

Human Cell Expressed TNF RII-Fc^{Hcx} Chimera Catalog Number 9011_

Source	A DNA sequence encoding the signal peptide and extracellular domain of human TNF Receptor II (TNFRII) (aa 1-253) 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 TNF RII-Fc ^{HCX} Chimera migrates as a band between 55 and 80 kDa in SDS- PAGE due to post-translation modifications, in particular glycosylation. This compares with the unmodified TNF RII-Fc Chimera that has a predicted molecular mass of 51.8 kDa.
pl	Symansis TNF RII-Fc ^{HCX} Chimera separates into a number of isoforms with a pl between 4.5 and 7.0 in 2D PAGE due to post-translational modifications, in particular glycosylation. This compares with the unmodified TNF RII-Fc Chimera that has a predicted pl of 7.9.
% Carbohydrate Symansis purified TNF RII-FcHCX Chimera consists of 5-35% carbohydrate by weight.	
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 ND50 of TNF RII-Fc ^{HCX} Chimera is typically 20-50 ng/ml as measured by its ability to neutralize TNF-a mediated cytoxicity in murine WEHI 164 cells in the presence of actinomycin D.
Background Information	Tumor necrosis factor alpha (TNF-alpha ; TNF alpha) and the structurally related lymphotoxin-alpha (LT-a) are pro-inflammatory cytokines produced by a variety of immune and non-immune cells in response to multiple stimuli. These stimuli can include bacteria and bacterial products such as LPS, viral infections, T-cell receptor activation and cross-linking of surface immunoglobulin (Ig) on B cells. Expression of TNF-a has been detected in macrophages/monocytes, B-cells, T-cells, basophils, eosinophils, glial cells and mast cells.
	The effects of TNF-alpha and LT-a are mediated through the TNF receptors, TNF RI and TNF RII.
	These receptors are type I transmembrane glycoproteins belonging to the TNF receptor superfamily, which is defined by the presence of repeating units of cysteine clusters. TNF RI is present on virtually all cell types except for red blood cells.
	For a review of the mechanisms that regulate TNF RII expression refer to Carpentier I, et al (2004) <i>Curr Med Chem.</i> 11 (16): 2205-12.



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



LPAQVAFTPYAPEPGSTCRLREYYDQTAQMCCSKCSPGQHAKVFCTKTSDTVCDSCED STYTQLWNWVPECLSCGSRCSSDQVETQACTREQNRICTCRPGWYCALSKQEGCRLC APLRKCRPGFGVARPGTETSDVVCKPCAPGTFSNTTSSTDICRPHQICNVVAIPGNASMD AVCTSTSPTRSMAPGAVHLPQPVSTRSQHTQPTPEPSTAPSTSFLLPMGPSPPAEGIPK VDKKVEPKSCDKTHTCPPCPAPELLGGPSVFLFPPKPKDTLMISRTPEVTCVVVDVSHED PEVKFNWYVDGVEVHNAKTKPREEQYNSTYRVVSVLTVLHQDWLNGKEYKCKVSNKAL PAPIEKTISKAKGQPREPQVYTLPPSRDELTKNQVSLTCLVKGFYPSDIAVEWESNGQPE NNYKTTPPVLDSDGSFFLYSKLTVDKSRWQQGNVFSCSVMHEALHNHYTQKSLSLSPG K



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