

Specification

**5,5'-Dithio-bis(2-Nitrobenzoic Acid) BioChemica**

**A3378**

<b>Physical Description:</b>	Solid
<b>Product Code:</b>	A3378
<b>Product Name:</b>	5,5'-Dithio-bis(2-Nitrobenzoic Acid) <i>BioChemica</i>
<b>Specifications:</b>	Assay (titr.): min. 99 % E 1 %/1 cm (307 nm; 0.001 %; MeOH): 330 - 370 Heavy metals (as Pb): max. 0.001 % Water (K.F.): max. 1.0 % Fe: max. 0.001 %
<b>WGK:</b>	1
<b>Storage:</b>	RT
<b>Molecular Formula:</b>	$C_{14}H_8N_2O_8S_2$
<b>M:</b>	396.36 g/mol
<b>CAS:</b>	69-78-3
<b>EINECS:</b>	200-714-4
<b>CS:</b>	29309098
<b>Comment</b>	<p>When an enzyme-catalyzed reaction creates a product with a reactive grouping not present in the substrate, it may be possible to include a reagent which will react directly with the product to form a chromophore. The chromogenic reactant should not interfere with the enzymic reaction. 5,5'-Dithiobis-2-nitrobenzoic acid (DTNB, Ellman's reagent) reacts with free thiols in an exchange reaction that produces the yellow 4-nitrothiolate anion. For the assay of acetylcholine esterase (E.C. 3.1.1.7), acetylthiocholine iodide (see product no. A3754) is used as a synthetic substrate, replacing the natural substrate acetylcholine. This enzyme hydrolyses the substrate to yield acetate and thiocholine. The free thiol group of thiocholine reacts with DTNB included in the assay mixture, producing the yellow 4-nitrothiolate. The release of this yellow anion is followed at 412 nm. The assay may be performed at 30°C in 0.1 M sodium phosphate buffer with 10 mM DTNB and 12.5 mM acetylthiocholine iodide (1). <b>Stability:</b> Solutions of DTNB are not very stable. You may keep them refrigerated for 3-4 days. Freezing improves stability. It is recommended to prepare the solution fresh each time.</p>
<b>Bibliography</b>	<p>(1) John, R.A. (1993) Photometric assays in <b>Enzyme Assays: A Practical Approach</b> p. 81 (Eisenthal, R. &amp; Danson, M.F. eds.) Oxford University Press; Oxford, New York, Tokyo.</p>