked to downregulation of ion exchangers that have been implicated in the pathogenesis of diabetic cardiac and renal disease (37,38).

#### Clinical Evidence Supporting the Proposed Conceptual Framework

Three lines of clinical evidence support the hypothesis that SGLT2 inhibitors exert their effects by the activation of low-energy sensors, which are responsible for mimicking a fasting transcriptional paradigm.

First, SGLT2 inhibitors induce a loss of calories in the urine, and glycosuria is accompanied by a decrease of glucagon synthesis (often with the promotion of glycolysis), increased fatty acid oxidation, and the shrinkage of adipose tissue depots, including the alleviation of organ steatosis (27). Viewed from this perspective, the ketonemia seen with these drugs is not the source of an efficient fuel but instead is a biomarker of a fasting-like transcriptional state.

Second, SGLT2 inhibitors may not only deceive cells into believing that they are fasting but also that they are hypoxic. Oxygen deprivation (like nutrient deprivation) stimulates AMPK and SIRT1 (39,40). The latter activates hypoxia-inducible factor-2 $\alpha$  (HIF-2 $\alpha$ ) (41) and possibly also hypoxia-inducible factor-1 $\alpha$  (HIF-1 $\alpha$ ) under certain conditions (42,43); these represent the principal stimuli for erythropoietin synthesis. Thus, the erythrocytosis that is seen with SGLT2 inhibitors may represent a biomarker for enhanced SIRT1 signaling and its organ-protective effects (Fig. 1). Such a relationship may

explain why, in statistical mediation analyses, erythrocytosis has been the most powerful predictor of the action of SGLT2 inhibitors to reduce heart failure events in large-scale trials (44,45).

Third, metformin also stimulates autophagy, primarily by activating AMPK and SIRT1 and suppressing Akt/mTOR (46). Metformin exerts both cardioprotective and renoprotective effects in experimental models, and it favorably influences the evolution of heart failure and nephropathy in cohort studies. The overlap in the mechanism of action between metformin and SGLT2 inhibitors (with respect to AMPK/SIRT1 activation and autophagy) may explain why the magnitude of the benefit of SGLT2 inhibitors in large-scale trials may be attenuated in patients receiving metformin (47). However, metformin suppresses HIF-1 $\alpha$  (48), thus distinguishing its action from that of SGLT2 inhibitors; this effect may explain why metformin modestly decreases hematocrit, whereas SGLT2 inhibitors induce erythrocytosis. However, since both HIF-1 $\alpha$  and HIF-2 $\alpha$  appear to induce autophagy (49,50) in a manner than is independent of AMPK (39), it is possible that enhanced HIF-1 $\alpha$ /HIF-2 $\alpha$  signaling by SGLT2 inhibitors may amplify the autophagic flux that is already augmented by AMPK/SIRT1, thereby contributing importantly to the striking cardiorenal benefits of these drugs, which are not seen with other glucose-lowering agents.

The conceptual framework described in this Commentary provides a powerful molecular basis for the model described by Avogaro et al. (13) in the current issue. Their proposal for a “dormancy state” is analogous to the fasting transcriptional paradigm that is delineated in this Commentary. However, regardless of how their actions are envisioned, it is now critical for physicians to reconceptualize SGLT2 inhibitors as organ-protective agents rather than glucose-lowering drugs. The antihyperglycemic action of these drugs represents a tiny fraction of their broad portfolio of effects, which (when fully exercised) cause an adaptive reprogramming of stressed cells in a manner that promotes homeostasis and survival.

**Duality of Interest.** M.P. has recently consulted for AbbVie, Actavis, Akcea, Amgen, AstraZeneca, Boehringer Ingelheim, Cardiorientis, Daiichi Sankyo, Johnson & Johnson, Novo Nordisk, Pfizer, Relypsa,

Sanofi, Synthetic Biologics, and Theravance. No other potential conflicts of interest relevant to this article were reported.

#### References

- Zelniker TA, Wiviott SD, Raz I, et al. SGLT2 inhibitors for primary and secondary prevention of cardiovascular and renal outcomes in type 2 diabetes: a systematic review and meta-analysis of cardiovascular outcome trials. *Lancet* 2019; 393:31–39
- Neuen BL, Young T, Heerspink HJL, et al. SGLT2 inhibitors for the prevention of kidney failure in patients with type 2 diabetes: a systematic review and meta-analysis. *Lancet Diabetes Endocrinol* 2019;7:845–854
- Nassif ME, Windsor S, Tang F, et al.; DEFINE-HF Investigators. Dapagliflozin effects on biomarkers, symptoms, and functional status in patients with heart failure with reduced ejection fraction: the DEFINE-HF trial. *Circulation* 2019; 140:1463–1476
- Ferrannini E, Mark M, Mayoux E. CV protection in the EMPA-REG OUTCOME trial: a “thrifty substrate” hypothesis. *Diabetes Care* 2016;39:1108–1114
- Mudaliar S, Alloju S, Henry RR. Can a shift in fuel energetics explain the beneficial cardiorenal outcomes in the EMPA-REG OUTCOME study? A unifying hypothesis. *Diabetes Care* 2016;39:1115–1122
- Samuel VT, Shulman GI. The pathogenesis of insulin resistance: integrating signaling pathways and substrate flux. *J Clin Invest* 2016;126:12–22
- Wang XX, Levi J, Luo Y, et al. SGLT2 protein expression is increased in human diabetic nephropathy: SGLT2 protein inhibition decreases renal lipid accumulation, inflammation, and the development of nephropathy in diabetic mice. *J Biol Chem* 2017;292:5335–5348
- Aubert G, Martin OJ, Horton JL, et al. The failing heart relies on ketone bodies as a fuel. *Circulation* 2016;133:698–705
- Zhang D, Yang H, Kong X, et al. Proteomics analysis reveals diabetic kidney as a ketogenic organ in type 2 diabetes. *Am J Physiol Endocrinol Metab* 2011;300:E287–E295
- Verma S, Rawat S, Ho KL, et al. Empagliflozin increases cardiac energy production in diabetes: novel translational insights into the heart failure benefits of SGLT2 inhibitors. *JACC Basic Transl Sci* 2018;3:575–587
- Nosadini R, Trevisan R, Fioretto P, et al. Kidney hemodynamics after ketone body and amino acid infusion in normal and IDDM subjects. *Diabetes* 1989;38:75–83
- Guh JY, Chuang TD, Chen HC, et al. Beta-hydroxybutyrate-induced growth inhibition and collagen production in HK-2 cells are dependent on TGF- $\beta$  and Smad3. *Kidney Int* 2003;64: 2041–2051
- Avogaro A, Fadini GP, Del Prato S. Reinterpreting cardiorenal protection of renal sodium-glucose cotransporter 2 inhibitors via cellular life history programming. *Diabetes Care* 2019;43: 501–507
- Alcendor RR, Gao S, Zhai P, et al. Sirt1 regulates aging and resistance to oxidative stress in the heart. *Circ Res* 2007;100:1512–1521
- Meijles DN, Zoumpoulidou G, Markou T, et al. The cardiomyocyte “redox rheostat”: Redox signalling via the AMPK-mTOR axis and regulation of

- gene and protein expression balancing survival and death. *J Mol Cell Cardiol* 2019;129:118–129
16. Chang C, Su H, Zhang D, et al. AMPK-dependent phosphorylation of GAPDH triggers Sirt1 activation and is necessary for autophagy upon glucose starvation. *Mol Cell* 2015;60:930–940
17. Luo G, Jian Z, Zhu Y, et al. Sirt1 promotes autophagy and inhibits apoptosis to protect cardiomyocytes from hypoxic stress. *Int J Mol Med* 2019;43:2033–2043
18. Ren H, Shao Y, Wu C, Ma X, Lv C, Wang Q. Metformin alleviates oxidative stress and enhances autophagy in diabetic kidney disease via AMPK/SIRT1-FoxO1 pathway. *Mol Cell Endocrinol* 2020;500:110628
19. Levine B, Packer M, Codogno P. Development of autophagy inducers in clinical medicine. *J Clin Invest* 2015;125:14–24
20. Kanamori H, Takemura G, Goto K, et al. Autophagic adaptations in diabetic cardiomyopathy differ between type 1 and type 2 diabetes. *Autophagy* 2015;11:1146–1160
21. Szejder M, Piwkowska A. AMPK signalling: implications for podocyte biology in diabetic nephropathy. *Biol Cell* 2019;111:109–120
22. Kitada M, Kume S, Takeda-Watanabe A, Kanasaki K, Koya D. Sirtuins and renal diseases: relationship with aging and diabetic nephropathy. *Clin Sci (Lond)* 2013;124:153–164
23. Paula-Gomes S, Gonçalves DA, Baviera AM, Zanoni NM, Navegantes LC, Kettelhut IC. Insulin suppresses atrophy- and autophagy-related genes in heart tissue and cardiomyocytes through AKT/FOXO signaling. *Horm Metab Res* 2013;45:849–855
24. Karbasforooshan H, Karimi G. The role of SIRT1 in diabetic cardiomyopathy. *Biomed Pharmacother* 2017;90:386–392
25. Takahashi A, Takabatake Y, Kimura T, et al. Autophagy inhibits the accumulation of advanced glycation end products by promoting lysosomal biogenesis and function in the kidney proximal tubules. *Diabetes* 2017;66:1359–1372
26. Umino H, Hasegawa K, Minakuchi H, et al. High basolateral glucose increases sodium-glucose cotransporter 2 and reduces sirtuin-1 in renal tubules through glucose transporter-2 detection. *Sci Rep* 2018;8:6791
27. Osataphan S, Macchi C, Singhal G, et al. SGLT2 inhibition reprograms systemic metabolism via FGF21-dependent and -independent mechanisms. *JCI Insight* 2019;4:123130
28. Inoue MK, Matsunaga Y, Nakatsu Y, et al. Possible involvement of normalized Pin1 expression level and AMPK activation in the molecular mechanisms underlying renal protective effects of SGLT2 inhibitors in mice. *Diabetol Metab Syndr* 2019;11:57
29. Chang YK, Choi H, Jeong JY, et al. Dapagliflozin, SGLT2 inhibitor, attenuates renal ischemia-reperfusion injury. *PLoS One* 2016;11:e0158810
30. Aragón-Herrera A, Feijóo-Bandín S, Otero Santiago M, et al. Empagliflozin reduces the levels of CD36 and cardiotoxic lipids while improving autophagy in the hearts of Zucker diabetic fatty rats. *Biochem Pharmacol* 2019;170:113677
31. Mizuno M, Kuno A, Yano T, et al. Empagliflozin normalizes the size and number of mitochondria and prevents reduction in mitochondrial size after myocardial infarction in diabetic hearts. *Physiol Rep* 2018;6:e13741
32. Vallon V, Rose M, Gerasimova M, et al. Knockout of Na-glucose transporter SGLT2 attenuates hyperglycemia and glomerular hyperfiltration but not kidney growth or injury in diabetes mellitus. *Am J Physiol Renal Physiol* 2013;304:F156–F167
33. Zhou H, Wang S, Zhu P, Hu S, Chen Y, Ren J. Empagliflozin rescues diabetic myocardial microvascular injury via AMPK-mediated inhibition of mitochondrial fission. *Redox Biol* 2018;15:335–346
34. Terami N, Ogawa D, Tachibana H, et al. Long-term treatment with the sodium glucose cotransporter 2 inhibitor, dapagliflozin, ameliorates glucose homeostasis and diabetic nephropathy in db/db mice. *PLoS One* 2014;9:e100777
35. Udwan K, Abed A, Roth I, et al. Dietary sodium induces a redistribution of the tubular metabolic workload. *J Physiol* 2017;595:6905–6922
36. Huang DY, Gao H, Boini KM, Osswald H, Nürnberg B, Lang F. In vivo stimulation of AMP-activated protein kinase enhanced tubuloglomerular feedback but reduced tubular sodium transport during high dietary NaCl intake. *Pflugers Arch* 2010;460:187–196
37. Shi H, Zhao X, Ding Z, et al. Na<sup>+</sup>/H<sup>+</sup> exchanger regulates amino acid-mediated autophagy in intestinal epithelial cells. *Cell Physiol Biochem* 2017;42:2418–2429
38. Packer M. Activation and inhibition of sodium-hydrogen exchanger is a mechanism that links the pathophysiology and treatment of diabetes mellitus with that of heart failure. *Circulation* 2017;136:1548–1559
39. Li H, Satriano J, Thomas JL, et al. Interactions between HIF-1 $\alpha$  and AMPK in the regulation of cellular hypoxia adaptation in chronic kidney disease. *Am J Physiol Renal Physiol* 2015;309:F414–F428
40. Balaiya S, Khetpal V, Chalam KV. Hypoxia initiates sirtuin1-mediated vascular endothelial growth factor activation in choroidal endothelial cells through hypoxia inducible factor-2 $\alpha$ . *Mol Vis* 2012;18:114–120
41. Dioum EM, Chen R, Alexander MS, et al. Regulation of hypoxia-inducible factor 2 $\alpha$  signaling by the stress-responsive deacetylase sirtuin 1. *Science* 2009;324:1289–1293
42. Joo HY, Yun M, Jeong J, et al. SIRT1 deacetylates and stabilizes hypoxia-inducible factor-1 $\alpha$  (HIF-1 $\alpha$ ) via direct interactions during hypoxia. *Biochem Biophys Res Commun* 2015;462:294–300
43. Laemmle A, Lechleiter A, Roh V, et al. Inhibition of SIRT1 impairs the accumulation and transcriptional activity of HIF-1 $\alpha$  protein under hypoxic conditions. *PLoS One* 2012;7:e33433
44. Inzucchi SE, Zinman B, Fitchett D, et al. How does empagliflozin reduce cardiovascular mortality? Insights from a mediation analysis of the EMPA-REG OUTCOME trial. *Diabetes Care* 2018;41:356–363
45. Li JW, Woodward M, Perkovic V, et al. Mediators of the effects of canagliflozin on heart failure in patients with type 2 diabetes. *JACC Heart Fail* 2020;8:57–66
46. Chen SC, Brooks R, Houskeeper J, et al. Metformin suppresses adipogenesis through both AMP-activated protein kinase (AMPK)-dependent and AMPK-independent mechanisms. *Mol Cell Endocrinol* 2017;440:57–68
47. Rådholm K, Figtree G, Perkovic V, et al. Canagliflozin and heart failure in type 2 diabetes mellitus. *Circulation* 2018;138:458–468
48. Li X, Li J, Wang L, et al. The role of metformin and resveratrol in the prevention of hypoxia-inducible factor 1 $\alpha$  accumulation and fibrosis in hypoxic adipose tissue. *Br J Pharmacol* 2016;173:2001–2015
49. Schönenberger MJ, Krek W, Kovacs WJ. EPAS1/HIF-2 $\alpha$  is a driver of mammalian pexophagy. *Autophagy* 2015;11:967–969
50. Gui L, Liu B, Lv G. Hypoxia induces autophagy in cardiomyocytes via a hypoxia-inducible factor 1-dependent mechanism. *Exp Ther Med* 2016;11:2233–2239