

AMP-activated protein kinase (AMPK) - the key role in metabolic regulation and control of food intake

Proteína quinase ativada por AMP (AMPK) - o papel-chave na regulação metabólica e controle da ingestão alimentar

ABSTRACT

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Obesity is a clinical condition which has been viewed as a serious and growing public health problem. Excessive body weight has been shown to predispose to several diseases, mainly cardiovascular diseases, type 2 diabetes, and metabolic syndrome. AMP-activated protein kinase (AMPK) plays a potential role in food intake control in the hypothalamus and peripheral tissues. In vivo administration of leptin, which leads to a reduction of food intake, decreases hypothalamic AMPK activity. In peripheral tissues, AMPK regulates a variety of metabolic pathways that result in the suppression of ATP consumption (anabolic pathway) and the induction of ATP production (catabolic pathway). These include stimulating fatty acid uptake and oxidation and glucose uptake in multiple tissues: stimulating mitochondrial biogenesis in the skeletal muscle, stimulating glycolysis in the heart, inhibiting fatty acid synthesis in the liver and adipocyte, inhibiting cholesterol synthesis and glucogenesis in the liver, and inhibiting insulin secretion from pancreatic β -cells. AMPK is activated in response to environmental or nutritional stress factors, including thermal shock, hypoxia, glucose deprivation, calorie restriction, fasting, epigallocatechin-3-gallate consumption, resveratrol, exercising, ethanol consumption, n-3 (PUFA), statins, or troglitazone. These outcomes demonstrate that AMPK plays a key role in the regulation of feeding, indicating AMPK as a new target in the study of the anti-metabolic syndrome. Particularly, this review with recent findings show how AMPK activation coordinates the metabolic regulations and food intake control under different metabolic and nutritional circumstances.

Keywords: Calcium-calmodulin-dependent. Protein kinases. Anti-obesity agents. Leptin. Central nervous system. Weight loss. Obesity.

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RESUMEN

La obesidad es una condición clínica vista como un serio y creciente problema de salud pública. Está demostrado que el exceso de peso corporal es un factor que predispone para muchas enfermedades, particularmente las cardiovasculares, diabetes mellitus y síndrome metabólico. La proteína quinasa AMP-activada (AMPK) en los tejidos periféricos y el hipotálamo tiene un papel importante sobre el control de la ingestión de alimentos. La administración de leptina in vivo conduce a la reducción de la ingestión energética, disminuyendo la actividad hipotalámica de la AMPK. En los tejidos periféricos, la AMPK regula una variedad de vías metabólicas provocando supresión del consumo de ATP (vía anabólica) e inducción de la producción de ATP (vía catabólica). Incluyendo estímulo a la captación de ácidos grasos y oxidación y captación de glucosa en diversos tejidos: estimulación de la glucosa en el corazón, inhibición de la síntesis de ácidos grasos en hígado y en los adipocitos, inhibición de la síntesis de colesterol y de la glicogénesis en el hígado e inhibición de la secreción de insulina por las células beta del páncreas. La AMPK es activada en respuesta a factores de stress ambiental o nutricional, incluyendo: choque térmico, hipoxia, privación de glucosa, restricción calórica, ayuno, epigallocatequina-3-galato, estatinas o troglitazona. Esos datos demuestran la AMPK como nuevo objeto de estudio anti-síndrome metabólico. En especial, esta revisión sobre pesquisas recientes muestra como la activación de la AMPK coordina las regulaciones metabólicas y el control de la ingestión alimentar en diferentes circunstancias metabólicas y nutricionales.

Palabras clave: Proteína Quinasas Dependiente de Calcio (+2)-Calmodulino. Agentes anti-obesidad. Leptina. Sistema Nervioso Central. Pérdida de peso. Obesidad.

RESUMO

A obesidade é uma condição clínica que é vista como um sério e crescente problema de saúde pública. O excesso de peso corporal tem demonstrado ser fator predisponente para várias doenças, particularmente as doenças cardiovasculares, diabetes tipo 2 e síndrome metabólica. A proteína quinasa AMP-ativada (AMPK) nos tecidos periféricos e hipotálamo tem um papel importante no controle do consumo alimentar. A administração de leptina in vivo conduz à redução da ingestão energética, diminuindo a atividade hipotalâmica da AMPK. Nos tecidos periféricos, a AMPK regula uma variedade de rotas metabólicas que resultam na supressão do consumo de ATP (rota anabólica) e na indução da produção de ATP (rota catabólica). Estas rotas incluem estimulação da captação de ácidos graxos e oxidação e captação de glicose em diversos tecidos: estimulação da biogênese mitocondrial no músculo esquelético, estimulação da glicólise no coração, inibição da síntese de ácidos graxos no fígado e nos adipócitos, inibição da síntese de colesterol e da glicogênese no fígado, e inibição da secreção de insulina pelas células β -pancreática. A AMPK é ativada em resposta a fatores de estresse ambiental ou nutricional, incluindo: choque térmico, hipóxia, privação de glicose, restrição calórica, jejum, epigallocatequina-3-galato, resveratrol, exercício físico, consumo de etanol/álcool, n-3 (PUFA), estatinas ou troglitazona. Esses dados demonstram que a AMPK tem um papel chave na regulação da alimentação, indicando a AMPK como um novo alvo no estudo da antissíndrome metabólica. Em especial, esta revisão com recentes pesquisas mostra como a ativação da AMPK coordena as regulações metabólicas e o controle da ingestão alimentar em diferentes circunstâncias metabólicas e nutricionais.

Palavras-chave: Proteínas Quinase Dependente de Ca(2+)-Calmodulina. Agentes antiobesidade. Leptina. Sistema Nervoso Central. Perda de peso. Obesidade.

INTRODUCTION

Obesity is a condition associated with the accumulation of excessive body fat resulting from chronic imbalance of energy whereby the intake of energy exceeds expenditure. Obesity and overweight are major risk factors for chronic diseases, including type 2 diabetes, cardiovascular disease, and certain forms of cancer (WASAN; LOOIJE, 2005).

The obesity epidemic has been recognized by the World Health Organization (WHO) as one of the top 10 global health problems. All around the world, there are more than 1 billion overweight adults and more than 300 million out of them are obese. Most of the developed countries, including the United States, Canada and England have been experiencing dramatic increases in obesity (WORLD HEALTH ORGANIZATION, 2007; WASAN; LOOIJE, 2005). According to the Brazilian Institute of Geography and Statistics (IBGE), only over 40 percent of the Brazilian adult population is overweight.

This review summarizes the current knowledge concerning the 5'AMP-activated protein kinase (AMPK) and the appetite and energy intake regulation, with a particular focus on 1- the mechanisms (gut hormones) that modulate energy intake, 2- the potential role of metabolic regulation and food intake control, and 3- implications for the pathophysiology and prevention of obesity.

METHODS

The technique used was the literature review, with the following criterion of inclusion: experimental or clinical research characterizing AMPK, based on metabolic regulation and food intake control.

The search for articles was carried out through the following data bases: Medline, Lilacs, and Scopus (1996 – 2008). In this search, the following keywords were used: calmodulin-dependent protein kinase, AMPK, anti-obesity agents, hormones, leptin, central nervous system, weight loss, obesity, and diet versus AMPK.

AMPK STRUCTURE

AMPK exists in the cell as a heterotrimeric enzyme complex with a catalytic (α) (63kDa) and two regulatory subunits (β and γ) (WOODS et al., 1996) that is activated by phosphorylation of threonine 172 (Thr 172) within the α -subunit, being each subunit encoded by a different gene (Figure 1).

The β subunits have a molecular mass of 30kDa [β 1 (38kDa) and β 2 (34kDa)], whereas the three γ isoforms have molecular masses of 37kDa (γ 1), 63kDa (γ 2), and 55 kDa (γ 3), respectively (CHEUNG et al., 2000). While AMPK complexes containing α 2 (the first catalytic isoform to be cloned) (HARDIE; CARLING, 1997) are predominant in the skeletal and cardiac muscle, approximately equal levels of α 1 and α 2 are present in the liver (WOODS et al.,

2000). In the adipose tissue, the $\alpha 1$ catalytic subunit is the predominant isoform expressed and accounts for the major part of AMPK activity (DAVAL et al., 2005). However, the specific isoforms or subunit combinations that provide preferential control of lipid oxidation when compared to glucose metabolism are unknown (OSLER; ZIERATH, 2008).

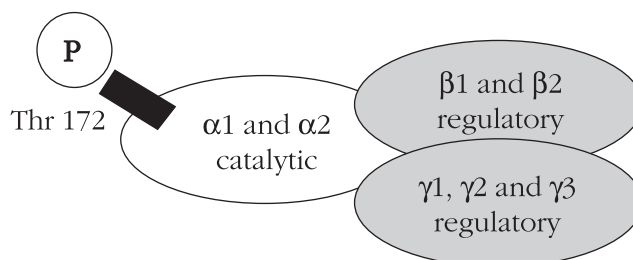


Figure 1 - Structure of AMPK

CONDITIONS THAT AFFECT AMPK ACTIVITY

Some energetic conditions and/or nutrients are necessary for the AMPK activation, such as calorie restriction or reduced growth-hormone (GH) signaling by the insulin-like growth factor (IGF-1), herbal weight-loss supplements, copper deficiency, high-protein diet, fasting, polyphenols and α -lipoic acid consumption. These energetic conditions and/or nutrients induce AMPK to play a lead role in the regulation of lipid metabolism by inhibiting the regulatory enzymes involved in biosynthetic pathways, such as acetyl-CoA carboxylase (ACC) and 3-hydroxy-3-methylglutaryl-CoA reductase. ACC is a key regulatory enzyme for the synthesis of fatty acids, and phosphorylation of hepatic ACC in vivo is mostly achieved by AMPK, rendering the enzyme inactive. ACC activity modulation is also essential for the control of carnitine palmitoyltransferase (CPT-1), which is a key regulatory step of hepatic ketogenesis. AMPK enhances lipid oxidation in the skeletal muscle, as well as in the liver, by inactivating ACC via phosphorylation, thereby reducing the synthesis of malonyl-CoA. This relieves the inhibition of CPT-1 activity and enhances fatty acid oxidation in the mitochondria (increases lipid β -oxidation in the muscle) (MERRIL et al., 1997; OSLER; ZIERATH, 2008). In other words, ACC acts as a metabolic sensor, which is regulated allosterically (via citrate activation and fatty acid inhibition) and by phosphorylation at Ser218 (WATT et al., 2006).

In summary, the biochemical regulation through those nutrients can favor the AMPK activation and reduce malonyl-CoA levels, increasing fatty acids oxidation; besides, they can 1- increase insulin sensitivity, 2- transport glucose, adiponectin secretion, 3- decrease lipogenesis, triglyceride synthesis, TNF- α , and IL-6 secretion, 4- reduce the risk of type 2 diabetes, obesity, metabolic syndrome, and cardiac diseases. The regulation of energy balance by AMPK induced by nutrients/diets is described in table 1.

Table 1 – Multiple nutrients/diets that altered the energy balance through AMPK activity in different sites (central nervous system or peripheral tissues)

Author (year)/Country	Humam/Animal	Nutrient/Diet	Results
Ropelle et al. (2008)/Brazil	Rats	Leucine (50g/kg) supplemented diet and high-protein (50% protein-enriched diet) (HPD) vs control (chow diet) for 3 weeks	Leucine and HPD ↓AMPK and ↑mTOR activity in the hypothalamus, leading to the inhibition of NPY and ↑POMC expression
Gybina e Prohaska (2008)/USA	Rats	Copper (Cu) deficiency evidenced by reduced brain Cu concentrations	AMPK activation and acetyl-CoA carboxylase phosphorylation in the cerebellum
Pang, Chi e Park (2008)/South Korea	Rats	<i>Ilex paraguariensis</i> (yerba mate) supplementation, 2.4g/kg for 60 days	Protection against a high-fat-diet-induced obesity in a rodent model through enhanced UCP2 and UCP3 expression, and ↑fatty acid oxidation through AMPK phosphorylation in the VAT
To et al. (2007)/Japan	Rats	30% calorie restriction (CR) diet vs fed ad libitum (AL) for 6 months	In QFM: Thr ¹⁷² phosphorylated AMPK-α level was slightly ↑ in the CR-fasted phase, but greatly ↑ in the AL-fasted phase. This suggests that CR down-regulates the AMPK activity in the liver
Zang et al. (2006)/USA	Mice	Polyphenol (resveratrol, apigenin, and S17834), containing 130mg/kg/day for 6 weeks	↑ phosphorylation of AMPK and ↑ activity of AMPK (200x the potency of metformin). Prevented: lipid accumulation (liver), hyperlipidemia and the acceleration of aortic lesion
Dasgupta e Milbrandt (2007)/USA	<i>In vitro</i>	10μM of resveratrol	↑AMPK (from 2h until up to 72h) in Neuro2a cells and stimulated mitochondrial biogenesis, suggests that these activations could affect neuronal energy homeostasis
Gabler et al. (2009)/USA	Pigs	PFO: 21% of total fat as n-3 (PUFA), with a 13% EPA and 13% as DHA for 15-19 days during gestation	n-3 during gestation: ↑ glucose uptake by two mechanisms: 1- ↑ total jejunum SGLT1 protein content due to PFO feeding, 2- in an acute manner, the ↑ AMPK resulting in the ↑ translocation of GLUT2 to the BBM
García-Villafranca, Guillén e Castro (2008)/Spain	Rat	Ethanol consumption 14.2±1.1g/kg for 6 weeks	Impairs regulation of fatty acid metabolism by ↓ activity of AMPK, facilitating triacylglycerol accumulation (↓AMPK may go the alcoholic fatty liver)
Collins et al. (2007)/USA	Mice	EGCG consumption ~1μM	EGCG inhibited hepatic gluconeogenesis at 1μM without cytotoxicity. It did not activate the insulin signaling pathway, but ↑AMPK was mediated by the CaMKK
Liu et al. (2006)/China	Rats	Three groups: 1st maintenance diet (control group); 2nd isocaloric rich-fat diet; 3rd isocaloric rich-fat diet with metformin (300mg during the last month) for 5 months	High-fat feeding impaired: the expression of AMPKa, while activating AMPKa by metformin obviously ameliorated high-fat ↑ insulin resistance, indicating a possible role of AMPKa in lipotoxicity
Kim et al. (2004a)/Korea	Rodents	α-lipoic acid (α-LA) consumption	↓hypothalamic AMPK activity and causes profound ↓weight by ↓food intake and ↑energy expenditure
Minokoshi et al. (2004)/USA	Letters to Nature	Fasting or refeeding	Fasting increased AMPK activity in multiple hypothalamic regions, whereas refeeding inhibited it

AMPK: AMP-activated protein kinase; mTOR: mammalian target of rapamycin; NPY: neuropeptide Y; POMC: proopiomelanocortin; VAT: visceral adipose tissue; QFM: quadriceps femoris muscle; S17834: a synthetic polyphenol.

PFO: protected fish oil; EPA: eicosapentaenoic acid; DHA: docosahexaenoic acid; PUFA: polyunsaturated fatty acids; SGLT1: sodium-dependent glucose transporter 1; BBM: brush border membrane; EGCG: epigallocatechin-3-gallate; CaMKK: Ca²⁺/calmodulin-dependent protein kinase kinase.

Moreover, AMPK is activated in response to environmental or nutritional stress factors which deplete intracellular ATP levels, including thermal shock, hypoxia, glucose deprivation, prolonged exercising, statins, or troglitazone (BORGER et al., 2008; FEDIUC et al., 2008; KOSHINAKA et al., 2008; SIMLER et al., 2007; SUN et al., 2006; VIOLLET et al., 2007), and also reduces the risk of type 2 diabetes and metabolic syndrome.

Another protein kinase which activates AMPK is calmodulin-dependent protein kinase kinase beta (CaMKK- β), which phosphorylates and activates AMPK in response to elevated intracellular calcium ions (Ca²⁺). CaMKK- β is intensively expressed in the central nervous system (CNS), and lower levels are detected in other tissues such as the skeletal muscle and the liver, suggesting that AMPK pathways are regulated by multiple mechanisms that are likely to take place in particular tissues (BIRNBAUM, 2005).

AMPK AND OBESITY: PERIPHERAL AND CENTRAL MECHANISMS

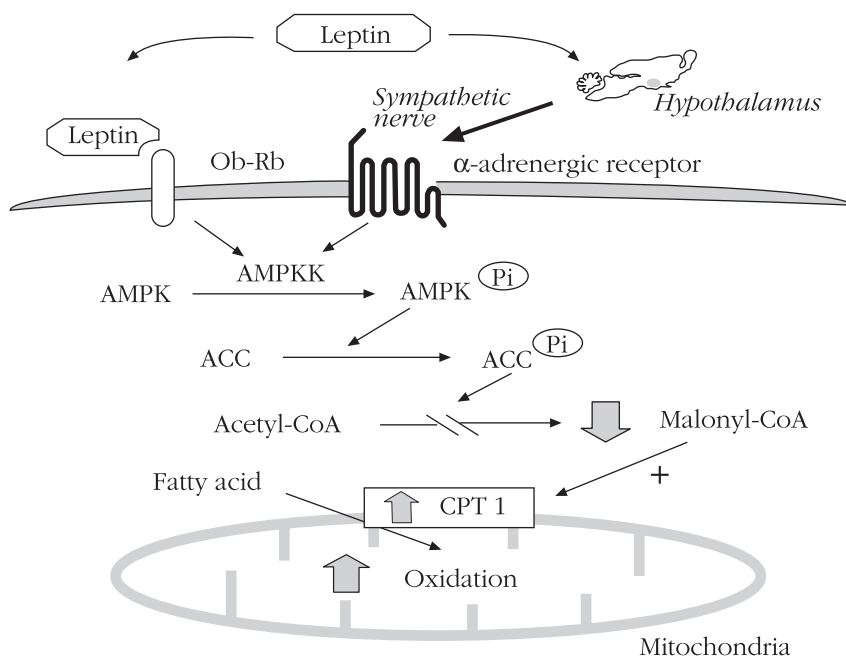
So far, most of the studies examining the role of AMPK have focused on its response to acute changes in energy levels within individual cells. Obesity is associated with leptin production and high plasma leptin concentration. Leptin activates AMPK in the muscle, inhibiting acetyl coenzyme A (acetyl-CoA), which is a thiol chemical that can react with carboxylic acids to form thiol-esters. Therefore, acetyl-CoA behaves as an acyl-group carrier. It also catalyzes a key step in fat synthesis, while adiponectin (also produced in the adipose tissue) activates AMPK both in the liver and in the skeletal muscle. This kinase acts as a lipogenic suppressor, promotes lipid oxidation, and inhibits glucose production in the liver (MINOKOSHI et al., 2002; YAMAUCHI et al., 2002) (Figure 2). However, the increase of body fat (obesity) is associated with high leptin and insulin levels, which induce leptin and insulin resistance partially through suppressor of cytokine signaling 3 (SOCS3) induction, resulting in decreased AMPK activity; stimulates the formation of metabolites (stearoyl-CoA desaturase 1, ACC and sterol response element binding protein-SREBP); and increases lipid synthesis and steatosis (AHIMA et al., 2006). Therefore, the increase of leptin and insulin levels in overweight individuals is not enough to inhibit the high prevalence of obesity in the world once such individuals are resistant to the effect of these hormones.

As a result of this mechanism, the energy sources that take part in fat formation are shifted into an oxidative pathway providing energy for the muscle cells. In the liver, leptin turns down the activity of the gene for stearoyl-CoA desaturase-1 (SCD-1), which has a similar role to the one of acetyl-CoA (COHEN et al., 2002).

SCD-1 is a lipogenic enzyme that catalyzes the synthesis of monounsaturated fatty acids (MUFA) and plays an important role in the development of obesity. SCD-1 deficiency activates metabolic pathways that promote MUFA β -oxidation and decrease lipogenesis in the liver (DOBRZYN et al., 2008; MACDONALD et al., 2008; MAR-HEYMING et al., 2008).

Dobrzyn et al. (2008) showed in mice that an increase in cardiac insulin sensitivity and glucose utilization due to SCD-1 deficiency could prove to be therapeutic in pathological conditions, such as non-communicable disease (e.g. obesity and type 2 diabetes). Moreover,

SCD-1 activity has been implicated in the metabolic syndrome. MacDonald et al. (2008) showed that the lack of the SCD-1 gene product reduces plasma triglycerides and weight gain in severely hyperlipidemic LDL-receptor-deficient mice challenged with a western diet. In the liver, SCD-1 deficiency also reduces lipid accumulation (avoid non-alcoholic steatohepatitis) dramatically while causing modest reduction in plasma apolipoproteins. This suggests that in conditions of sustained hyperlipidemia, SCD-1 works primarily as a mediator of the lipid stores, which leads to an increase of the adipose tissue. However, it can cause hypertriglyceridemia, insulin and leptin resistance. Thus, both lipogenic enzymes (acetyl-CoA and SCD-1) are synthesized as fat.



The therapeutic agent could activate AMPKK/AMPK in the muscle through two different mechanisms: one is a direct effect and the other one is mediated by the hypothalamic-sympathetic nervous system. It activates AMPK, phosphorylates and inhibits ACC activity. Malonyl-CoA synthesis is inhibited, either therapeutically or through the action of leptin resulting in activation of CPT1, thereby increasing mitochondrial import and fatty acid oxidation in the muscle. Ob-Rb: Ob-Rb gene; AMPK: AMP-activated protein kinase; ACC: acetyl-CoA carboxylase; CPT-1: carnitine palmitoyltransferase (MINOKOSHI; KAHN, 2003).

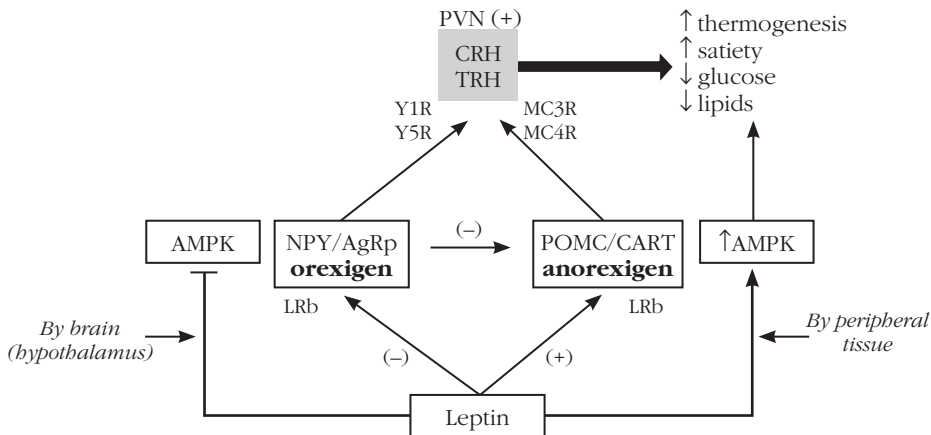
Figure 2 - Leptin inhibits malonyl-CoA synthesis, activating CPT1, thereby increasing mitochondrial import and fatty acid oxidation in the muscle

In addition, the hypothalamus and the dorsal vagal complex appear to be the main regions within the CNS that directly regulates the appetite. The arcuate nucleus (ARC) and the paraventricular nucleus (PVN) of the hypothalamus have been shown to play an integrative role in appetite regulation (SPIEGELMAN; FLIER, 2001). Neurons within the hypothalamus respond to the different neuro-endocrine and metabolic signals coordinating the body's

response to changes in energy intake and energy expenditure. The mechanisms involved are complex but depend at least in part on hormones derived from either adipose tissue, e.g. leptin, or the gastrointestinal tract, e.g. ghrelin.

Leptin, a hormone derived from adipocytes, with 167 amino acids and a molecular mass of 16 kDa, acts on neurons within the ARC, decreasing the release of orexigenic neuropeptides and increasing the release of the anorexigenic ones, which results in decreased food intake (SPIEGELMAN; FLIER, 2001). Leptin is also present in low levels in the gastric fundic epithelium, intestine, placenta, skeletal muscle, mammary epithelium, and the brain (FLIER, 2004; MANCINI et al., 1997).

Leptin regulates specific neuronal groups within the hypothalamus, brainstem and other regions of the CNS (Figure 3). Various leptin receptor isoforms (LRa to LRc) are derived from alternate splicing of *lepr* transcript; however, the effects of leptin on energy homeostasis and other systems are thought to involve the long receptor LRb (especially in the brain). LRb expression is present in the ARC, dorsomedial, ventromedial and PVN, besides the premammillary hypothalamic nucleus. LRb is also present in the periventricular region and posterior hypothalamic nucleus, whereas low levels are found in the PVN and lateral hypothalamic area (FLIER, 2004).



Leptin directly suppresses NPY and AgRp and stimulates POMC/CART neurons in the ARC, leading to inhibition of feeding, increased thermogenesis and reduced glucose and lipids. Leptin is produced by the white adipose tissue, decreased food intake and decreased hypothalamic AMPK. NPY controls feeding through Y1 and Y5 receptors, AgRp antagonize the anorectic action of α -MSH at MC4 receptors.

(-): inhibited; (+): activated; NPY: neuropeptide Y; AgRp: agouti-related protein; AMPK: AMP-activated protein kinase; POMC: proopiomelanocortin; CART: cocaine and amphetamine-regulated transcript; LRb: leptin receptor isoform b; Y1R/Y2R: neuropeptide Y receptor 1 and 2; MC3R/MC4R: melanocortin 3 and 4 receptor; PVN: paraventricular nucleus; CRH: corticotrophin-releasing hormone; TRH: thyrotrophin-releasing hormone.

Adapted: Ahima, 2005; Kim et al., 2004a,b.

Figure 3 - Leptin is inhibited by AMPK in the brain, but it is active in the peripheral tissue (e.g. skeletal muscle), resulting in the stimulation of fatty acid oxidation, increased thermogenesis, satiety and weight loss

Long leptin receptor isoform b (LRb) is intensively expressed in hypothalamic regions involved in energy balance and neuroendocrine function and has been colocalized with targets, e.g. and activator of transcription (STAT3), neuropeptide Y (NPY), proopimelanocortin (POMC), and agouti-related protein (AgRp). NPY and AgRp stimulate feeding, whereas POMC inhibit feeding (BASKIN; HAHN; SCHWARTZ, 1999; SCHWARTZ et al., 1996) and all these hormones are present in the same neurons in the medial ARC.

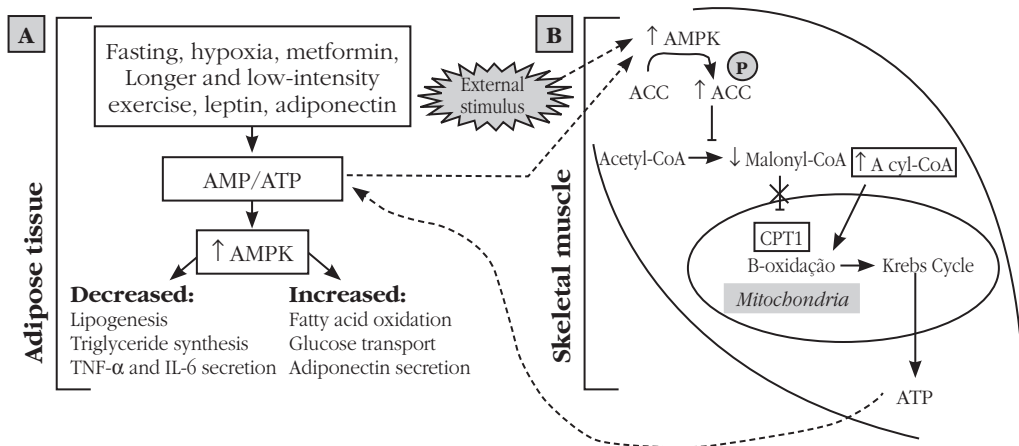
Short leptin receptor isoforms a (LRa) are intensively expressed in the brain microvessels, upregulated in response to hyperleptinemia in diet-induced obesity, and have been suggested as potential mediators of leptin transport into the brain (BJORBAEK et al., 1998). An increase in leptin directly suppresses peptides (NPY and AgRp). Other orexigenic peptides, such as melanin-concentrating hormone (MCH) and orexins, are synthesized in the lateral hypothalamic area and are inhibited indirectly by leptin. Because MCH and orexin neurons project to the cerebral cortex, they might provide a way of transducing the effect of leptin to higher CNS centers in order to coordinate feeding with sleep-wake cycles and other complex functions. Leptin increases the levels of anorectic peptides, α -melanin-stimulating hormone (α -MSH) derived from proopimelanocortin (POMC) and cocaine and amphetamine-regulated transcript (CART), in the lateral ARC. Second order neurons that synthesize corticotropin-releasing hormone (CRH), thyrotropin-releasing hormone (TRH) and oxytocin in the PVN are indirectly controlled by leptin targets in the ARC, and mediate the inhibitory effects of leptin on food intake, stimulation of thermogenesis and neuroendocrine secretion (AHIMA et al., 2000).

AMPK is another leptin target of interest. AMPK is phosphorylated and activated in response to energy deficits during cellular stress or fasting, leading to the stimulation of fatty acid oxidation (KIM et al., 2004a,b) in the skeletal muscle and adipose tissue (DAVALL; FOUFELLE; FERRE, 2006; OSLER; ZIERATH, 2008). Conversely, feeding inactivates AMPK and promotes fatty acid synthesis (KIM et al., 2004a,b).

In the hypothalamus, AMPK is colocalized with signal transducer and transcription (STAT3) (that is translocated to the nucleus, and regulates expression of neuropeptides and other genes), NPY and other peptides implicated in energy balance. The leptin signal is transmitted by the Janus kinase (JAK-STAT) pathway. The binding of leptin to LRb results in auto-phosphorylation of JAK1 and JAK2, tyrosyl-phosphorylation of the cytoplasmic domain of LRb, and phosphorylation and activation of STAT3. The activity of AMPK is causally linked to leptin-mediated changes in neuropeptide synthesis and feeding (MINOKOSHI et al., 2004). Moreover, when AMPK is decreased in hypothalamus, it has an anorexigenic effect.

AMPK is also activated in conditions of increased lipolysis, such as exercising and fasting (Figure 4 A and B) (KADENBACH, 2003; OSLER; ZIERATH, 2008). This activation inhibits fatty acid and triglyceride synthesis and could limit lipolysis. This latter finding might seem counter-intuitive if one considers AMPK as an enzyme which, in case of energy shortage, should enhance energy availability (here understood as fatty acids through

lipolysis) for cells. However, a high rate of lipolysis could be very demanding for adipocyte energy homeostasis since part of the fatty acids can be reactivated into acyl-CoA, a reaction which consumes ATP and generates AMP (KADENBACH, 2003).



B: External stimulus [e.g. exercising (longer and low-intensity, more than 35 minutes), hypoxia and fasting] causes an increase in AMP/ATP ratio. Moreover, phosphor-AMPK phosphorylates and inhibits ACC activity, thereby inhibiting malonyl-CoA synthesis. This leads to the activation of CPT-1 activity and increases mitochondrial import and β -oxidation of fatty acids in the muscle.

ACC: acetyl-CoA carboxylase; CPT-1: carnitine palmitoyltransferase-1.

Figure B - adapted: Seer e Zierath, 2008.

Figure 4 – AMPK activation in the adipose tissue (A) and regulation of fatty-acid oxidation in the skeletal tissue (B)

In high-intensity exercising, the activation of AMPK responds to the depletion of ATP and increases glucose transport (HAYASHI et al., 1998). As showed by Koshinaka et al. (2008), which evaluated insulin sensitivity after high-intensity intermittent swimming (HIS-for 20 second with a weight equal to 18% of body weight) compared to low-intensity continuous swimming exercise (LIS-swam with no load) for 3 hours in rat epitrochlearis muscle. In the HIS group, the phosphorylation of AMPK Thr¹⁷² was increased (13-fold) and the phosphorylation of ACC Ser⁷⁹ was reduced (6-fold), whilst in the LIS group AMPK was increased (2.8-fold) and ACC was reduced (2-fold). In contrast, insulin-stimulated 2-deoxyglucose uptake measured 4 hours post exercise was 73% and 46% higher ($p < 0.05$) in LIS and HIS groups, respectively, compared to the rest. Thus, HIS exercising resulted in greater activation of AMPK compared to LIS, but insulin sensitivity was higher after LIS compared with HIS, suggesting that HIS is more effective than LIS in enhancing insulin sensitivity (KOSHINAKA et al., 2008). With lower intensity exercising, the persistent AMPK activation in the contracted muscle favors lipid metabolism. Furthermore, after training, the occurring changes make the muscle primed for lipid oxidation (WINDER; HARDIE, 1996).

The uncoupling mitochondrial protein (UCP-1) is overexpressed in adipocytes leading to an increase in the AMP/ATP ratio, activation of AMPK, inactivation of ACC and a decreased lipogenesis (MATEJKOVA et al., 2004), preventing the non-communicable diseases. In the adipose tissue of these hyper-leptinized rats, UCP-1 and UCP-2 expression is increased, AMPK activity is induced and leads to the phosphorylation and inactivation of ACC. Therefore, hyperleptinaemia induces a depletion in body fat storages (SHIMABUKURO et al., 1997), suggesting that this is due to the oxidation of fatty acids within the adipocytes inasmuch as these adipocytes release glycerol but no fatty acids (WANG; LEE; UNGER, 1999).

Effects of AMPK on the liver, adipose tissue, muscle, pancreatic islets, hypothalamus and their relation with metabolism are described in figure 5.

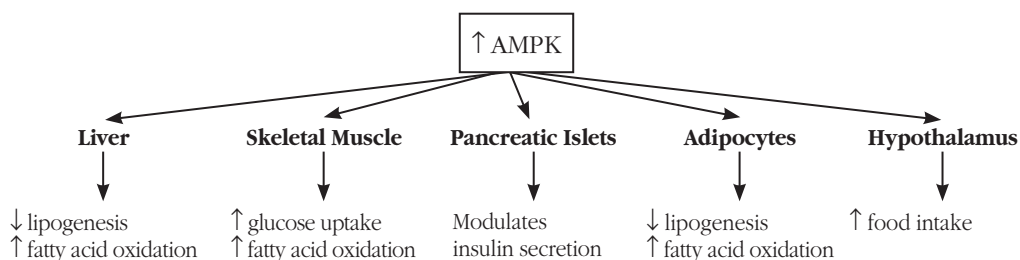


Figure 5 – Effects of AMPK on the liver, adipose tissue, muscle metabolism, pancreatic islets and hypothalamus.

Furthermore, AMPK also inhibits the gene encoding peroxisome proliferator-activated receptor γ (PPAR γ), a receptor for insulin-sensitizing drugs and a key element in the adipocyte differentiation and fat deposition. Moreover, PPAR γ mutations lead to insulin resistance, type 2 diabetes, and hypertension (non-communicable diseases). In addition, these observations suggest that the activation of AMPK and PPAR γ activation apparently are beneficial. Thus, when PPAR γ is activated, reduction of lipogenesis, inflammation and synthesis of proinflammatory cytokines occur (DUSHKIN et al., 2007; FERNANDEZ, 2008; ORASANU et al., 2008).

CONCLUSIONS

AMPK is part of a mechanism which coordinates the changes in lipid metabolism from anabolism to catabolism in case of energy shortage. Therefore, AMPK can lead to an inhibition of fatty acid synthesis and an activation of oxidation mediated by a decreased malonyl-CoA content, which is due to an inhibition of ACC activity, thus reducing the risk of type 2 diabetes, metabolic syndrome and cardiac diseases. These findings demonstrate that AMPK plays a role in the regulation of feeding and indicate AMPK as a new target in the study of anti-metabolic syndrome.

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