

**DESCRIPTION**

**Source** Chinese Hamster Ovary cell line, CHO-derived  
Phe17-Gln317, with a C-terminal 10-His tag  
Accession # AAC95490

**N-terminal Sequence Analysis** Phe17

**Predicted Molecular Mass** 34.7 kDa

**SPECIFICATIONS**

**SDS-PAGE** 75-90 kDa, reducing conditions

**Activity** Measured by the ability of the immobilized protein to support the adhesion of the MCF-7 human breast cancer cells.  
When  $5 \times 10^4$  cells/well are added to recombinant human IBSP-coated plates (3  $\mu\text{g}/\text{mL}$  with 100  $\mu\text{L}/\text{well}$ ), approximately 60-80% will adhere after 30 minutes at 37° C.  
**Optimal concentration depends on cell type as well as the application or research objectives.**

**Endotoxin Level** <1.0 EU per 1  $\mu\text{g}$  of the protein by the LAL method.

**Purity** >90%, by SDS-PAGE under reducing conditions and visualized by silver stain.

**Formulation** Lyophilized from a 0.2  $\mu\text{m}$  filtered solution in MES and NaCl. See Certificate of Analysis for details.

**PREPARATION AND STORAGE**

**Reconstitution** Reconstitute at 100  $\mu\text{g}/\text{mL}$  in sterile PBS.

**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.

**BACKGROUND**

IBSP (integrin-binding sialoprotein; also BSP or bone sialoprotein (II)) is a 55 - 75 kDa, secreted, variably glycosylated, monomeric noncollagenous member of the SIBLING family of extracellular matrix (ECM) proteins (1 - 3). It is principally associated with the early stages of bone mineralization. BSP is synthesized as a 317 amino acid (aa) precursor that contains a 16 aa signal sequence and a 301 aa mature region (4 - 6). The mature segment is divided into a basic N-terminus (aa 17 - 62), a central region (aa 63 - 233), and an acidic C-terminus (aa 234 - 317) (7).

Functional segments associated with the mature molecule include a type I collagen binding domain (aa 19 - 46), two non-RGD cell binding sites (aa 30 - 57 and 261 - 281), an RGD  $\alpha\beta_3$  integrin-binding site (aa 286 - 288) and two potential hydroxyapatite (HAP) nucleation domains (aa 76 - 83 and 151 - 158) (3, 4, 8 - 11). HAP formation requires a BSP nucleation site composed of at least eight consecutive glutamic acid residues and, likely, a contribution from a BSP-associated co-nucleator (10, 12). BSP is highly glycosylated, sulfated, and phosphorylated. Phosphorylation may impact HAP growth, while carbohydrate may regulate cell adhesion (1, 3, 13). Mature human BSP is 70%, 72%, 78%, and 72% aa identical to porcine, rat, canine, and mouse BSP, respectively. BSP is synthesized by megakaryocytes/platelets, osteoblasts, osteocytes, odontoblasts, osteoclasts, and bone marrow stromal cells (14 - 17).

**References:**

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