SYMANSIS
CELL SIGNALING SCIENCE

Product Data Sheet

human cell expressed IL-1ra^{HCX}

Source	A DNA sequence encoding the human Interleukin-1 receptor antagonist (IL-1ra) protein sequence (including the signal peptide sequence, and the mature human Interleukin-1 receptor antagonist sequence) was expressed in modified human 293 cells.
Molecular Mass	Symansis IL-1ra^{HCX} migrates as a broad band between 18 and 23 kDa in SDS-PAGE due to post-translation modifications, in particular glycosylation. This compares with the unmodified IL-1ra that has a predicted molecular mass of 17.1 kDa.
pI	Symansis IL-1ra^{HCX} separates into a number of isoforms with a pI between 5.4 and 6.3 in 2D PAGE due to post-translational modifications, in particular glycosylation. This compares with the unmodified IL-1ra that has a predicted pI of 5.4.
% Carbohydrate	Symansis purified IL-1ra^{HCX} consists of 5-25% carbohydrate by weight.
Glycosylation	Symansis IL-1ra^{HCX} has N-linked oligosaccharides.
Purity	>95%, as determined by SDS-PAGE and visualized by silver stain.
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 and longer-term storage of aliquots at –18 to -20°C. Repeated freeze thawing is not recommended.
Activity	The ED50 of IL-1ra^{HCX} is typically 30-100 ng/ml as measured by its ability to inhibit IL-1 mediated proliferation using the murine D10S cell line.
Background Information	IL-1 receptor antagonist (IL-1ra) is a naturally occurring protein that plays an important role in regulating the activity of IL-1 by competitively binding to the IL-1 receptor and thereby inhibiting IL-1 signaling activation in target cells. The expression of IL-1ra has been detected in bone marrow monocytes, macrophages, neutrophils and T cells. IL-1 is a major cytokine involved in immune responses to infection, including both innate and acquired immunity. It promotes the activation of macrophages, and is a co-stimulatory molecule for lymphocyte proliferation, antigen presentation and T cell dependent antibody production. IL-1 is also involved in inflammatory disease states. Maintenance of a balance between IL-1 and IL-1ra is important in preventing the development or progression of inflammatory disease states. IL-1ra may exist as a number of different isoforms and these may be either secreted or intracellular forms. Additionally, IL-1ra shares 25-30% structural homology with IL-1a and IL-1b and includes one potential N-linked glycosylation site. For further information on the relationship between IL-1 and IL-1ra, please refer to Arend WP. (2002) *Cytokine Growth Factor Rev.* **13**(4-5): 323-40.

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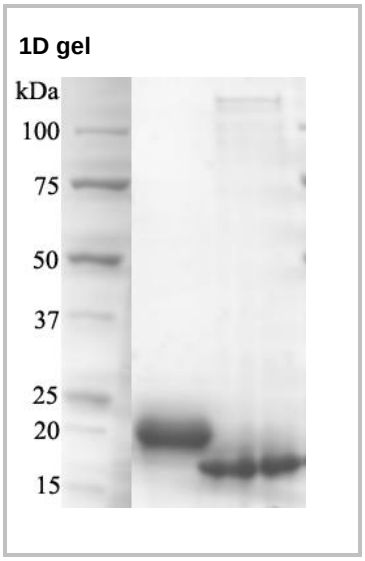


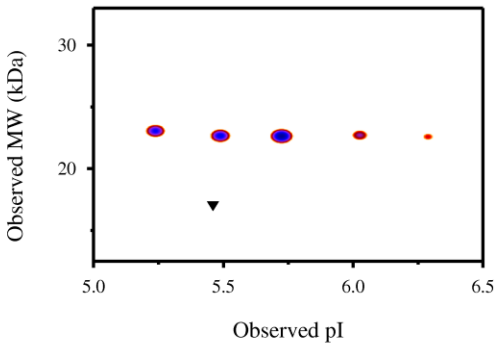
1D gel data

Lane 1 – MW markers; Lane 2 – IL-1ra^{HCX}; Lane 3 – IL-1ra^{HCX} treated with PNGase F to remove potential N-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. Faint 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 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

RPSGRKSSKMQAFRIWDVNQKTFYLRNNQLVAGYLQGPNVNLEEKIDVVPIEPHALF
LGIHGGKMCLSCVKSGDETRLQLEAVNITDLSENRKQDKRFAFIRSDSGPTTSFESAA
CPGWFLCTAMEADQPVSLTNMPDEGVMVTKFYFQEDE

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