

## PDE12, Active

Full-length recombinant protein expressed in Sf9 cells

Catalog # P100-310G

Lot # T4241-14

### Product Description

Recombinant full-length human PDE12 was expressed by baculovirus in Sf9 insect cells using an N-terminal GST tag. The gene accession number is [NM\\_177966](#).

### Alternative Name(s)

2'-PDE

### Formulation

Recombinant protein stored in 50mM Tris-HCl, pH 7.5, 150mM NaCl, 10mM glutathione, 0.1mM EDTA, 0.25mM DTT, 25% glycerol.

### Storage and Stability

Store product at -70°C. For optimal storage, aliquot target into smaller quantities after centrifugation and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles.

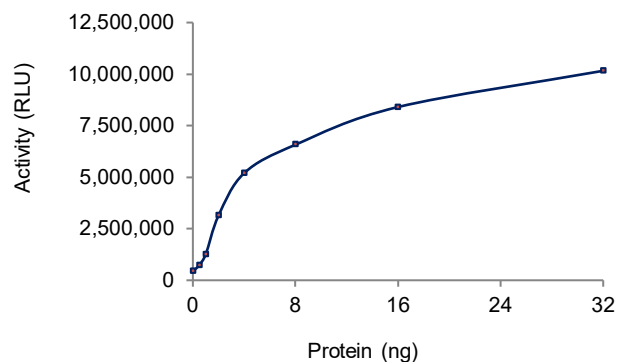
### Scientific Background

PDE12 is 2', 5'-phosphodiesterase 12 protein that belongs to the CCR4/nocturin family. PDE12 cleaves the 2',5'-phosphodiester bond linking adenosines of the 5'-triphosphorylated oligoadenylates. Triphosphorylated oligoadenylates, which are referred to as 2-5A, modulates in the 2-5A system that is one of the major pathways for antiviral and antitumor functions induced by interferons (IFNs) (1). Suppression of PDE12 induces reduction of viral replication in Hela cells, thus counteracting the antiviral pathway probably by inhibiting the 2-5A system.

### References

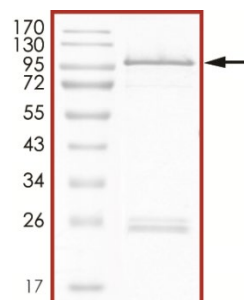
1. Kubota K. et.al: "Identification of 2'-phosphodiesterase, which plays a role in the 2-5A system regulated by interferon." J. Biol. Chem. 279:37832-37841,2004.

### Specific Activity



The specific activity of PDE12 was determined to be **1,242 nmol/min/mg** as per activity assay protocol.

### Purity



The purity of PDE12 was determined to be **>90%** by densitometry. Approx. MW **100kDa**.

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Full-length recombinant protein expressed in Sf9 cells

Catalog #	P100-310G
Specific Activity	1,242 nmol/min/mg
Lot #	T4241-14
Purity	>90%
Concentration	0.1 µg/µl
Stability	1 yr at -70°C from date of shipment
Storage & Shipping	Store product at -70°C. For optimal storage, aliquot target into smaller quantities after centrifugation and store at recommended temperature. For most favorable performance, avoid repeated handling and multiple freeze/thaw cycles. Product shipped on dry ice.

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# Activity Assay Protocol

## Reaction Components

### Active Enzymes

Active PDE12 (Catalog#: P100-310G)

Active OAS1 (Catalog #: O711-30H)

### Activator

Poly I:C (Sigma Cat# P1530) dissolved in 200  $\mu$ L saline (150mM NaCl) to make 5 mg/mL working stock.

### 4X OAS/PDE Reaction Buffer

40mM Tris-HCl, pH 7.5, 100mM NaCl, 20mM MgCl<sub>2</sub>, 0.04% (w/v) Prionex, 4mM DTT (added fresh)

### AMP-Glo™ Assay (Promega, Catalog #: V5011)

AMP, 10 mM

Ultra Pure ATP, 10 mM

AMP-Glo™ Reagent I

AMP-Glo™ Reagent II

Kinase-Glo™ One Solution

## Assay Protocol

**Step 1.** Thaw active OAS1 and PDE12 enzymes on ice. Equilibrate the reaction buffer, Poly I:C and ATP to ambient temperature.

**Step 2.** Prepare the following working solutions with 1X Reaction Buffer:

- o 4X final concentration of Active OAS1 (Catalog #: O711-30H)
- o 4X final concentration of Active PDE12 (Catalog #: P100-310G)
- o 2X final concentration of substrate solution containing Poly I:C (20  $\mu$ g/mL) and ATP (200  $\mu$ M)

**Step 3.** In a half-area solid white 96-well plate, add the following components to bring the reaction volume to 20  $\mu$ L:

**Component 1.** 5  $\mu$ L of 4X Active OAS1 (100 nM OAS1 used for PDE12 activity titration)

**Component 2.** 10  $\mu$ L of 2X substrate solution

**Component 3.** 5  $\mu$ L of serial diluted 4X Active PDE12

*Note 1:* A blank control can be set up as outlined in step 3 by replacing the enzyme working solution with an equal volume of reaction buffer.

*Note 2:* For PDE12 inhibition assay, replace component 3 with a pre-incubated PDE12/compound mix.

**Step 4.** Mix the plate for 1 minute on a tabletop orbital shaker. Seal the assay wells with a plate sealer and incubate at 37°C for 30 minutes.

*Note:* A series of AMP standard solutions can be dispensed to the same plate at the end of the incubation period in order to determine the specific activity of the enzyme.

**Step 5.** Equilibrate plate to room temperature. Add 20  $\mu$ L of AMP-Glo™ Reagent I to all wells, mix by shaking for 1 minute. Incubate plate at room temperature for 60 minutes.

**Step 6.** Prepare AMP Detection Solution by adding AMP-Glo™ Reagent II to Kinase-Glo™ One Solution at a 1:100 volume ratio. Add 40  $\mu$ L of the Detection Solution to all wells. Mix for 1 minute and incubate at room temperature for 45 minutes.

**Step 7.** Read the plate on a microplate luminometer.

**Step 8.** Using the AMP standard curve, determine the concentration of AMP produced ( $\mu$ M) and calculate the enzyme specific activity as outlined below. For a detailed protocol of how to determine AMP amount from RLU, see AMP-Glo™ Assay protocol at Promega's website: [www.promega.com/protocols](http://www.promega.com/protocols).

$$\text{Enzyme Specific Activity (SA) (nmol/min/mg)} \\ = \frac{[\text{AMP}] (\mu\text{M}) \times \text{Reaction Volume} (\mu\text{L})}{\text{Reaction Time (min)} \times \text{Enzyme Amount (mg)}} \times 10^{-3}$$

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# SAFETY DATA SHEET

## Article 1 – Product Identification

**Product Name: PDE12, Active**

**Catalog # P100-310G**

*This product is sold only for research use by qualified laboratory personnel, and is not to be used as a drug, medical device, food additive, cosmetic, nor household chemical. It is not to be used in diagnostic, therapeutic, consumer, agricultural, nor pesticidal applications.*

Manufacturer's Name: SignalChem Biotech Inc.  
 Street Address: 110-13120 Vanier Place  
 City, Prov. Postal Code: Richmond, BC, V6V 2J2  
 Fax: 604-232-4601  
 EMERGENCY PHONE: 604-232-4600

## Article 2 - Hazard Identification

- **WHMIS Classification:** Not WHMIS controlled.
- **GHS classification:** Skin irritation (Category 3); Eye irritation (Category 2B).
- **Hazard Pictograms:** none.
- **Signal words:** Warning.
- **Hazard statements:** Causes mild skin irritation (H316); Causes eye irritation (H320).
- **Precautionary statements:** IF IN EYES: Rinse cautiously with water for several minutes. Remove contact lenses, if present and easy to do. Continue rinsing. (P305 + P351 + P338).
- **Other hazards:** none known.

## Article 3 – Composition/Information on Ingredients

**Chemical Characterization:** Mixtures.

**Description:** This product consists of the substances listed below.

Common name	Chemical name	CAS-No.	Concentration
Glycerol	Glycerol	56-81-5	≤25%
NaCl	Sodium chloride	7647-14-5	≤1.753 %
Tris-HCl; Tris (hydroxymethyl) aminomethane hydrochloride	2 - Amino - 2 - (hydroxymethyl) propane - 1, 3 - diol hydrochloride	1185-53-1	<0.8%
Glutathione	Glutathione	70-18-8	0.307%
Protein		No data available	≤0.02%
DTT; Dithiothreitol	(R*,R*)-1,4-Dimercaptobutane-2,3-diol	3483-12-3	0.0038%
EDTA	Ethylenediaminetetracetic acid	6381-92-6	0.0037%

## Article 4 – First-aid Measures

- **General information:** Consult a physician by providing the SDS.
- **After inhalation:** Breathe in fresh air. If cannot breathe, give artificial respiration and consult a physician.
- **After skin contact:** Immediately wash with soap and plenty of water and rinse thoroughly. Consult a physician.
- **After eye contact:** Rinse opened eyes with plenty of water for at least 15 minutes. Consult a physician.
- **After swallowing:** rinse the mouth with plenty of water and consult a physician.

## Article 5 - Fire-fighting Measures

- **Suitable extinguishing media:** Use water spray, extinguishing powder, carbon dioxide, or other appropriate measure that is suitable to the environment.
- **Specific hazards arising from the substance or mixture:** None known.
- **Special protective equipment and precautions for fire-fighters:** Self-contained breathing apparatus if necessary.

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## Article 6 – Accidental Release Measures

- **Personal precautions, protective equipment and emergency procedures:** Apply standard laboratory practices and personal protective equipment. Avoid breathing vapors, mist, or gas. Ensure adequate ventilation.
- **Environmental precautions:** Do not allow to enter drains.
- **Methods and materials for containment and cleaning up:** Absorb on sand or vermiculite and place in closed containers for disposal.

## Article 7 - Handling and Storage

- **Precautions for safe handling:** Wear chemical safety goggles and compatible chemical-resistant gloves. Avoid inhalation, contact with eyes, skin or clothing.
- **Conditions for safe storage:** Store in a dry and well-ventilated place in -70 °C. Keep container upright and tightly closed.

## Article 8 - Exposure Controls/Personal Protection

- **Components with limit monitoring values at workplace:**  
Glycerol (CAS-No: 56-81-5)

Values	Control parameters	Regulations
TWA	10 mg/m <sup>3</sup> for mist	British Columbia, Canada
TWA	3 mg/m <sup>3</sup> for respirable mist	British Columbia, Canada
TWA	10 mg/m <sup>3</sup>	Alberta, Canada
TWAEV	10 mg/m <sup>3</sup>	Ontario, Canada
TWAEV	10 mg/m <sup>3</sup>	Quebec, Canada
TWA	10 mg/m <sup>3</sup>	USA

- **Appropriate engineering controls:**  
Apply adequate ventilation including mechanical exhaust or laboratory fume hood. Follow standard laboratory practices.
- **Individual protection measures:**  
**Respiratory protection:**  
Use appropriate respirator if there is inadequate ventilation by following the government standards.  
**Hand protection:**  
Wear gloves and use proper glove removal technique to avoid skin contact. Discard gloves after use by following the applicable laboratory regulations. Wash and dry hands.  
**Eye/face protection:**  
Safety goggles with side-shields approved under appropriate government standards.  
**Skin/body protection:**  
Use appropriate clothing, footwear and any additional protection measures to protect from splashing or contamination.

## Article 9 – Physical and Chemical Properties

<b>Appearance:</b> Colorless fluid.	<b>Danger of explosion:</b> Product does not present an explosion hazard.
<b>Odour/Odour Threshold:</b> Not determined.	<b>Explosion limits:</b> Lower: 0.9 Vol %; Upper: 0.0 Vol %.
<b>pH:</b> Not available.	<b>Decomposition temperature:</b> Not available.
<b>Melting point/freezing point:</b> Not determined.	<b>Vapor pressure at 20 °C:</b> 0.1 hPa
<b>Boiling point/Boiling range:</b> 100 °C.	<b>Density:</b> Not determined.
<b>Flash point:</b> > 100 °C.	<b>Relative density:</b> Not determined.
<b>Flammability (solid, gaseous):</b> Not determined.	<b>Vapor density:</b> Not determined.
<b>Ignition temperature:</b> 400 °C.	<b>Evaporation rate:</b> Not determined.
<b>Auto-igniting:</b> Product is not self-igniting.	<b>Solubility in / Miscibility with Water:</b> Fully miscible.

## Article 10 - Stability and Reactivity

- **Reactivity:** Stable under recommended transport and storage conditions.
- **Chemical stability:** Stable under recommended transport and storage conditions.
- **Possible hazardous reactions:** No dangerous reactions known.
- **Conditions to avoid:** Heat and moisture.
- **Incompatible materials:** Strong acids/bases, strong oxidizing/reducing agents.
- **Hazardous decomposition products:** Carbon oxides may formed under fire conditions; no known decomposition information for other decomposition products.

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## Article 11 - Toxicological Information

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- **Acute toxicity:** Not available.
- **LD/LC50:** Not available.
- **Skin corrosion/irritation:** Not available.
- **Serious eye damage/eye irritation:** Not available.
- **Respiratory or skin sensitization:** Not available.
- **Germ cell mutagenicity:** Not available.
- **Carcinogenicity:** No components are listed in IARC, or NTP, or OSHA, or ACGIH.
- **Reproductive toxicity:** Not available.
- **Teratogenicity:** Not available.
- **Specific target organ toxicity - single exposure/ - repeated exposure (GHS):** Not available.
- **Aspiration hazard:** Not available.
- **Potential health effects:**  
**Inhalation:** May be harmful if inhaled. May cause respiratory tract irritation.  
**Ingestion:** May be harmful if swallowed.  
**Skin:** May be harmful if absorbed through skin. May cause skin irritation.  
**Eyes:** May cause eye irritation.
- **Signs and Symptoms of Exposure:**  
Prolonged or repeated exposure can cause: Nausea, Dizziness.
- **Synergistic effects:** Not available.

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## Article 12 - Ecological Information

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- **Eco-toxicity:** Not applicable.
- **Biodegradability:** Not applicable.
- **Bio-accumulative potential:** Not applicable.
- **Mobility in soil:** Not applicable.
- **PBT and vPvB assessment:** Not applicable.
- **Other adverse effects:** Not applicable.

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## Article 13 - Disposal Considerations

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- **Disposal methods:** In accordance to applicable national, regional, or local laws and regulations. For additional handling information and protection of employees please refer to Article 7 and 8.
- **Contaminated packaging:** Disposal should be made in accordance to official regulations. Use water or cleansing agents to clean the area.

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## Article 14 - Transport Information

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- **DOT:** Not dangerous goods.
- **IMDG:** Not dangerous goods.
- **IATA:** Not dangerous goods.

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## Article 15 - Regulatory Information

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- **WHMIS Classification:** Non-hazardous.
- **GHS label elements:** Not applicable.
- **Signal word:** Not applicable.
- **Hazard statements:** Not applicable.

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## Article 16 - Other Information

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The above information is believed to be correct but does not purport to be all-inclusive and shall be used only as a guide. SignalChem shall not be held liable for any damage resulting from handling or from contact with the above product. See the Technical Specification, Packing Slip, Invoice, and Product Catalog for additional terms and conditions of sale.

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