Product Data Sheet

Human Cell Expressed IL-2 R gamma-Fc^{HCX} Chimera Catalog # **4132**

Source	A DNA sequence encoding the signal peptide and extracellular domain of human Interleukin 2 receptor gamma chain (aa 1-259) was fused to the Fc region of human IgG1 (aa 93-330). The chimeric protein was expressed in modified human 293 cells.
Molecular Mass	Symansis IL-2 R gamma-Fc ^{HCX} Chimera migrates as a band between 75 and 105 kDa in SDS-PAGE due to post-translation modifications, in particular glycosylation. This compares with the unmodified IL-2 R gamma-Fc that has a predicted molecular mass of 55.1 kDa.
pI	Symansis IL-2 R gamma-Fc ^{HCX} Chimera separates into a number of isoforms with a pI between 4.9 and 7.2 in 2D PAGE due to post-translational modifications, in particular glycosylation. This compares with the unmodified IL-2 R gamma-Fc that has a predicted pI of 6.68.
% Carbohydrate	Symansis purified IL-2 R gamma-Fc ^{HCX} Chimera consists of 25-50% carbohydrate by weight.
Glycosylation	Symansis IL-2 R gamma-Fc ^{HCX} Chimera has N-linked and O-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.
Background Information	Interleukin-2 (IL-2) is a pleiotropic glycoprotein that mediates the production of regulatory T cells and promotes T cell dependent tolerance. Additionally, IL-2 promotes the proliferation of antigen-activated T lymphocytes, facilitates the cytolytic activity of natural killer (NK) cells and stimulates anti-tumor activity in monocytes by inducing synthesis of GM-CSF, IL-1b and IL-6. IL-2 also promotes the proliferation of large granular lymphocytes and the growth and differentiation of mitogen activated B lymphocytes *in vitro*. The biological effects of IL-2 are mediated through the IL-2 receptor, which is predominantly expressed on activated T cells, B cells and monocytes. There are three subunits that may associate in different combinations to form the IL-2 receptor. These subunits include IL-2Ra, IL-2Rb, which is shared with IL-15 receptor and IL-2 R gamma (IL-2Rg) which is also known as the common cytokine receptor gamma chain as it is shared with receptors for IL-4, IL-7, IL-9, IL-15 and IL-21. Different associations of these subunits form low affinity, intermediate affinity or high affinity IL-2 receptors. The low affinity receptor comprises the IL-2Ra glycopeptide, the intermediate affinity receptor is formed by the association of IL-2Rb and the IL-2R gamma while the high affinity receptor comprises IL-2Ra, IL-2Rb and IL-2 R gamma chains. Mutations in the gene encoding IL-2 R gamma results in X-linked severe combined immunodeficiency, which is characterised by an absence of T and NK cells, and near normal numbers of functionally deficient B cells. For information on IL-2 R gamma and X-linked severe combined immunodeficiency please refer to Ginn SL *et al.,* (2004) *Hum Mutat.* **23**(5): 522-3.

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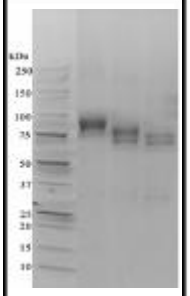
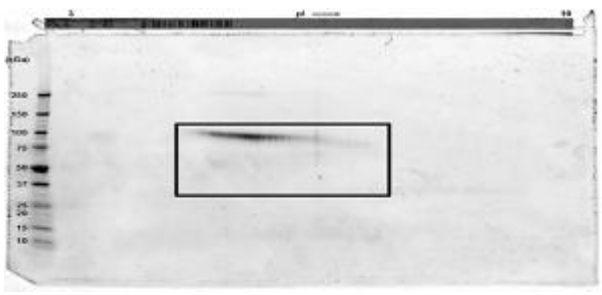
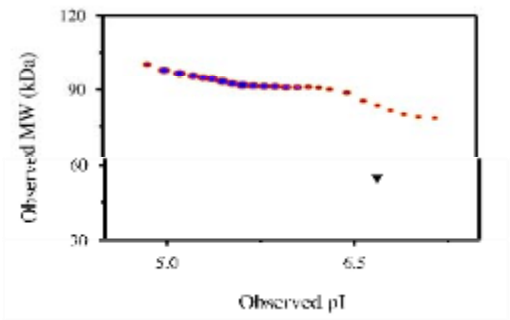
	1D gel  **2D gel** 
1D gel data	Lane 1 – MW markers; Lane 2 – IL-2 R gamma-Fc ^{HCX} Chimera; Lane 3 – IL-2 R gamma-Fc ^{HCX} Chimera treated with PNGase F to remove potential N-linked glycans; Lane 4 – IL-2 R gamma-Fc ^{HCX} Chimera treated with a glycosidase cocktail to remove potential N- and O-linked glycans. 10 µg protein loaded per lane; Deep Purple™ stained. Drop in MW after treatment with PNGase F indicates presence of N-linked glycans. Subsequent drop in MW after treatment with glycosidase cocktail indicates presence of O-linked glycans. Faint bands in lane 3 and lane 4 are glycosidase enzymes.
2D gel data	A sample of IL-2 R gamma-Fc ^{HCX} Chimera without carrier protein was reduced and alkylated and focused on a 3-10 IPG strip then run on a 4-20% Tris-HCl 2D gel. 40 µg protein loaded per lane; Deep Purple™ stained. Spot train indicates presence of multiple isoforms of IL-2 R gamma-Fc ^{HCX} Chimera. Spots within the spot train were cut from the gel and identified as IL-2 R gamma-Fc ^{HCX} Chimera by protein mass fingerprinting. Experimental details and results are available upon request.
Densitometry	Post-translational modifications result in protein heterogeneity. The densitometry scan, derived from the 2D gel image above, 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. 
Theoretical Sequence	LNTTILTPNGNEDTTADFFLTTMPTDSLSVSTLPLPEVQCFVFNVEYMNCTWNSSS EPQPTNLTLHYWYKNSDNDKVQKCSHYLFSEEITSGCQLQKKEIHLYQTFVVQLQD PREPRRQATQMLKLQNLVIPWAPENLTLHKLSESQLELNWNNRFLNHCLEHLVQY RTDWDHSWTEQSVDYRHKFSLPSVDGQKRYTFRVRSRFNPLCGSAQHWSEWSH PIHWGSNTSKENPFLFAWIPKVDKKVEPKSCDKTHTCPPCPAPELLGGPSVFLFPP KPKDTLMISRTPEVTCVVVDVSHEDPEVKFNWYVDGVEVHNAKTKPREEQYNSTY RVVSVLTVLHQDWLNGKEYKCKVSNKALPAPIEKTISKAKGQPREPQVYTLPPSRD ELTKNQVSLTCLVKGFYPSDIAVEWESNGQPENNYKTTPPVLDSDGSFFLYSKLTV DKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPGK