

human cell expressed EPO<sup>hex</sup>

<b>Source</b>	A DNA sequence encoding the human EPO protein sequence (containing the signal peptide sequence, and the mature Epo sequence) was expressed in modified human 293 cells
<b>Molecular Mass</b>	Apollo EPO <sup>hex</sup> migrates as a broad band between 25 and 40 kDa in SDS-PAGE due to post-translation modifications, in particular glycosylation. This compares with the unmodified EPO that has a predicted molecular mass of 18.4 kDa.
<b>pI</b>	The Apollo EPO <sup>hex</sup> separates into a number of isoforms in 2D PAGE due to the presence of post-translational modifications, in particular glycosylation. The pI of the isoforms range between 4.1 and 8.5. This compares with a predicted pI of 7.88 for the unmodified EPO.
<b>% Carbohydrate</b>	Purified Apollo EPO <sup>hex</sup> consists of 25-55% carbohydrate by weight.
<b>Glycosylation</b>	Apollo EPO <sup>hex</sup> has N-linked and O-linked oligosaccharides. All 3 N-linked sites are verified by peptide mass fingerprinting.
<b>Purity</b>	>95%, as determined by SDS-PAGE and visualized by silver stain.
<b>Formulation</b>	When reconstituted in 0.5 ml sterile phosphate-buffered saline, the solution will contain 1% human serum albumin (HSA) and 10% trehalose.
<b>Reconstitution</b>	It is recommended that 0.5 ml of sterile phosphate-buffered saline be added to the vial.
<b>Storage</b>	Lyophilized products should be stored at 2 to 8°C. Following reconstitution short-term storage at 4°C is recommended, and longer-term storage of aliquots at -18 to -20°C. Repeated freeze thawing is not recommended.
<b>Activity</b>	The ED50 of EPO <sup>hex</sup> is typically 0.2 - 0.8 ng/ml as measured in a cell proliferation assay using the human growth factor-dependent TF-1 cell line.
<b>Background Information</b>	<p>Erythropoietin (EPO) is a hormone produced primarily by the kidney and is the main regulator of red blood cell production. Its major functions are to promote the differentiation and development of red blood cells and to initiate the production of hemoglobin. EPO acts by binding to a specific erythropoietin receptor (EPOR) present on target cells, the red cell precursors in the bone marrow, stimulating them to transform into mature erythrocytes.</p> <p>Human EPO cDNA encodes a 193 amino acid residue precursor protein that is processed to yield a 165 amino acid residue mature protein. EPO contains one O-linked and three N-linked glycosylation sites. Glycosylation of EPO is required for EPO biological activities <i>in vivo</i>. Recombinant EPO has been approved for the treatment of anemia associated with chronic renal failure, anemia secondary to AZT treatment of AIDS, and anemia associated with cancer. There is recent evidence that recombinant EPO may be useful for cardioprotection and neuroprotection.</p> <p>For recent reviews on EPO, please refer to Bartesaghi S, <i>et al.</i> (2005) <i>Neurotoxicology</i>. <b>26</b>(5): 923-8; Brines M &amp; Cerami A, (2005) <i>Nat Rev Neurosci</i>. <b>6</b>(6): 484-94; Engert A, (2005) <i>Ann Oncol</i>. <b>16</b>(10): 1584-95.</p>

FOR RESEARCH USE ONLY

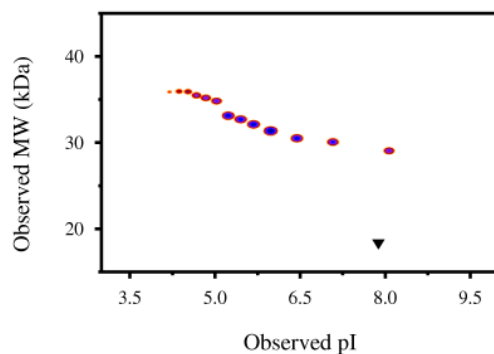


## human cell expressed EPO<sup>hex</sup>

### Densitometry

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

The 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

APPRLICDSRVLERYLLEAKEAENITTGCAEHCSLNENITVPDTKVNIFYAWKRMEVGQQAVE  
VWQGLALLSEAVLRGQALLVNSSQPWEPLQLHVDKAVSGLRSLTLLRALGAQEEAISPPD  
AASAAPLRTITADTFRKLFRVYSNFLRGKCLKLYTGEACRTGDR