

human cell expressed PIGF^{hcx}

Source	A DNA sequence encoding the human PIGF protein sequence (containing the signal peptide sequence, and the mature human PIGF sequence) was expressed in modified human 293 cells.
Molecular Mass	Symansis PIGF ^{hcx} migrates as two broad bands between 17 and 30 kDa on SDS-PAGE due to post-translational modifications, in particular glycosylation. This compares with the unmodified PLGF that has a predicted molecular mass of 16.7kDa.
pI	Symansis PIGF ^{hcx} separates into a number of glycoforms with varying pI values on 2D PAGE due to post-translational modifications, in particular glycosylation. This compares with the unmodified PLGF that has a predicted pI of 6.3. (See 2D densitometry image below)
% Carbohydrate	Symansis purified PIGF ^{hcx} consists of 0-38% carbohydrate by weight.
Glycosylation	Symansis PIGF ^{hcx} has N-linked and O-linked oligosaccharides.
Purity	>95%, as determined by SDS-PAGE and visualized by Coomassie Brilliant Blue.
Formulation	When reconstituted in 0.5 ml sterile phosphate-buffered saline, the solution will contain 1% human serum albumin (HSA) and 10% trehalose.
Reconstitution	It is recommended that 0.5 ml of sterile phosphate-buffered saline be added to the vial.
Storage	Lyophilized products should be stored at 2 to 8°C. Following reconstitution short-term storage at 4°C is recommended, with longer-term storage in aliquots at -18 to -20°C. Repeated freeze thawing is not recommended.

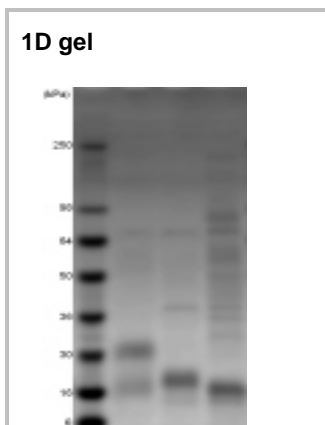
Background Information

PIGF (Placental growth Factor) was isolated initially as a cDNA from a human placenta cDNA library. PIGF is expressed also in human umbilical vein endothelial cells colon and mammary carcinomas. Placenta growth factor (PIGF) is a member of the vascular endothelial growth factor (VEGF) family of growth factors. PIGF and VEGF share primary structural as well as limited amino acid sequence homology with the A and B chains of PDGF. The biologically active form of this protein is a disulfide-linked dimer. The N-glycosylated dimeric protein is secreted and stimulates the proliferation of endothelial cell lines and supports angiogenesis.

PIGF has been renamed PIGF-1 after the discovery of PIGF-2, which has a 21 amino acid insertion not present in PIGF. PIGF-1 and PIGF-2 compete in a dose-dependent way with the 165 amino acid form of VEGF for receptor binding on endothelial cells. The PIGF proteins bind with high affinity to Flt1, but not to Flk1/KDR. Neuropilin-1 probably functions as a receptor for PIGF-2.

human cell expressed PLGF^{hex}

1D gel data

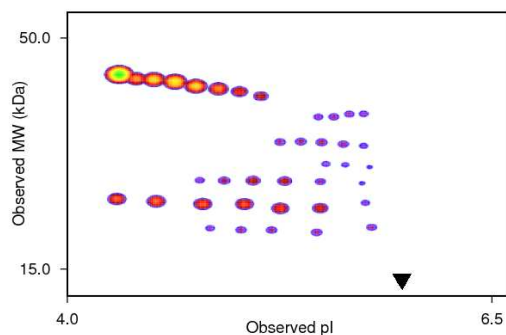


Lane 1 – MW markers; Lane 2 – PLGF^{hex}; Lane 3 – PLGF^{hex} treated with PNGase F to remove potential N-linked glycans; Lane 4 – PLGF^{hex} treated with a glycosidase cocktail to remove potential N- and O-linked glycans. 10 µg protein loaded per lane; Deep Purple™ stained.

Drop in MW after treatment with PNGase F indicates presence of N-linked glycans. A further drop in MW after treatment with the glycosidase cocktail indicates the presence of O-linked glycans. Additional bands in lane 3 and lane 4 are glycosidase enzymes.

Densitometry

Post-translational modifications result in protein heterogeneity. The densitometry scan demonstrates the purified human cell expressed protein exists in multiple glycoforms, which differ according to their level of post-translational modification. Expression of these glycoforms is highly significant for cell biology, as they more closely resemble the native human proteins.



Triangle indicates theoretical pI and MW of the protein. The original 2D gel from which the densitometry scan was derived is available upon request.

Theoretical Sequence

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MPVMRLFPCFLQLLAGLALPAVPPQQWALSAGNGSSEVEVVPFQEVWGRSYCRAL
ERLVDVVSEYPSEVEHMFSPSCVLLRCTGCCGDENLHCVVETANVTMQLLKIRS
GDRPSYVELTFSQHVRCECRPLREKMKPERCGDAVPRR
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