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Tumor Necrosis Factor and Tumor Necrosis Factor Receptors in Coelacanth genes

Kierra Brown

Leyete Winfield

Alan Christoffels

Barbara Picone
School of Chemistry, Spelman College, Atlanta, GA

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Kierra Brown, Dr. Leyete Winfield, Prof. Alan Christoffels, Dr. Barbara Picone

South African National Bioinformatics Institute.

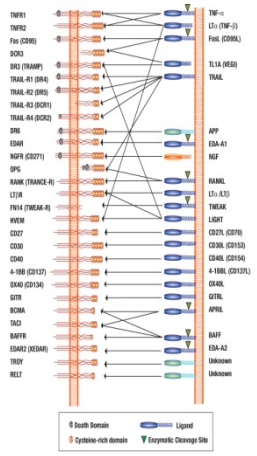
School of Chemistry, Spelman College, Atlanta, GA

Abstract

Protein superfamilies incorporate any of a group of proteins having similar structure and functionality which descend from the same ancestral gene. The Tumor Necrosis Factor, superfamily composed of Tumor Necrosis Factor, TNF, and Tumor Necrosis Factor Receptors, TNFR, were investigated by utilizing bioinformatics software. In this study, protein families were characterized by analyzing the similarities and differences between groups in the TNF and superfamily. Using bioinformatics techniques to analyze protein structures of the superfamily found in this study were Coelacanth, Fugu, and Homo sapiens. A computer database, ENSEMBL, was used to collect data for the Coelacanth, Fugu, and Human TNF and TNFR proteins. ENSEMBL browser provides a variety of genomes with complete explanations using an automated genome annotation system. After determining whether the protein was the Tumor Necrosis Factor or Tumor Necrosis Factor receptor subfamily different groups were analyzed to determine similarities and inconsistencies in each group. Several genes were hypothesized to be artifacts or pseudo genes after analyzing the collected data. Computational tools or gene prediction tools mistake similar proteins that are not functionally related so they require manual manipulation to verify actual genes and not artifacts. Additional studies are needed to further analysis to verify all member of the TNF superfamily.

Introduction

Tumor necrosis factor is a proinflammatory cytokine, or protein that is produced by many immune and nonimmune cells types such as macrophages, t cells, mast cells, granulocytes, natural killer (NK) cells, fibroblasts, neurons, keratinocytes, and smooth muscle cells (Partsch, G., et al., 1997). Their main purpose is to prevent or inhibit the growth of cancer cells. The first tumor necrosis factor superfamily members to be identified were tumor necrosis factor-α and lymphotoxin-α, the superfamily in humans consist of 19 ligands or binding molecules. The tumor necrosis factor receptor superfamily has 29 receptors in humans with three additional receptors being found in mice. TNF ligands and receptors are still being studied in the Coelacanth. The tumor necrosis factor superfamily has 472 amino acids and a conserved C-terminal in the TNF homology domain (Wang et al., 2012). Fig. 1 shows the different TNF ligand and TNFR.



TNF superfamily ligands and receptors are important for normal developmental processes. The TNF ligand family structure consists of a stalk connecting the transmembrane domain to the core region where the THD, TNF homology domain, is located ("Research topics TNF," 2012) . The THD is an anti-parallel beta-pleated sheet. Most TNF superfamily (TNFSF) ligands are type II transmembrane proteins. TACE, TNF-alpha-converting enzyme cleaves TNFSF at extracellular or outer domains (Tracey, D et al, 2008). The cleaved form of TNF ligand exists as soluble tumor necrosis factor. At small concentrations in tissue TNF is beneficial. TNF assists in lymphoid tissue development and homeostatic host defense against bacterial infections or injury. The main problem with tumor necrosis factor is that it causes inflammation and in some cases organ failure if the concentration of the protein is too high. TNF is at the head of the inflammatory cascade network mediating a variety of pathogenic effects which.

The five different mechanisms of action that TNF and TNFR interact in are reverse signaling, apoptosis, immune regulation, inflammation, and bone and cartilage. Reverse signaling is initiated by tumor necrosis factor antagonist binding and is important to apoptosis, cytokine suppression, and cellular events. Apoptosis is programmed cell death in the cell. In regard to immune regulation tumor necrosis factor antagonist role in the immune system is to maintain homeostasis. In inflammation tumor necrosis factor is the top of the proinflammatory cytokine network and plays a central role while tumor necrosis factor antagonists reduce the cellularity of inflamed tissues. In bone and cartilage tumor necrosis factor causes inflammation in the joints that can then lead to joint damage and loss of bone in which tumor necrosis factor antagonist reduces or stops bone destruction.

Methods

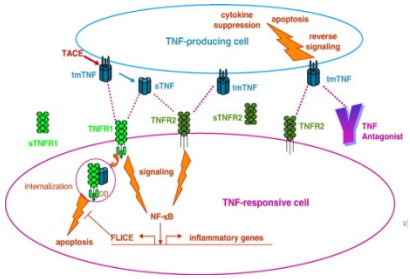


Fig. Biology of TNF production (Tracey, D et al, 2008) Firstly the cell produces tmTNF which is cleaved to become sTNF by the TACE, TNF-alpha-converting-enzyme. Both sTNF and tmTNF can bind to two different types of receptors TNFR1 or TNFR2 on a TNF- responsive cell. The TNF-responsive cell sends a signaling pathway that can lead to apoptosis, NF-κB activation, or inflammatory gene activation. Through internalization of the ligand-receptor complex, addition of the death domains and blockage of the FLICE enzyme apoptosis is induced. Cytokine suppression or apoptosis of the producing cell can be cause by reverse signaling from TNFR2 or TNF antagonist. sTNFR1 and sTNFR2 receptors are released from TNF-responsive cell after enzyme cleavage.

TNF and TNFR were analyzed using the ENSEMBL browser. The ENSEMBL browser uses genomics, proteomics, and transcriptomics to analyze genes of different organisms in a biological database. Genomics is the branch of molecular biology dealing with the structure, function, evolution, and mapping of genomes. Proteomics is the branch of genetics that studies the full set of proteins encoded by a genome. Transcriptomics is the set of all RNA molecules, including mRNA, rRNA, tRNA, and non-coding RNA. Biological databases are libraries of life sciences information, collected from scientific experiments and published literature. ENSEMBL uses genomic Annotation to characterize different genes. The main task of biological databases like ENSEMBL is data storage, retrieval, and analysis.

ENSEMBL was first created to annotation the human genome now it has genomic data from a variety of vertebrates, bacteria, plants etc. ENSEMBL browser provides a variety of genomes with complete explanations using an automated genome annotation system. GeneTreeView allows the user to see the genetic family or phylogenetic relationship. This eliminates sequences similarity analysis. DAS (distributed annotation system) helps users upload data to compare to the data in ENSEMBL genome browser. All TNF and TNFR protein molecules were analyzed from Coelacanth, Human, and Fugu genes. In the study we will be analyzing what these protein families look like. Exon numbers are predicted by analyzing the messenger RNA on the protein gene.

Results

OG	Gene name	Organism	Summary OG	# of Exons	Protein Domain	Splice Variant	Protein length	Genome Contig Scaffold	Start position	Stop position	Direction
					ENSLACP00000018056						
OG6134	ENSLACG00000018375	latimeria_chalumnae	TNFR	15	6.1	1	284	JH426768.1	1953,597	1,954,936	Reverse
					ENSLACP00000018967						
OG6134	ENSLACG00000018403	latimeria_chalumnae	TNFR	4	4.1	1	163	JH426488.1	418,020	440,875	Forward
OG6134	ENSTFUG00000004668	takifugu_rubripes	TNFR								
OG6134	ENSG00000067582	homo_sapiens	TNFR								
OG6134	ENSG00000218768	homo_sapiens	TNFR								
					ENSLACP00000018742						
OG6441	ENSLACG00000018347	latimeria_chalumnae	TNF	4	2.1	1	281	JH427097.1	1,235,710	1,242,801	Forward
OG6441	ENSTFUG00000018475	takifugu_rubripes	TNF								
OG6441	ENSG00000107561	homo_sapiens	TNF								
					ENSLACP00000018936						
OG6823	ENSLACG00000018638	latimeria_chalumnae	TNFR	6	6.1	1	285	JH426563.1	1,662,479	1,671,368	Reverse
					ENSLACP00000018253						
OG6823	ENSLACG00000018927	latimeria_chalumnae	TNFR	7	7.1	1	274	JH426562.1	5,327,799	5,346,975	Forward
					ENSLACP00000018253						
OG6823	ENSLACG00000018919	latimeria_chalumnae	TNFR	6	6.1	1	286	JH426562.1	5,368,569	5,391,121	Forward
					ENSLACP00000018253						
OG6823	ENSLACG00000018925	latimeria_chalumnae	TNFR	8	8.1	1	230	JH426562.1	5,398,286	5,412,404	Forward
OG6823	ENSG00000057873	homo_sapiens	TNFR								
					ENSLACP00000018035						
OG7536	ENSLACG00000018169	latimeria_chalumnae	TNFR	7	55.1	1	555	JH426568.1	72,089	114,306	Reverse
OG7536	ENSTFUG00000018306	takifugu_rubripes	TNFR								
OG7536	ENSG00000177804	homo_sapiens	TNFR								
					ENSLACP00000018237						
OG6747	ENSLACG00000018093	latimeria_chalumnae	TNF	6	2.1	1	223	JH426522.1	544,269	553,571	Reverse
OG6747	ENSG00000105810	homo_sapiens	TNF								
					ENSLACP00000000418						
OG6329	ENSLACG00000003889	latimeria_chalumnae	TNFR	7	7.1	2	161	JH427355.1	52,570	124,809	Reverse
					ENSLACP00000000418						

The data that was collected from the *Latimeria chalumnae, Takifugu rubripes, and Homo sapiens* genes to identity whether the portions in the genes were TNF or TNFR were analyzed. After determining whether the protein was the TNF or TNF receptor subfamily different groups were analyze to determine similarities and inconsistencies in each group. The table below was constructed and displays the different TNF and TNFR groups. The highlighted area signifies a TNF or TNFR that is not consistent with the data collect for the other members of that group.

Using the computational device can cause artifact or undesired alteration in data, introduced by a technique and/or technology to occur. The table illustrates genes that are hypothesized to be artifacts or pseudo genes. These genes are characterized for not having intron coding resulting in a nonfunctional gene. Computational tools or gene prediction tools mistake similar proteins that are not functionally related so they require manual manipulation to verify that it is a gene and not an artifact. This verification will be conducted at SANBI, so that no artifacts will be in the final phylogenetic tree.

Conclusion

The data collected will be complied with other genomic information from other organisms that have TNF in their genomic composition. Further analysis will be conducted by the South African National Bioinformatics Institute to determine TNF superfamily genes main function in all of these organisms to create a TNF superfamily phylogenetic tree. A phylogenetic tree or evolutionary tree is a branching diagram showing the hypothesized evolutionary relationships between various biological species based upon similarities and differences in their genetic characteristics.

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