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Review Article

Adenosine Monophosphate Activated Kinase (AMPK) the Breakthrough Target for Metabolic Syndrome

Abhishek B. Jha
Department of Pharmacology, Saraswati Institute of Pharmaceutical Sciences, Dhanap, Gandhinagar- **382355**, Gujarat, India

***Corresponding author**
Abhishek B. Jha
Email: infinite.ab62@gmail.com

Abstract: Metabolic syndrome is the challenging condition in current scenario in developing countries. To overcome it, the best target identification is the most challenging task. The enzyme Adenosine Monophosphate Activated Protein Kinase (AMPK) could be one of the targets to battle against such intricate conditions. The Adenosine Monophosphate Activated Protein Kinase (AMPK), a heterotrimeric serine/threonine protein kinase. Activated AMPK switches cells from an anabolic to a catabolic state, shutting down the ATP-consuming synthetic pathways and restoring energy balance, regulates key metabolic enzymes to reduce the activity of ATP-utilizing biosynthetic pathways and increase the activity of ATP-generating pathway. The fuel-sensing enzyme 5'-AMP-activated protein kinase (AMPK) has a major role in the regulation of cellular lipid and protein metabolism in response to stimuli such as exercise, changes in fuel availability and the adipocyte-derived hormones leptin and adiponectin. Abnormalities in cellular lipid metabolism are involved in the pathogenesis of the metabolic syndrome, possibly because of dysregulation of AMPK and Malonyl-CoA.It is now recognized that pharmacological activation of AMPK improves blood glucose homeostasis, lipid profile and blood pressure in insulin-resistant conditions. Indeed, AMPK activation mimics the beneficial effects of physical activity or those of calorie restriction by acting on multiple cellular targets. In addition, AMPK is one of the probable targets of major antidiabetic drugs including, the biguanides (metformin) and thiazolidinedione, as well as of insulin sensitizing adipokines (e.g., adiponectin). Taken together, such findings highlight the logic underlying the concept of targeting the AMPK pathway for the treatment of metabolic syndrome.
Keywords: Metabolic syndrome, AMPK, ACC-2, HMGCo-A Reductase, CPT.

INTRODUCTION

Metabolic syndrome is a complex disorder defined by a cluster of interconnected factors like, insulin resistance, hyperglycaemia, hypertension, low High Density Lipoprotein (HDL), and raised Very Low Density Lipoprotein (VLDL)-triglycerides. The term "metabolic" refers to the biochemical processes involved in the body's normal functioning. Risk factors are traits, conditions, or habits that increase your chance of developing a disease. In general, a person who has metabolic syndrome is twice as likely to develop heart disease and five times as likely to develop diabetes as someone who doesn't have metabolic syndrome [1-3]. It is labeled as syndrome-X [4]. It is estimated that 20%–25% of South Asians have developed Metabolic Syndrome and many more may be prone to it [3]. The five conditions described below are metabolic risk factors. There might be any one of these risk factors by itself but they tend to occur together.

- Abdominal obesity or Excess fat in the stomach area is a greater risk factor for heart disease than excess fat in other parts of the body, such as on the hips.
- A high triglyceride. Triglycerides are a type of fat found in the blood.
- A low HDL cholesterol level. HDL sometimes is called "good" cholesterol. This is because it helps remove cholesterol from your arteries. A low HDL cholesterol level raises risk for heart disease.
- High blood pressure. Blood pressure is the force of blood pushing against the walls of your arteries as your heart pumps blood. If this pressure rises and stays high over time, it can damage your heart and lead to plaque build-up.
- High fasting blood sugar. Mildly high blood sugar may be an early sign of diabetes. Increased Blood glucose level is associated with Diabetes related Hypertension, Glaucoma, Vascular damage, Atherosclerosis.

Other risk factors, besides those described above, also increase risk for heart disease. For example, a high LDL cholesterol level and smoking are major risk factors for heart disease, but they aren't part of metabolic syndrome. The risk of having metabolic syndrome is closely linked to overweight and obesity and a lack of physical activity. Insulin resistance also may increase your risk for metabolic syndrome. Insulin resistance is a condition in which the body can't use its insulin properly. Insulin is a hormone that helps move blood sugar into cells where it's used for energy. Insulin resistance can lead to high blood sugar levels, and it's closely linked to overweight and obesity. Genetics (ethnicity and family history) and older age are other factors that may play a role in causing metabolic syndrome.

ADENOSINE MONOPHOSPHATE ACTIVATED PROTEIN KINASE (AMPK)

The AMP-activated protein kinase (AMPK) is a heterotrimeric serine/threonine protein kinase consisting of the catalytic subunit (α) and 2 regulatory subunits (β and γ) that exist as multiple isoforms and splice variants, resulting in the generation of 12 possible heterotrimeric combinations. As its name suggests, the AMPK is activated in many different cell types by increased intracellular concentrations of AMP and is generally referred to as a "metabolite-sensing kinase." Indeed, the AMPK is activated following heat shock, vigorous exercise, hypoxia/ischemia, and starvation and appears to be a metabolic master switch, phosphorylating key target proteins that control flux through metabolic pathways of hepatic ketogenesis, cholesterol synthesis, lipogenesis, triglyceride synthesis, adipocyte lipolysis, skeletal muscle fatty acid oxidation, and protein synthesis [5, 6]. The AMPK determines whole body insulin sensitivity and may prevent insulin resistance, in part, by inhibiting pathways that antagonize insulin signaling. Through its effects on signaling, metabolism, and gene expression, the AMPK enhances insulin sensitivity and may reduce the risk of type 2 diabetes [7].

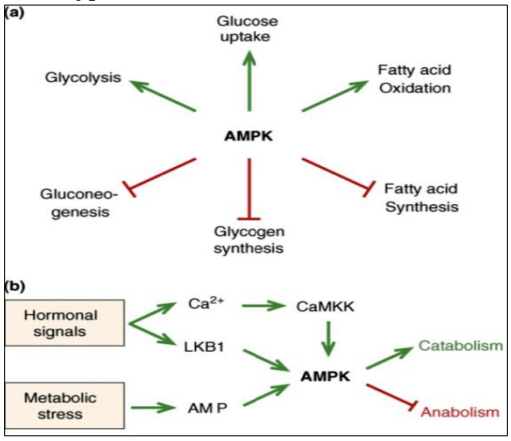

Fig. 1: AMPK and the regulation of intracellular and whole-bodymetabolism.

(a) Effects of AMPK activation on whole-body parameters related to diabetes. Glycolysis, glucose uptake and fatty acid oxidation are stimulated in multiple tissues in the body, while the biosynthesis of glucose, glycogen and lipids is inhibited. In the diabetic state, these effects conspire to improve reduced blood glucose levels and increase peripheral insulin sensitivity.

(b) The intracellular effects of AMPK activation. The kinase is activated by metabolic stresses, such as nutrient deprivation or by hormonal signals such as the antidiabetic hormone adiponectin. These signals produce an increase in intracellular AMP (which acts directly on AMPK), and/or activate upstream kinases that activate AMPK by direct phosphorylation. The activated kinase then phosphorylates numerous targets in the cytoplasm and nucleus to elicit changes in intracellular metabolism that reduce ATP consumption, increase ATP production and re-establish a proper energy balance in the cell.

AMPK ACTIVATION

Some chemicals like, Metformin, Berberin, Resveratrol, Thiazolidinediones, Cannabinoids, Adeponectin, Leptin, α-Lipoic Acid, Thiazolidinedione activate AMPK [8]. AMPK is activated by increase in the intracellular AMP: ATP ratio resulting from imbalance between ATP production and consumption. Activation of AMPK involves AMP binding to regulatory sites on the γ subunits. This causes conformational changes that allosterically activates the enzyme and inhibits Dephosphorylation of Thr172 within the activation loop of the catalytic α subunit. AMPK activation requires phosphorylation on Thr172 by upstream kinases, identified as the tumor suppressor serine-threonine kinase 11/ Liver Kinase B1 (STK11/LKB1) and Calmodulin Kinase Kinase (CaMKKβ), which is further stimulated by the allosteric activator AMP [9]. Moreover, it has been recently shown that the ADP: ATP ratio could also play a regulatory role on AMPK by binding to specific domains on the γ subunit [10, 11]. Drug such as Metformin inhibits mitochondrial respiratory chain complex-1 and AMP Deaminase, hence increased AMP: ATP ratio activates AMPK [12]. The γ -subunit of AMPK contains three AMP-binding sites, these are formed in the interface of its two pairs of Cystathionine beta synthase (CBS) domains, also called Bateman domains. CBS domains giving AMPK its ability to sensitively detect shifts in the AMP: ATP ratio. Two of these binding sites can bind either AMP or ATP, whereas a third site contains a tightly bound AMP that does not exchange. When AMPK is inactive under physiological conditions, it binds two ATP and one AMP molecule, while in low energy states it binds three AMP molecules. It is now proposed that the interaction of the catalytically active kinase domain with the AMP-bound γ -subunit protects the phosphorylated Thr172 residue from Dephosphorylation [13]. Metformin also

activates Liver Kinase B1 (LKB1) which activates AMPK which in turn reduces fatty acid synthesis [14]. Metformin inactivate enzyme AMP deaminase and prevent degradation of AMP to IMP (Inosine Mono Phosphate) and NH3. The enzyme AMPK needs to be phosphorylated, which is catalyzed principally by LKB1 in liver and muscle [15]. Activated AMPK switches cells from an anabolic to a catabolic state, shutting down the ATP-consuming synthetic pathways and restoring energy balance [9]. The activated kinase phosphorylates and regulates key metabolic enzymes to reduce the activity of ATP-utilizing biosynthetic pathways and increase the activity of ATP-generating pathway [16]. As a result, glucose, lipid and protein synthesis as well as cell growth are inhibited whereas fatty acid oxidation and glucose uptake are stimulated.

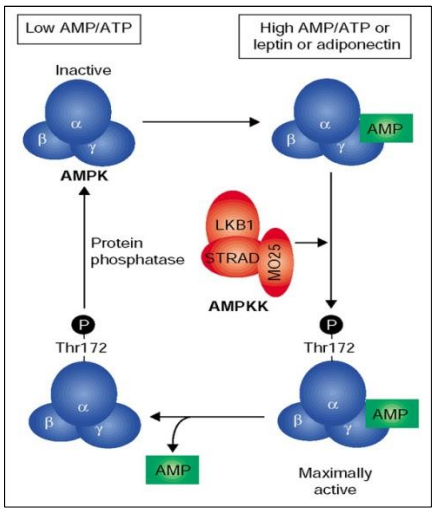

Fig. 2: Regulation of AMPK. AMPK (blue) becomes activated under conditions of high AMP/ATP (metabolic depletion), or in response to the hormones leptin and adiponectin. Under these circumstances, AMP binds to AMPK, facilitating phosphorylation at Thr172 and activation, in a reaction catalyzed by the LKB1-STRAD-MO25 complex (AMPKK; red). AMP also prevents dephosphorylation and deactivation of AMPK and serves as an allosteric activator of AMPK.

AMPK AND REGULATION OF CHOLESTEROL BIOSYNTHESIS

The AMPK system acts as a protective system for individual cells during metabolic stress by acting as a "fuel gauge". Once activated, AMPK initiates energy-saving and energy generating Systems. In this regard, AMPK phosphorylates and inhibits 3-hydroxy-3-methylglutaryl CoA (HMG- CoA) Reductase and Acetyl- CoA Carboxylase (ACC) [17]. As the Activated AMPK inhibits the HMG- CoA Reductase; a rate limiting enzyme for *De novo* biosynthesis of Cholesterol, inhibition of cholesterol bio synthesis leads to depletion of cholesterol and activation of SCAP (SREBP-cleavage activating protein)-SREBP (Sterol Regulatory Element Binding Protein) transportation activity. In inactive state, SREBP resides in the endoplasmic reticulum and associated with another transmembrane protein SCAP which provides conditional chaperon activity to SREBP. SCAP contains cholesterol sensing domain which responds to depletion of sterol with activation of SCAP-SREBP transporting activity. Under such condition, SCAP transports SREBP to the Golgi apparatus, where the N-terminal of transcription activation domain of the SREBP is released from the precursor protein through specific cleavage. The active form of SREBP trans-locates to the nucleus and binds to its cognate SRE-1(Sterol Regulatory Element-1) site and activates transcription of LDLR (Low Density Lipoprotein Receptor) gene. Increased hepatic LDLR Expression results in improved clearance of plasma LDL through receptor mediated endocytosis and this has been strongly associated with decreased risk of developing cardiovascular associated disease in Humans [18].

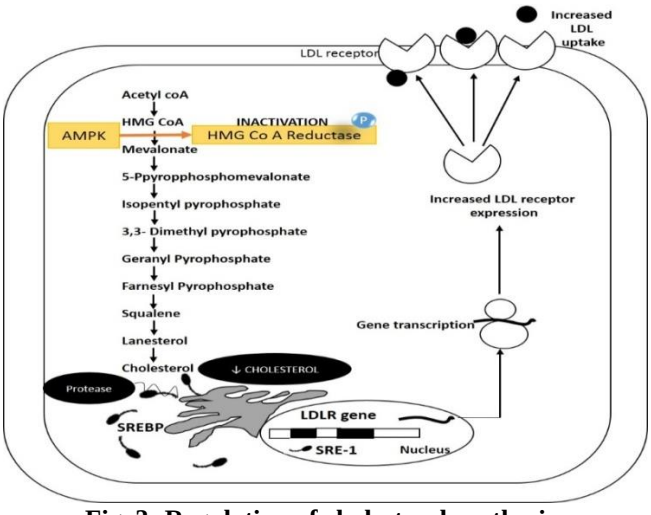

Fig. 3: Regulation of cholesterol synthesis.

Inhibition of HMG CoA Reductase reduces intracellular cholesterol levels, thus activating a protease, which in turn cleaves sterol regulatory element-binding proteins (SREBPs) from the endoplasmic reticulum. The SREBPs translocate to the nucleus where they up regulate expression of the LDL receptor gene. Enhanced LDL receptor expression increases receptor-mediated endocytosis of LDL and thus lowers serum LDL. Inhibition of HMG CoA Reductase also reduces intracellular levels of isoprenoids, which are intermediates in cholesterol biosynthesis.

AMPK AND FATTY ACID OXIDATION

Skeletal muscle is a dynamic tissue that preferentially utilizes fatty acids as a fuel source during postprandial conditions. Defects in skeletal muscle fatty acid oxidation contribute to the pathogenesis of insulin resistance and obesity [19], therefore an understanding of the signalling pathways mediating fatty acid oxidation may yield therapeutic targets for the treatment of insulin resistance and associated disorders. The AMP-activated protein kinase (AMPK) is thought to regulate fatty acid oxidation in response to energy demand, nutrients and hormones by directly phosphorylating the muscle-specific isoform of acetyl-CoA carboxylase-2 (ACC2) on Ser-221 (corresponding to Ser-79 in ACC1) [20]. ACC2 resides on the mitochondrial membrane catalyzing the carboxylation of acetyl CoA to Malonyl CoA, an allosteric inhibitor of the mitochondrial long-chain fatty-acyl CoA shuttle, carnitine palmitoyltransferase-1 (CPT-1), which is part of a family of enzymes called carnitine acyltransferases [21, 22]. Phosphorylation of ACC-2 lessens enzyme activity, causing a reduction in Malonyl CoA levels, hence prevention of the Malonyl CoA-mediated inhibition of CPT-1, and an increase in fatty acid β-oxidation by mitochondria. [23] Thiokinase or acyl CoA synthetase activates fatty acid to Acyl CoA. Fatty acid reacts with ATP to form acyladenylate which then combines with Coenzyme A to produce Acyl Co-A, which then transferred to Carnitine catalyzed by CPT-1; present on the outer surface of the inner mitochondrial membrane. The Acyl- Carnitine complex is then transported across the membrane to mitochondrial matrix by carrier protein; carnitine: acylcarnitinetranslocase. CPT-2, found on the inner surface of inner mitochondrial membrane, converts Acyl- Carnitine to Acyl CoA. Carnitine returns to cytosol for reuse. Acyl Co A gets converted in to Acetyl CoA by β- oxidation. Acetyl Co A can enter cyclic acid cycle and get oxidized to CO_2 and H_2O [24].

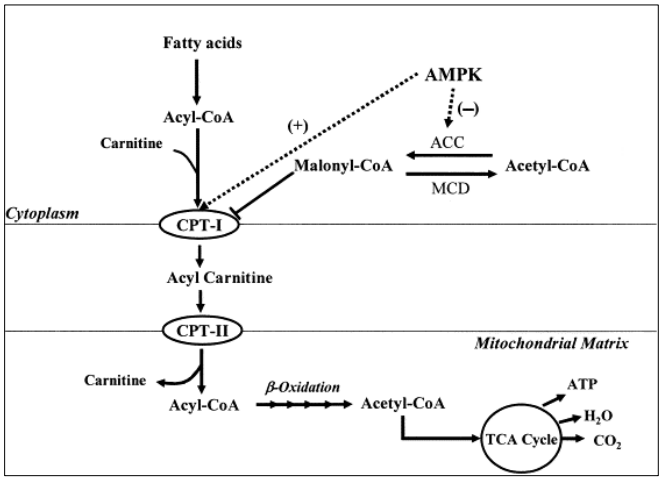

Fig. 4: Fatty acid oxidation mechanism of AMPK

AMPK AND GLUT-4

After a meal or during the euglycaemic hyperinsulinemic clamp, both situations with high circulating levels of insulin, skeletal muscle is the main site for glucose disposal in the body. This is sustained by the insulin-dependent translocation of glucose transporter GLUT4 from intracellular vesicles to the cell surface, which is impaired in T2D patients. Muscular AMPK activation, either by exercise or by drugs, stimulates muscle glucose uptake. Interestingly, even if AMPK and insulin acts through phosphorylation of downstream target of Akt (Akt substrate of 160kDa, AS160) [25], AMPK-dependent and insulin-dependent GLUT4 translocations are distinct pathways [26]. Additionally, exercise-induced muscular AMPK activation and AS160 phosphorylation are both reduced in obese non-diabetic and obese type 2 diabetes subjects [27] but maintained in lean type 2 diabetes patients [28] suggesting that dysregulation of muscular AMPK is more dependent of obesity than of hyperglycaemia. Under conditions of exercise, however, blood sugar levels are not necessarily high, and insulin is not necessarily activated, yet muscles are still able to bring in glucose. AMPK seems to be responsible in part for this exercise-induced glucose uptake. Two proteins are essential for the regulation of GLUT-4 expression at a Transcriptional level Myocyte enhancer Factor 2 (MEF2) and GLUT-4 enhancer factor (GEF). Mutations

in the DNA binding regions for either of these proteins results in ablation of transgene GLUT-4 expression [29, 30]. AMPK activation by 5-aminoimidazole-4-carboxamide riboside (AICAR) treatment has been shown, however, to increase transport of both proteins into the nucleus, as well as increase the binding of both to the GLUT-4 promoter region [31].

CONCLUSION

Lifestyle modifications are recognized as an important preventive and therapeutic intervention for impaired glucose tolerance, insulin resistance and type 2 diabetic patients. By manipulating the activity of AMPK and its upstream kinases, LKB1and CaMKK-β, in differenttissues there is a profound impact on feeding, body weight,glucose homeostasis and insulin sensitivity in rodents andhumans. Altogether, this evidence strengthensthe idea that, besides its role as cellular sensor, AMPK hasa crucial role in the regulation of energy balance at thewhole-body level, opening avenues for future therapeuticand medical intervention not just for insulin resistance,obesity and related disorders but also for feeding andmetabolic alterations associated with disease. AMPK activators are potential new therapeutic agents for the treatment of type 2 diabetes by mimicking the beneficial effects of physical activity and of calorie restriction. Accordingly, AMPK-activating agents could also be used as regulators of hyperglycaemia, obesity, lipids disorders, lipotoxicity and cardiovascular risk by targeting specific cellular pathways. Resveratrol, metformin, TZDs (thiazolidinediones), adiponectin and leptin are now considered as AMPK activators. But still advancement is needed for better safe target specific drugs.

REFERENCES

1. Kassi E, Pervanidou P, Kaltsas G, Chrousos G; Metabolic syndrome: definitions and controversies. BMC Medicine, 2011; 9: 48.
2. Eckel RH, Grundy SM, Zimmet PZ; The metabolic syndrome. Lancet, 2005; 365(9468): 1415–1428.
3. Sawant A, Mankeshwar R, Shah S, Raghavan R, Dhongde G, Raje H *et al*.; Prevalence of Metabolic Syndrome in Urban India. Cholesterol, 2011. Available from http://dx.doi.org/10.1155/2011/920983
4. Reaven G; Role of insulin resistance in human disease. Diabetes, 1988; 37(12):1595-1607.
5. Hardie DG; AMP-activated/SNF1 protein kinases: conserved guardians of cellular energy. Nat Rev Mol Cell Biol., 2007; 8(10): 774 –785.
6. Sanz P; AMP-activated protein kinase: structure and regulation. Curr Protein Pept Sci., 2008; 9(5): 478–492.
7. Kahn BB, Alquier T, Carling D, Hardie DG; AMP-activated protein kinase: ancient energy gauge provides clues to modern understanding of metabolism. Cell Metab., 2005; 1(1): 15–25.
8. Zhang BB, Zhou G, Li C; AMPK: An Emerging Drug Target for Diabetes and Metabolic Syndrome. Cell metabolism, 2009; 9(5): 407-416.
9. Viollet B, Guigas B, Leclerc J, Hebrard S, Lantier L, Mounier R *et al*.; AMP-activated protein kinase in the regulation of hepatic energy metabolism: from physiology to therapeutic perspectives. Acta Physiol. (Oxf), 2009; 196(1): 81–98.
10. Oakhill JS, Steel R, Chen ZP, Scott JW, Ling N, Tam S *et al*.; AMPK is a direct adenylate charge-regulated protein kinase. Science, 2011; 332(6036):1433–1435.
11. Xiao B, Sanders MJ, Underwood E, Heath R, Mayer FV, Carmena D *et al*.; Structure of mammalian AMPK and its regulation by ADP. Nature, 2011; 472: 230–233.
12. Viollet B, Guigas B, Garcia NS, Leclerc J, Foret M, Andreelli F; Cellular and molecular mechanisms of metformin: an overview. Clinical Science, 2012; 122(6), 253–270.
13. Xiao B, Heath R, Saiu P, Leiper FC, Leone P, Jing C *et al*.; Structural basis for AMP binding to mammalian AMP-activated protein kinase. Nature, 2007; 449(7161): 496–500.
14. Jalving M, Gietema JA, Lefrandt JD, de Jong S, Reyners AK, Gans RO *et al*.; Metformin: Taking away the candy for cancer? European Journal of Cancer, 2010; 46(13): 2369-2380.
15. Ouyang J, Parakhia RA, Ochs RS; Metformin Activates AMP Kinase through Inhibition of AMP Deaminase. Journal of Biological Chemistry, 2010; 286(1): 1-11.
16. Todd L; AMP-activated protein kinase as a therapeutic target for diabetes. Discovery Today: Therapeutic Strategies, 2007; 4(2): 123-126.
17. Sarah LL, Richard BW, Hannah LP, Roger WB, Michael FA; 5-Aminoimidazole-4-carboxamide 1-β-D- ribofuranoside (AICAR) stimulates myocardial glycogenolysis by allosteric mechanisms. Am J Physiol Regul Integr Comp Physiol., 2003; 284(4): R936–R944.
18. Kong W, Wei J, Abidi P, Lin M, Inaba S, Li C *et al*.; Berberine is a novel cholesterol-lowering drug working through a unique mechanism distinct from statins. Nature Medicine, 2004; 10(12): 1344.
19. Savage DB, Petersen KF, Shulman GI; Disordered lipid metabolism and the pathogenesis of insulin resistance. Physiol Rev., 2007; 87(2): 507–520.
20. Steinberg GR, Bonen A, Dyck DJ; Fatty acid oxidation and triacylglycerol hydrolysis are enhanced following chronic leptin treatment in rats. Am J Physiol Endocrinol Metab., 2002; 282(3): 593–600.
21. Abu-Elheiga L, Brinkley WR, Zhong L, Chirala SS, Woldegiorgis G, Wakil SJ; The subcellular localization of acetyl-CoA carboxylase 2. Proc Natl Acad Sci USA, 2000; 97(4): 1444–1449.
22. Tong L, Gerwald J; Crystal Structure of Carnitine Acetyltransferase and Implications for the Catalytic

Mechanism and Fatty Acid Transport. Cell, 2003; 112 (1): 113–122.

23. Dzamko N, Schertzer JD, Ryall JG, Steel R, Macaulay SL, Wee S *et al*.; AMPK-independent pathways regulate skeletal muscle fatty acid oxidation. The Journal of Physiology, 2008; 586 (23): 5819-5831.
24. Schulz H; Oxidation of fatty acids in eukaryotes. Chapter 5, In Vance DE, Vance JE editors; Biochemistry of Lipids, Lipoproteins and Membranes. 4th edition, Elsevier Science, 2002: 130-131.
25. Dreyer HC, Drummond MJ, Glynn EL, Fujita S, Chinkes DL, Volpi E *et al*.; Resistance exercise increases human skeletal muscle AS160/TBC1D4 phosphorylation in association with enhanced leg glucose uptake during post-exercise recovery. J Appl Physiol., 2008; 105(6): 1967–1974.
26. Treebak JT, Birk JB, Rose AJ, Kiens B, Richter EA, Wojtaszewski JF; AS160 phosphorylation is associated with activation of alpha2beta2gamma1- but not alpha2beta2gamma3-AMPK trimeric complex in skeletal muscle during exercise in humans. Am J Physiol Endocrinol Metab., 2007; 292(3): E715–722.
27. Musi N, Fujii N, Hirshman MF, Ekberg I, Fröberg S, Ljungqvist O *et al*.; AMP-activated protein kinase (AMPK) is activated in muscle of subjects with type 2 diabetes during exercise. Diabetes, 2001; 50(5): 921–927.
28. Bruce CR, Mertz VA, Heigenhauser GJ, Dyck DJ; The stimulatory effect of globular adiponectin on insulin-stimulated glucose uptake and fatty acid oxidation is impaired in skeletal muscle from obese subjects. Diabetes, 2005; 54(11): 3154–3160.
29. Hayashi T, Hirshman MF, Fujii N, Habinowski SA, Witters LA, Goodyear LJ; Metabolic stress and altered glucose transport: activation of AMP-activated protein kinase as a unifying coupling mechanism. Diabetes, 2000; 49(4): 527–531.
30. Kurth-Kraczek EJ, Hirshman MF, Goodyear LJ, Winder W; 5' AMP-activated protein kinase activation causes GLUT4 translocation in skeletal muscle. Diabetes, 1999; 48(8): 1667–1671.
31. Merrill GF, Kurth EJ, Hardie DG, Winder WW; AICA riboside increases AMP-activated protein kinase, fatty acid oxidation, and glucose uptake in rat muscle. Am J Physiol., 1997; 273 (6 Pt 1): 1107–1112.