Alternative splicing regulation in tumor necrosis factor-mediated inflammation (Review)

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Abstract. It is generally accepted that alternative splicing has an effect on disease when it leads to conspicuous changes in relevant proteins, but that the combinatorial effect of several small modifications can have marked outcomes as well. Inflammation is a complex process involving numerous signaling pathways, among which the tumor necrosis factor (TNF) pathway is one of the most studied. Signaling pathways are commonly represented as intricate cascades of molecular interactions that eventually lead to the activation of one or several genes. Alternative splicing is a common means of controlling protein expression in time and space; therefore, it can modulate the outcome of signaling pathways through small changes in their elements. Notably, the overall process is tightly regulated, which is easily overlooked when analyzing the pathway as a whole. The present review summarizes recent studies of the alternative splicing of key players of the TNF pathway leading to inflammation, and hypothesizes on the cumulative results of those modifications and the impact on cancer development.

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1. Introduction

The majority of eukaryotic genes undergo alternative splicing; once believed to be a peculiarity of a few genes, it is now closer to being the rule rather than the exception. Alternative splicing is a proteome-diversifying process through which several mature RNA messengers are obtained from a single gene, each of these transcripts potentially coding for a different protein isoform with a potentially different function (1).

The specialized molecular machinery that performs splicing is known as the spliceosome, a macromolecular complex composed of four small nuclear ribonucleoproteins (snRNPs) (U1, U2, U4/U6 and U5) and >100 non-snRNP splicing factors. The spliceosome identifies individual splice sites along immature transcripts through the recognition of consensus sequences located in the exon/intron boundaries, which are complementary with the RNA present in the snRNPs. Next, through a series of spatial rearrangements, it facilitates two trans-esterification reactions that result in the excision of the intronic sequence; the process takes place at each intron to yield a mature mRNA (1).

On average, human genes have 8.8 introns (2) and ~90% of them produce more than one mature transcript (3). Alternative splicing can remove introns from a pre-mRNA in a number of different combinations, giving rise to different mRNAs. Known patterns of alternative splicing include exon skipping (removal of an exon along with the surrounding introns), usage of alternative intron donor (5’) or acceptor (3’) sites, intron retention and mutually exclusive exon splicing (Fig. 1). Alternative exons may possess splice sites with diverging sequences, thus are less likely to be recognized through RNA-RNA interaction; such sites are considered sub-optimal and their recognition is aided by the recruitment of trans-acting splicing factors that bind to cis regulatory elements in the vicinity of splice sites and recruit the main spliceosome components. These cis elements are classified in exonic/intronic splicing enhancers/silencers (ESE, ESS, ISE and ISS) according to their positions and functions, while the factors that bind to them mainly belong to two families, namely the SR and heterogeneous (hn)RNP proteins, which usually promote and prevent splice site recognition, respectively (Fig. 2). This way, the alternative splicing of a given precursor (pre)-mRNA is regulated by the abundance...
of the pre-mRNA itself, its sequence (presence/absence of splicing factor recognition sites) and the relative abundance of the splicing factors (4-6). So, it is now understood that the main components of the spliceosome remain largely unchanged and that the majority of the regulatory nature of alternative splicing relies on protein-protein, protein-nucleic acid interactions and their kinetics (7). Furthermore, RNA splicing is coupled to transcription physically and dynamically. Spliceosome factors are recruited to the carboxy-terminal domain of the RNA polymerase during RNA elongation, from where they are in turn recruited to the splice sites along the nascent transcript as they become available, thus making splicing dependent on the processivity of the RNA polymerase elongation complex (8).

Alternative splicing has been observed to have an effect on disease. There are a considerable number of hereditary diseases caused by point mutations, which in general, disrupt splice sites or ESE/ISEs and modify the aforementioned interactions preventing the expression of one particular isoform of a protein (9,10). The role that alternative splicing plays in the generation and/or maintenance of complex pathological conditions, such as inflammation, is not as straightforward, since these conditions are the result of several alterations in pathways that often comprise of dozens of proteins with diverse functions, each of which is prone to regulation through alternative splicing. High-throughput sequencing and bioinformatics have made it possible to assess alternative splicing modifications on a global scale (3,11); however, these methods are not yet without limitations and require complementing with particular, functional studies. The present review summarizes the impact of alternative splicing in the core components of the TNF signaling pathway.

2. Tumor necrosis factor (TNF) pathway

TNF-a (also known as cachectin) is a strong pro-inflammatory cytokine, which plays an important role in certain processes, including inflammation, cell proliferation, differentiation and apoptosis. Inflammation is an extremely important part of innate immunity and is regulated in a number of steps (12).

TNF-a binds two distinct receptors: TNFR1 and TNFR2. Activation of TNFR1 leads to the formation of signal complexes that activate pathways that lead to: i) Expression of pro-inflammatory genes through the recruitment of receptor-interacting protein 1 (RIP1) and TNF-receptor-associated factor 2 (TRAF2); and ii) apoptosis and cell death by recruiting Fas-associated death domain protein (FADD) and caspase 8. Binding to TNFR2 induces only inflammation through the direct recruitment of TRAF2, which in turn recruits TRAF1. The two pro-inflammatory complexes lead to IKK (inhibitor of nuclear factor κ-B kinase and mitogen-activated protein kinase (JNK and p38) activation (Fig. 3) (12).

The balance between TNF-activated inflammation and apoptosis is regulated on several levels, including signal strength, expression of signaling molecules and regulating proteins, and crosstalk with other cell signals (13), all of which can be heavily influenced by the alternative splicing of each element.

3. Known isoforms in TNF signaling

A search on the Ensmbl database (14) for the splice variants originated from the transcripts of the receptor-proximal elements of TNF signaling shows that all but FADD have at least 2 potential protein variants (Table I). Experimental evidence is not available for all of them; and thus far, it comprises mostly association data, rather than molecular evidence that offers an understanding of their regulation. In the following section, experimental evidence concerning alternative splicing of the key players in TNF signaling is reviewed.

TNFR. Two recent studies suggested that the TNF receptor superfamily member 1A (TNFRSF1A) transcript balance may depend on TNFRSF1A alleles. Gregory et al (15) investigated a single nucleotide polymorphism (rs1800693, c.625+10A>G) in the TNFRSF1A gene that was previously
identified as a susceptibility marker for multiple sclerosis through genome-wide association studies. In in vitro splicing assays, only the G allele resulted in skipping of exon 6. TNFR1 exon 6 skipping results in a frameshift and a premature stop codon, which translates into a soluble form of TNFR [D6-TNFR, comprising only the amino-terminal domain of TNFR1, followed by a novel 45-amino acid (aa) sequence] for which a TNFR-antagonistic role was suggested. Another detailed study described a TNFR1 transcript lacking exon 2 (TNFR1-d2) and its association with a specific haplotype at 3 single nucleotide polymorphisms (SNPs) previously associated with TNF-receptor-associated periodic syndrome (TRAPS). These SNPs have distant locations along the TNFR1A gene: rs4149570 lies at the promoter region (c.610G>T), rs767455 at exon 1 (c.36A>G), and rs1800692 at exon 4 (c.473-33C>T), so it is plausible that an interplay of transcription and splicing dynamics determines transcript outcome (16). This evidence strongly suggests that these SNPs disrupt splicing enhancers/silencers along intronic or exonic sequences, an aspect yet to be studied for other reported SNPs associated with TRAPS (17) or other inflammatory conditions, including Crohn's disease (18).

TNFR2 isoforms have been described as well. Seitz et al (19) characterized hicpTNFR, a TNFR2 splice variant with an alternate exon 1 sequence that results from the usage of an alternate transcription start site and alternative splicing. This variant is mostly retained in the trans-Golgi network and in endosomes where it could function as a storage pool of preformed p75TNFR that is not affected by shedding. Upon emerging on the cell surface, hicp75TNFR is functionally no longer distinguishable from p75TNFR; furthermore, hicpTNFR colocalizes with endogenous TNF, hinting at intracellular activation of the hicp75TNFR by endogenous TNF (20). There are a number of studies on a soluble TNFR2 isoform lacking exons 7 and 8, known as differentially spliced (DS)-TNFR; as a consequence of splicing, it lacks the transmembrane and cytoplasmic domains. The data gathered suggests that it regulates TNFR function by antagonizing its biological activity. This soluble receptor was detected at increased levels in patients with sepsis and at higher concentrations in patients with rheumatoid arthritis, relative to the levels detected in the sera of healthy individuals (21). In another study, performed in insulin-resistant patients, this same isoform was found in 26% of samples from patients with type 2 diabetes and in 44% of non-diabetic subjects. An increase in waist size was associated with a progressive decrease in DS-TNFR2 concentration. Moreover, it was suggested that DS-TNFR2 exhibits anti-inflammatory activity, based on observations of the correlation between DS-TNFR2 and circulating adiponectin (22). This novel isoform and its potential anti-inflammatory properties have been associated with markers of liver injury (23) and with a favorable outcome in patients with rheumatoid arthritis (24), although no information is currently available on the dynamics of this splicing event.

Figure 2. Splicing cis elements influence splice site recognition through recruitment of different spliceosome components. Generally, splicing enhancers recruit SR proteins, which in turn recruit the splicing machinery; splicing suppressors recruit hnRNP proteins, which hinder these interactions. ESE, exonic splicing enhancer; ISE, intronic splicing enhancer; ISS, intronic splicing suppressor; ESS, exonic splicing suppressor; hnRNP, heterogeneous ribonucleoprotein; pre-mRNA, precursor-mRNA; snRNP, small nuclear RNP.

Figure 3. Proximal components of the TNF signaling pathway. TNFR1 transduces apoptotic signals through the recruitment of FADD and caspase 8, and inflammatory signals through the recruitment of RIP1 and TRAF2. TNFR2 binds TRAF1 and TRAF2 in order to transduce inflammation signals. IKK is an enzyme involved in the degradation of the inhibitor IKB, which binds NF-κB to inhibit its function. NF, nuclear factor; TNF, tumor necrosis factor; TNFR1, TNF receptor type 1; FADD, Fas-associated death domain protein; RIP1, receptor-interacting protein 1; TRAF1, TNF-receptor-associated factor 1; IKK, inhibitor of NF-κB kinase.
Alternative splicing has been described in the RIP family only in the RIP3 gene; two splice variants (RIP3-β and RIP3-γ) with truncated N-terminal kinase domains and novel, shorter C termini have been found. These variants abrogate nucleocytoplasmic shuttling and are therefore not able to induce apoptosis. Additionally, they downregulate RIP3 pro-apoptotic activity (25). Suppression of RIP3-dependent apoptotic TNF signaling could potentially upregulate RIP1-dependent pro-inflammatory pathways.

Splice variants of the human RIP1 gene should not be ruled out, since it has been demonstrated that the associated Xenopus RIP1 gene produces at least one alternative splicing-derived isoform (26). According to the Ensembl database annotations, human RIP1 10 exon-pre-mRNA potentially produces 5 alternative transcripts, which are yet to be experimentally described.

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**TRAF.** A splice variant of TRAF2 was described as early as 1998, this variant (TRAF2A) contains a 7-aa insertion within the RING finger domain, presumably produced by alternative usage of splice donor sites present at the 3' end of exon 1. TRAF2A is incapable of mediating the activation of nuclear factor (NF)-κB, thus it can act as a dominant inhibitor of TNFR2-mediated NF-κB activation; this way, cells can regulate NF-κB activation through TNF family receptors by modifying alternative splicing of primary transcripts from the TRAF2/TRAF2A gene to produce different ratios of TRAF2A and TRAF2 mRNAs (27). In a comparison of the structure of the human, murine and Drosophila TRAF genes, the TRAF2A transcript was only found to be expressed in mice, and not in humans or rats, although only Ramos (human B lymphoma) and HEK293 (human embryonic kidney) cells were assayed (28).

TRAF3 is another member of the TRAF family of proteins, (currently six genes have been identified, TRAF1 to 6). Three isoforms produced though alternative splicing of this gene were initially identified; they differ in the number of Zn fingers remaining from the five contained in the full-length TRAF3. The TRAF3b isoform (Δ25 aa) contains four Zn fingers with a C-terminal finger formed by the fusion of the N-terminal half of the 3rd and the C-terminal half of the 4th finger, the TRAF3c isoform (Δ52 aa) contains three Zn fingers with a C-terminal finger formed by the fusion of the 2nd and the 4th finger, while the TRAF3d isoform (Δ56 aa) contains three complete Zn fingers and the N-terminal portion of the 5th finger. Additionally, the study detected three variant
5′-untranslated regions (UTRs) and two variant 3′ UTRs among the isolated clones, although they did not establish whether they corresponded to particular isoform-coding open reading frames (29).

A further study identified five additional TRAF-3 protein isoforms with alterations in the Zn finger domains (Δ27aa, Δ83aa, Δ103aa, Δ130aa and Δ221aa). TRAF3 splice-deletion variants, including Δ25 aa, Δ52 aa and Δ56 aa, were found to induce NF-κB activation in 293T cells, while full-length TRAF3 and TRAF3 Δ221 failed to do so. Unexpectedly, when full-length TRAF3 was co-expressed with each of the 7 TRAF3 splice variants capable of activating NF-κB alone, a 1.4- to 5-fold augmentation of their NF-κB activation was observed (30). All but the Δ130 aa were found to be expressed in four different lymphoma cell lines (Jurkat D1.1, BJAB, Daudi and Raji). Overexpression experiments of individual isoforms revealed that only the Δ27 aa, Δ103 aa or Δ130 aa isoforms are able to induce NF-κB activation in BJAB cells in contrast to full-length TRAF-3 or Δ221aa, which could not; notably, TRAF-3 Δ25 aa, Δ52 aa, Δ56 aa and Δ83 aa variants also failed to induce NF-κB activity, contrary to their activity in 293T cells (31). The difference in the TRAF-3 splice variants activation properties in different cell lines suggests an association between TRAF-3 isoforms and the cellular environment. The two examples provide evidence of the regulation of NF-κB activation in TRAF family genes through alternative splicing: The TRAF2 and TRAF3 shorter splice variants have this ability, while the full-length proteins lack them.

4. Splicing potential

The demonstration by Rittore et al (16) that SNPs can have an effect on the alternative splicing pattern of the TNFRI1 gene may be the cornerstone that joins two groups of association studies, namely, transcript diversity and SNPs, and sheds light on their association. The Rittore group identified that SNPs rs4149570, rs767455 and rs1800692 have a combined effect on the TNFRI1 transcript output through regulation of alternative splicing. When considering that the aforementioned TNFRI1 SNPs have already been found to be associated with inflammation-related conditions, including susceptibility to develop invasive pulmonary aspergillosis (32), the prognosis of peripheral T-cell non-Hodgkin lymphoma (33), radiation-induced toxicity following treatment for non-small cell lung cancer (34), the risk of breast cancer (35), inflammatory demyelinating diseases (36), the response to Crohn's disease treatment (37) and adult onset Still's disease (38), it is plausible to consider whether it is the differential balance of TNFRI1 isoforms produced by alternative splicing that causes these associations.

There is conclusive evidence that supports the fact that SNPs can modify the outcome of alternative splicing by disrupting canonical splice sites (39), or by modifying cis elements (enhancers or silencers) along both exonic and intronic sequences, as Pagani et al (40) found in a number of synonymous mutations that cause exon 12 skipping of the cystic fibrosis transmembrane conductance regulator. Curated repositories that gather SNP association data, such as SNPedia (41), pinpoint relevant SNPs worth analyzing for their potential effect on alternative splicing. A search in SNPedia reveals several disease-associated polymorphisms in virtually all of the receptor-proximal elements of TNF signaling (Table II).

Regarding human cancer, evidence points out that specific functions of certain genes regulating the TNF signaling pathway can be affected by alternative splicing, with an impact on tumor phenotype. In this sense, the TNF-apoptosis inhibitor c-FLIP (cellular FLICE inhibitory protein) has distinct alternative splicing variants (c-FLIPs and c-FLIPL) that have distinct roles in the TNFα-induced signaling cascade. Park et al (42) demonstrated that c-FLIP alternative variants activate either Erk or NF-κB through the association with Raf and TRAF2, which contributes to TNF-induced cell cycle promotion. For instance, c-FLIPL showed stronger affinity to Raf in order to activate Erk and PI3K. Meanwhile, c-FLIP, showed strong affinity to TRAF2 to mediate activation of the TRAF-JNK pathway. Moreover, each splicing variant is regulated differentially at transcriptional level, that is, after TNF-α stimulation, a delayed c-FLIPL response is induced, whereas c-FLIP shows a rapid response to the stimulation (42). Thus, alternative splicing enables protein diversity, generating structurally and functionally distinct proteins from the same gene. Later, Haag et al (43) reported the expression of these splicing variants in pancreatic cancer tissue. It was observed that c-FLIP is underexpressed in pancreatic intraepithelial neoplasm and pancreatic ductal adenocarcinomas compared with normal pancreatic tissues. Moreover, in pancreatic cancer cell line ULA-PaC, the downregulation of these isoforms by RNA interference enhances apoptosis, indicating that c-FLIP is an important regulator of death receptor-induced apoptosis (43).

In another study, in neoplastic and more notably, in non-neoplastic cells, two novel TNF-associated apoptosis-inducing ligand (TRAIL) splice variants were reported: TRAIL-β, lacking exon 3, corresponding to loss of 98 aa, and TRAIL-γ, lacking exons 2 and 3, corresponding to loss of 52 aa. These two variants result in a truncation of the extra-cellular domain, which is important for trimeric stability and ligand-receptor binding; consequently, these splice variants fail to trigger apoptosis signaling. The study suggested that these novel TRAIL variants may have implications for deepening our understanding of TRAIL-mediated apoptosis in neoplastic and non-neoplastic human cells (44). Recently, Krieg et al (45) reported the first study that describes the expression of TRAIL splice variants in 41 gastric carcinoma tissue samples by reverse transcription-quantitative polymerase chain reaction. Notably, all three TRAIL-splice variants could be detected in non-malignant and malignant tissues, but only TRAIL-γ had a prognostic value, since it was associated with a significantly higher survival rate (45).

5. Concluding remarks

TNF-mediated inflammation, as with numerous other signaling pathways, involves the concerted expression of a fairly well known number of genes; however, the human transcriptome has turned out to be far more complex than initially conceived, and the contribution of alternative splicing to the transcriptomic variability through the diversification of mature transcripts produced from the same gene, is becoming clearer and more
important (3). A considerable amount of research is currently being conducted into the search for transcriptomic signatures associated with inflammation-related diseases (among numerous other conditions), including osteoarthritis (46), atherosclerotic plaque progression (47), multiple sclerosis, systemic lupus erythematosus, juvenile rheumatoid arthritis, Crohn’s disease, ulcerative colitis and type 1 diabetes (48), to name a few. Incorporation of alternative splicing data into this information can shed light on how these transcriptomic signatures come to be, and on the possible scenarios that they lead to (49).

The present study has reviewed the literature showing that protein isoforms derived from alternative transcripts of the genes involved in TNF signaling can have different, mainly antagonistic, functions. We therefore find it reasonable to hypothesize that, although TNF signaling ultimately leads to regulation through alternative splicing of its elements, this process can have different, mainly antagonistic, functions. We therefore find it reasonable to hypothesize that, although TNF signaling ultimately leads to inflammatory response, the pathway itself can be subject to regulation through alternative splicing of its elements, potentially modifying its end result.

References

1. Sharp PA: Split genes and RNA splicing. Cell 77: 805-815, 1994.
2. Lande R, Linton LM, Birren B, Nusbaum C, Zody MC, Baldwin J, Devon K, Dewar K, Doyle M, FitzHugh W, et al. Initial sequencing and analysis of the human genome. Nature 409: 860-921, 2001.
3. Pan Q, Shao O, Lee LJ, Frey BJ and Blencowe BJ: Deep surveying of alternative splicing complexity in the human transcriptome by high-throughput sequencing. Nat Genet 40: 1413-1415, 2008.
4. Black DL: Mechanisms of alternative pre-messenger RNA splicing. Annu Rev Biochem 72: 291-336, 2003.
5. Stamm S, Ben-Ari S, Rafalska I, Thanaraj TA and Soreq H: Function of alternative splicing. Gene 344: 1-20, 2005.
6. Smith CW and Valcárcel J: Alternative pre-mRNA splicing: The logic of combinatorial control. Trends Biochem Sci 25: 381-388, 2000.
7. Matlin AJ, Clark F and Smith CW: Understanding alternative splicing: Towards a cellular code. Nat Rev Mol Cell Biol 6: 4129-4134, 1998.
8. Komiyama M, Takahashi T, Sasaki T, Tanaka M, Uemura T, et al. Identification and characterization of a novel spliced variant that encodes human soluble tumor necrosis factor alpha receptor type 2. J Immunol 162: 2765-2771, 1999.
9.张 P, Bota M, Casals L, Monroy N, Ruberte E and Botas J: The tumor necrosis factor receptor-associated periodic syndrome (TRAPS): Emerging concepts of an autoinflammatory disorder. Medicine (Baltimore) 81: 349-368, 2002.
10. Wassef KM, Villani AC, Vermeire S, Defresne L, Chen T, Bitton A, Cohen A, Thomson AB and Wild GE: Tumor necrosis factor receptor gene polymorphisms in crohn’s disease: Association with clinical phenotypes. Am J Gastroenterol 100: 1126-1133, 2005.
11. Seitz C, Muller P, Krieg RC, Mannel DN and Hehlgans T: A novel p75TNF receptor isoform mediating NF kappa B activation. J Biol Chem 276: 19390-19395, 2001.
12. Scherübl C, Schneider-Bracht W, Schütte S, Hehlgans T and Mannel DN: Colocalization of endogenous TNF with a functional intracellular splice form of human TNF receptor type 2. J Immunol 178: 7-15, 2002.
13. Lainez B, Fernandez-Real JM, Romero X, Esplugues E, Cao John JD, Ricart W and Engels P: Identification and characterization of a novel splice variant that encodes soluble tumor necrosis factor receptor 2. Int Immunol 16: 169-177, 2004.
14. Fernandez-Real JM, Straczkowski M, Lainez B, Chacon MR, Kowalska I, Lopéz-Bermejo A, Garcia-Espana A, Nikolajuk A, Kinalska I and Ricart W: An alternative spliced variant of circulating soluble tumor necrosis factor-alpha receptor-2 is paradoxically associated with insulin action. Eur J Endocrinol 154: 723-730, 2006.
15. Esteve E, Botas P, Delgado E, Lopez-Bermejo A, Lainez B, Engels P, Ricart W and Fernandez-Real JM: Soluble TNF-alpha receptor 2 produced by alternative splicing is paradoxically associated with markers of liver injury. J Immunol 173: 894-904, 2004.
16. Cañete JD, Albaladejo C, Hernández MV, Lainez B, Pinto JA, Ramírez J, López-Armada MJ, Rodríguez-Cros JR, Engels P, Blanco FJ and Sanmartí R: Clinical significance of high levels of soluble tumor necrosis factor alpha-receptor-2 produced by alternative splicing in rheumatoid arthritis: A longitudinal prospective cohort study. Rheumatology (Oxford) 50: 721-728, 2011.
17. Yang Y, Hu W, Feng S, Ma J and Wu M: RIP3 beta and RIP3 gamma, two novel splice variants of receptor-interacting protein 3 (RIP3), downregulate RIP3-induced apoptosis. Biochem Biophys Res Commun 332: 181-187, 2005.
18. Ishizawa YH, Tamura K, Yamaguchi T, Matsumoto K, Komiyama M, Takamatsu N, Shiba T and Ito M: Xenopus death-domain-containing proteins FADD and RIP1 synergistically activate JNK and NF-kappabB. Biol Cell 98: 465-478, 2006.
19. Brink R and Lodish HF: Tumor necrosis factor receptor (TNFR)-associated factor-2A (TRAF2A), a TRAF2 splice variant with an extended RING finger domain that inhibits TNFR2-mediated NF-kappabB activation. J Biol Chem 273: 4129-4134, 1998.
20. Grech A, Quinn R, Srivinvasan D, Badoux X and Brink R: Complete structural characterisation of the mammalian and Drosophila TRAF genes: Implications for TRAF evolution and the role of RING finger splice variants. Mol Immunol 37: 721-734, 2000.
21. Van Eyndhoven WG, Frank D, Kalachichov S, Cleary AM, Hong DI, Cho E, Nazer S, Perzaj AJ, Mackus WJ, Cayanis E, et al: A single gene for human TRAF-3 at chromosome 1q42.3 encodes a variety of mRNA species by alternative polyadenylation, mRNA splicing and transcription initiation. Mol Immunol 35: 1189-1206, 1998.
22. Van Eyndhoven WG, Gamper CJ, Cho E, Mackus WJ and Lederman S: TRAF3 mRNA splice-deletion variants encode isoforms that induce NF-kappabB activation. Mol Immunol 36: 647-658, 1999.
23. Gamper C, Omene CO, Van Eyndhoven WG, Glassman GD and Lederman S: Expression and function of TRAF-3 splice-variant isoforms in human lymphoma cell lines. Hum Immunol 62: 1167-1177, 2001.
24. Sainz J, Salas-Alvadado I, Lopez-Fernández E, Olmedo C, Comino A, García F, Blanco A, Gómez-Lopera S, Oyarte S, Bueno P and Jurado M: TNFR1 mRNA expression level and TNFR1 gene polymorphisms are predictive markers for susceptibility to develop invasive pulmonary aspergillosis. Int J Immunopathol Pharmacol 23: 423-436, 2010.
25. Heemann C, Kreuz M, Stoller I, Schoof N, von Bonin F, Ziepert M, Löfler M, Jung W, Pfreundschuh M, Trümper L and Kühn D: Circulating levels of TNF receptor II are prognostic for patients with peripheral T-cell Non-Hodgkin lymphoma. Clin Cancer Res 18: 3637-3647, 2012.
34. Hildebrandt MA, Komaki R, Liao Z, Gu J, Chang JY, Ye Y, Lu C, Stewart DJ, Minna JD, Roth JA, et al: Genetic variants in inflammation-related genes are associated with radiation-induced toxicity following treatment for non-small cell lung cancer. PLoS One 5: e12402, 2010.

35. Madeleine MM, Johnson L, Malkki M, Resler AJ, Petersdorf EW, McKnight B and Malone KE: Genetic variation in proinflammatory cytokines IL6, IL6R, TNF-region, and TNFRSF1A and risk of breast cancer. Breast Cancer Res Treat 129: 887-899, 2011.

36. Park TI, Kim HI, Kim JH, Bae JS, Cheong HS, Park BL and Shin HD: Associations of CD6, TNFRSF1A and IRR8 polymorphisms with risk of inflammatory demyelinating diseases. Neuropathol Appl Neurobiol 39: 519-530, 2013.

37. Matsukara H, Ikeda S, Yoshimura N, Takazoe M and Muramatsu M: Genetic polymorphisms of tumour necrosis factor receptor superfamily 1A and 1B affect responses to infliximab in Japanese patients with Crohn's disease. Aliment Pharmacol Ther 27: 765-770, 2008.

38. Cosan F, Emrence Z, Erbag G, Azakli H, Yilmazer B, Yazici A, Ekmekci SS, Abaci N, Ustek D and Cefle A: The association of TNFRSF1A gene and MEFV gene mutations with adult onset Still's disease. Rheumatol Int 33: 1675-1680, 2013.

39. Shimada MK, Hayakawa Y, Takeda J, Gojobori T and Imanishi T: A comprehensive survey of human polymorphisms at conserved splice dinucleotides and its evolutionary relationship with alternative splicing. BMC Evol Biol 10: 122, 2010.

40. Pagani F, Raponi M and Baralle FE: Synonymous mutations in CFTR exon 12 affect splicing and are not neutral in evolution. Proc Natl Acad Sci USA 102: 6368-6372, 2005.

41. Carioso M and Lennon G: SNPedia: A wiki supporting personal genome annotation, interpretation and analysis. Nucleic Acids Res 40 (Database Issue): D1308-D1312, 2012.

42. Park SJ, Kim YY, Ju JW, Han BG, Park SI and Park BJ: Alternative splicing variants of c-FLIP transduce the differential signal through the Raf or TRAF2 in TNF-induced cell proliferation. Biochem Biophys Res Commun 289: 1205-1210, 2001.

43. Haag C, Stadel D, Zhou S, Bachem MG, Möller P, Debatin KM and Fulda S: Identification of c-FLIP(L) and c-FLIP(S) as critical regulators of death receptor-induced apoptosis in pancreatic cancer cells. Gut 60: 225-237, 2011.

44. Krieg A, Krieg T, Wenzel M, Schmitt M, Ramp U, Fang B, Gabbert HE, Gerharz CD and Mahotka C: TRAIL-beta and TRAIL-gamma: Two novel splice variants of the human TNF-related apoptosis-inducing ligand (TRAIL) without apoptotic potential. Br J Cancer 88: 918-927, 2003.

45. Krieg A, Mersch S, Wolf N, Stoecklein NH, Verde PE, Am Esch JS II, Heikaus S, Gabbert HE, Knoefel WT and Mahotka C: Expression of TRAIL-splice variants in gastric carcinomas: Identification of TRAIL-γ as a prognostic marker. BMC Cancer 13: 384, 2013.

46. Ritter SY, Subbaiah R, Bebek G, Grish J, Scanzello CR, Krastins B, Sarracino D, Lopez MF, Crow MK, Aigner T, et al: Proteomic analysis of synovial fluid from the osteoarthritic knee: Comparison with transcriptome analysis of joint tissues. Arthritis Rheum 65: 981-992, 2013.

47. Nührenberg T, Langwieser N, Binder H, Kurz T, Stratz C, Kienzle RP, Trenk D, Zohlhöfer-Momm D and Neumann FJ: Transcriptome analysis in patients with progressive coronary artery disease: Identification of differential gene expression in peripheral blood. J Cardiovasc Transl Res 6: 81-93, 2013.

48. Fuller T, Atar S, Ruppin E, Gurevich M and Achiron A: Common and specific signatures of gene expression and protein-protein interactions in autoimmune diseases. Genes Immun 14: 67-82, 2013.

49. Frankish A, Mudge JM, Thomas M and Harrow J: The importance of identifying alternative splicing in vertebrate genome annotation. Database (Oxford) 2012: bas014, 2012.