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Product Information

Phosphatase Inhibitor Cocktail 1

For Serine/Threonine Protein Phosphatases and L-Isozymes of Alkaline Phosphatases

Product Number **P2850**

Storage Temperature 2-8 °C

Product Description

Crude cell extracts contain a number of endogenous enzymes, such as proteases and phosphatases, which are capable of modifying the proteins present in the extract. The best way to improve the yield of intact proteins is to add inhibitors of these enzymes known to be present in the source material. This phosphatase inhibitor cocktail has been optimized and tested for L-isozymes of alkaline phosphatase as well as serine/threonine protein phosphatases, such as protein phosphatases 1 and 2A.

Components

The individual components of this proprietary formulation have specific inhibitory properties. A description of each inhibitor is given below.

Cantharidin (Product Code C7632) inhibits protein phosphatase 2A (PP-2A).^{1,2}

Bromotetramisole (Product Code B8132) inhibits alkaline phosphatases.

Microcystin LR (Product Code M2912) inhibits protein phosphatases 1 and 2A (PP-1 and PP-2A).³

Precautions and Disclaimer

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

Preparation Instructions

The cocktail is supplied as a clear solution in dimethyl sulfoxide (DMSO). The product has been sterile filtered through a 0.2 µm membrane and the bottles are aseptically filled.

Storage/Stability

The cocktail is shipped on wet ice and storage at 2-8 °C is recommended. The product, as supplied, is stable for four years from the date of manufacture.

Procedure

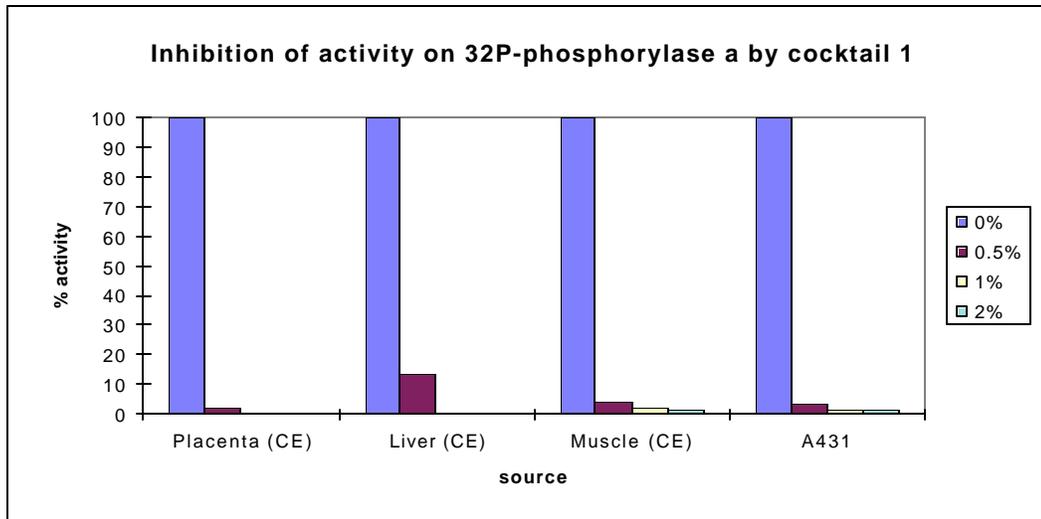
One ml will inhibit phosphatase activities found in the 100,000 x g supernatant from human placenta, bovine liver, rabbit muscle, A431 or Jurkat cell extracts at a protein concentration of approximately 5 mg/ml.

One ml of cocktail solution is used to prepare 100 ml of supernatant that contains a maximum of 500 mg of protein. Therefore, 1 ml of cocktail should be added per 500 mg of protein extracted from the tissue used or 1 ml of cocktail solution per 100 ml of extraction buffer.

This product has been tested on cell extracts from various animal tissues (cytosolic and Triton X-100 extracts of bovine liver and human placenta; cytosolic extract of rabbit muscle; Triton X-100 extracts of A431 and Jurkat cells). It was found to inhibit L-isozymes of alkaline phosphatase as measured with p-nitrophenyl phosphate (pNPP) at pH 10.4, and serine/threonine protein phosphatase activities as measured by dephosphorylation of ³²P-Ser/Thr-myelin basic protein or ³²P-Ser phosphorylase at pH 7.6.

References

1. Jain, M.K. (1982) Handbook of Enzyme Inhibitors, pp. 222. John Wiley and Sons, New York, NY.
2. Jain, M.K. (1982) Handbook of Enzyme Inhibitors, pp. 334. John Wiley and Sons, New York, NY.
3. Nishiwaki, S. et al., FEBS, 279, **No. 1** 115-118, Feb., 1992.

Appendix

Sigma brand products are sold through Sigma-Aldrich, Inc.

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