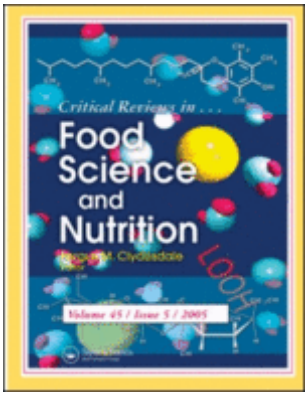

N1-Methylnicotinamide: Is It Time to Consider as a Sport Supplement?

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N1-Methylnicotinamide: Is It Time to Consider as a Sport Supplement?

Running title: N1-Methylnicotinamide and Sport

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Abstract

The lowering of risk for metabolic disorders, such as diabetes and obesity, by exercise can be considered "medicinal". The intensity and duration of exercise determine the mechanism of energy production by various tissues of the body, especially by muscles, in which the requirement for adenosine triphosphate (ATP) increases by as much as 100-fold. Naturally, athletes try to improve their exercise performance by dietary supplementation with, e.g., vitamins, and amino acids. Nicotinamide N-methyltransferase (NNMT) transfers a methyl group from S-adenosylmethionine (SAM) to nicotinamide (NAM) to generate N1-methylnicotinamide (MNAM), a metabolite of vitamin B3. MNAM induces lipolysis in adipocytes, while at the same time reducing both serum and hepatic levels of triglycerides and cholesterol. In addition, in the liver this compound stimulates gluconeogenesis and attenuates cholesterol and fatty acid synthesis by up-regulating the expression of sirtuin1 (SIRT1). The NNMT-MNAM axis is activated more potently in the muscles of subjects who perform high-volume, low-intensity exercise combined with caloric restriction. Therefore, during exercise and/or fasting, MNAM could act as a key myokine that helps provide the substrates required for supplying energy for the contracting muscle, *i.e.* through the promotion of lipolysis by adipocytes and gluconeogenesis by hepatocytes. These observations, among others, lead us to propose that dietary supplementation with MNAM could boost both exercise capacity and improve performance, especially in connection with endurance training. However, a definitive recommendation along these lines requires more detailed insight into the effects of MNAM on various cell types and tissues.

Keyword: MNAM, exercise, sports medicine, dietary supplements

Current Status of Knowledge

1. Metabolism of Exercise

Exercise is well known to exert a variety of beneficial effects on whole-body metabolism, with physical inactivity being considered one of the key factors leading to a wide spectrum of metabolic disorders such as obesity and diabetes (Moghetti et al. 2016; Plowman and Smith 2013; Egan and Zierath 2013). From this perspective, exercise is sometimes referred to as "medicine".

The type, intensity and duration of exercise are major determinants of how energy is provided in the form of adenosine triphosphate (ATP) (Mougios 2019) to the different tissues of the body, and especially to muscles, where the requirement for energy can increase as much as 100-fold during exercise (Moghetti et al. 2016; Mougios 2019). Synthesis of ATP occurs primarily through three different pathways – anaerobic synthesis, with or without associated production of lactate (the latter being referred to as alactic anaerobic synthesis) and aerobic synthesis by mitochondria (Moghetti et al. 2016).

1.1. Alactic Anaerobic ATP Synthesis

The main metabolite involved in alactic anaerobic ATP synthesis is phosphocreatine, stored primarily in muscle cells at levels higher than those of ATP under resting conditions (Tiidus, Tupling, and Houston 2012). During the very first seconds of exercise, phosphate is transferred

from phosphocreatine to adenosine diphosphate (ADP) in order to produce ATP (Moghetti et al. 2016; Tiidus, Tupling, and Houston 2012).

1.2. Anaerobic Glycolysis

Production of ATP through glycolysis requires glucose, either initially present itself or derived metabolically from glycogen, triacylglycerol, and/or amino acids (McArdle, Katch, and Katch 1991). The final metabolite in the glycolytic pathway is pyruvate, which, under conditions where the supply of oxygen is adequate, indirectly supplies electrons for aerobic production of ATP (McArdle, Katch, and Katch 1991). However, when the supply of oxygen is inadequate, as may often be the case in exercising muscles, the pyruvate formed by anaerobic glycolysis is converted to lactate in order to ensure the availability of the cofactor (NADH) required for continued glycolysis (Moghetti et al. 2016).

1.3. Aerobic (Mitochondrial) ATP Synthesis

Following conversion of pyruvate into acetyl-CoA in the presence of adequate levels of oxygen, the latter enters the Krebs cycle, which is designed mainly to reduce the co-enzymes nicotinamide adenine dinucleotide (NAD+) and flavin adenine dinucleotide (FAD) (Stryer 1995). These reduced co-enzymes then funnel electrons into the mitochondrial electron transport chain for oxygen-dependent production of ATP (Stryer 1995).

2. Different Forms of Exercise

The elevated requirement of exercising muscles for oxygen and sources of energy (primarily fatty acids originating from lipolysis in adipose tissue and glucose supplied by glycogen

breakdown and gluconeogenesis in the liver) is met by substantially increasing the blood flow through these tissues (Moghetti et al. 2016). At the same time, the muscle cells regulate their own uptake and metabolism of these substances (Moghetti et al. 2016). The manner in which this metabolism is regulated is determined largely by the type of exercise being performed, basically, endurance versus strength training, which activate muscles in different ways (Moghetti et al. 2016).

2.1. Endurance Exercise

Endurance exercise requires a substantial supply of oxygen, as reflected in the enhanced biogenesis of mitochondria, as well as extensive angiogenesis induced by this form of training (Moghetti et al. 2016). In this context, the master regulator of mitochondrial biogenesis is co-activator 1a of the peroxisome proliferator-activated receptor (PPAR)-γ (abbreviated as PGC-1α) (Spiegelman 2007), the expression of which can be up-regulated by SIRT1 and HIF-1 (Moghetti et al. 2016). At the same time, accumulating evidence indicates that endurance training stimulates 5' adenosine monophosphate-activated protein kinase (AMPK) by elevating cellular levels of Ca^{2+}, the AMP/ATP ratio, and reactive oxygen species (ROS) (Hardie and Sakamoto 2006; Moghetti et al. 2016). Coordination of all of these molecular events (Fig. 1a) is required for generation of the ATP required by muscles performing endurance exercise.

2.2. Strength Training

The muscle hypertrophy that results from strength training (Kvorning et al. 2015) requires enhanced synthesis of the protein components of muscle myofibrils, which may be partially dependent on androgens and insulin-like growth factor (IGF)-1 (Kvorning et al. 2015) (Fig. 1b).

However, the primary molecular regulator of such hypertrophy is the mammalian target of rapamycin (mTOR), which acts through phosphatidylinositol 3-kinase (PI3K) and Akt (Cleasby et al. 2007; Kimball and Jefferson 2010) (Fig. 1b).

3. Metabolism during Fasting

During prolonged fasting, animals utilize endogenous energy stores (Champagne et al. 2012) and, at the same time, reduce energy usage by lowering body temperature, physical activity, and the overall metabolic rate (Secor and Carey 2011). The first phase of fasting, which is very short, is associated with highly pronounced reductions in body mass, metabolic rate, and glycogen reserves (Secor and Carey 2011; Arnould, Green, and Rawlins 2001; Pinheiro et al. 2006). In connection with the second relatively long and adaptive phase the normal utilization of carbohydrates for energy metabolism switches to use of lipids, with an accompanying elevation in circulating levels of ketone bodies (Cherel, Charrassin, and Challet 1994; David et al. 2014; Secor and Carey 2011). During this same phase synthesis of glucose from glycerol and amino acids is activated in order to continue to be able to supply the central nervous system with this energy source (Arnould, Green, and Rawlins 2001; Farley and Robbins 1995; Pernia, Hill, and Ortiz 1980). The third phase is highly critical, involving switching from the use of lipid to produce energy to amino acid catabolism for this purpose, which, if prolonged, may lead to organ failure and death (Nordøy et al. 1992; Robin et al. 1988; Secor and Carey 2011).

4. Dietary Supplements

Dietary supplements such as vitamins, amino acids, and metabolites (Edenfield 2020) are consumed orally in the forms of powders, liquids, capsules, and gels and can be divided broadly into sport, medical, and ergogenic supplements (Edenfield 2020).

4.1. Sport Supplements

Sport supplements are typically consumed as drinks, gels, other liquids or powders (Edenfield 2020). Sport gels and drinks usually contain carbohydrates, sodium, and potassium in varying relative amounts designed to help replenish glycogen depots and improve osmolar homeostasis and hydration (Marriott, Nesheim, and Rosemont 1990; Maughan et al. 2018; Kerksick et al. 2018). Protein supplements, usually in the form of liquids or powders, are designed to stimulate protein synthesis and thereby help enhance muscle mass (Kerksick et al. 2018).

4.2. Medical Supplements

A medical supplement is used to compensate for a deficiency in some essential nutrient or other factor, such as vitamin D, iron, probiotics, and omega 3 (Edenfield 2020). In addition to eliminating the deficiency in question, such supplements may exert beneficial effects on performance, e.g., by strengthening the immune system (vitamin D, and probiotics) (Gleeson et al. 2011; Hao, Dong, and Wu 2015; Institute of Medicine Committee on Military Nutrition 1994; Owens, Allison, and Close 2018; Shuler et al. 2012), improving energy efficiency (iron) (Dellavalle and Haas 2014; Rubeor et al. 2018), or through anti-inflammatory and antioxidant actions (omega 3) (Dellavalle and Haas 2014; Rubeor et al. 2018).

4.3. Ergogenic Supplements

Ergogenic supplements are designed essentially to improve performance during exercise (Edenfield 2020). For example, caffeine improves performance by stimulating mobilization of free fatty acids and calcium (Burke 2008; Ganio et al. 2009; Talanian and Spriet 2016; Wellington, Leveritt, and Kelly 2017; Wiles et al. 2006), while creatine promotes resynthesis of ATP, especially during intensive training (Edenfield 2020). Moreover, creatine raises the level of phosphocreatine in muscle cells, thereby improving short-term high-intensity performance, which could, in turn, augment muscle mass and strength (Branch 2003; Buford et al. 2007; Lanhers et al. 2017; Peeling et al. 2018). Beta-alanine, another widely used supplement, attenuates exercise-induced fatigue by elevating the synthesis of carnosine (Chung et al. 2012; Junior et al. 2015). Nitrate improves tolerance during repeated high-intensity training, possibly by elevating the level of nitric oxide (NO) (Bailey et al. 2015; Thompson et al. 2015; Wylie, Bailey, et al. 2016), while also lowering oxygen cost in a dose- and time-dependent manner (Wylie, Ortiz de Zevallos, et al. 2016).

Therefore, all dietary supplements that improve general health, as well as the performance and tolerance for exercise may be categorized as sports supplements and, unfortunately, almost all of these also have side-effects. Consequently, extensive ongoing research focuses on the discovery and testing of novel supplements that are both more effective and exhibit fewer side-effects. In this context, N1-methylnicotinamide (MNAM) is receiving considerable attention, for the reasons described in detail below.

5. Nicotinamide N-Methyltransferase (NNMT), MNAM and Energy Metabolism

First discovered by Gulio Cantoni in 1951 (Cantoni 1951, 1952), nicotinamide N-methyltransferase (NNMT) transfers methyl groups from s-adenosylmethionine (SAM) to nicotinamide (NAM) to produce MNAM (Nejabati et al. 2018; Nejabati et al. 2019; Pissios 2017; Nejabati et al. 2020). This pathway is involved in multiple processes, including energy metabolism, in a variety of tissues (Nejabati et al. 2018; Nejabati et al. 2019; Pissios 2017; Nejabati et al. 2020).

5.1. NNMT, MNAM and the Liver

The MNAM generated by NNMT in the liver, the organ that expresses this enzyme in highest amounts (Aksoy, Szumlanski, and Weinshilboum 1994), promotes glucose production by inducing gluconeogenic enzymes such as glucose-6-phosphatase (G6Pc) and phosphoenolpyruvate carboxykinase (Pck1) (Hong et al. 2015). At the same time, the synthesis of cholesterol and fatty acid is attenuated, thereby lowering both serum and hepatic levels of these compounds (Hong et al. 2015). MNAM achieves these effects by up-regulating the expression of sirtuin1 (SIRT1) (Hong et al. 2015; Peng et al. 2015).

5.2. NNMT, MNAM and Adipose Tissue

In contrast to the situation in the liver, knockdown of NNMT in adipocytes elevates oxygen consumption and the expenditure of energy by elevating the SAM/SAH ratio and subsequently activating polyamine metabolism (Kraus et al. 2014). Accumulating evidence indicates that NNMT and MNAM are correlated with insulin resistance, especially in individuals with metabolic disorders such as diabetes and obesity, and adipose-specific inhibitors of NNMT are

currently attractive candidates for the prevention of such disorders (Kannt et al. 2015; Kraus et al. 2014; Liu et al. 2015; Riederer et al. 2009).

The consumption of cellular acetyl-coA in connection with polyamine catabolism is primarily responsible for the reduction in adipogenesis and elevated usage of oxygen and energy expenditure (Jell et al. 2007; Koponen et al. 2012; Pirinen et al. 2007). In this connection, the enhanced SAM/SAH ratio is directly related to the methylation of histones that control expression of genes that synthesize, catabolize, and excrete polyamines (Kraus et al. 2014). Furthermore, adipose knockdown of NNMT activates AMPK, visfatin, and SIRT1 (Kraus et al. 2014). It seems that MNAM is not responsible for the actions of NNMT in adipose tissues as MNAM inhibits adipose NNMT activity and acts like the inhibitors of NNMT activity (Kraus et al. 2014). MNAM stimulation is similar to NNMT-knockdown as it increases oxygen consumption, energy expenditure and polyamine catabolism (Kraus et al. 2014).

5.3. NNMT, MNAM and Muscle

It is now clear that the NNMT-MNAM axis is activated both during fasting and exercise (Li et al. 2017; Ström et al. 2018). Li and colleagues (Li et al. 2017) concluded that anaerobic exercise stimulates the expression of NNMT more potently than aerobic exercise, especially when glucose is being utilized as the main source of energy (Li et al. 2017). Furthermore, with either of these types of exercise, NNMT expression was higher in fast- than slow-twitch muscle (Li et al. 2017).

Focusing on the combined effects of exercise and fasting, we found that activation of the NNMT-MNAM axis is particularly enhanced in subjects performing high-volume, low-intensity exercise under conditions of caloric restriction and also by fasting alone. In addition, MNAM

induces lipolysis in rat adipocytes without altering the secretion of insulin or glucagon (Ström et al. 2018). Moreover, expression of NNMT and MNAM secretion by human myotube cells were found to be significantly greater when the culture medium was enriched in amino acids (Ström et al. 2018).

Interestingly, the level of MNAM is strongly correlated to that of α-hydroxybutyrate (α-HB) (Ström et al. 2018), a compound whose level usually increases in response to oxidative stress and a demand for glutathione (GSH) production (Gall et al. 2010; Lord and Bralley 2008) and has been suggested as a biomarker for insulin resistance. In support, catabolism of MNAM is accompanied by transient elevation of the cellular level of ROS and potential subsequent activation of AMPK, as well as elevation of the levels of antioxidants such as catalase and GSH (Schmeisser et al. 2013).

Consequently, under conditions of energy stress such as those caused by exercise and/or fasting, MNAM may act as a key myokine that promotes the usage of amino acids for gluconeogenesis in the liver and stimulates lipolysis in adipose tissue, thereby providing the energy required by muscles.

6. MNAM as a Potential Dietary Supplement for Athletes

According to the Food Standards Agency of the United Kingdom, MNAM is safe and non-toxic (Osęka 2013). It is well documented that MNAM increases the capacity of diabetic mice to exercise via the PGI2-NO pathway and also reduces cardiovascular risk during exercise in individuals with metabolic disorders such as diabetes (Przyborowski et al. 2015). In addition, MNAM is also cardio-protective in connection with other diseases, such as PCOS (Nejabati et al.

2019). Thus, since MNAM improves both endothelial functions and the cardiovascular system (Nejabati et al. 2018; Nejabati et al. 2019), this compound could be considered as an ergogenic supplement.

In addition, MNAM exerts clear anti-inflammatory and anti-thrombotic effects (Nejabati et al. 2018; Nejabati et al. 2019; Pissios 2017) which could improve resistance to mild stress (Schmeisser et al. 2013). However, intake of antioxidants in combination with MNAM may attenuate the resistance against stress induced by the latter (Brown et al. 2001; Nordestgaard et al. 2007).

As mentioned above, SIRT1 and AMPK are the key enzymes involved in the mitochondrial biogenesis and angiogenesis that occurs in response to endurance training (Hardie and Sakamoto 2006; Moghetti et al. 2016). MNAM activates both of these and, moreover, stimulates the gluconeogenesis and lipolysis required to provide glucose and fatty acids, respectively, for the production of energy by muscle cells during training (Hong et al. 2015; Kraus et al. 2014). Therefore, this compound could well be both an effective and safe supplement in connection with endurance training.

7. Conclusions

Induction of the myokine MNAM in response to fasting and/or high-volume, low-intensity exercise enhances lipolysis in adipocytes and gluconeogenesis in hepatocytes. Accordingly, dietary supplementation with MNAM may boost capacity and improve performance, especially in connection with endurance exercise. However, the multiple effects of MNAM in adipose

tissue, liver, and muscle, and perhaps in other tissues as well, remain to be clarified in detail before such supplementation can be definitively recommended.

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References

Aksoy, Saime, Carol L Szumlanski, and Richard M Weinshilboum. 1994. "Human liver nicotinamide N-methyltransferase. cDNA cloning, expression, and biochemical characterization." *Journal of Biological Chemistry* 269 (20):14835-40.

Arnould, John PY, JA Green, and DR Rawlins. 2001. "Fasting metabolism in Antarctic fur seal (Arctocephalus gazella) pups." *Comparative Biochemistry and Physiology Part A: Molecular & Integrative Physiology* 129 (4):829-41.

Bailey, Stephen J, Richard L Varnham, Fred J DiMenna, Brynmor C Breese, Lee J Wylie, and Andrew M Jones. 2015. "Inorganic nitrate supplementation improves muscle oxygenation, O2 uptake kinetics, and exercise tolerance at high but not low pedal rates." *Journal of applied physiology* 118 (11):1396-405.

Branch, J David. 2003. "Effect of creatine supplementation on body composition and performance: a meta-analysis." *International journal of sport nutrition and exercise metabolism* 13 (2):198-226.

Brown, B Greg, Xue-Qiao Zhao, Alan Chait, Lloyd D Fisher, Marian C Cheung, Josh S Morse, Alice A Dowdy, Emily K Marino, Edward L Bolson, and Petar Alaupovic. 2001. "Simvastatin and niacin, antioxidant vitamins, or the combination for the prevention of coronary disease." *New England Journal of Medicine* 345 (22):1583-92.

Buford, Thomas W, Richard B Kreider, Jeffrey R Stout, Mike Greenwood, Bill Campbell, Marie Spano, Tim Ziegenfuss, Hector Lopez, Jamie Landis, and Jose Antonio. 2007. "International Society of Sports Nutrition position stand: creatine supplementation and exercise." *Journal of the International Society of Sports Nutrition* 4 (1):6.

Burke, Louise M. 2008. "Caffeine and sports performance." *Applied physiology, nutrition, and metabolism* 33 (6):1319-34.

Cantoni, GL. 1951. *Methylation of nicotinamide with a soluble enzyme system from rat liver*: Waberly Press.

———. 1952. "The nature of the active methyl donor formed enzymatically from l-methionine and adenosinetriphosphate1, 2." *Journal of the American Chemical Society* 74 (11):2942-3.

Champagne, Cory D, Daniel E Crocker, Melinda A Fowler, and Dorian S Houser. 2012. "Fasting physiology of the pinnipeds: the challenges of fasting while maintaining high energy expenditure and nutrient delivery for lactation." In *Comparative physiology of fasting, starvation, and food limitation*, 309-36. Springer.

Cherel, YVES, JEAN-BENOfT Charrassin, and ETIENNE Challet. 1994. "Energy and protein requirements for molt in the king penguin Aptenodytes patagonicus." *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology* 266 (4):R1182-R8.

Chung, Weiliang, Greg Shaw, Megan E Anderson, David B Pyne, Philo U Saunders, David J Bishop, and Louise M Burke. 2012. "Effect of 10 week beta-alanine supplementation on competition and training performance in elite swimmers." *Nutrients* 4 (10):1441-53.

Cleasby, M. E., T. A. Reinten, G. J. Cooney, D. E. James, and E. W. Kraegen. 2007. "Functional studies of Akt isoform specificity in skeletal muscle in vivo; maintained insulin sensitivity despite reduced

insulin receptor substrate-1 expression." *Mol Endocrinol* 21 (1):215-28. doi: 10.1210/me.2006-0154.

David, Lawrence A, Corinne F Maurice, Rachel N Carmody, David B Gootenberg, Julie E Button, Benjamin E Wolfe, Alisha V Ling, A Sloan Devlin, Yug Varma, and Michael A Fischbach. 2014. "Diet rapidly and reproducibly alters the human gut microbiome." *Nature* 505 (7484):559-63.

Dellavalle, Diane M, and Jere D Haas. 2014. "Iron supplementation improves energetic efficiency in iron-depleted female rowers." *Medicine & Science in Sports & Exercise* 46 (6):1204-15.

Edenfield, Katherine M. 2020. "Sports Supplements: Pearls and Pitfalls." *Primary Care: Clinics in Office Practice* 47 (1):37-48.

Egan, Brendan, and Juleen R Zierath. 2013. "Exercise metabolism and the molecular regulation of skeletal muscle adaptation." *Cell metabolism* 17 (2):162-84.

Farley, Sean D, and Charles T Robbins. 1995. "Lactation, hibernation, and mass dynamics of American black bears and grizzly bears." *Canadian Journal of Zoology* 73 (12):2216-22.

Gall, Walter E, Kirk Beebe, Kay A Lawton, Klaus-Peter Adam, Matthew W Mitchell, Pamela J Nakhle, John A Ryals, Michael V Milburn, Monica Nannipieri, and Stefania Camastra. 2010. "α-Hydroxybutyrate is an early biomarker of insulin resistance and glucose intolerance in a nondiabetic population." *PLoS One* 5 (5).

Ganio, Matthew S, Jennifer F Klau, Douglas J Casa, Lawrence E Armstrong, and Carl M Maresh. 2009. "Effect of caffeine on sport-specific endurance performance: a systematic review." *The Journal of Strength & Conditioning Research* 23 (1):315-24.

Gleeson, Michael, Nicolette C Bishop, Marta Oliveira, and Pedro Tauler. 2011. "Daily probiotic's (Lactobacillus casei Shirota) reduction of infection incidence in athletes." *International journal of sport nutrition and exercise metabolism* 21 (1):55-64.

Hao, Qiukui, Bi Rong Dong, and Taixiang Wu. 2015. "Probiotics for preventing acute upper respiratory tract infections." *Cochrane Database of Systematic Reviews* (2).

Hardie, D. G., and K. Sakamoto. 2006. "AMPK: a key sensor of fuel and energy status in skeletal muscle." *Physiology (Bethesda)* 21:48-60. doi: 10.1152/physiol.00044.2005.

Hong, Shangyu, Jose M Moreno-Navarrete, Xiaojing Wei, Yusuke Kikukawa, Iphigenia Tzameli, Deepthi Prasad, Yoonjin Lee, John M Asara, Jose Manuel Fernandez-Real, and Eleftheria Maratos-Flier. 2015. "Nicotinamide N-methyltransferase regulates hepatic nutrient metabolism through Sirt1 protein stabilization." *Nature medicine* 21 (8):887.

Institute of Medicine Committee on Military Nutrition, Research. 1994. *Fluid Replacement and Heat Stress*, edited by B. M. Marriott. Washington (DC): National Academies Press (US).

Jell, Jason, Salim Merali, Mary L Hensen, Richard Mazurchuk, Joseph A Spernyak, Paula Diegelman, Nicholas D Kisiel, Carlos Barrero, Kristin K Deeb, and Leena Alhonen. 2007. "Genetically altered expression of spermidine/spermine N1-acetyltransferase affects fat metabolism in mice via acetyl-CoA." *Journal of Biological Chemistry* 282 (11):8404-13.

Junior, Antonio Herbert Lancha, Vitor de Salles Painelli, Bryan Saunders, and Guilherme Giannini Artioli. 2015. "Nutritional strategies to modulate intracellular and extracellular buffering capacity during high-intensity exercise." *Sports medicine* 45 (1):71-81.

Kannt, Aimo, Anja Pfenninger, Lenore Teichert, Anke Tönjes, Arne Dietrich, Michael R Schön, Nora Klöting, and Matthias Blüher. 2015. "Association of nicotinamide-N-methyltransferase mRNA expression in human adipose tissue and the plasma concentration of its product, 1-methylnicotinamide, with insulin resistance." *Diabetologia* 58 (4):799-808.

Kerksick, Chad M, Colin D Wilborn, Michael D Roberts, Abbie Smith-Ryan, Susan M Kleiner, Ralf Jäger, Rick Collins, Mathew Cooke, Jaci N Davis, and Elfego Galvan. 2018. "ISSN exercise & sports nutrition review update: research & recommendations." *Journal of the International Society of Sports Nutrition* 15 (1):38.

Kimball, S. R., and L. S. Jefferson. 2010. "Control of translation initiation through integration of signals generated by hormones, nutrients, and exercise." *J Biol Chem* 285 (38):29027-32. doi: 10.1074/jbc.R110.137208.

Koponen, Taina, Marc Cerrada-Gimenez, Eija Pirinen, Esa Hohtola, Jussi Paananen, Susanna Vuohelainen, Maija Tusa, Sini Pirnes-Karhu, Sami Heikkinen, and Antti Virkamäki. 2012. "The activation of hepatic and muscle polyamine catabolism improves glucose homeostasis." *Amino acids* 42 (2-3):427-40.

Kraus, Daniel, Qin Yang, Dong Kong, Alexander S Banks, Lin Zhang, Joseph T Rodgers, Eija Pirinen, Thomas C Pulinilkunnil, Fengying Gong, and Ya-chin Wang. 2014. "Nicotinamide N-methyltransferase knockdown protects against diet-induced obesity." *Nature* 508 (7495):258.

Kvorning, T., F. Kadi, P. Schjerling, M. Andersen, K. Brixen, C. Suetta, and K. Madsen. 2015. "The activity of satellite cells and myonuclei following 8 weeks of strength training in young men with suppressed testosterone levels." *Acta Physiol (Oxf)* 213 (3):676-87. doi: 10.1111/apha.12404.

Lanhers, Charlotte, Bruno Pereira, Geraldine Naughton, Marion Trousselard, François-Xavier Lesage, and Frédéric Dutheil. 2017. "Creatine supplementation and upper limb strength performance: A systematic review and meta-analysis." *Sports medicine* 47 (1):163-73.

Li, J-H, L-Q Qiu, X-J Zhu, and C-X Cai. 2017. "Influence of exercises using different energy metabolism systems on NNMT expression in different types of skeletal muscle fibers." *Science & Sports* 32 (1):27-32.

Liu, Ming, Lihua Li, Jihong Chu, Boyu Zhu, Qingtao Zhang, Xueyan Yin, Weimin Jiang, Guoliang Dai, Wenzheng Ju, and Zhenxing Wang. 2015. "Serum N 1-methylnicotinamide is associated with obesity and diabetes in Chinese." *The Journal of Clinical Endocrinology & Metabolism* 100 (8):3112-7.

Lord, Richard S, and J Alexander Bralley. 2008. "Clinical applications of urinary organic acids. Part I: Detoxification markers." *Altern Med Rev* 13 (3):205-15.

Marriott, Bernadette M, Robert O Nesheim, and Connie Rosemont. 1990. *Fluid Replacement and Heat Stress: A Report of the Proceedings of a Workshop*: National Academy Press.

Maughan, Ronald J, Louise M Burke, Jiri Dvorak, D Enette Larson-Meyer, Peter Peeling, Stuart M Phillips, Eric S Rawson, Neil P Walsh, Ina Garthe, and Hans Geyer. 2018. "IOC consensus statement: dietary supplements and the high-performance athlete." *International journal of sport nutrition and exercise metabolism* 28 (2):104-25.

McArdle, William D, Frank I Katch, and Victor L Katch. 1991. "Exercise physiology: energy, nutrition, and human performance." In.: LWW.

Moghetti, Paolo, Elisabetta Bacchi, Corinna Brangani, Silvia Donà, and Carlo Negri. 2016. "Metabolic effects of exercise." In *Sports Endocrinology*, 44-57. Karger Publishers.

Mougios, Vassilis. 2019. *Exercise biochemistry*: Human Kinetics Publishers.

Nejabati, Hamid Reza, Aynaz Mihanfar, Masoud Pezeshkian, Amir Fattahi, Zeinab Latifi, Naser Safaie, Mohammad Valiloo, Ahmad Reza Jodati, and Mohammad Nouri. 2018. "N1-methylnicotinamide (MNAM) as a guardian of cardiovascular system." *Journal of cellular physiology* 233 (10):6386-94.

Nejabati, Hamid Reza, Nasser Samadi, Leila Roshangar, and Mohammad Nouri. 2019. "N1-methylnicotinamide as a possible modulator of cardiovascular risk markers in polycystic ovary syndrome." *Life sciences*:116843.

Nejabati, Hamid Reza, Nasser Samadi, Vahideh Shahnazi, Aynaz Mihanfar, Amir Fattahi, Zeinab Latifi, Zahra Bahrami-asl, Leila Roshangar, and Mohammad Nouri. 2020. "Nicotinamide and its metabolite N1-Methylnicotinamide alleviate endocrine and metabolic abnormalities in adipose and ovarian tissues in rat model of Polycystic Ovary Syndrome." *Chemico-Biological Interactions*:109093. doi: https://doi.org/10.1016/j.cbi.2020.109093.

Nordestgaard, Børge G, Marianne Benn, Peter Schnohr, and Anne Tybjærg-Hansen. 2007. "Nonfasting triglycerides and risk of myocardial infarction, ischemic heart disease, and death in men and women." *Jama* 298 (3):299-308.

Nordøy, ES, DE Stijfhoorn, A Råheim, and AS Blix. 1992. "Water flux and early signs of entrance into phase 111 of fasting in grey seal pups." *Acta physiologica scandinavica* 144 (4):477-82.

Osęka, Foodie-G. 2013. "APPLICATION FOR THE APPROVAL OF 1-METHYLNICOTINAMIDE CHLORIDE (1-MNA) AS A NOVEL FOOD INGREDIENT FOR USE IN THE MANUFACTURE OF FOOD SUPPLEMENTS PURSUANT TO EU NOVEL FOODS REGULATION (EC) 258/97."

Owens, Daniel J, Richard Allison, and Graeme L Close. 2018. "Vitamin D and the athlete: current perspectives and new challenges." *Sports medicine* 48 (1):3-16.

Peeling, Peter, Martyn J Binnie, Paul SR Goods, Marc Sim, and Louise M Burke. 2018. "Evidence-based supplements for the enhancement of athletic performance." *International journal of sport nutrition and exercise metabolism* 28 (2):178-87.

Peng, Lirong, Zhigang Yuan, Yixuan Li, Hongbo Ling, Victoria Izumi, Bin Fang, Kenji Fukasawa, John Koomen, Jiandong Chen, and Edward Seto. 2015. "Ubiquitinated sirtuin 1 (SIRT1) function is modulated during DNA damage-induced cell death and survival." *Journal of Biological Chemistry* 290 (14):8904-12.

Pernia, Sherman D, Allan Hill, and C Leo Ortiz. 1980. "Urea turnover during prolonged fasting in the northern elephant seal." *Comparative Biochemistry and Physiology, B* 65 (4):731-4.

Pinheiro, Eliana C, Valdir A Taddei, Renato H Migliorini, and Isis C Kettelhut. 2006. "Effect of fasting on carbohydrate metabolism in frugivorous bats (Artibeus lituratus and Artibeus jamaicensis)." *Comparative Biochemistry and Physiology Part B: Biochemistry and Molecular Biology* 143 (3):279-84.

Pirinen, Eija, Teemu Kuulasmaa, Marko Pietilä, Sami Heikkinen, Maija Tusa, Paula Itkonen, Susanna Boman, Joanna Skommer, Antti Virkamäki, and Esa Hohtola. 2007. "Enhanced polyamine catabolism alters homeostatic control of white adipose tissue mass, energy expenditure, and glucose metabolism." *Molecular and cellular biology* 27 (13):4953-67.

Pissios, Pavlos. 2017. "Nicotinamide N-methyltransferase: more than a vitamin B3 clearance enzyme." *Trends in Endocrinology & Metabolism* 28 (5):340-53.

Plowman, Sharon A, and Denise L Smith. 2013. *Exercise physiology for health fitness and performance*: Lippincott Williams & Wilkins.

Przyborowski, Kamil, Marta Wojewoda, Barbara Sitek, Agnieszka Zakrzewska, Agnieszka Kij, Krystyna Wandzel, Jerzy Andrzej Zoladz, and Stefan Chlopicki. 2015. "Effects of 1-methylnicotinamide (MNA) on exercise capacity and endothelial response in diabetic mice." *PLoS One* 10 (6).

Riederer, Monika, Wolfgang Erwa, Robert Zimmermann, Saša Frank, and Rudolf Zechner. 2009. "Adipose tissue as a source of nicotinamide N-methyltransferase and homocysteine." *Atherosclerosis* 204 (2):412-7.

Robin, JEAN-PATRICE, MICHEL Frain, CLAUDE Sardet, RENE Groscolas, and YVON Le Maho. 1988. "Protein and lipid utilization during long-term fasting in emperor penguins." *American Journal of Physiology-Regulatory, Integrative and Comparative Physiology* 254 (1):R61-R8.

Rubeor, Amity, Carmen Goojha, Jeffrey Manning, and Jordan White. 2018. "Does iron supplementation improve performance in iron-deficient nonanemic athletes?" *Sports health* 10 (5):400-5.

Schmeisser, Kathrin, Johannes Mansfeld, Doreen Kuhlow, Sandra Weimer, Steffen Priebe, Ines Heiland, Marc Birringer, Marco Groth, Alexandra Segref, and Yariv Kanfi. 2013. "Role of sirtuins in lifespan regulation is linked to methylation of nicotinamide." *Nature chemical biology* 9 (11):693.

Secor, Stephen M, and Hannah V Carey. 2011. "Integrative physiology of fasting." *Comprehensive Physiology* 6 (2):773-825.

Shuler, Franklin D, Matthew K Wingate, G Hunter Moore, and Charles Giangarra. 2012. "Sports health benefits of vitamin D." *Sports health* 4 (6):496-501.

Spiegelman, B. M. 2007. "Transcriptional control of energy homeostasis through the PGC1 coactivators." *Novartis Found Symp* 286:3-6; discusssion -12, 162-3, 96-203.

Ström, Kristoffer, David Morales-Alamo, Filip Ottosson, Anna Edlund, Line Hjort, Sine W Jörgensen, Peter Almgren, Yuedan Zhou, Marcos Martin-Rincon, and Carl Ekman. 2018. "N 1-methylnicotinamide is a signalling molecule produced in skeletal muscle coordinating energy metabolism." *Scientific reports* 8 (1):1-12.

Stryer, Lubert. 1995. "Fatty acid metabolism." *Biochemistry* 3:469-93.

Talanian, Jason L, and Lawrence L Spriet. 2016. "Low and moderate doses of caffeine late in exercise improve performance in trained cyclists." *Applied physiology, nutrition, and metabolism* 41 (8):850-5.

Thompson, Christopher, Lee J Wylie, Jonathan Fulford, James Kelly, Matthew I Black, Sinead TJ McDonagh, Asker E Jeukendrup, Anni Vanhatalo, and Andrew M Jones. 2015. "Dietary nitrate improves sprint performance and cognitive function during prolonged intermittent exercise." *European journal of applied physiology* 115 (9):1825-34.

Tiidus, Peter M, A Russell Tupling, and Michael E Houston. 2012. *Biochemistry primer for exercise science*: Human Kinetics.

Wellington, Brandon M, Michael D Leveritt, and Vincent G Kelly. 2017. "The effect of caffeine on repeat-high-intensity-effort performance in rugby league players." *International journal of sports physiology and performance* 12 (2):206-10.

Wiles, Jonathan D, Damian Coleman, Michael Tegerdine, and Ian L Swaine. 2006. "The effects of caffeine ingestion on performance time, speed and power during a laboratory-based 1 km cycling time-trial." *Journal of sports sciences* 24 (11):1165-71.

Wylie, L. J., J. Ortiz de Zevallos, T. Isidore, L. Nyman, A. Vanhatalo, S. J. Bailey, and A. M. Jones. 2016. "Dose-dependent effects of dietary nitrate on the oxygen cost of moderate-intensity exercise: Acute vs. chronic supplementation." *Nitric Oxide* 57:30-9. doi: 10.1016/j.niox.2016.04.004.

Wylie, Lee J, Stephen J Bailey, James Kelly, James R Blackwell, Anni Vanhatalo, and Andrew M Jones. 2016. "Influence of beetroot juice supplementation on intermittent exercise performance." *European journal of applied physiology* 116 (2):415-25.

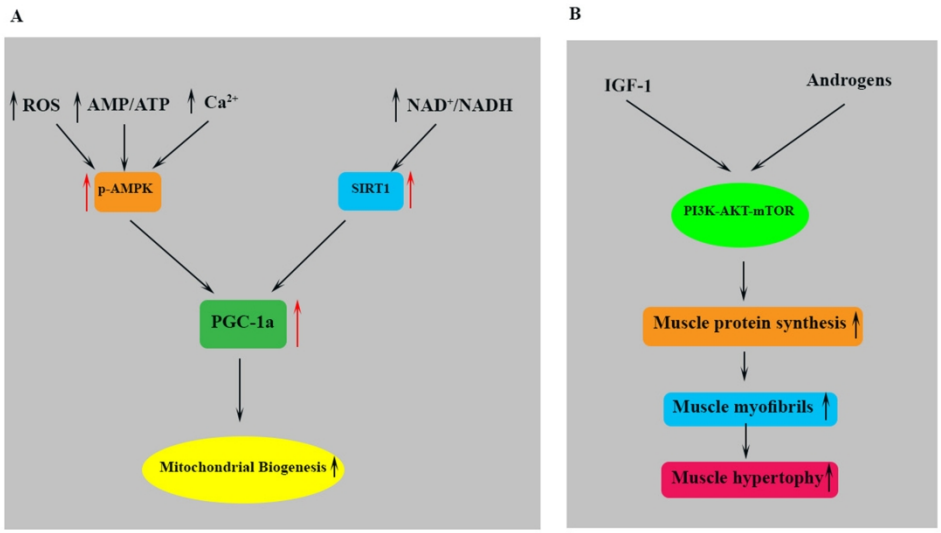

Figure 1. Molecular mechanisms underlying responses to endurance and strength training. A. Following endurance training, elevation of the intracellular NAD+/NADH and AMP/ATP ratios, as well as the levels of ROS, followed by activation of SIRT1 and AMPK induce the master regulator of mitochondrial biogenesis, PGC-1a. B. In response to strength training, androgens and IGF-1 lead to muscle hypertrophy via the PI3K-AKT-mTOR pathway. NADH, nicotinamide adenine dinucleotide; ATP, adenosine triphosphate; ROS, reactive oxygen species; SIRT1, sirtuin1; AMPK, 5'-adenosine monophosphate-activated protein kinase, PGC-1a, coactivator 1a of the peroxisome proliferator-activated receptor (PPAR)-γ; IGF-1, insulin-like growth factor-1.

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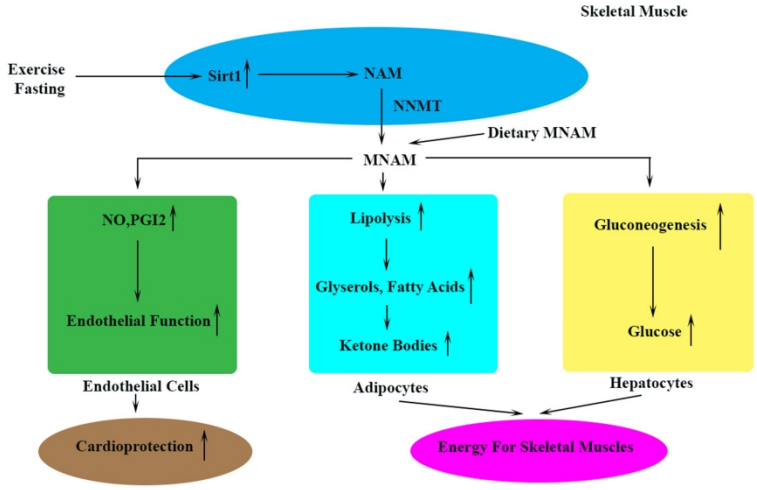

Figure 2. Regulation of energy metabolism during exercise and/or fasting by MNAM. Exercise and/or fasting activate SIRT1 in skeletal muscles, leading to the release of NAM and its subsequent conversion by NNMT into MNAM. MNAM acts as a myokine that induces lipolysis in adipocytes and gluconeogenesis in hepatocytes, thereby providing the circulating free fatty acids and glucose, respectively, required for energy production by muscles. In addition, by elevating the levels of PGI2 and NO, MNAM improves the functions of endothelial cells, thereby providing cardioprotection. NAM, nicotinamide; NNMT, nicotinamide N-methyltransferase; MNAM, N1-methylnicotinamide; SIRT1, sirtuin1; PGI2, prostaglandin I2; NO, nitric oxide.

529x272mm (300 x 300 DPI)