Review

The role of myeloperoxidase in the pathogenesis of systemic vasculitis

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ABSTRACT

Wegener's granulomatosis, microscopic polyangiitis, Churg-Strauss syndrome and idiopathic pauci-immune necrotizing crescentic glomerulonephritis are strongly associated with the presence of anti-neutrophil cytoplasmic antibodies (ANCA). These ANCA-associated vasculitides can serologically be separated into myeloperoxidase (MPO)-ANCA and proteinase 3 (PR3)-ANCA positive patients. The unique properties of the antigen targeted by the anti-MPO antibodies could help to explain the specific characteristics of MPO-ANCA associated disease. Recently, an animal model has been developed that proves that anti-mouse MPO immunoglobulins alone are capable of causing disease similar to that in humans. Also, the in vitro *pathologic effects of binding of MPO-ANCA to MPO are better understood.*

MPO-ANCA can activate (primed) neutrophils directly causing extensive reactive oxygen species formation and degranulation of neutrophil constituents, including MPO, resulting in a destructive inflammatory response towards the vessel wall. MPO-ANCA can prevent the clearing and inactivation of MPO by ceruloplasmin as well, resulting in increased myeloperoxidase activity. Myeloperoxidase produces not only the strong oxidant bleach (hypochlorous acid) out of hydrogen peroxide and chloride ions but also oxidizes LDL into a macrophage high-uptake form, inactivates protease inhibitors, and consumes nitric oxide. These may contribute to endothelial dysfunction and add to the chronic renal lesions observed in patients with MPO-ANCA. MPO levels are influenced by genetic factors including two, MPO463 and MPO129, single nucleotide polymorphisms. The MPO 463 polymorphism has been associated with an increased risk of development of MPO-ANCA associated disease.

Introduction

Vasculitis is an inflammatory process of blood vessels resulting in damage and eventually destruction and occlusion of vessels. Vasculitis may be primary or secondary to underlying connective tissue disease, infection, malignancy, or drugs. Clinical presentation largely depends on the type and size of vessels involved. Primary vasculitides can be subdivided according to the 1993 Chapel Hill Consensus Conference definitions into large vessel vasculitides, medium-sized vessel vasculitides, small vessel vasculitides not associated with anti-neutrophil cytoplasmic antibodies (ANCA), and small vessel vasculitides associated with ANCA(1, 2).

The ANCA associated small vessel vasculitides can be separated into Wegener's granulomatosis (WG), microscopic polyangiitis (MPA), Churg-Strauss syndrome (CSS) and idiopathic pauci-immune necrotizing crescentic glomerulonephritis (iNCGN) (Table I). Generally, ANCA detected in patients with these small vessel vasculitides are either directed against the myeloid lysosomal enzymes proteinase 3 (PR3) or myeloperoxidase (MPO) (3). There is increasing awareness that MPO-ANCA and PR3-ANCA positive patients represent separate diseases within the spectrum of the ANCA associated vasculitides (4). The reason why MPO-ANCA positive patients develop a different type of disease is largely unknown. The unique properties of the enzyme targeted by anti-MPO antibodies could perhaps help to explain the specific characteristics of MPO-ANCA associated disease.

This review will focus on the clinical characteristics of MPO-ANCA associated vasculitis and will discuss recently developed animal models. Also, the role and function of the enzyme will be discussed as well as promoter polymorphisms that influence MPO expression levels and disease risk.

Clinical characteristics of MPO-ANCA associated vasculitis

The ANCA associated vasculitides are characterized by a necrotizing vasculitis of the smaller vessels and as such can give rise to arteriolitis, capillaritis, venulitis and glomerulonephritis. However, larger vessels can sometimes participate in the disease process as well. According to the 1993 Chapel Hill Consensus Conference, the types of tissue affected, the presence or absence of granulomatous inflammation and the presence or absence of asthma and eosinophilia distinguishes between the different ANCAassociated vasculitides (Table I). The commonly used generalized term 'ANCA-associated vasculitides' for these diseases fails to distinguish the differences between patients with PR3-ANCA positivity and those with MPO-ANCA(Table II).

Studies looking at disease associations between patients with PR3-ANCA and MPO-ANCAhave shown that most patients with WG have PR3-ANCA and most iNCGN as well as CSS patients have MPO-ANCA. However, in MPA positivity is more evenly distributed (Table I) (5-13).

Renal disease is a hallmark of ANCA associated disease, most studies report renal involvement between 75-90% of patients (5,7,10,14). For CSS reported percentages are somewhat lower, between 26 and 68% of patients (15, 16). The prevalence of renal disease generally does not differ between MPO- and PR3-ANCApositive patients (4). Also, the type of histopathological lesions is essentially the same and consist of fibrinoid necrosis of the capillary wall with extracapillary proliferation and crescent formation. However, Franssen *et al*. found that at the time of diagnosis, anti-MPO positive patients have less acute renal lesions and more chronic lesions (17). Recently, these findings were confirmed by Hauer *et al*. in a large cohort of 173 patients with ANCA associated renal disease. MPO-ANCA compared to PR3-ANCA positive patients had the same occurrence of fibrinoid necrosis and crescents, but more glomerulosclerosis (25% versus 15%, respectively), more interstitial fibrosis, more tubular atrophy, and more arteriosclerosis (18). These findings suggest a different pathogenesis of renal disease in MPO-ANCA positive patients compared to PR3-ANCA patients. In line with these histopathological differences in renal lesions, pretreatment deterioration of renal function is slower in MPO-ANCA patients compared to PR3-ANCA positive patients (17). Since at the time of diagnosis serum creatinine levels are the same in patients with MPO-ANCA associated glomerulonephritis (17, 19), we think that renal lesions in MPO-ANCA patients develop more slowly, and could therefore result in delayed diagnosis of MPO- versus PR3-ANCApositive patients. This, in turn, results in

Table I. The ANCA-associated small vessel vasculitides.

Disease	Pathologic definition	Clinical hallmarks	MPO-ANCA	PR3-ANCA	Ref.
Idiopathic pauci-immune necrotizing crescentic glomerulonephritis (iNCGN)	Necrotizing crescentic glomerulonephritis, with few or no immune deposits. Absence of small vessel vasculitis in other organs.	Nephritis, malaise	70-80%	10-30%	(6, 24)
Microscopic polyangiitis (MPA)*	Necrotizing vasculitis affecting small vessels with absence of granulomatous inflammation of the respiratory tract.	Nephritis, purpura, hemoptysis	50- 80%	20-50%	(5, 7, 69-71)
Churg-Strauss syndrome (CSS)*	Eosinophil-rich and granulomatous inflammation involving the respiratory tract. Necrotizing vasculitis affecting small to medium-sized vessels.	Asthma, eosinophilia, neuropathy	50-80%	3-10%	(3, 8, 16, 72)
Wegener's granulomatosis (WG)*	Granulomatous inflammation involving the respiratory tract. Necrotizing vasculitis affecting small to medium-sized vessels.	Nose bleeds, nephritis, lung infiltrates, hemoptysis	10-30%	70-90%	(73-76)

* (Necrotizing) arteritis involving medium-sized or larger arteries may be occasionally found (1, 4, 7). PR3-ANCA: anti-proteinase 3 neutrophil cytoplasmic antibodies; MPO-ANCA: anti-myeloperoxidase neutrophil cytoplasmic antibodies.

Table II. Differences between MPO-ANCAassociated disease and PR3-ANCA.

	MPO-ANCA	PR3-ANCA
Disease association (inorderofprevalence)	iNCGN, CSS, MPAand WG *For the relative frequency of encountered ANCA, see Table I*	WG, MPA, iNCGN and CSS
Age	Older age (57-63)	Younger age (45-56)
Extra-renal organ involvement	Less involved (average 2.2)	More involvement (average 3.9)
Upper respiratory 21 tract disease	Less frequent, if present mild, characterized by rhinitis and polyps	Frequently present, characterized by destructive and granulomatous inflammation
Granuloma	Rare, with the exception of CSS	Granulomas hallmark of disease
Renal characteristics	Pre-treatment renal deterioration rate lower More chronic lesions in kidney biopsy Renal outcome depends on persistent high MPO-ANCAtitres and proteinuria at diagnosis and during follow-up	Pre-treatment renal deterioration rate higher More acute lesions in kidney biopsy Renal outcome associated with renal relapses
Relapse	Lower relapse rate	Higher relapse rate

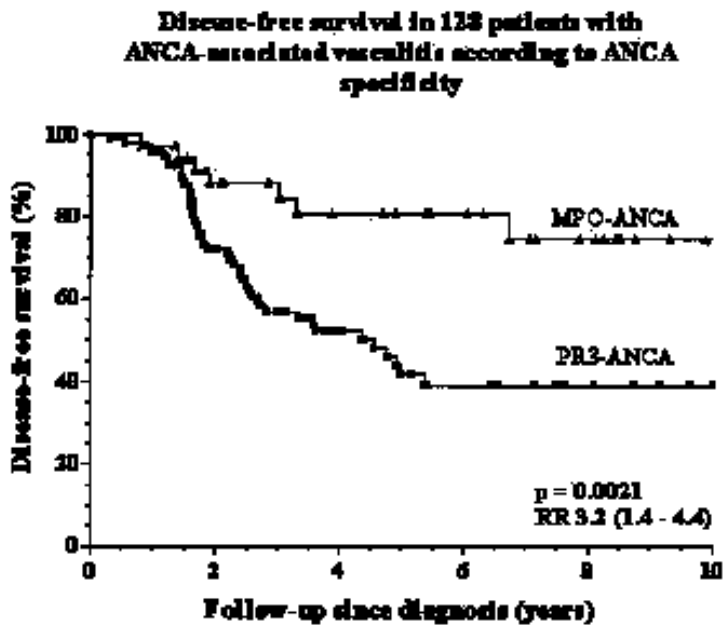

Fig. 1. Disease-free survival (%) in 128 patients with ANCApositive vasculitis according to ANCA specificity (PR3-ANCAn = 93; MPO-ANCAn = 35).

increased chronic lesions in MPO-ANCApatients.

Also, despite good initial response to treatment progressive deterioration of renal function leading to end-stage renal disease has been observed in a substantial proportion of patients with MPO-ANCApositivity, but is extremely infrequent in patients with PR3-ANCA (7, 20). Risk factors for decrease of renal function in patients with MPO-ANCA are proteinuria at diagnosis and during follow-up as well as persistent high levels of MPO-ANCA (21). This is contrary to the risk factors found in PR3-ANCA patients, where long term renal survival was determined by renal relapses during follow-up (20). The presence of hypertension was not found to be an independent risk factor for renal function deterioration in both MPO and PR3-ANCA positive patients (20, 21). Progressive deterioration of renal function without relapses, as found in the MPO-ANCA patients, is rarely observed in PR3-ANCA associated glomerulonephritis (20). Moreover, relapses are more frequently observed in patients with PR3-ANCA as compared to MPO-ANCA(Fig.1) (22). In most PR3-ANCA positive patients the upper respiratory tract is characterized by an aggressive, destructive inflammatory process, whilst in MPO-ANCA positive patients upper respiratory tract involvement is usually much milder and characterized by nasal polyps, rhinitis and/or sinusitis. Except for patients with MPO-ANCA and CSS, granuloma are almost exclusively found in PR3-ANCApatients (10,11,23,24).

Next to these differences, MPO-ANCA positive patients are older at the time of diagnosis (17,25) and generally have fewer extrarenal organs involved in the disease process than PR3-ANCA patients (7,10, 11,25). The frequency of pulmonary disease on the other hand, is equal in both patient groups. However, MPO-ANCA patients less frequently have nodular and/or pneumonia-like lesions typical for PR3-ANCA patients on their chest x-rays and will have more patchy lesions consistent with pulmonary hemorrhage (5,6,12). The absence of nodular lesions is probably due to the absence of granulomatous inflammation in most of the MPO-ANCA patients.

This review will further discuss the evidence for the pathogenecity of MPO-ANCA focusing on a recently developed animal model. For a pathogenetic comparison between MPO-ANCA and PR3-ANCA associated vasculitis we would like to refer to another review article (4). We will also discuss the functions of the enzyme-antigen MPO, as well as promoter polymorphisms that influence MPO expression and disease risk.

MPO-ANCA: Animal models

Animal models for MPO-ANCA associated vasculitis have been developed to prove that MPO-ANCA are pathogenic *in vivo* as well as to address specific questions about pathogenic mechanisms and to study the effects of novel therapies *in vivo*. Existing animal models are summarized in Table III. The two recent models are one developed by Smyth *et al*. and one by Xiao *et al*. In preliminary studies, Smyth *et al*. reported that in Wistar Kyoto rats immunized with human MPO, cross-reacting rat anti-rat MPO antibodies develop accompanied by pauci-immune crescentic glomerulonephritis and alveolar hemorrhage (26). The investigators suggested that the rat strain used is most likely to be very important since other groups have failed to induce disease after immunization with human MPO in different strains of rat. It will be interesting to genetically compare these different rat strains to investigate disease specific genes.

Xiao *et al.* made use of the recently generated MPO knockout mouse. Since these mice lack MPO, no immunological tolerance for this enzyme has developed. After immunization in this otherwise immunocompetent mouse a strong humoral and cellular response will develop. Xiao *et al.* showed that $Rag2^{-/-}$ (T, B cell deficient) mice injected with anti-MPO splenocytes developed circulating MPO-ANCA and a dose-dependent deterioration of renal function, histologically characterized by severe necrotizing and crescentic glomerulonephritis (27). Interestingly, 7 of 16 injected animals developed extrarenal vasculitis, including pulmonary alveolar capillaritis and granulomatous inflammation was present in only 1 of 16 mice.

Also, passive transfer of MPO-ANCA IgG was capable of transferring disease. Within 6 days after injection of the antibodies into MPO sufficient mice, focal necrotizing glomerulone-

Table III. Animal models for MPO-ANCAassociated disease.

Animal model	Animals used	Characteristics	Comment	MPO-ANCA	Ref.
Mercuric chloride exposure	Brown Norway	Necrotizing vasculitis of the small to medium sized vessels accompanied by fibrinoid necrosis of the vessel wall	Mostly Tcell dependent disease, no disease by serum transfer	Spontaneous	(77)
Immunization	Brown Norway	Necrotizing vasculitis. Organ distribution depends on the site of the neutrophil extract injection	Only disease induction after additional perfusion with products of activated neutrophils, including MPO and H_2O_2	After immunization with human MPO	(78)
Passive transfer with rabbit anti-rat MPO and GBM		Glomerulonephritis with influx of neutrophils and fibrin deposition	The presence of rabbit anti-rat MPO aggravates rabbit anti-rat GBM disease	Rabbit-anti rat MPO	(79)
Active immunization with human MPO + passive transfer with sub-nephritogenic anti-GBM antibodies	Brown Norway	Severe glomerulonephritis with fibrinoid necrosis, extensive fibrin deposition, influx of monocytes/macrophages and crescent formation		After immunization with human MPO, crossreacting with rat MPO	(80)
Direct immunization with human MPO	Wistar Kyoto rat	Small vessel vasculitis with pauci-immune focal segmental glomerulonephritis and alveolar hemorrhage			(26)
Passive transfer with mouse anti-mouse MPO	C57BL/6J mice	Focal necrotizing and crescentic glomerulonephritis with a paucityof glomerular Ig deposition	Production of mouse anti-mouse MPO by use of MPO knockout mouse	Mouse-MPO-ANCA	(27)

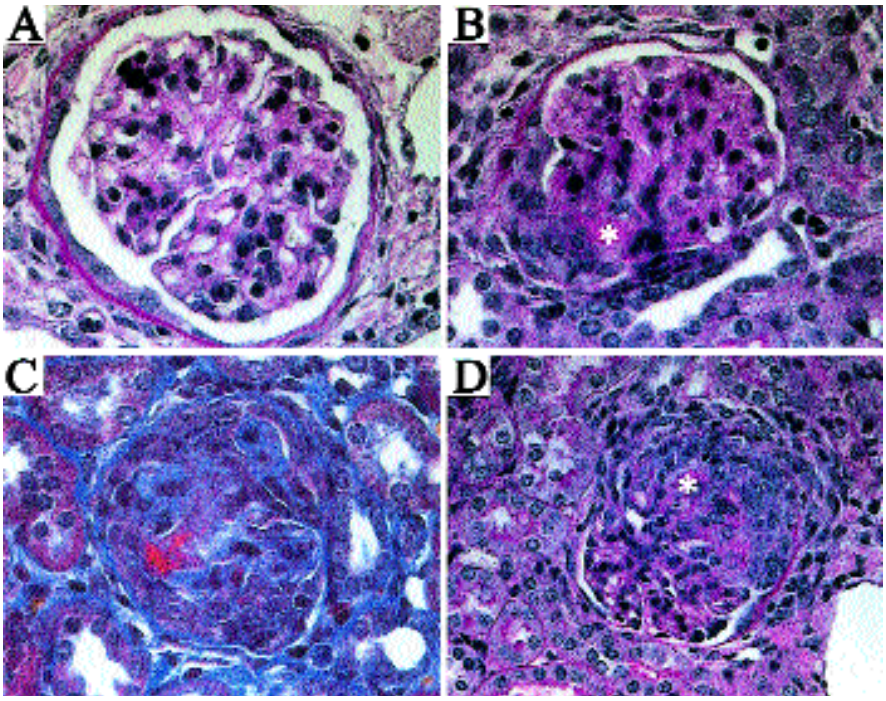

Fig. 2. The development of necrotizing crescentic glomerulonephritis in a wildtype C57BL/6j mouse by passive transfer with MPO-ANCA. MPO-ANCA were produced by immunization of a MPO knock-out mouse with mouse MPO. This figure shows 4 glomeruli cut from the same kidney of a mouse 6 days after injection with MPO-ANCA. (**A**) shows a normal glomerulus, (**B**) shows fibrinoid necrosis (*****) and extracapillary proliferation, (**C**) shows fibrinoid necrosis (**red**) and crescent formation, (**D**) shows fibrinoid necrosis (*****) surrounded by a large crescent with inflammatory involvement of Bowman's capsule. (**A**), (**B**) and (**D**) are PAS stained sections, (**C**) is a Martius Scarlet Blue stained section.

phritis developed in all injected mice, although the number of glomeruli affected was much lower than in the splenocytes transfer. The histopathological similarities to the human form of MPO-ANCA disease is striking. In collaboration with Xiao *et al*. we could reproduce this model of murine anti-MPO associated glomerulonephritis as well (Fig. 2). This disease model offers unique opportunities to study pathogenic mechanisms and to test novel therapies. Whether it will be possible to mimic aspects of the chronic disease like in humans remains to be seen.

MPO-ANCApathologic mechanisms

Although it still remains to be determined what the exact pathological mechanisms are, the experimental animal models described above strongly support the hypothesis that MPO-ANCA directly cause disease. Many *in vitro* studies have tried to unravel the pathologic mechanisms involved in MPO-ANCA associated disease [reviewed in (28)]. In short, priming of neutrophils by TNFα (e.g. as a result of an upper respiratory tract infection) results in mitogen-activated protein kinase

(MAPK) dependent translocation of MPO to the cell surface (29). Stimulation of these primed neutrophils with MPO-ANCA results in neutrophil activation, with reactive oxygen species generation (including O_2^- and H_2O_2), degranulation, adhesion to and killing of endothelial cells (28). Also, degranulated MPO can bind to unprimed neutrophils making them directly vulnerable to activation by MPO-ANCA (30). Monocytes can be activated as well, resulting in enhanced cytokine and chemokine production (e.g. IL-8, Il-6, TNF and MCP-1) (28). Most of these processes are thought to be Fc and Fcγ-receptor dependent (28).

Circulating MPO has been detected in patients with MPO-ANCA associated vasculitis in higher concentrations than in healthy controls (31). Endothelial cells can bind and take up this degranulated MPO resulting in an increased intracellular oxidant radical formation (32). MPO can also form complexes with circulating MPO-ANCA that bind and dimerize Fcγ-receptors on neutrophils resulting in activation (28). MPO present outside inflammatory sites is normally cleared and inactivated by ceruloplasmin (33). The presence of MPO-ANCA however, interferes with these mechanisms, leaving a circulating highly reactive enzyme (34). The potential pathologic significance of this circulating functional enzyme will be discussed in the next paragraph.

Functions of MPO and pathophysiological implications for MPO-ANCA associated vasculitis

Myeloperoxidase (MPO; EC 1.11.1.7) is a heme containing peroxidase expressed and stored in monocyte and neutrophil granulocyte precursors. In mature resting neutrophils, MPO is the most abundant protein and upon neutrophil activation MPO is readily released into phagocytic vacuoles as well in the extracellular space where it catalyses the formation of bleach (hypochlorous acid; HOCl) out of chloride ions and hydrogen peroxide (H_2O_2). As summarized in Table IV and Figure 3 the effects and pathological consequences of MPO are multiple. Firstly, HOCl can react with a variety of cellular substrates including thiols, nucleotides and amines and, as such, it contributes to innate immunity against bacteria and fungi by neutrophils (35). HOCl can however, also damage host tissue and catalytically active MPO has been detected in glomerular and atherosclerotic lesions in conjunction with HOCL-modified proteins (36, 37). HOCl is directly cytotoxic in high concentrations, whereas in lower concentrations it is able to activate MAPK pathways and can cause growth arrest and apoptosis (38). Apoptosis of neutrophils could contribute to the presentation of the normally concealed MPO to the immune system (39). Moreover, exposure to large numbersof apoptotic cells, exceeding the phagocytic capacity of the reticuloendothelial system to clear apoptotic material, is able to elicit an autoantibodyresponse (40).

Secondly, in addition to the conventional view on the role of the MPO/HOCl system recent evidence suggests that MPO has an important role in the oxidative conversion of LDL into a high-uptake form for macrophages, leading to foam cell formation (41). In this respect, Swets *et al*. observed that patients with ANCA associated vasculitis had increased levels of autoantibodies against oxidized LDL, suggesting that patients were more susceptible to oxidation of LDL(42).

Thirdly, MPO can increase protease activity at inflammatory sites by inactivating protease inhibitors and by acting as a detoxicant for H_2O_2 thereby protecting proteolytic enzymes from inactivation (43-45). In ANCA associated vasculitis protease/anti-protease imbalance has been proposed to play a role in vasculitic lesion development (46).

Finally, MPO can act as a leukocyte-derived vascular nitric oxide (NO) oxidase, thereby reducing NO bioavailability and contributing to endothelial dysfunction (47-49). Endothelial dysfunction has been shown to be an important characteristic of ANCAassociated disease, especially during active disease (50, 51). Interestingly, chronic blockade of NO synthesis in rats results in hypertension, proteinuria, glomerular sclerotic injury, tubulointersitial da-

Table IV. The effects and pathological consequences of active myeloperoxidase (MPO).

Function	Effect	Pathologic significance
Production of HOCl (bleach)	Bactericidal (35)	Aids in clearing bacteria
	Activate pro-MMP(81) MAPK activation (38)	Contributes to matrix degradation Contributes to neutrophil membrane expression of MPO (29)
	Inhibition of Tcell proliferation (82) Depending on concentration pro-apoptotic or cytotoxic	– Apoptotic overload may cause antigen presentation and auto-antibody production (40) Apoptosis of endothelial cells (83)
Consumption of H_2O_2	Prevent protease inactivation (43, 44)	Contributes to protease mediated vascular damage
Inactivate protease inhibitors	Prevent protease inactivation (45, 84)	Contributes to protease mediated vascular damage
Oxidation	Oxidize LDL(41), MPO-ANCApatients have high anti-oxLDLtiters (42)	Pro-apoptotic, stimulates monocyte-endothelial cell interactions, and stimulates proliferation of smooth muscle cells and monocytes/macrophages (85)
Consume NO (47)	Lower NO bioavailability at inflammatory sites	Contributes to endothelial dysfunction (50, 51). Promotes renal injury e.g. through pathogenic properties of unopposed normal angiotensin II levels (53)

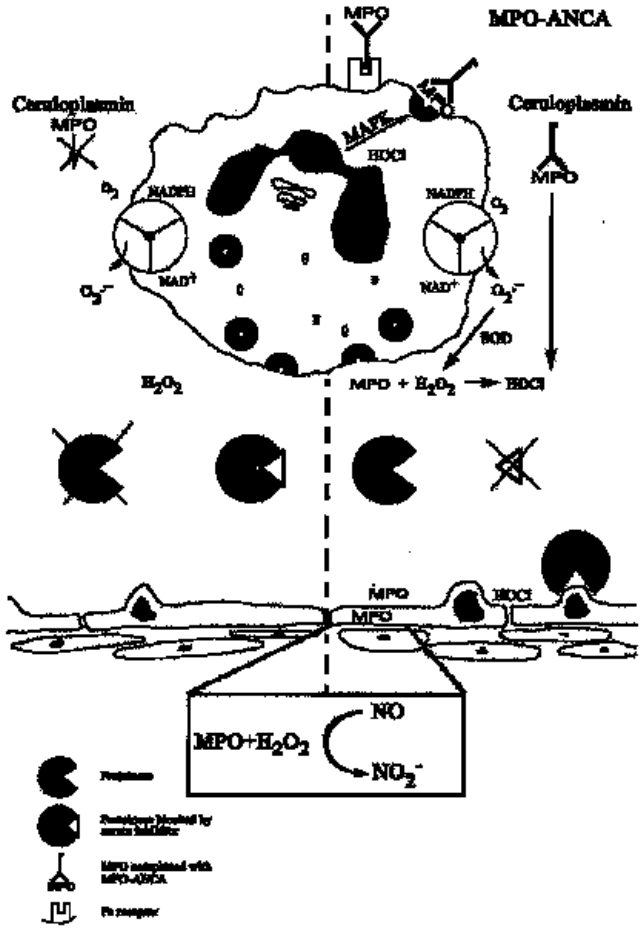

Fig. 3. Schematic presentation of the functions of MPO. Left-side of the figure represents the situation without ANCA, right-side figure represents the situation where MPO-ANCAcan activate primed neutrophils directly or via the Fcγ receptor and prevent inhibition of MPO by ceruloplasmin. See also Table IV.

mage and infiltration of the interstitium by mononuclear cells (52,53). Moreover, in glomerulonephritis it has been shown that NO synthase is reduced and that mice lacking a functional gene for endothelial NO synthase (eNOS) develop more severe glomerulonephritis than wildtypes (54, 55).

Levels of MPO expression

Increased neutrophil MPO content has been linked to coronary atherosclerosis suggesting a link between MPO protein levels and atherosclerosis (48). Also, in patients with MPO-ANCA associated vasculitis higher circulating levels of serum MPO have been found compared to control subjects (31). Higher levels of MPO could result in more antigen present for binding by MPO-ANCA and more active enzyme circulating uninhibited by ceruloplasmin due to binding of MPO-ANCA. In this respect two MPO promoter polymorphisms influencing MPO levels are of particular interest.

Expression of MPO during myelopoiesis is tightly regulated and multiple transcription factors are involved in transcription of MPO (56). Two genetic promoter polymorphisms have been shown to influence MPO expression levels, the MPO463 and MPO129 single nucleotide polymorphisms (Fig. 4). The most studied polymorphism is the -463 G/A nucleotide change within an Alu-encoded hormone response element. The GG genotype is the most common, at 61% of the Northern European population. The AA genotype occurs 3-7% in Caucasians and thus the GAin 32-36% (57).

The G allele contains a SP1 binding site associated with a stronger promoter activity. The A allele destroys this SP1 binding site, but creates an estrogen binding motif making the promoter responsive to estrogens (57). Reynolds *et al.* found that in 10 tested patients MPO protein level expression was higher for the GG than the GA or AA genotypes (58). In contrast, however, Hoy *et al*. were not able to demonstrate an association between MPO protein levels and the 463 G/A polymorphism (59).

Interesting is the association of the MPO463 polymorphism with an impaired vasodilatative response to the administration of nitroglycerine, a direct NO donor, in females (60). This implies that higher levels of MPO (i.e. GG genotype) correlate with impaired vasodilatation. The MPO463 polymorphism has been investigated in MPO-ANCA associated disease as well. Reynolds established that the MPO GG genotype was overrepresented in fe-

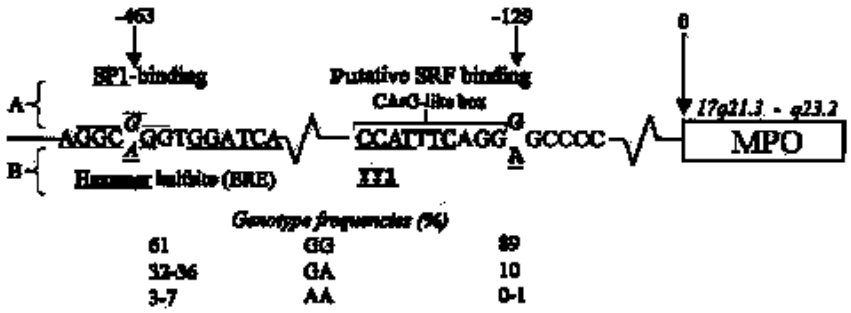

Fig. 4. Figure showing two important single nucleotide polymorphisms in the promoter of myeloperoxidase at position –463 and –129. (**A**) shows (putative) binding sites (black line on top of text) when both polymorphisms have their wild type genotype (i.e. G allele), (**B**) shows the (putative) binding sites (underlined) when both sites are polymorphic (i.e. A allele). Note the single nucleotide mismatch with the consensus binding site of yin yang-1 (YY1) transcription factor (dotted underlined) (86). ERE: Estrogen response element; SRF: serum response factor.

male, but not male, patients with MPO-ANCA associated vasculitis. Furthermore, we found that GA/AA genotypes were diagnosed at an earlier age compared to the GG genotype (41 versus 56 years, respectively). Interestingly, relapse-free survival is significantly different between patients with the 463-GG compared to those with the 463GA or AA genotype (Fig. 5). However, the latter finding was not confirmed in a recent abstract (61). Taken together, these data suggest that the high MPO expression genotype (GG) is a risk factor for development of disease, but once disease is present it is possibly associated with a lower relapse rate (62).

A second MPO promoter polymorphism (129G/A) has been described recently. This G to A single nucleotide polymorphism at position –129 has been associated with decreased levels of MPO in the serum of healthy controls. About 10% of people in a French population was heterozygous for this polymorphism (59). To date the 129GA polymorphism has been evaluated for disease in only one population of sarcoidosis patients (63) and it will be interesting to learn its role in vasculitic disease.

Thus, genetic factors influence MPO activity by controlling MPO expression. However, MPO activity can also be changed directly by other factors. Estrogens for instance, can have a stimulatory effect on the enzymatic activity of MPO (64-66), whereas, for example non-steroidal anti inflammatory drugs can inhibit MPO activity *in vitro* (67). Whether the latter has relevance *in vivo* is doubted (68).

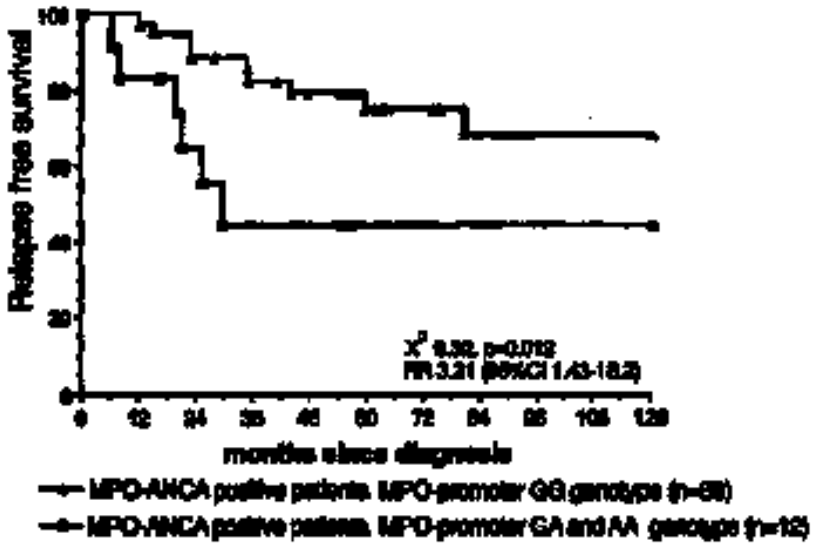

Fig. 5. Relapse-free survival is significantly different between patients with the 463GG (n = 38) genotype compared to 463 GAor AAgenotypes (n = 12) (p = 0.012). Reproduced from (62) with permission.

Conclusions

MPO-ANCA induces vasculitis and glomerulonephritis in mice. *In vitro*, MPO-ANCA can activate (primed) neutrophils directly causing extensive reactive oxygen species formation and degranulation of neutrophil constituents, including MPO, resulting in an aggravated immune response towards the vessel wall. MPO-ANCA can prevent the clearing and inactivation of MPO by ceruloplasmin as well, resulting in increased MPO activity. MPO produces not only the strong oxidant bleach (hypochlorous acid) out of hydrogen peroxide and chloride ions but also inactivates protease inhibitors, oxidizes LDL into a macrophage high-uptake form, and consumes nitric oxide. These may contribute to endothelial dysfunction and add to the chronic renal lesions observed in patients with MPO-ANCA. MPO levels are further influenced by hormonal and genetic factors including two, MPO463 and MPO129, single nucleotide polymorphisms. The MPO 463 polymorphism has been associated with an increased risk of development of MPO-ANCA associated disease.

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