Regulation of AMP catabolism by AMP-activated Protein Kinase (AMPK)-mediated Phosphorylation of the Cytosolic 5'Nucleotidase Type I (cN-I) and Type II (cN-II) Isoforms.

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AMPK is a ubiquitously expressed mammalian Serine/Threonine protein kinase that protects the cell against ATP depletion. AMPK is activated under physiological or pathological stress when the AMP: ATP ratio increases. AMPK is allosterically activated by AMP and covalently by AMPK kinase phosphorylation (1). Acutely, AMPK phosphorylates and inhibits enzymes that consume ATP while stimulating fatty acid release and glucose uptake for ATP re-synthesis. Chronically, AMPK regulates transcription by phosphorylating nuclear transcriptional factors (2). 5' Nucleotidases dephosphorylate non-cyclic nucleoside monophosphates to nucleosides and Pi. cN-I hydrolyzes AMP to adenosine while cN-II generates inosine from IMP which arises from the action of AMP deaminase on AMP. Both cN-I and cN-II are known to be regulated allosterically by ADP (activation) and Pi (inhibition) (3) but as both isoforms contain conserved consensus sequences for phosphorylation by AMPK (2) (L [K, A] Y H [T168] N L Y L on cN-IA; L [F, R] S G [S445] R G T L on cN-II) we are investigating whether phosphorylation also regulates their activities. So far, we have found that AMPK phosphorylates cN-II and inhibits its IMP-hydrolysing activity. Ongoing studies will determine whether cN-I activity is similarly inhibited. If so, cN-I and cN-II would be novel AMPK substrates that help orchestrate cell energy homeostasis by inhibiting adenosine and inosine release from cells and maybe sustaining AMPK activity. Further, as 5' Nucleotidases are implicated in drug resistance to nucleoside analogues, used to treat leukaemia (4), their inhibition by AMPK might increase the drug efficacy of such compounds.

1. Moore F., Weekes J. and Hardie D. G. (1991) AMP triggers phosphorylation as well as direct allosteric activation of rat liver AMP- activated protein kinase: A sensitive mechanism to protect the cell against ATP depletion. Eur. J. Biochem. 199, 691-697.

2. Hardie D. G., Carling D. and Carlson M. (1998) The AMP-activated/SNF1 protein kinase subfamily: metabolic sensors of the eukaryotic cell? Annu. Rev. Biochem. 67, 821-855.

3. Bianchi V. and Spychala J. (2003) Mammalian 5-nucleotidases. J. Biol. Chem. 278, 46195-46198.

4. Galmarini, C M; Mackey, J R; Dumontet, C. (2001) Nucleoside analogues: mechanisms of drug resistance and reversal strategies. Leukemia, 15, 875-890.