

## Product Information

### Anti-MAP Kinase, Activated

#### (Diphosphorylated ERK1&2) antibody, Mouse monoclonal

clone MAPK-YT, purified from hybridoma cell culture

Product Number **M 9692**

### Product Description

Anti-MAP Kinase, Activated (Diphosphorylated ERK-1&2) antibody, Mouse monoclonal (mouse IgG1 isotype) is derived from the MAPK-YT hybridoma produced by the fusion of mouse myeloma cells (NS1) and splenocytes from BALB/c mice immunized with a synthetic phosphorylated peptide containing 11 amino acids (HTGFLT-pEYpVAT) corresponding to the phosphorylated form of ERK-activation loop, conjugated to KLH.<sup>1</sup> The isotype is determined by a double diffusion immunoassay using Mouse Monoclonal Antibody Isotyping Reagents, Product Number ISO2.

Monoclonal Anti-MAP Kinase, Activated (Diphosphorylated ERK-1&2) is specific for the active, dually-phosphorylated form of MAP kinase (ERK-1 and ERK-2, 44 kDa and 42 kDa, respectively).<sup>1</sup> The epitope recognized by the antibody contains the phosphorylated threonine and tyrosine residues within the regulatory site of active MAP kinase (Thr<sup>183</sup> and Tyr<sup>185</sup> in ERK-2). It does not recognize the non-phosphorylated or the monophosphorylated forms of the MAP kinase molecule or the diphosphorylated form of Jun-kinase (JNK) and p38 MAP kinase. The antibody may be used for immunoblotting<sup>1,9</sup> (cultured cells and tissue extracts), immunocytochemistry,<sup>14</sup> immunoprecipitation, immunohistochemistry<sup>12</sup> (formalin and formaldehyde-fixed sections), and ELISA. Reactivity has been observed with human,<sup>14</sup> bovine, rat, mouse, hamster,<sup>15</sup> *Xenopus*,<sup>12</sup> *Drosophila*, *Spodoptera frugiperda*, *C. elegans*,<sup>13</sup> and yeast.

MAP kinase (MAPK, mitogen-activated protein kinase, also termed ERK, extracellular regulated protein kinase),<sup>2,3</sup> consists of a family of protein kinases that are considered to play a crucial role in various signal transduction pathways leading signals of growth factor, as well as G protein-coupled receptors to their intracellular targets.<sup>4,5</sup> MAP kinase was shown to regulate several cellular processes among them proliferation, differentiation, cellular morphology, and oncogenesis.<sup>4,5</sup> Molecular cloning has established that MAP kinase (ERKs) consists of at least four isoforms: ERK-1 (p44<sup>mapk</sup>), ERK-2 (p42<sup>mapk</sup>), ERK-3,

and ERK-5.<sup>2</sup> Activation of ERK-1 and -2 in mitogen-stimulated cells is directly mediated by MAP kinase kinase (MAPKK or MEK), a dual-specificity protein kinase, which phosphorylates both threonine and tyrosine residues in the regulatory sites of MAP kinase.<sup>6,7</sup>

Following activation, MAP kinase phosphorylates several nuclear targets, including transcription factors as well as membrane and cytoskeletal proteins.<sup>4,5</sup> Termination of MAP kinase signaling appears to be mediated by MAP kinase phosphatase, MKP-1, a dual specificity Thr/Tyr phosphatase which dephosphorylates and inactivates MAP kinase.<sup>8</sup> MAP kinase isoforms appear to be widely expressed (central nervous system, thymus, spleen, heart, lung, and kidney) and are expressed in high levels in PC12 cells and fibroblasts.

Antibodies that react specifically with the active form of MAP kinases can be used for studying specific activation requirements, differential tissue expression, and intracellular localization of the active form of MAP kinase in normal and neoplastic tissue.

### Reagent

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

Antibody Concentration: Approx. 1.5-2 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 at -20 °C. 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 dilutions should be discarded if not used within 12 hours.

## Product Profile

Immunoblotting: a working antibody concentration of 0.5-1 µg/ml is recommended using a whole cell extract of the RAT-1 cell line treated with vanadate and H<sub>2</sub>O<sub>2</sub>.

**Note:** In order to obtain the best results using various techniques and preparations, we recommend determining optimal working dilutions by titration.

## References

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