

**DESCRIPTION**

**Source** *E. coli*-derived human Sonic Hedgehog/Shh protein  
Cys24-Gly197 (Cys24Ile-Ile), with an N-terminal Met  
Accession # NP\_000184

**N-terminal Sequence Analysis** Met

**Predicted Molecular Mass** 20 kDa

**SPECIFICATIONS**

**SDS-PAGE** 22 kDa, reducing conditions

**Activity** Measured by its ability to induce alkaline phosphatase production by C3H10T1/2 mouse embryonic fibroblast cells. Nakamura, T. *et al.* (1997) *Biochem. Biophys. Res. Commun.* **237**:465.  
The ED<sub>50</sub> for this effect is 0.1-0.4 µg/mL.

**Endotoxin Level** <0.01 EU per 1 µg of the protein by the LAL method.

**Purity** >95%, by SDS-PAGE visualized with Silver Staining and quantitative densitometry by Coomassie® Blue Staining.

**Formulation** Lyophilized from a 0.2 µm filtered solution in PBS and NaCl with BSA as a carrier protein. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

**Reconstitution** Reconstitute at 100-200 µg/mL in sterile PBS containing at least 0.1% human or bovine serum albumin, and allow up to 24 hours at 2 to 8 °C for complete reconstitution.

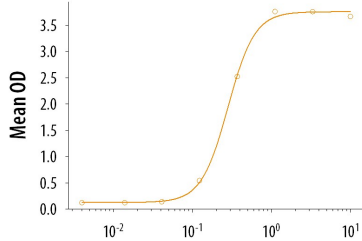
**Shipping** The product is shipped at ambient temperature. Upon receipt, store it immediately at the temperature recommended below.

**Stability & Storage** Use a manual defrost freezer and avoid repeated freeze-thaw cycles.

- 12 months from date of receipt, -20 to -70 °C as supplied.
- 1 month, 2 to 8 °C under sterile conditions after reconstitution.
- 3 months, -20 to -70 °C under sterile conditions after reconstitution.

**DATA**

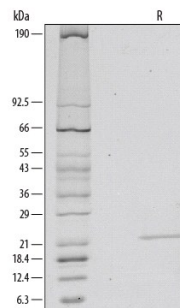
**Bioactivity**



Recombinant Human Sonic Hedgehog (C24II) N-Term (µg/mL)

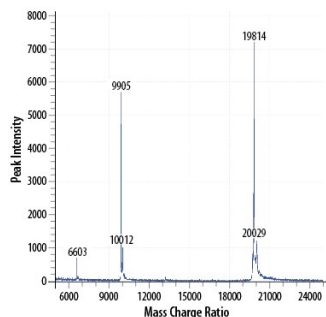
Recombinant Human Sonic Hedgehog/Shh (C24II), N-Terminus (Catalog # 1845-SH) induces alkaline phosphatase production by the C3H10T1/2 mouse embryonic fibroblast cell line. The ED<sub>50</sub> for this effect is 0.1-0.4 µg/mL.

**SDS-PAGE**



1 µg/lane of Recombinant Human Sonic Hedgehog/Shh (C24II), N-Terminus was resolved with SDS-PAGE under reducing (R) conditions and visualized by silver staining, showing a single band at 22 kDa.

**Mass Spectrometry**



MALDI-TOF analysis of Recombinant Human Sonic Hedgehog/Shh (C24II), N-Terminus. The major peak corresponds to the calculated molecular mass, 19814 Da. The minor peak at 20029 is a matrix-associated artifact of the MALDI-TOF.

## BACKGROUND

Sonic Hedgehog (Shh) is expressed in embryonic tissues that are critical for the patterning of the developing central nervous system, somite, and limb. It is also involved in whisker, hair, foregut, tooth, and bone development. Shh regulates neural and hematopoietic stem cell fate and is important for thymocyte differentiation and proliferation as well as T cell determination. In adult tissue Shh is associated with cancer development and tissue remodeling following injury (1-3). Human Shh encodes a 462 amino acid (aa) precursor protein that is autocatalytically processed to yield a non-glycosylated 19 kDa N-terminal fragment (Shh-N) and a glycosylated 25 kDa C-terminal protein (Shh-C) (4). Shh-C, which is responsible for the intramolecular processing of Shh, is rapidly degraded following Shh proteolysis (5). Shh-N is highly conserved, sharing >98% aa identity between mouse, human, rat, canine, porcine, and chicken Shh-N. Shh-N can be palmitoylated at its N-terminal cysteine and modified by cholesterol addition at its C-terminus (6). These modifications contribute to the membrane tethering of Shh as well as its assembly into various sized multimers (6-9). Lipid modification and multimerization greatly increase Shh-N receptor binding affinity and signaling potency (5, 6, 8, 9). Monomeric and multimeric Shh can be released from the plasma membrane by the cooperative action of DISP1, SCUBE2, and TACE/ADAM17 (10-12). Modifications also extend the effective range of Shh functionality and are required for the development of protein gradients important in tissue morphogenesis (9, 13). Canonical signaling of Shh is mediated by a multicomponent receptor complex that includes Patched (PTCH1, PTCH2) and Smoothened (SMO) (14). The binding of Shh to PTCH releases the basal repression of SMO by PTCH. Shh activity can also be regulated through interactions with heparin, glypicans, and membrane-associated Hip (hedgehog interacting protein) (13, 15, 16).

## References:

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