The Long Pentraxin 3 and Its Role in Autoimmunity

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Objectives: To review the physiological and physiopathological roles of pentraxin 3 (PTX3), focusing on autoimmunity and vascular pathology.

Methods: A systematic literature review using the keywords "pentraxin 3," "innate immunity," "apoptosis," "autoimmunity," and "endothelial dysfunction" from 1990 to 2007 was performed. All relevant articles and pertinent secondary references in English were reviewed.

Results: PTX3 has a large number of multiple functions in different contexts. PTX3 plays an important role in innate immunity, inflammation, vascular integrity, fertility, pregnancy, and also in the central nervous system. In innate immunity, its normal function is to increase the immune response to selected pathogens while also exerting control over potential autoimmune reactions. It maintains a tightly homeostatic equilibrium in the local immune microenvironment by avoiding an exaggerated immune response and controlling peripheral tolerance to self-antigens. In contrast, in some autoimmune diseases, PTX3 appears to be involved in the development of autoimmune phenomena. A possible explanation for these apparent paradoxical functions may be related to the highly polymorphic *PTX3* gene.

Conclusion: PTX3 is physiologically a protective molecule. However, in several autoimmune diseases PTX3 appears to facilitate the development of autoimmunity. The *PTX3* gene could influence the development of autoimmune reactions and vascular involvement in human pathology.

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Keywords: *pentraxin 3, innate immunity, apoptosis, autoimmunity, endothelial dysfunction, genetics*

Pentraxins are a family of evolutionarily conserved, soluble, multifunctional, pattern recognition proteins characterized by a cyclic multimeric structure (1). Pentraxins are divided into 2 groups: short pentraxins and long pentraxins. The 2 short pentraxins are C-reactive protein (CRP) and serum amyloid P-component (SAP) (2). The prototype protein in the long pentraxin group is pentraxin 3 (PTX3). This molecule shares similarities with the classical, short pentraxins but differs from them by the presence of an unrelated long N-terminal domain as well as in the fields of gene organization, cellular source, and ligands that it recognizes (3). PTX3 production is induced by lipopolysaccharide (LPS), interleukin-1 (IL-1), and tumor necrosis factor-alpha (TNF-α) but not by interleukin-6 (IL-6) (4). PTX3 has different functions depending on the context. PTX3 plays a nonredundant role in resistance to selected pathogens. It acts as a predecessor of antibodies by activating complement and facilitating pathogen recognition by phagocytes (5). In addition, PTX3 is essential in female fertility since it acts as a nodal point for the assembly of the cumulus oophorus hyaluronan-rich extracellular matrix (6). PTX3 also acts as a scavenger of cell debris and as an immunosuppressant (7). It specifically binds to late apoptotic cells and subsequently inhibits their uptake by dendritic cells (DC), thus acting as an additional tool to maintain immune tolerance (8).

PTX3 represents a newly discovered marker of vascular bed involvement (9). Nonvascular autoimmune diseases have vascular complications in the long term but the exact mechanisms involved in their pathogenesis have not been elucidated. However, plasma levels of PTX3 are higher in

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Abbreviations	
ACS	acute coronary syndrome
AMI	acute myocardial infarction
AT	adenine-thymine
C1q	first component of the classical complement pathway
CNS	central nervous system
COC	cumulus cell oocyte complex
CRP	C-reactive protein
DC	dendritic cells
EGF	Epidermal growth factor
ELISA	enzyme-linked immunosorbent assay
FGF-2	fibroblast growth factor-2
FSH	follicle stimulating hormone
GM-CSF	granulocyte macrophage-colony stimulating factor
HA	hyaluronic acid
HC	heavy chains
IαI	inter-α-trypsin inhibitor
IL	interleukin
imDC	immature dendritic cells
INF-γ	interferon-γ
LDL	low-density lipid lipoprotein
LPS	lipopolysaccharide
MCP-1	monocyte chemotactic protein-1
M-CSF	macrophage-colony stimulating factor
MHC	major histocompatibility complex molecules
MNB10	monoclonal antibody to PTX3
NF-IL6	nuclear factor-IL-6
NF-kβ	nuclear factor kappa-β
Ox-LDL	oxidized-low density lipid lipoprotein
PAF	platelet activator factor
PGE2	prostaglandin E2
PTX3	pentraxin 3
SAP	serum amyloid P-component
SLE	systemic lupus erythematosus
SNP	single nucleotide polymorphism
SSc	systemic sclerosis
TF	tissue factor
TGF-β	β-transforming growth factor
TLR	toll like receptor
TNF-α	tumor necrosis factor-α
TSG-6	tumor necrosis factor-α-induced protein-6
UAP	unstable angina pectoris
VSMC	vascular smooth muscle cells

vascular disorders including myocardial infarction, and in those disorders concerned only with autoimmunity such as small-vessel vasculitis (10,11); in those contexts, PTX3 levels correlate with the clinical outcome and disease activity (12). Several markers (short pentraxins) are thought to correlate well with the vascular inflammatory state. However, PTX3 is the earliest inflammatory marker that can be tested (10). The normal levels of PTX3 range from undetectable to 1.5 ng/mL, and concentrations can be measured with noncommercial assay kits based on a monoclonal antibody (MNB10) or rabbit antiserum (11).

In this review, we describe the complex functions of PTX3 and its potential role in the induction or protection of autoimmunity. We also discuss their clinical significance in different settings and their possible role in the development of vascular manifestations in some autoimmune diseases.

METHODS

PubMed and MEDLINE databases were researched for the key words "pentraxin 3," "innate immunity," "apoptosis," "autoimmunity," and "endothelial dysfunction" from 1990 to 2007. All relevant articles in English and their pertinent secondary references were also reviewed. Our approach was to find all information about PTX3 and its role in innate immunity and in autoimmune diseases.

RESULTS

Molecular Structure

The PTX3 protein is 381 amino acids long including a signal peptide of 17 amino acids. The mature secreted protein has a predicted molecular weight of 40,165 D and an apparent molecular mass which corresponds to a decamer (13). PTX3 consists of 203 amino acids, composed of a C-terminal, pentraxin-like domain coupled with an N-terminal portion of 178 amino acids unrelated to other known proteins (14). PTX3 is much longer than SAP and CRP at its NH2 terminus portion (15) (Fig. 1).

The crystal structure of PTX3 has not been determined yet, but computer analysis shows that the PTX3 structure fits comfortably within the 3-dimensional structure of SAP (14). When comparing PTX3 to SAP, there are changes in all amino acids involved in interprotomer interactions. The protomers[1] are assembled to form multimers stabilized by disulfide bonds (15). PTX3 is characterized by a β-sheet secondary structure and an α-helical arrangement for the domain. The domain shares with the short pentraxins the typical pentraxin family signature sequence (HxCxS/TWxS, where x is any amino acid), which is 8 amino acids in length and is conserved in all the members of the pentraxin family (14) (Fig. 1). Two cysteines at amino acid positions 210 and 271 form the disulfide bridge and are responsible for determining the secondary structure of the domain (Fig. 2).

In addition, pentraxins are described to be variably glycosylated among species (15,16). The presence of certain oligosaccharides may affect protein activity in various ways. They can mask a large region of the molecule to ligand recognition, participate in protein folding, and define binding sites on the protein surface (16,17). PTX3 is mainly N-linked to fucosylated and sialylated, biantennary, complex-type sugars (17). The site of sugar fucosylation attachment is asparagine 220 (Asn 220), which is

[1] Protometer: a subunit from which a larger protein structure is built. Multimer: a protein made up of more than one peptide chain. Glycosylation: the process of adding sugar units such as glycan chains to protein. Fucosylation: the process of adding manose sugar to proteins.

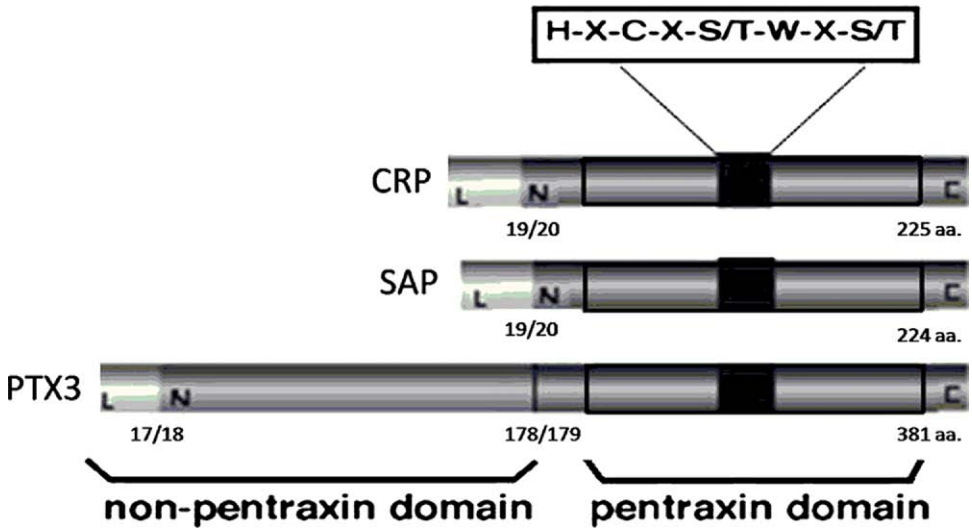

Figure 1 Structural organization of short and long pentraxins. The figure shows a comparison in sequence homology between C-reactive protein (CRP), serum amyloid protein (SAP), and pentraxin 3 (PTX3). Each one is divided into 3 sections: L- (leader peptide), N- (amino terminal portion); C- (carboxy-terminal portion); the longer nonpentraxin 3 domain is unrelated to other known proteins. The sequence on top is the pentraxin signature and contains a canonical pentraxin sequence 8 amino acids in length (HxCxS/TWxS), where x represents any amino acid. The pentraxin signature is highly conserved in the whole pentraxin family. The numbers represent amino acid long portions. [Adapted from reference (14).] (Color version of figure is available online.)

located within the pentraxin domain and is known to mediate C1q recognition (Fig. 2). C1q represents the first described and best characterized PTX3 ligand (18) and each protomer is capable of binding to one C1q molecule (18) (Fig. 3). In addition, sialic acid interacts with polar residues at specific positions (Fig. 2). Deglycosylation or desialylation equally enhance PTX3 binding to C1q, but the lack of sialic acid is the main determinant of this effect (17). PTX3 glycosylation represents a mechanism for the modulation of complement activation via the classical

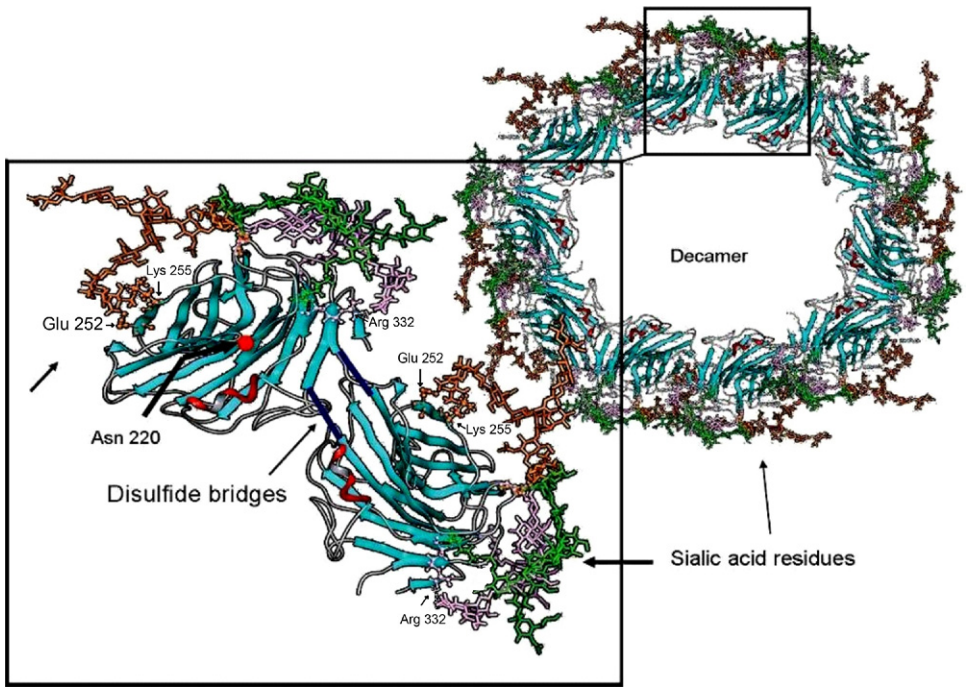

Figure 2 Tridimensional hypothetical model of pentraxin 3. Alpha-helix interacts with β-strength forming protomers, which are organized in a decamer structure. The spatial proximity of 2 cysteine residues in the loop regions of each protomer permits the formation of unstrained disulphide bridges between them (highlighted in the left quarter). These constitute a stabilized decameric protein. Sialic acids join to specific residues (lys214, glu252, lys255, and arg332) and these are exposed in the external face of the protein. Fucosylation site is located in the pentraxin domain which interacts with the C1q complement component. [Adapted from reference (17).]

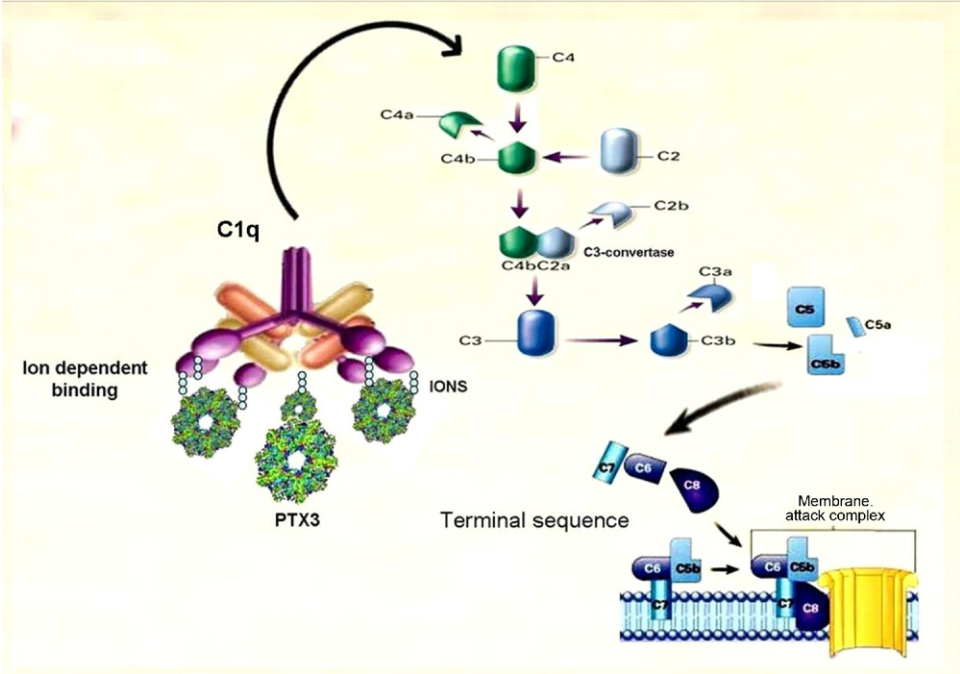

Figure 3 PTX3 and complement interaction. Specific residues located at the C1q globular head interact closely with PTX3 for binding. Ionic interactions should be performed before (H+). Once binding occurs, a change in the configuration of the C1q component structure induces complement activation via the classical pathway.

pathway (Fig. 3). Otherwise, the degree of glycosylation also plays an important role in regulating PTX3 functions during acute immune responses to pathogens. Protein-conjugated oligosaccharides can be processed in the extracellular environment by specific exoglycosidases. Among these, neuraminidases appear to be particularly relevant (19). Different pathogens express neuraminidases on their surface so that they can modulate PTX3 binding to their membranes. Polymorphonuclear leukocytes also move intracellular neuraminidases to their plasma membrane on inflammatory stimuli (17) (Fig. 4).

The N-terminal portion of PTX3, which is unrelated to other known proteins, appears to be related to the binding of the fibroblast growth factor-2 (FGF2), a potent angiogenic growth factor. FGF2 stimulates the migration and proliferation of fibroblasts during inflammation and of small muscles cells during atherosclerosis. PTX3 binds FGF2 with high affinity and selectivity and without showing any interaction with other growth factors, cytokines, chemokines, or other members of the FGF family (20). A loop region consisting of a 97 to 110 amino acid long sequence located on the N-terminal portion has been identified as a possible site for binding. It includes an α2-helix residue (glutamic 97), β-turn residues (alanine-104, proline-105, glycine-106, alanine-107), and several α3-helix residues (alanine-109, glutamic acid-110) (20). The capacity of PTX3, but not of CRP, to bind FGF2 suggests that the 178-amino-acid, NH2-terminal portion, absent in short pentraxins, is largely involved in this process. Moreover, C1q does not compete with FGF2 for binding to PTX3, which confirms that distinct domains are involved in the interaction with the two ligands (17). PTX3 inhibits FGF2-dependent endothelial cell proliferation and angiogenesis in vivo. It inhibits smooth muscle cell activation in vitro and intima thickening after arterial injury in vivo (21). In all these conditions, the concomitant production of PTX3 might be modulating the activity exerted by FGF2 on these cells in all contexts (21) (Table 1).

PTX3 Functions

PTX3 has a special role in the clearance of apoptotic cells and in the regulation of self-antigen presentation by DC (22). When apoptotic cells are not soon cleared, they undergo secondary necrosis and release intracellular content in the context of inflammatory signals (22,23). These modifications may also unmask epitope T-cell-dependent, dominant auto-antigens (24,25). In this context, PTX3 binds to apoptotic cells, inhibiting their recognition by dendritic cells but not by macrophages (Fig. 4) (26). Normal binding occurs late in the apoptotic process but before the appearance of a necrotic state (26) and, in contrast to what happens with classic short pentraxins, it is calcium independent (27,28). PTX3 binds to apoptotic cells better than to necrotic ones in vitro (26). It fails to recognize nuclear domains. The estimated half residence time on the apoptotic plasma membrane is longer than 36 hours in vitro (26). However, the exact binding site has not been defined (29). Binding occurs at a definite mo-

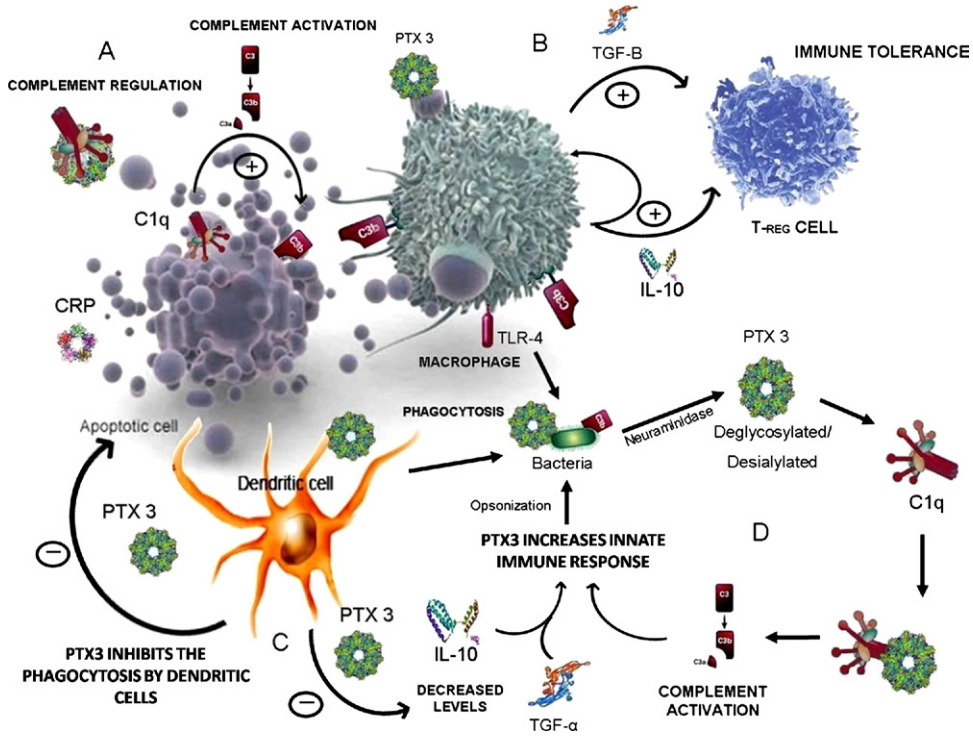

Figure 4 Pentraxin 3 functions. (A) Apoptotic cells are opsonized by complement components and pentraxins, thus facilitating prompt uptake of cellular debris to maintain peripheral immune tolerance. PTX3 regulates complement activation to avoid an exaggerated innate immune response. (B) PTX3 induces macrophages to secrete immunosuppressive cytokines, such as TGF-β and IL-10. (C) PTX3 edits cross presentation of self-epitopes expressed by apoptotic cells by inhibiting phagocytosis by dendritic cells and down-regulating the expression of immunosuppressive cytokines, which increases normal innate immune response to pathogens. (D) The PTX3 glycosylation state affects the interaction with C1q. On their surfaces, pathogens and polymorphonuclear leukocytes express neuraminidases, which are capable of removing sugars from protein surfaces, thus modulating the activation of the complement via the classical pathway. PTX3 also acts as an opsonin by enhancing phagocytosis of pathogens by macrophages and dendritic cells. C1q: first component of the complement cascade; CRP: C-reactive protein; TLR-4: toll-like receptor-4; TGF-β: transforming growth factor beta; IL-10: interleukin-10; PTX3: Pentraxin 3; TGF-α: transforming growth factor-alpha. (Color version of figure is available online.)

ment during the apoptotic process (30), thus preventing the onset of autoimmune reactions in inflamed tissues (31,32) (Fig. 4).

PTX3 also edits antigen presentation by imDC during acute phase immune response. It only edits cross-presentation of epitopes expressed by apoptotic cells to autoreactive CD8 T-lymphocytes and this effect does not depend on the features of the antigen (26,33). ImDCs phagocytose antigens more efficiently and present them at the local lymphoid node by both class II and I major histocompatibility complex molecules (34). Usually, particulate antigens, such as cell-associated antigens, are several-fold more efficiently cross-presented than soluble ones (35). It is important to note that PTX3 does not influence the presentation of exogenous, soluble antigens (36).

PTX3 also influences the local cytokine profile. It inhibits the release of TNF-α and IL-10 by LPS-challenged dendritic cells (DCs), but in the presence of apoptotic cells, PTX3 reverses the inhibition (Fig. 4). PTX3 also increases the production of tissue growth factor-α (TGF-α) by DCs when challenged by apoptotic cells (37). PTX3 enhances the immune response to pathogens but attenuates this when it derives from a necrotic context (36). PTX3 consistently inhibits the up-regulation of membrane molecules (CD86, HLA-ABC, and HLA-DR) on an inflammatory cell surface induced by LPS regardless of the apoptotic stage. At the same time, PTX3 also induces macrophages to secrete immunosuppressive cytokines, such as TGF-β and IL-10 (38,39) (Fig. 4). PTX3 is a key molecule that maintains the slight equilibrium between infection and autoimmunity.

Additional functions of PTX3 are represented in another context. PTX3 appears to be a structural and essential constituent of the COC matrix which allows fecundation to occur (Table 1) (40). PTX3 has recently been shown to bind to the HC derived form the IαI (41). The HCs transfer from IαI to hyaluronic acid (HA) by the catalytic activity of the α-induced protein (TSG6) and bind to distinct protomers in the PTX3 protein; each protomer might interact with a single HCs molecule (41). Although PTX3 does not interact directly with HA, it might cross-link several HA strands by binding HCs that are covalently linked to each protomer in the PTX3 molecule (42). In addition it has been shown that HCs inter-

act only with the N-terminal portion of PTX3 molecule and that this portion is required and sufficient for organizing and for enabling matrix formation. Those findings suggest that the essential role played by PTX3 in matrix assembly is specifically related to its N-terminal portion (42). The COC matrix organization might be responsible for the formation of a postulated gradient of sperm chemoattractants, both by determining the tighter arrangement of corona radiata cells and likely by regulating the diffusion of molecules synthesized by the oocyte and cumulus cells (43). Thus, PTX3 probably is related to matrix deposition, tissue repair, remodeling, and fertility.

PTX3 has an additional role in innate resistance to selected pathogens. It binds to the outer membrane of protein A, a conserved constituent of enterobacteria (*Pseudomonas aeruginosa* and *Salmonella typhimurium*) (44), conidia of *Aspergillus fumigatus* (45), zymosan, and *Paracoccidioides brasiliensis* but not to the outer membrane proteins of *Listeria monocytogenes*, *Escherichia coli*, or *Candida albicans* (Table 1) (46). Mycobacterium tuberculosis induces PTX3 expression in monocytes (47). Blood levels are useful for monitoring disease activity in infected individuals (48). Recently, single nucleotide polymorphisms (SNPs) at the *PTX3* gene have been studied for susceptibility to pulmonary tuberculosis (49).

All described functions point to a complex interaction between all components of innate immunity during inflammatory processes. Patients with systemic autoimmune diseases show a remarkable defect in the generation of pentraxins during flare-ups (50). In contrast to its normal function, in some vascular inflammatory diseases, PTX3 seems to inhibit rather than facilitate the uptake of late apoptotic cells, thus leading to local autoimmune reactions (Table 1). This has led to the suggestion that at times it might facilitate autoimmunity, and 1 clue might come from the PTX3 gene variants.

PTX3 Gene

Pentraxins belong to the animal lectins family and have been found in many vertebrate and some invertebrate species. In *Limulus polyphemus* (horseshoe crab), the ancient "living fossil," pentraxins have been shown to play an important role in innate host defense (51). Horseshoe crab serum pentraxins and mammalian long pentraxins cluster to form a clade distinct from other pentraxins (52). However, the evolutionary relationship between the short and long pentraxins is not clear. PTX3 appears to represent a fusion between regions encoding a specific amino-terminal polypeptide of unknown function and a carboxy-terminal polypeptide homologous to the short pentraxins. These similarities suggest a common ancestral protein, which was certainly a pentraxin. Furthermore, PTX3 is highly conserved (82% identity, 92% similarity) between mouse and man in terms of primary sequence and gene regulation (53).

PTX3 was the first cloned member of the newly emerging group of long pentraxins. It is located on human chromosome 3q25.6. In contrast, *SAP* and *CRP* genes are located on chromosome 1 (53). The exact size of the PTX3 transcript is 1861 nucleotides. The complete nucleotide sequence of the PTX3 gene begins with a 68-bp 5′-untranslated region followed by the site of translation initiation at nucleotide 92 (ATG boxed), where the first methionine is located (Fig. 5). The transcript starts with a 1143-nucleotide-long open reading frame and ends with a TAA stop codon at position 1211. It is followed by a long AT-rich 3′-untranslated region (70% AT), which is a polyadenylation signal, located at position 1802 (Fig. 5). The 3′-untranslated portion contains, in addition, 2 consensus sequences for mRNA instability (53).

The organization of the PTX3 gene is 3 exons separated by 2 introns. The first 2 exons encode, respectively, for the leader peptide and the N-terminal domain of the protein, while the third exon encodes the pentraxin domain. Thus, the third exon matches the other members of the pentraxin family exactly, while the first 2 exons are divergent (Fig. 5) (54).

The promoter has a number of potential binding sites for transcription factors. One nuclear factor-IL-6 (NF-IL6), two nuclear factor kappa-β (NF-kβ), one AP-1, two Pu1, three PEA 3, one Ets-1, and two Sp1 consensus sequences have been identified (Fig. 5). The NF-kβ in the human and mouse promoter is essential for the induction by IL-1 and TNF-α. No obvious TATA or CAAT consensus box has been found (54). TNF-α exposure increases NF-kβ activity, while the minimal construct carrying an inactivating mutation at this site loses the TNF-inducibility in cultured cells.

PTX3 mRNA is strongly induced by both IL-l and TNF-α but not by IL-6 (54), which is a potent inducer of CRP and SAP in the liver. The presence of only one NF-IL6 binding site on the PTX3 gene and the absence of multiple copies of signal transducers and activators of transcription might explain the lack of response to IL-6 (Fig. 5). The PTX3 gene also lacks consensus sites for other hepatic transcription factors (54).

SNPs influence baseline pentraxin-levels (55). Some authors have suggested that reducing levels of other pentraxins, for instance CRP, allows the development of antinuclear antibodies, leading to the appearance of SLE (56-58). Prospective studies have indicated an association between CRP levels, polymorphisms in the CRP gene, and the long-term risk of cardiovascular disease (59). Several SNPs in the PTX3 gene have also been reported (Fig. 5), but none has been studied in vascular pathology. PTX3 polymorphisms might explain the apparently opposite functions in different settings. To date, two main features characterize PTX3-deficient mice: they are sterile and more susceptible to infection by selected pathogens (Table 1) (60).

Table 1 Pentraxin 3 and Disease

Microenvironment	Stimuli	Down Regulation	Cellular Source
Inflammation	LPS	IFN-γ	Dendritic cells
	IL-10[a] IL-1β TNF-α Toll-like receptors ligands	TGF-β	Fibroblasts Endothelial cells
	LAM		
			Mononuclear phagocytes
		IL-4, IL-13, and IL-10[c]	
Apoptosis	LPS	TGF-β?	Dendritic cells, fibroblast, macrophages
	IL-1 TNF-α	IL-10?	
			Neutrophils in dermal tissue around blood vessels
Central nervous system	LPS		Glial cells
	IL-1 TNF-α	?	
	Seizures		
Vascular bed	Oxidized LDL	IFN-γ	Endothelial cells
	IL-1β TNF-α	TGF-β	
			Hypertrophied human cardiomyocytes
			Advanced atherosclerotic plaques
			VSMC
			Human mesangial cells

Ligands	Functions	Disease	Ref.
Outer membrane protein A of enterobacteria	Mediation of microbicidal activity	Sepsis	(5)
			(36)
Microbial moieties		Infection by enterobacteria	(43)
			(45)
Conidia of *Aspergillus fumigatus* (in vitro)	Increasing of complement opzonization	Invasive pulmonary aspergillosis in PTX3-deficient mice	(46)
			(47)
TLRs		*Mycobacterium tuberculosis* infection (correlation with disease activity)	(48)
	Modulation of mononuclear phagocyte functions (opzonization and phagocytosis)		(49)
			(74)
			(84)
Late apoptotic cells but the domain is unknown (the binding is calcium independent)	Clearance of apoptotic cells	SLE (short pentraxins)	
			(12)
C1q, complement component	Peripheral immune tolerance	Small-vessel vasculitis	(18)
			(30)
		Wegener's granulomatosis	(88)
	Leukocytoclastic phenomena (inhibition of the clearance of apoptotic neutrophils)		(89)
		Microscopic polyangiitis	(92)
			(97)
		Churg–Straus syndrome	
Seizure-challenged neurons	Attraction of leukocytes from vasculature to neural loss areas	Limbic seizures	
			(68)
	Rescue of dying neurons from irreversible damage		(69)
Apoptotic neurons?		EAE	(70)
FGF-2 (*N*-terminal portion of PTX3)	Inhibition of binding of growth factor to tyrosine-kinase receptors	Atherosclerosis and restenosis	
			(9)
			(10)
Endothelial cells	Increasing in the expression of TF and procoagulant activity		(11)
(Modulation of procoagulant activity)		Unstable angine pectoris	(20)
			(21)
C1q, complement component	Mediation of vascular damage? (complement activation)		(71)
			(72)
	Clearance of lipid-load apoptotic macrophages	AMI	(73)
		(Early indicator of a cardiovascular event in acute coronary syndromes)	(81)
			(82)
			(83)
	Synthesis of proinflammatory lipid mediator-PAF by MC	IgA, type I membranoproliferative GN	

Table 1 Pentraxin 3 and Disease (continued)

Microenvironment	Stimuli	Down Regulation	Cellular Source
Kidney			
	TNF-α	?	Monocyte/macrophages
	IgA		
			Renal interstitium
			Peritubular capillaries
Vascular placental cells	Proinflammatory cytokines		Stroma of the stem villi and in the anchoring
			Villi in normal and preeclamptic
		?	Placentas
Joint	TNF-α	TNF-β	Sinovial tissue
		INF-γ	(Type B sinovial cells and endothelial cells)
		IL-4[d]	
Skin	Constitutive expression[b]	TNF-β	Skin scleroderma fibroblast
		TGF-β	
		INF-γ	Cutaneous psoriasis
		IL-4[d]	
Ovary	FSH		Cumulus oophorus cells (ovary) in the preovulatory follicle and it is located in the extracellular matrix
	EGF	?	
	PGE2		
	hCG		
	(in vitro)		

AMI, acute myocardial infarction, EAE, experimental allergic encephalomyelitis, EGF, epidermal growth factor; FGF-2, fibroblast growth factor-2; FSH, follicle-stimulating hormone; FSGS, focal segmental glomerular sclerosis; GN, glomerulonephritis; HCG, human chorionic gonadotropins; IFN-γ, interferon gamma; IGR, intrauterine growth restriction; LDL, low-density lipid lipoprotein; LPS, lipopolysaccharide; LAM, lipoarabinomannan; MC, mesangial cells; PAF, platelet activating factor; PE, preeclampsia; PGE2, prostaglandin E2; SLE, systemic lupus erythematosus; TF, tissue factor; TGF-β, β-transforming growth factor; PSo, psoriasis; RA, rheumatoid arthritis; SSc, systemic sclerosis; TLRs, toll-like receptors; TNF-α, tumor necrosis factor-alpha; TSG-6, tumor necrosis factor α-induced protein 6; VSMC, vascular smooth muscle cells.

[a]IL-10 is a weak and inconsistent stimulus of PTX3 production (4).

[b]NF-α and IL-1β increase gene expression (14).

[c]These cytokines do not affect PTX3 production during *M. tuberculosis* infection (44).

[d]Inhibits constitutive but not stimulated expression (84).

Cellular Sources and Regulation of Production

PTX3 is produced at inflammatory sites by several cell types of which DC, macrophages, fibroblasts, activated endothelial cells, renal cells, and smooth muscle cells are the primary ones (61-63) (Table 1). However, the production of PTX3 by DC is subjected to a distinct regulatory network. IFN-γ (inhibitory) and IL-10 (stimulatory) exert opposite effects (Table 1) (64). IL-10 stimulates B-cell differentiation and participates in the activation of the humoral arm of innate (PTX3), and adaptive (antibodies) immunity (64). Anti-inflammatory cytokines and cytokines associated with type II responses (IL-4, IL-13) do not affect PTX3 production, although IL-4 seems to inhibit its constitutive expression by scleroderma fibroblasts (Table 1) (65). Other cytokines, including IL-6, monocyte chemotactic protein-1, M-CSF, granulocyte macrophage-colony stimulating factor, and IFN-γ are not able to induce PTX3 gene expression. Indeed PTX3 represents the activation of innate immunity and modulation of the adaptive immune response (66,67). PTX3 is not consti-

Ligands	Functions	Disease	Ref.
			(63)
	Mesangial cell contraction	Diffuse proliferative lupus GN	
Mesangial cell			(90)
	Interstitial damage	FSGS	
	Glomerular and tubular injury?		
Apoptotic placental cells?	Placental vascular damage?	PE	(100)
Inhibition of the recognition of apoptotic cells by dendritic cells?	Atherothrombosis		
		IGR	(101)
	Complement activation Amplification of the complement-mediated inflammation and joint damage?		
Not well recognized		RA	(85)
	A marker of scleroderma fibroblast activation	SSc	(65)
They have not been defined			
	An early marker of endothelial dysfunction vs ethyopathogenic role?		
		PSo	(87)
			(86)
Heavy chains derived from hyaluronic acid and thereby acting as a node to cross-link multiple hyaluronic acid chains	Female fertility	Infertility	(6)
	Assembly of the ovarian cumulus oophorus hyaluronan-rich extracellular matrix		(8)
			(40)
			(60)

tutively expressed in the central nervous system but might be inducible in response to inflammatory signals (68,69). It is found in central nervous system regions where seizure-induced neurodegeneration and local autoimmune reactions occur (Table 1). PTX3 appears to bind dying neurons and rescue them from otherwise irreversible damage (Fig. 6) (70).

PTX3 also represents the cross-linking between inflammation and coagulation where endothelial cells represent a common ground for both (71). Endothelial cells participate actively at sites of vascular injury by expressing tissue factor on their membrane. When generated by endothelial cells, PTX3 potentiates the expression of tissue factor at vascular sites by itself (Fig. 6) (72). Hence, PTX3 is of particular relevance for the amplification of the clotting activation, which is involved in thrombogenesis, wound healing, and vascular ischemia (Table 1) (72). All these processes have been demonstrated on advanced atherosclerotic plaques where PTX3 has also been found (73). PTX3 is stored in specific granules within neutrophils

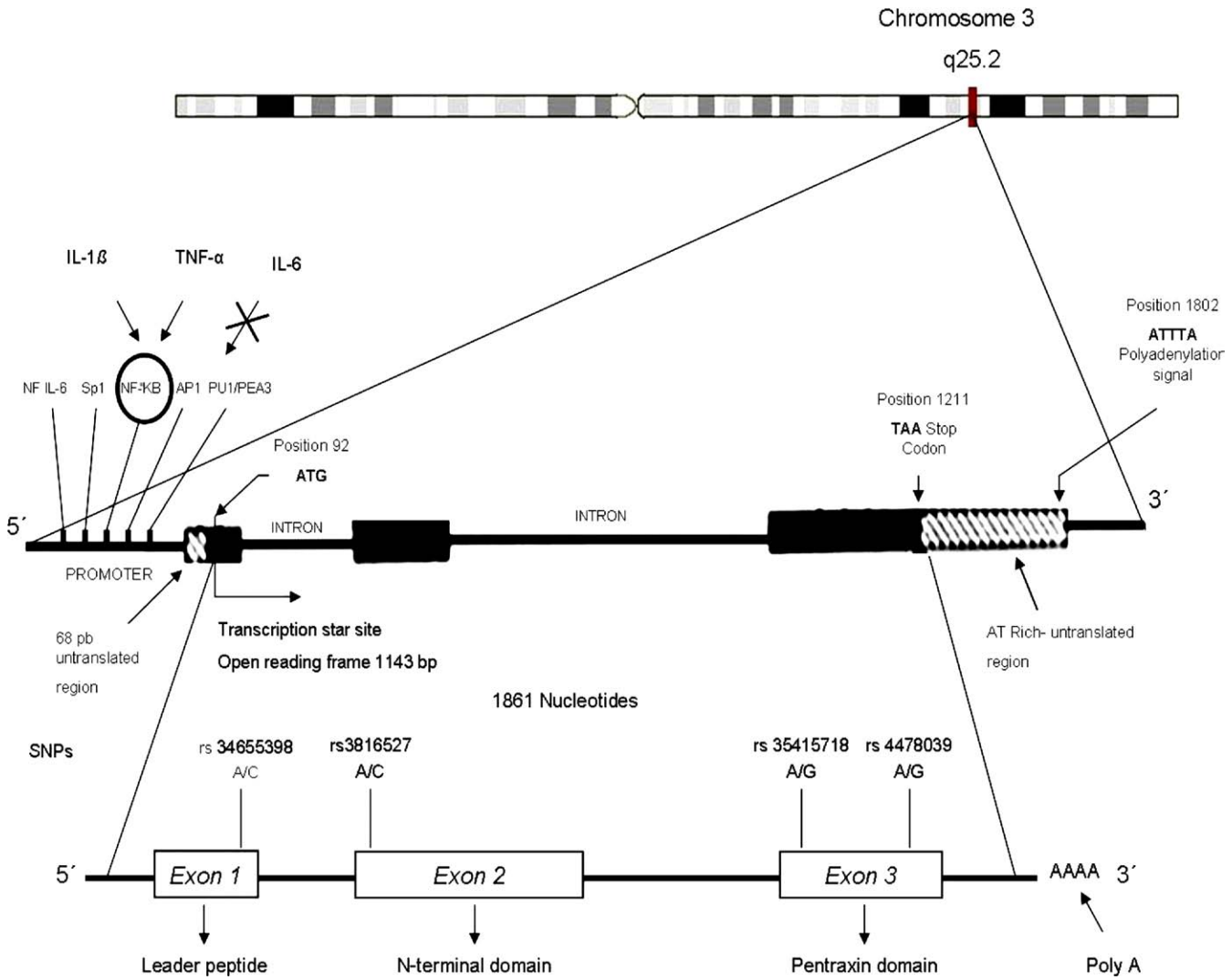

Figure 5 Pentraxin 3 gene. The organization of the human PTX3 gene (at 3q25) is 3 exons separated by 2 introns. The first 2 exons, respectively, encode for the leader peptide and the N-terminal domain of the protein. The third exon encodes the pentraxin domain. A genomic fragment of 1317 bp located 59 bp upstream from the transcription start site corresponds to the NF-kβ transcription factor binding site, which responds to TNF-α and IL-1β stimulation. The PTX3 promoter does not response to IL-6 stimulation because only one NF-IL 6 binding site is present. Sp1 and AP-1 transcriptional binding sites do not seem to play a major role in the cytokine inducibility of the gene. Transcription star site (ATG boxed), stop codon (TAA), and the AT 3′-rich untranslated region are also shown. SNPs are labeled above the gene. There are almost 4 validated SNPs (nonsynonymous). (Color version of figure is available online.)

near blood vessels and is released in response to microbial recognition and inflammatory signals (Fig. 7) (74).

PTX3 and Atherosclerosis

Atherosclerosis is considered an immune system-mediated inflammatory process. The presence of macrophages and activated lymphocytes within atherosclerotic plaques supports this concept (75). Acute inflammation can aggravate atherosclerosis just as autoimmunity and chronic infections do (76,77). The association of increased serum levels of acute-phase proteins with the progression of atherosclerosis in systemic autoimmune diseases has been demonstrated (78). The occurrence of related adverse events, such as coronary heart disease and myocardial infarction, has been well documented in several epidemiological studies (Table 1) (79,80).

The main autoantigen involved in the development of atherosclerosis is the LDL, which on reaching the oxidative stage is recognized and internalized by macrophages through scavenger receptors (81). Oxidized LDL increases PTX3 mRNA expression by vascular smooth muscle cells more than 70-fold (81). In contrast, native LDL does not demonstrate any effect on its expression (Table 1). Vascular smooth muscle cells actively participate in the atherogenic process by promoting local inflammatory reactions, which lead to in situ vascular damage by the activation of complement via the classical pathway (81). PTX3 also contributes to the clearance of lipid-loaded

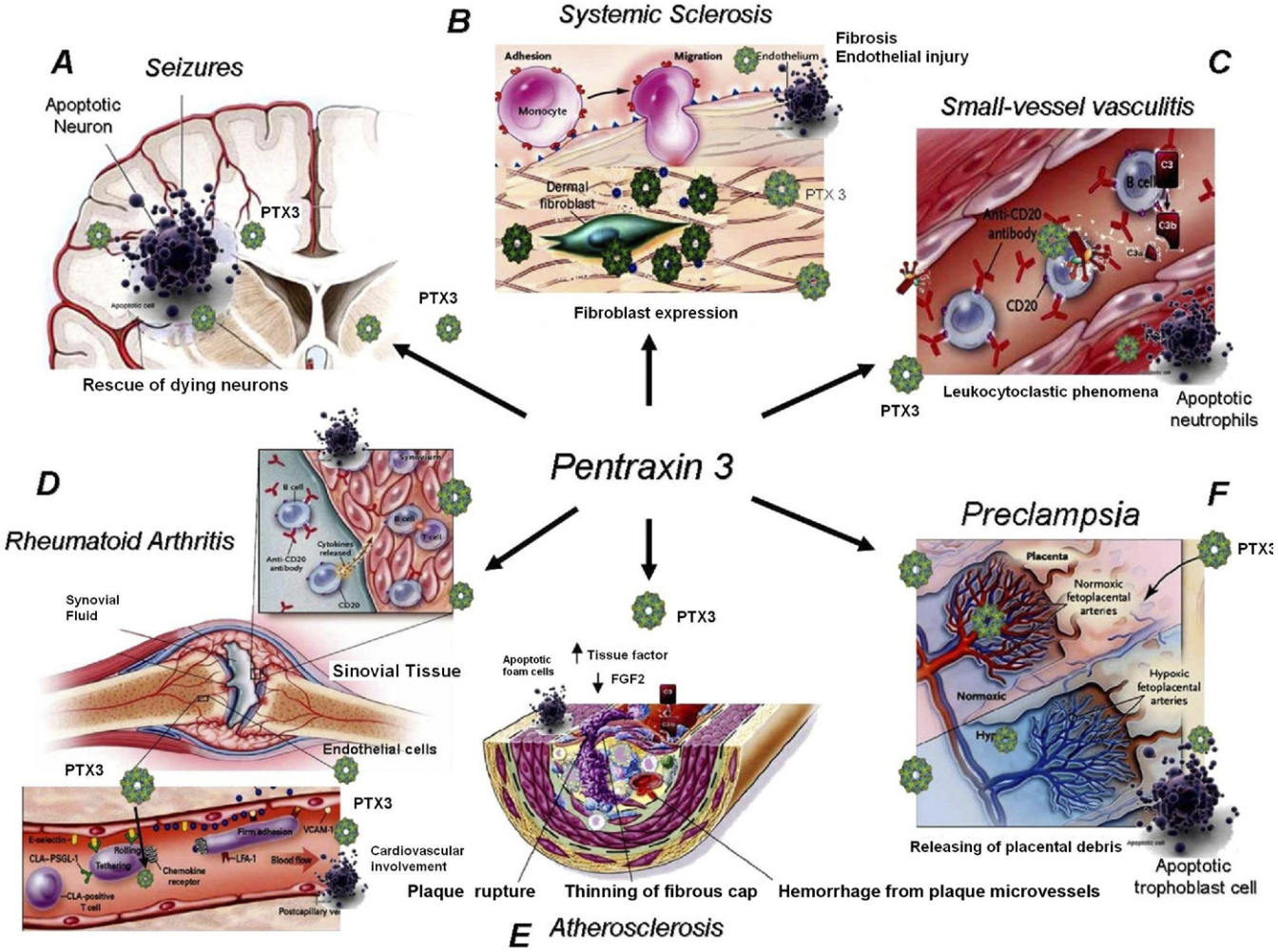

Figure 6 Autoimmune disorders and organs investigated for PTX3. (A) In the central nervous system, its expression is inducible by infection and seizures. PTX3 binds to dying neurons to rescue them from irreversible damage. (B) Scleroderma fibroblasts constitutively express PTX3, although it could be used as a vascular marker of endothelial dysfunction. (C) In small vessel vasculitis, PTX3 seems to disturb the normal clearance of apoptotic neutrophils. (D) Synovial pannus represents the main source of PTX3 in rheumatoid arthritis. It acts by amplifying complement-mediated injury but could also represent a common link between joint damage and vascular involvement. (E) Atherosclerotic plaque is another source of PTX3. Oxidized LDL induces protein expression by vascular smooth muscle cells and increases phagocytosis of dead foam cells. PTX3 also inhibits fibroblast growth factor and up-regulates tissue factor expression by endothelial cells. (F) PTX3 has been identified in the stroma of stem villi and anchoring villi in preeclamptic placentas. However, the role of elevated levels in preclampsia is not fully understood.

macrophages and vascular smooth muscle cells foam apoptotic cells, thus mediating their removal by mature DC (Fig. 6) (81). PTX3 also represents an early indicator of atherosclerotic complications such as acute myocardial infarction (AMI) among others (Table 1). Since PTX3 was demonstrated to be present in normal and hypertrophied human cardiomyocytes, cellular distribution in the myocardium of patients with infarctions was clearly distinct from that of CRP (82,83). Early disappearance of PTX3 from myocytes represents irreversible injury and ischemic cardiomyopathy. Increased blood PTX3 correlates with AMI. The peak time is much earlier than that of CRP, thus behaving like an earlier indicator of AMI (11,83). PTX3 reaches peak values after 6 to 8 hours compared with 24 hours for CRP and in patients with unstable angina pectoris, plasma levels are significantly elevated more than 3-fold (6.20 ng/mL) (9,83). These results indicate that PTX3 might be a better early predictive marker of acute coronary syndrome and severity of infection in critically ill patients (83,84).

Autoimmune Vascular Involvement

Autoimmune disorders in which PTX3 has been studied include rheumatoid arthritis (85), cutaneous psoriasis (86), systemic sclerosis (SSc) (87), small vessel vasculitis (88,89), immunoglobulin A glomerulonephritis (90), Churg–Strauss syndrome (91), and Wegener's granulomatosis (92) (Table 1). The main source of PTX3 in rheumatoid arthritis is the synovial pannus, which is rich in monocytes/macrophages and endothelial cells. Synovial tissue contains endothelial cells and synoviocytes ex-

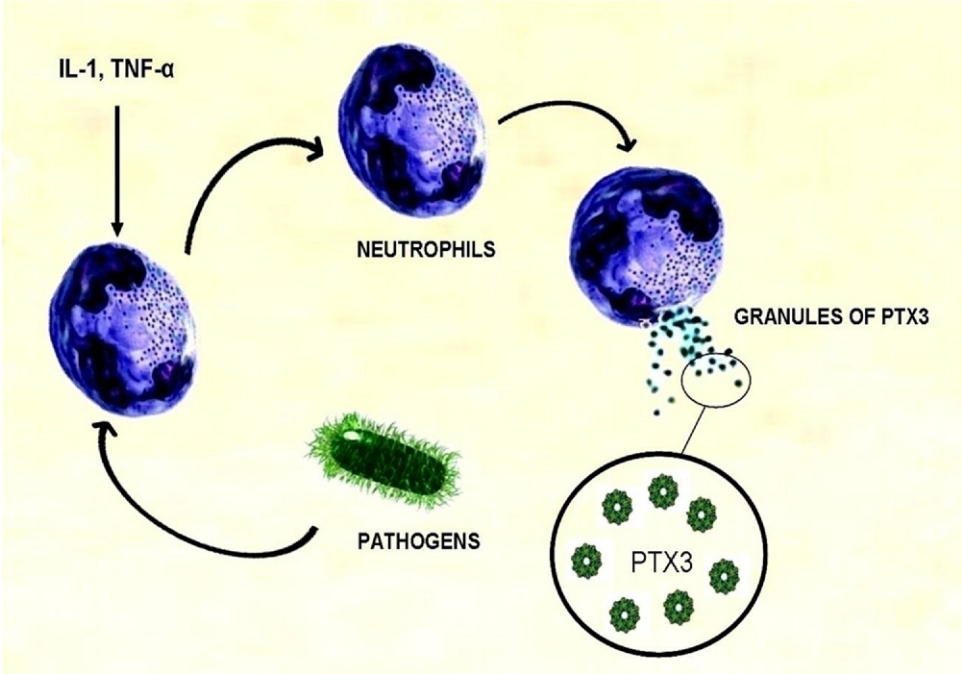

Figure 7 Neutrophils, the main source of PTX3 for rapid release. Only neutrophils and no other polymorphonuclear cells store preformed PTX3. Pathogen recognition and inflammatory signals stimulate neutrophils to release granules containing PTX3. Neutrophils represent the main reservoir when an inflammatory process begins. PTX3, which is released early, avoids an exaggerated immune response and participates in peripheral immune tolerance to self antigens. (Color version of figure is available online.)

pressing PTX3 (85). It is tempting to speculate that PTX3 participates in synovial membrane injury in early stages of the disease (amplifying complement-mediated tissue damage) (Table 1) and in the onset of extra-articular manifestations such as cardiovascular involvement at later stages (93). PTX3 levels might indirectly reflect the inflammatory state of the vasculature in rheumatoid arthritis as other pentraxins usually do (Fig. 6) (94).

PTX3 is absent in normal skin. However, biopsy from patients with severe cutaneous psoriasis shows that the expression of PTX3 by endothelial cells and high plasma levels correlate well with disease activity and severity. In mild cutaneous psoriasis, PTX3 is only produced by macrophages, but its physiopathological role in endothelial damage has not been studied (86).

In SSc, fibroblasts constitutively express high levels of PTX3 (the main feature of its activated status). This constitutive expression cannot be modified by anti-TNF-α antibodies, IL-1 receptor antagonist, or both. However, PTX3 production is suppressed by IFN-γ and TGF-β in fibroblast cultured cells (87) (Table 1). Vascular dysfunction has been suggested as one of the earliest events in SSc (Fig. 6) (65). In addition, C1q binding immune complexes have been thought to be another initial step for thrombosis in SSc (95). The possibility of PTX3 association with these phenomena in SSc has not been studied.

Early apoptotic neutrophils are normally engulfed by phagocytes, particularly in inflammatory tissue (96-98). In contrast, histopathologic features of small-vessel vasculitis almost always comprise leukocytoclastic vasculitis. This is clearly demonstrated by pyknotic nuclei and nuclear fragments of infiltrated leukocytes scattered throughout the tissue (Fig. 6) (97). PTX3 inhibits the clearance of late apoptotic neutrophils by macrophages, and therefore, it is a candidate factor for the development of leukocytoclastic phenomena (Table 1) (97). PTX3 has a typical pattern of distribution located in the area around blood vessels, but it is also present in a more diffuse pattern throughout the dermis, particularly in areas with large infiltrates. The up-regulated expression of PTX3 may favor the accumulation of unscavenged dead neutrophils on skin, which is characteristic of small-vessel vasculitis (Fig. 6) (96,97). In this case, PTX3 levels also correlate with disease activity and response to therapy (88,97). The detection of PTX3 in the renal tissue of patients with glomerulonephritis suggests a potential role in the modulation of glomerular and tubular injury (90). PTX3 might participate in the activation of endothelial and mesangial cells, by inducing them to produce proinflammatory mediators and glomeruli damage (90). PTX3 appears to increase the synthesis of PAF by mesangial cells, thus favoring the deposit of immune complexes on glomeruli and leading to the deterioration of the glomerular filtration rate. PTX3 has shown a specific pattern of distribution on interstitium, perivascular vessels, and glomeruli, depending on the glomerular disease (Table 1) (90).

Preclampsia has been associated with high levels of apoptosis in the placenta (98) and PTX3 levels are higher in

maternal blood in preclampsia. Some authors suggest that it is caused by an inflammatory response of the mother against placental debris released during pregnancy (Fig. 6) (99). Thus elevated maternal plasma levels might represent a marker of altered endothelial function or perhaps an initial step for vascular damage (99). Moreover, spontaneous preterm delivery is associated with significantly increased levels in maternal plasma and PTX3 appears to be a marker of placenta vasculopathy (100).

DISCUSSION

The evidence suggests that PTX3 is a two-edged sword having both a protective effect against autoimmunity and favoring autoimmune reactions, depending on milieu, disposal structure, cytokine profile, and gene variants. Short pentraxin polymorphisms have been associated with accelerated atherosclerosis (101) and with an increased occurrence of vascular arterial events in patients with autoimmune diseases (102). We hypothesize that PTX3 gene polymorphisms may be associated with the occurrence of vascular complications in patients with specific autoimmune diseases. PTX3 SNPs might cause a change in amino acid sequence and, thereby, in its normal function. A dysfunctional variant protein may explain its physiopathological role in some vascular settings (102-104). PTX3 is an early marker of endothelial dysfunction and may reflect the vascular inflammatory state. However, It is currently unclear if it really has a physiopathological role in vascular damage (105,106).

In terms of clinical application, PTX3 appears to be a stronger predictor of cardiovascular events than CRP, and it adds prognostic information regarding response to treatment of vascular involvement in some autoimmune diseases. Since CRP is an hepatically derived pentraxin, it does not indicate the extent of vascular compromise as well as local production of PTX3 does (83,107,108).

Otherwise, in the setting of AMI and unstable angina pectoris, the role of CRP has been largely described (109). Studies in acute coronary ischemia have shown that CRP levels are predictors of early and late mortality, just as is cardiac troponin (110). In AMI, PTX3 levels increase rapidly and earlier than CRP levels (83). It is noteworthy that no correlation was found between PTX3 and CRP levels in AMI (83). In infectious and degenerative disorders pentraxin levels also are increased. PTX3 levels correlate with clinical severity scores in sepsis and high levels are associated with an unfavorable outcome. Similarly, in autoimmune diseases, elevated levels of PTX3 in RA and SVV correlate with disease activity and response to therapy (85,88,89).

In conclusion, PTX3 may represent a new diagnostic approach unrelated to CRP for the study of inflammatory and autoimmune diseases. Further research is warranted to assess the influence of PTX3 polymorphisms on the risk of acquiring autoimmune diseases and their vascular complications.

REFERENCES

1. Introna M, Alles VV, Castellano M, Picardi G, De Gioia L, Bottazzi B, et al. Cloning of mouse ptx3, a new member of the pentraxin gene family expressed at extrahepatic sites. Blood 1996;87:1862-72.
2. Coe JE, Margossian SS, Slayter HS, Sogn JA. Hamster female protein. A new Pentraxin structurally and functionally similar to C-reactive protein and amyloid P component. J Exp Med 1981; 153:977-91.
3. Bottazzi B, Garlanda C, Salvatori G, Jeannin P, Manfredi A, Mantovani A. Pentraxins as a key component of innate immunity. Curr Opin Immunol 2006;18:10-5.
4. Han B, Mura M, Andrade CF, Okutani D, Lodyga M, dos Santos CC, et al. TNF alpha induced long pentraxin PTX3 expression in human lung epithelial cells via JNK. J Immunol 2005;175:8303-11.
5. Garlanda C, Hirsch E, Bozza S, Salustri A, De Acetis M, Nota R, et al. Nonredundant role of the long pentraxin PTX3 in antifungal innate immune response. Nature 2002;420:182-6.
6. Varani S, Elvin JA, Yan C, DeMayo J, DeMayo FJ, Horton HF, et al. Knockout of pentraxin 3, a downstream target of growth differentiation factor-9, causes female subfertility. Mol Endocrinol 2002;16:1154-67.
7. Kravitz MS, Pitashny M, Shoenfeld Y. Protective molecules—C-reactive protein (CRP), serum amyloid P (SAP), pentraxin3 (PTX3), mannose-binding lectin (MBL), and apolipoprotein A1 (Apo A1), and their autoantibodies: prevalence and clinical significance in autoimmunity. J Clin Immunol 2005;25:582-91.
8. Garlanda C, Bottazzi B, Bastone A, Mantovani A. Pentraxins at the crossroads between innate immunity, inflammation, matrix deposition, and female fertility. Annu Rev Immunol 2005;23: 337-66.
9. Inoue K, Sugiyama A, Reid PC, Ito Y, Miyauchi K, Mukai S, et al. Establishment of a high sensitivity plasma assay for human pentraxin 3 as a marker for unstable angina pectoris. Arterioscler Thromb Vasc Biol 2007;27:161-7.
10. Peri G, Introna M, Corradi D, Iacuitti G, Signorini S, Avanzini F, et al. PTX3. A prototypical long pentraxin, is an early indicator of acute myocardial infarction in humans Circulation 2000; 102:636-41.
11. Latini R, Maggioni AP, Peri G, Gonzini L, Lucci D, Mocarelli P, et al. Prognostic significance of the long pentraxin PTX3 in acute myocardial infarction. Circulation 2004;110:2349-54.
12. Fazzini F, Peri G, Doni A, Dell'Antonio G, Dal Cin E, Bozzolo E, et al. PTX3 in small vessel vasculitides: an independent indicator of disease activity produced at sites of inflammation. Arthritis Rheum 2001;44:2841-50.
13. Bottazzi B, Vouret-Craviari V, Bastone A, De Gioia L, Matteucci C, Peri G, et al. Multimer formation and ligand recognition by the long pentraxin PTX3. Similarities and differences with the short pentraxins C-reactive protein and serum amyloid P component. J Biol Chem 1997;272:32817-23.
14. Montovani A, Garlanda C, Otero K, Peri G, Vecchi A, Bottazzi B. Membrane and soluble pattern recognition receptors: unique functions of the long pentraxin PTX3. Clin Exp All Rev 2004; 4:S150-4.
15. Srinivasan N, White HE, Emsley J, Wood SP, Pepys MB, Blundell TL. Comparative analyses of pentraxins: implications for protomer assembly and ligand binding. Structure 1994;2: 1017-27.
16. Cross AS, Sakarya S, Rifat S, Held TK, Drysdale BE, Grange PA, et al. Recruitment of murine neutrophils in vivo through endogenous sialidase activity. J Biol Chem 2003;278:4112-20.
17. Inforzato A, Peri G, Doni A, Garlanda C, Mantovani A, Bastone A, et al. Structure and function of the long pentraxin PTX3 glycosidic moiety: fine-tuning of the interaction with C1q and complement activation. Biochemistry 2006;45:11540-51.

18. Nauta AJ, Bottazzi B, Mantovani A, Salvatori G, Kishore U, Schwaeble WJ, et al. Biochemical and functional characterization of the interaction between pentraxin 3 and C1q. Eur J Immunol 2003;33:465-73.
19. Camara M, Boulnois GJ, Andrew PW, Mitchell TJ. A neuraminidase from Streptococcus pneumoniae has the features of a surface protein. Infect Immun 1994;62:3688-95.
20. Camozzi M, Rusnati M, Bugatti A, Bottazzi B, Mantovani A, Bastone A, et al. Identification of an antiangiogenic FGF2-binding site in the N terminus of the soluble pattern recognition receptor PTX3. J Biol Chem 2006;281:22605-13.
21. Camozzi M, Zacchigna S, Rusnati M, Coltrini D, Ramirez-Correa G, Bottazzi B, et al. Pentraxin 3 inhibits fibroblast growth factor 2-dependent activation of smooth muscle cells in vitro and neointima formation in vivo. Arterioscler Thromb Vasc Biol 2005;25:1837-42.
22. Franz S, Gaipl US, Munoz LE, Sheriff A, Beer A, Kalden JR, et al. Apoptosis and autoimmunity: when apoptotic cells break their silence. Curr Rheumatol Rep 2006;8:245-7.
23. Navratil JS, Liu CC, Ahearn JM. Apoptosis and autoimmunity. Immunol Res 2006;36:3-12.
24. Mahnke K, Johnson TS, Ring S, Enk AH. Tolerogenic dendritic cells and regulatory T cells: a two-way relationship. J Dermatol Sci 2007;46:159-67.
25. Cocca BA, Cline AM, Radic MZ. Blebs and apoptotic bodies are B cell autoantigens. J Immunol 2002;169:159-66.
26. Rovere P, Peri G, Fazzini F, Bottazzi B, Doni A, Bondanza A, et al. The long pentraxin PTX3 binds to apoptotic cells and regulates their clearance by antigen presenting dendritic cells. Blood 2000;96:4300-6.
27. Bijl M, Horst G, Bijzet J, Bootsma H, Limburg PC, Kallenberg CG. Serum amyloid P component binds to late apoptotic cells and mediates their uptake by monocyte derived macrophages. Arthritis Rheum 2003;48:248-54.
28. Szalai AJ. C-Reactive protein (CRP) and autoimmune Disease: facts and conjetures. Clin Dev Immunol 2004;11:221-6.
29. Schwalbe RA, Dahlbäck B, Coe JE, Nelsestuen GL. Pentraxin family of proteins interact specifically with phosphorylcholine and/or phosphorylethanolamine. Biochemistry 1992;31: 4907-15.
30. Van Rossum AP, Fazzini F, Limburg PC, Manfredi AA, Rovere-Querini P, Mantovani A, et al. The prototypic tissue pentraxin PTX3, in contrast to the short pentraxin serum amyloid P, inhibits phagocytosis of late apoptotic neutrophils by macrophages. Arthritis Rheum 2004;50:2667-74.
31. Familian A, Zwart B, Huisman HG, Rensink I, Roem D, Hordijk PL, et al. Chromatin independent binding of serum amyloid P component to apoptotic cells. J Immunol 2001;167: 647-54.
32. Roos A, Xu W, Castellano G, Nauta AJ, Garred P, Daha MR, et al. Mini-review: a pivotal role for innate immunity in the clearance of apoptotic cells. Eur J Immunol 2004;34:921-9.
33. Patel VA, Longacre A, Hsiao K, Fan H, Meng F, Mitchell JE, et al. Apoptotic cells, at all stages of the death process, trigger characteristic signaling events that are divergent from and dominant over those triggered by necrotic cells: Implications for the delayed clearance model of autoimmunity. J Biol Chem 2006;281: 4663-70.
34. Wilson NS, El-Sukkari D, Villadangos JA. Dendritic cells constitutively present self antigens in their immature state in vivo and regulate antigen presentation by controlling the rates of MHC class II synthesis and endocytosis. Blood 2004;103: 2187-95.
35. Schnorrer P, Behrens GM, Wilson NS, Pooley JL, Smith CM, El-Sukkari D, et al. The dominant role of CD8+ dendritic cells in cross-presentation is not dictated by antigen capture. Proc Natl Acad Sci USA 2006;103:10729-34.
36. Baruah P, Propato A, Dumitriu IE, Rovere-Querini P, Russo V, Fontana R, et al. The pattern recognition receptor PTX3 is recruited at the synapse between dying and dendritic cells, and edits the cross-presentation of self, viral, and tumour antigens. Blood 2006;107:151-8.
37. Mantovani A, Garlanda C, Bottazzi B. Pentraxin 3, a non-redundant soluble pattern recognition receptor involved in innate immunity. Vaccine 2003;21:S43-7.
38. Aoki CA, Borchers AT, Li M, Flavell RA, Bowlus CL, Ansari AA, et al. Transforming growth factor beta (TGF-beta) and autoimmunity. Autoimmun Rev 2005;4:450-9.
39. Xu W, Roos A, Schlagwein N, Woltman AM, Daha MR, van Kooten C. IL-10-producing macrophages preferentially clear early apoptotic cells. Blood 2006;107:4930-7.
40. Wisniewski HG, Vilcek J. Cytokine-induced gene expression at the crossroads of innate immunity, inflammation and fertility: TSG-6 and PTX3/TSG-14. Cytokine Growth Factor Rev 2004; 15:129-46.
41. Mukhopadhyay D, Asari A, Rugg MS, Day AJ, Fülöp C. Specificity of the tumor necrosis factor-induced protein 6-mediated heavy chain transfer from inter-alpha-trypsin inhibitor to hyaluronan: implications for the assembly of the cumulus extracellular matrix. J Biol Chem 2004;279:11119-28.
42. Scarchilli LA, Camaioni A, Botazzi B, Negri V, Doni A, Deban L, et al. PTX3 interacts with inter-alpha-trypsin inhibitor: implications for hyaluronan organization and cumulus oophorus expansion. J Biol Chem 2007;282:30161-70.
43. Eisenbach M, Giojalas LC. Sperm guidance in mammals—an unpaved road to the egg. Nat Rev Mol Cell Biol 2006;7:276-85.
44. Jeannin P, Bottazzi B, Sironi M, Doni A, Rusnati M, Presta M, et al. Complexity and complementarity of outer membrane protein A recognition by cellular and humoral innate immunity receptors. Immunity 2005;22:551-60.
45. Gaziano R, Bozza S, Bellocchio S, Perruccio K, Montagnoli C, Pitzurra L, et al. Anti-aspergillus fumigatus efficacy of pentraxin 3 alone and in combination with antifungals. Antimicrob Agents Chemother 2004;48:4414-21.
46. Diniz SN, Nomizo R, Cisalpino PS, Teixeira MM, Brown GD, Mantovani A, et al. PTX3 function as an opsonin for the dectin-1-dependent internalization of zymosan by macrophages. J Leukoc Biol 2004;75:649-56.
47. Vouret-craviari V, Matteucci C, Peri G, Poli G, Introna M, Mantovani A. Expression of a long pentraxin. PTX3, by monocytes exposed to the mycobacterial cell wall component lipoarabinomannan. Infect Immun 1997;65:1345-50.
48. Azzurri A, Sow OY, Amedei A, Bah B, Diallo S, Peri G, et al. IFN-gamma-inducible protein 10 and pentraxin 3 plasma levels are tools for monitoring inflammation and disease activity in Mycobacterium tuberculosis infection. Microbes Infect 2005; 7:1-8.
49. Olesen R, Wejse C, Velez DR, Bisseye C, Sodemann M, Aaby P, et al. DC-SIGN (CD209). Pentraxin 3 and vitamin D receptor gene variants associate with pulmonary tuberculosis risk in west. Africans Genes Immun 2007;8:456-67.
50. Kravitz MS, Shoenfeld Y. Autoimmunity to protective molecules: is it the perpetuum mobile (vicious cycle) of autoimmune rheumatic diseases? Nat Clin Pract Rheumatol 2006;2:481-90.
51. Hughes AL. Protein phylogenies provide evidence of a radical discontinuity between arthropod and vertebrate immune systems. Immunogenetics 1998;47:283-96.
52. Shrive AK, Metcalfe AM, Cartwright JR, Greenhough TJ. C-reactive protein and SAP like pentraxin are both present in Limulus polyphemus haemolymph: crystal structure of Limulus SAP. J Mol Biol 1999;290:997-1008.
53. Lee TH, Lee GW, Ziff EB, Vilcek J. Isolation and characterization of eight tumor necrosis factor-induced gene sequences from human fibroblast. Mol Cell Biol 1990;10:1982-8.
54. Basile A, Sica A, d'Aniello E, Breviario F, Garrido G, Castellano M, et al. Characterization of the promoter for the human long

pentraxin PTX3. Role of NF kappa B in tumour necrosis factor-alpha and interleukin-1beta regulation. J Biol Chem 1997;272:8172-8.
55. Lange LA, Carlson CS, Hindorff LA, Lange EM, Walston J, Durda JP, et al. Association of polymorphisms in the CRP gene with circulating C-reactive protein levels and cardiovascular events. JAMA 2006;296:2703-11.
56. Vogt B, Führnrohr B, Müller R, Sheriff A. CRP and the disposal of dying cells: consequences for systemic lupus erythematosus and rheumatoid arthritis. Autoimmunity 2007;40:295-8.
57. Russell AI, Cunninghame Graham DS, Shepherd C, Roberton CA, Whittaker J, Meeks J, et al. Polymorphism at the C-reactive protein locus influences gene expression and predisposes to systemic lupus erythematosus. Hum Mol Genet 2004;13:137-47.
58. Gaipl US, Kuhn A, Sheriff A, Munoz LE, Franz S, Voll RE, et al. Clearance of apoptotic cells in human SLE. Curr Dir Autoimmun 2006;9:173-87.
59. Bansal S, Ridker PM. Comparison of characteristics of future myocardial infarctions in women with baseline high versus baseline low levels of high-sensitivity C reactive protein. Am J Cardiol 2007;99:1500-3.
60. Salustri A, Garlanda C, Hirsch E, De Acetis M, Maccagno A, Bottazzi B, et al. PTX3 plays a key role in the organization of the cumulus oophorus extracellular matrix and in vivo fertilization. Development 2004;131:1577-86.
61. Doni A, Peri G, Chieppa M, Allavena P, Pasqualini F, Vago L, et al. Production of the soluble pattern recognition receptor PTX3 by myeloid, but not plasmacytoid, dendritic cells. Eur J Immunol 2003;33:2886-93.
62. Shelton RC, Liang S, Liang P, Chakrabarti A, Manier DH, Sulser F. Differential expression of pentraxin 3 in fibroblasts from patients with major depression. Neuropsychopharmacology 2004;29:126-32.
63. Nauta AJ, de Haij S, Bottazzi B, Mantovani A, Borrias MC, Aten J, et al. Human renal epithelial cells produce the long pentraxin PTX3. Kidney Int 2005;67:543-53.
64. Doni A, Michela M, Bottazzi B, Peri G, Valentino S, Polentarutti N, et al. Regulation of PTX3, a key component of humoral innate immunity in human dendritic cells: stimulation by IL-10 and inhibition by IFN-gamma. J Leukoc Biol 2006;79:797-802.
65. Szucs G, Timar O, Szekanecz Z, Der H, Kerekes G, Szamosi S, et al. Endothelial dysfunction precedes atherosclerosis in systemic sclerosis, relevance for prevention of vascular complications. Rheumatology (Oxford) 2007;46:759-62.
66. Polentarutti N, Picardi G, Basile A, Cenzuales S, Rivolta A, Matteucci C, et al. Interferon-gamma inhibits expression of the long pentraxin PTX3 in human monocytes. Eur J Immunol 1998;28:496-501.
67. Du Clos TW, Mold C. C-reactive protein: an activator of innate immunity and a modulator of adaptive immunity. Immunol Res 2004;30:261-77.
68. Polentarutti N, Bottazzi B, Di Santo E, Blasi E, Agnello D, Ghezzi P, et al. Inducible expression of the long pentraxin PTX3 in the central nervous system. J Neuroimmunol 2000;106: 87-94.
69. Hossain MA, Russell JC, O'Brien R, Laterra J. Neuronal pentraxin 1: a novel mediator of hypoxic-ischemic injury in neonatal brain. J Neurosci 2004;24:4187-96.
70. Ravizza T, Moneta D, Bottazzi B, Peri G, Garlanda C, Hirsch E, et al. Dynamic induction of the long pentraxin PTX3 in the CNS after limbic seizures: evidence for a protective role in seizure-induced neurodegeneration. Neuroscience 2001;105: 43-53.
71. Napoleone E, Di Santo A, Bastone A, Peri G, Mantovani A, de Gaetano G, et al. Long pentraxin PTX3 up regulates tissue factor expression in human endothelial cells: a novel link between vascular inflammation and clotting activation. Arterioscler Thromb Vasc Biol 2002;22:782-7.
72. Napoleone E, Di Santo A, Peri G, Mantovani A, de Gaetano G, Donati MB, et al. The long pentraxin PTX3 up-regulates tissue factor in activated monocytes: another link between inflammation and clotting activation. J Leukoc Biol 2004;76:203-9.
73. Rolph MS, Zimmer S, Bottazzi B, Garlanda C, Mantovani A, Hansson GK. Production of the long pentraxin PTX3 in advanced atherosclerotic plaques. Arterioscler Thromb Vasc Biol 2002;22:10-4.
74. Soares AC, Souza DG, Pinho V, Vieira AT, Nicoli JR, Cunha FQ, et al. Dual function of the long pentraxin PTX3 in resistance against pulmonary infection with Klebsiella pneumoniae in transgenic mice. Microbes Infect 2006;8:1321-9.
75. Sherer Y, Shoenfeld Y. Atherosclerosis. Ann Rheum Dis 2002; 61:97-9.
76. Doria A, Sherer Y, Meroni PL, Shoenfeld Y. Inflammation and accelerated atherosclerosis: basic mechanisms. Rheum Dis Clin North Am 2005;31:355-62.
77. George J, Afek A, Gilburd B, Harats D, Shoenfeld Y. Autoimmunity in atherosclerosis: lessons from experimental models. Lupus 2000;9:223-7.
78. Hahn BH, Grossman J, Chen W, McMahon M. The pathogenesis of atherosclerosis in autoimmune rheumatic diseases: roles of inflammation and dyslipidemia. J Autoimmun 2007;28:69-75.
79. Manzi S, Meilahn EN, Rairie JE, Conte CG, Medsger TA, Jansen-McWilliams L, et al. Age-specific incidence rates of myocardial infarction and angina in women with systemic lupus erythematosus: comparison with the Framingham Study. Am J Epidemiol 1997;145:408-15.
80. Tabuchi M, Inoue K, Usui-Kataoka H, Kobayashi K, Teramoto M, Takasugi K, et al. The association of C-reactive protein with an oxidative metabolite of LDL and its implication in atherosclerosis. J Lipid Res 2007;48:768-81.
81. Klouche M, Peri G, Knabbe C, Eckstein HH, Schmid FX, Schmitz G, et al. Modified atherogenic lipoproteins induce expression of pentraxin-3 by human vascular smooth muscle cells. Atherosclerosis 2004;175:221-8.
82. Bosutti A, Grassi G, Zanetti M, Aleksova A, Zecchin M, Sinagra G, et al. Relation between the plasma levels of LDL-cholesterol and the expression of the early marker of inflammation long pentraxin PTX3 and the stress response gene p66ShcA in pacemaker-implanted patients. Clin Exp Med 2007;7:16-23.
83. Per G, Introna M, Corradi D, Iacuitti G, Signorini S, Avanzini F, et al. PTX3. A prototypical long pentraxin, is an early indicator of acute myocardial infarction in humans. Circulation 2000; 102:636-41.
84. Muller B, Peri G, Doni A, Torri V, Landmann R, Bottazzi B, et al. Circulating levels of the long pentraxin PTX3 correlate with severity of infection in critically ill patients. Crit Care Med 2001; 29:1404-7.
85. Luchetti MM, Piccinini G, Mantovani A, Peri G, Matteucci C, Pomponio G, et al. Expression and production of the long pentraxin PTX3 in rheumatoid arthritis (RA). Clin Exp Immunol 2000;119:196-202.
86. Bevelacqua V, Libra M, Mazzarino MC, Gangemi P, Nicotra G, Curatolo S, et al. Long pentraxin 3: a marker of inflammation in untreated psoriatic patients. Int J Mol Med 2006; 18:415-23.
87. Luchetti MM, Sambo P, Majlingová P, Svegliati Baroni S, Peri G, Paroncini P, et al. Scleroderma fibroblasts constitutively express the long pentraxin PTX3. Clin Exp Rheumatol 2004;22: S66-72.
88. Van Rossum AP, Pas HH, Fazzini F, Huitema MG, Limburg PC, Jonkman MF, et al. Abundance of the long pentraxin PTX3 at sites of leukocytoclastic lesions in patients with small-vessel vasculitis. Arthritis Rheum 2006;54:986-91.
89. Molloy ES, Langford CA. Advances in the treatment of small vessel vasculitis. Rheum Dis Clin North Am 2006;32: 157-72.

90. Bussolati B, Peri G, Salvidio G, Verzola D, Mantovani A, Camussi G. The long pentraxin PTX3 is synthesized in IgA glomerulonephritis and activates mesangial cells. J Immunol 2003; 170:1466-72.
91. Chen KR, Sakamoto M, Ikemoto K, Abe R, Shimizu H. Granulomatous arteritis in cutaneous lesions of Churg-Strauss syndrome. J Cutan Pathol 2007;34:330-7.
92. van Rossum AP, Limburg PC, Kallenberg CG. Activation, apoptosis, and clearance of neutrophils in Wegener's granulomatosis. Ann NY Acad Sci 2005;1051:1-11.
93. Solomon HD, Karlson WE, Rimm BE, Cannuscio CC, Mandl AL, Manson JE, et al. Cardiovascular morbidity and mortality in women diagnosed with rheumatoid arthritis. Circulation 2003; 107:1303-7.
94. Torres JL, Ridker PM. Clinical use of high sensitivity C-reactive protein for the prediction of adverse cardiovascular events. Curr Opin Cardiol 2003;18:471-8.
95. Enzenauer RJ, Collier DH, Lopez LR. Anticardiolipin antibodies in scleroderma. J Clin Rheumatol 2006;12:324-6.
96. Peng SL. Neutrophil apoptosis in autoimmunity. J Mol Med 2006;84:122-5.
97. Carlson JA, Chen KR. Cutaneous vasculitis update: small vessel neutrophilic vasculitis syndromes. Am J Dermatopathol 2006; 28:486-506.
98. Rovere-Querini P, Castiglioni MT, Sabbadini MG, Manfredi AA. Signals of cell death and tissue turnover during physiological pregnancy, pre-eclampsia, and autoimmunity. Autoimmunity 2007;40:290-4.
99. De Falco M, Penta R, Laforgia V, Cobellis L, De Luca A. Apoptosis and human placenta: expression of proteins belonging to different apoptotic pathways during pregnancy. J Exp Clin Cancer Res 2005;24:25-33.
100. Rovere-Querini P, Antonacci S, Dell'Antonio G, Angeli A, Almirante G, Cin ED, et al. Plasma and tissue expression of the long pentraxin 3 during normal pregnancy and preclampsy. Obstet Gynecol 2006;108:148-55.
101. Cetin I, Cozzi V, Pasqualini F, Nebuloni M, Garlanda C, Vago L, et al. Elevated maternal levels of the long pentraxin 3 (PTX3) in preclampsy and intrauterine growth restriction. Am J Obstet Gynecol 2006;194:1347-53.
102. Brull DJ, Serrano N, Zito F, Jones L, Montgomery HE, Rumley A, et al. Human CRP gene polymorphism influences CRP levels: implications for the prediction and pathogenesis of coronary heart disease. Arterioscler Thromb Vasc Biol 2003; 23:2063-9.
103. Toloza SM, Uribe AG, McGwin G, Alarcon GS, Fessler BJ, Bastian HM, et al. Systemic lupus erythematosus in a multiethnic US cohort (LUMINA). XXIII. Baseline predictors of vascular events. Arthritis Rheum 2004;50:3947-57.
104. Farzaneh-Far A, Roman MJ. Accelerated atherosclerosis in rheumatoid arthritis and systemic lupus erythematosus. Int J Clin Pract 2005;59:823-4.
105. Arici M, Walls J. End-stage renal disease, atherosclerosis, and cardiovascular mortality: is C-reactive protein the missing link? Kidney Int 2001;59:407-14.
106. Van Doornum S, McColl G, Wicks IP. Accelerated atherosclerosis: an extra-articular feature of rheumatoid arthritis? Arthritis Rheum 2002;46:862-73.
107. Ridker PM. High-sensitivity C-reactive protein: potential adjunct for global risk assessment in the primary prevention of cardiovascular disease. Circulation 2001;103:1813-18.
108. Ridker PM, Hennekens CH, Buring JE, Rifai N. C-reactive protein and other markers of inflammation in the prediction of cardiovascular disease in women. N Engl J Med 2000;342:836-43.
109. Sakkinen P, Abbott RD, Curb JD, Rodriguez BL, Yano K, Tracy RP. C-reactive protein and myocardial infarction. J Clin Epidemiol 2002;55:445-51.
110. Morrow DA, Rifai N, Antman EM, Weiner DL, McCabe CH, Cannon CP, et al. C-reactive protein is a potent predictor of mortality independently and in combination with troponin T in acute coronary syndromes: a TIMI 11A substudy. Thrombolysis in Myocardial Infarction. J Am Coll Cardiol 1998;31:1460-5.