

3050 Spruce Street, St. Louis, MO 63103 USA
Tel: (800) 521-8956 (314) 771-5765 Fax: (800) 325-5052 (314) 771-5757
email: techservice@sial.com sigma-aldrich.com

# **Product Information**

Anti-AMPKα antibody produced in rabbit affinity isolated antibody, buffered aqueous solution

Product Number A3730

## **Product Description**

Anti-AMPK $\alpha$  is produced in rabbit using a synthetic peptide corresponding to amino acids 2-19 located at the N-terminus of human AMPK $\alpha$ 1, conjugated to KLH, as immunogen. This sequence is highly conserved (single amino acid substitution, 2 amino acids deletion) in rat and mouse AMPK $\alpha$ 1 and in human, rat and mouse AMPK $\alpha$ 2. The antibody is affinity-purified using the immunizing peptide immobilized on agarose.

Anti-AMPK $\alpha$  recognizes AMPK $\alpha$ , 60 kDa, by immunoblotting. Staining of the AMPK $\alpha$  band is specifically inhibited with the AMPK $\alpha$  immunizing peptide, human, amino acids 2-19.

AMPK (AMP-activated protein kinase) is a serine/threonine protein kinase highly conserved in animals and yeast, and it plays a key role in the regulation of energy homeostasis. 1,2 AMPK appears to act as a metabolic stress-sensing protein kinase switching off biosynthetic pathways when cellular levels of ATP fall and AMP levels rise in response to cellular and environmental stress. AMPK is activated by stress conditions such as anoxia, ischemia, heat shock and exercise-induced skeletal muscle contraction. 2,3 AMPK can also be activated by hyperosmotic stress, by leptin and adiponectin, and by the anti-diabetic drugs metformin and rosiglitazone. 4-7 Once activated, AMPK acts to down-regulate several biosynthetic pathways by phosphorylating and inactivating multiple targets including acetyl-CoA carboxylase (ACC), hydroxymethylglutaryl-CoA (HMG-CoA) reductase, glycogen synthase and endothelial nitric oxide synthase (eNOS). AMPK inhibits protein synthesis by promoting the phosphorylation of eukaryotic elongation factor-2. It also inhibits the TOR (target of rapamycin) pathway by phosphorylating TSC2, thus inhibiting cell growth during cellular stress conditions. AMPK also activates pathways involved in ATP production. For example, in heart, AMPK activation stimulates glycolysis by increasing glucose uptake and by activating 6-phosphofructo-2-kinase. The increase in AMPK activity results in the stimulation of glucose uptake in muscle, fatty acid oxidation, inhibition of hepatic glucose

production, cholesterol and triglyceride synthesis, and lipogenesis. AMPK is a heterotrimeric protein complex consisting of a catalytic  $\alpha\text{-subunit}$  and two regulatory subunits,  $\beta$  and  $\gamma$ . Each subunit exists as multiple isoforms ( $\alpha$ 1-2,  $\beta$ 1-2, and  $\gamma$ 1-3), with different tissue distribution and subcellular localization.  $^{1,2}$  AMPK is regulated by phosphorylation by the upstream kinase, tumor suppressor LKB1.  $^{8\cdot10}$  The major regulatory phosphorylation site has been identified as Thr  $^{172}$  within the activation loop of the AMPK $\alpha$  subunits.

## Reagent

Supplied as a solution in 0.01 M phosphate buffered saline, pH 7.4, containing 15 mM sodium azide as a preservative.

Antibody concentration: ~1 mg/mL

#### **Precautions and Disclaimer**

This product is for R&D use only, not for drug, household, or other uses. Please consult the Safety Data Sheet for information regarding hazards and safe handling practices.

# Storage/Stability

For continuous use, store at 2-8 °C for up to one month. For extended storage freeze in working aliquots. Repeated freezing and thawing, or storage in "frost-free" freezers, is not recommended. If slight turbidity occurs upon prolonged storage, clarify the solution by centrifugation before use. Working dilution samples should be discarded if not used within 12 hours.

# **Product Profile**

 $\frac{Immunoblotting}{0.5\text{-}1.0~\mu\text{g/mL}} \ \text{is determined using a whole cell extract} \\ \text{of mouse 3T3-L1 adipocytes, and a whole cell extract} \\ \text{of human kidney 293 cell line.}$ 

**Note**: In order to obtain best results and assay sensitivity in different techniques and preparations we recommend determining optimal working concentrations by titration test.

#### References

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SG,ER,AH,PHC 05/17-1