

## PRODUCT DATA SHEET

<b>PRODUCT:</b>	<b>Phosphoinositide 3-kinase p110<math>\gamma</math> (human) (recombinant)</b> [PI(3)K p110 $\gamma$ (human)(recombinant)]
<b>PRODUCT NUMBER:</b>	P-1291
<b>LOT NUMBER:</b>	A8516
<b>SOURCE:</b>	Recombinant full length human phosphoinositide-3-kinase p110 $\gamma$ fused to a His-tag at the N-terminus. Produced in Sf9 cells.
<b>PURITY:</b>	$\geq 95\%$ by SDS-PAGE
<b>SPECIFIC ACTIVITY:</b>	Approx. <b>4.5 nmoles/min/mg</b> using phosphatidylinositol as the substrate according the following protocol:  <b>Lipid kinase activity test.</b> Kinase reaction buffer: 20 mM Tris-HCl (pH 7.4); 4 mM MgCl <sub>2</sub> ; 100 mM sodium chloride.  The reaction in a total volume of 100 $\mu$ l contained 1X reaction (final concentration), the enzyme (4 reactions with 1,2,3 and 4 $\mu$ g were performed), 10 $\mu$ g phosphatidylinositol, 20 $\mu$ M cold ATP and 10 $\mu$ Ci <sup>32</sup> P-ATP. The reaction was started with the addition of the <sup>32</sup> P-ATP and it was incubated for 10 min. at 37 °C.



The reaction was stopped by adding 150  $\mu$ l 1M HCl. The lipids were extracted with 400  $\mu$ l chloroform : methanol (1:1) by vortexing. The extract was washed three times with 200  $\mu$ l 1M HCl.

The incorporation of  $^{32}\text{P}$  in the extracted lipids was measured by mixing of the extract with 3 ml scintillation cocktail (ROTH) and counting in a liquid scintillation analyzer.

To determine the specific activity of PI3K p110 $\gamma$  the assay has been performed under the same conditions stated above by using 10  $\mu\text{Ci}$   $^{32}\text{P}$ -ATP, with a value of 10,000 units.

The average value of measurements of the lipid extracts of the performed 4 reactions was 227 units for 1  $\mu\text{g}$ . Dividing 10,000 by 227 a factor of 44 times less for the incorporation has been determined.

The total ATP (20  $\mu\text{M}$ ) was divided by 44 giving 0.454  $\mu\text{M}$  for 10 min. (the time of the reaction) or 45.4 nM/min/ $\mu\text{g}$ . Recalculation of this value for a 1 liter solution, (as this was a 100  $\mu\text{l}$  reaction) results in **4.5 nmoles/min/mg**.

## FORMULATION:

Liquid. 0.5 mg/ml in 10mM HEPES, pH 7.5, 100mM sodium chloride, 0.5 mM magnesium chloride and 50% glycerol.

## STORAGE & HANDLING:

Keep cool and dry at  $-20^{\circ}\text{C}$ .

CAUTION: For research use only. Not for human or drug use. The pharmacological and toxicological properties of this product have not been fully investigated. Use caution when handling. Do not use this compound if you are not fully trained or are unaware of the hazards involved.

Verified:

A handwritten signature in black ink, appearing to be 'JK'.