Clinical & Experimental Immunology
The Journal of Translational Immunology

Clinical and Experimental Immunology REVIEW ARTICLE doi:10.1111/j.1365-2249.2011.04355.x

Anti-neutrophil cytoplasmic antibody (ANCA)-associated vasculitis: where to go?

C. G. M. Kallenberg
Department of Rheumatology and Clinical Immunology, University Medical Center Groningen, University of Groningen, Groningen, the Netherlands

Accepted for publication 12 January 2011
Correspondence: C. G. M. Kallenberg, Department of Rheumatology and Clinical Immunology, AA21, University Medical Center Groningen, PO Box 30·001, 9700 RB Groningen, the Netherlands.
E-mail: c.g.m.Kallenberg@int.umcg.nl

Summary

Enormous progress has been made during the last 25 years in our understanding of the aetiopathogenesis of the anti-neutrophil cytoplasmic autoantibody (ANCA)-associated vasculitides (AAV). This has led to improvements in early diagnosis, treatment and secondary prevention of these diseases. Nevertheless, there are still unmet needs in the AAV. With respect to diagnosis and follow-up, sensitive biomarkers that reflect disease activity, also during smouldering disease, are needed. In the field of aetiopathogenesis, genetic and epigenetic studies are being performed not only directed at the autoimmune response but also at the expression of, possibly modified, autoantigens. Environmental factors, in particular microbial factors, are also being explored. This will enable analysis of gene–environment interactions in the AAV, so elucidating further their aetiopathogenesis. Explaining the differences in clinical presentation between proteinase 3 (PR3)-associated AAV and myeloperoxidase (MPO)-associated AAV requires an adequate animal model for PR3-ANCA disease, which is currently lacking. Although many large randomized controlled trials have built a base for a rational therapeutic approach in the AAV, late morbidity and mortality is still significant. The availability of new biologicals and the development of sensitive biomarkers for disease activity could further improve prognosis for patients suffering from AAV.

Keywords: ANCA-associated vasculitis, myeloperoxidase-ANCA, pathogenesis, proteinase 3-ANCA, treatment

Introduction

Since the discovery of anti-neutrophil cytoplasmic antibodies (ANCA) in 1982, enormous progress has been made in our understanding of the associated diseases and their treatment. From a diagnostic viewpoint, the elucidation of proteinase 3 (PR3) and myeloperoxidase (MPO) as the relevant antigenic specificities of ANCA in the ANCA-associated vasculitides (AAV) has strongly improved early diagnosis and treatment of these diseases. Major advances have been made in our insight into the aetiopathogenesis of AAV based on *in vitro* and *in vivo* experimental studies and, at least in part, confirmed by clinical observations. Finally, unique international collaborations have enabled large multi-centre randomized controlled trials (RCTs) in these relatively rare diseases which have resulted in evidence-based therapeutic approaches with higher efficacy and less toxicity compared to previous regimens [1]. Nevertheless, there are still unmet needs in the AAV [2].

Diagnosis and follow-up of the AAV

The AAV are currently classified either according to the American College of Rheumatology (ACR) criteria [3] or the Chapel Hill Consensus Conference definitions [4]. The former are classification and not diagnostic criteria with limited sensitivity and specificity, the latter definitions only. There is a strong need for diagnostic criteria that take into account the clinical presentation, pathogenic concept and response to treatment of a particular disorder. An international working party, including representatives of EUVAS (European Vasculitis Study Group) and VCRC (Vasculitis Clinical Research Consortium), has started a prospective study to achieve these goals. Also, nomenclature will be adapted, with the proposal to change the eponym Wegener's granulomatosis to granulomatosis with polyangiitis (GPA), based on the awareness of the Nazi connections of Friedrich Wegener.

Although the diagnostic sensitivity and specificity of PR3-ANCA and MPO-ANCA for the AAV are, in the right clinical

context, very high, their use as biomarkers of disease activity is still insufficient [5]. Recent uncontrolled data from B cell depletion treatment in PR3-ANCA AAV [6] suggest a direct relation between the reappearance of ANCA and disease reactivation, but further data are needed. Analysis of epitope specificities of ANCA could be useful, particularly in relation to functional characteristics, but these data are currently lacking or insufficient. In general, there is a strong need for biomarkers that reflect relapsing disease or even predict relapse early. Further insight into the mechanisms underlying relapse, with the higher relapse rate in PR3-ANCA disease than in MPO-ANCA disease taken into account, would probably lead to the availability of these biomarkers.

More recently, autoantibodies to human lysosomal-associated membrane protein 2 (hLAMP2) have been described as sensitive markers for ANCA-associated pauci-immune necrotizing glomerulonephritis [7]. These antibodies were suggested to be induced by molecular mimicry between hLAMP2 and FimH, and adhesin of Gram-negative bacteria. Furthermore, *in vitro* and *in vivo* experimental data support a pathogenetic role of anti-hLAMP2. As these antibodies were reported to disappear during remission, they could potentially be used as biomarkers for active disease as well. However, the clinical significance of anti-hLAMP2 in AAV awaits confirmation by other groups.

Aetiopathogenesis of the AAV

A multitude of *in vivo* and *in vitro* experimental studies have broadened our understanding of the pathophysiological pathways involved in lesion development in the AAV. The aetiology of these diseases is, however, far from known. Genetic and environmental factors are, as in many other autoimmune diseases, involved. The recent finding of a $CD8^+$ T cell transcription signature predicting a poor prognosis [8] together with data soon becoming available from the EUVAS-based GWAS study in AAV will further support a genetic base for the AAV. Interesting data from Chapel Hill [9] show that not only the autoimmune response *per se*, but also the availability and modification of the autoantigens PR3 and MPO as based on epigenetic mechanisms, are important factors determining disease induction and expression. It has been recognized that significant geographical differences in incidence and clinical presentation of AAV occurs. AAV and particularly PR3-ANCA associated GPA (Wegener's) are more frequent in Caucasians. Furthermore, PR3-ANCA-associated GPA (Wegener's) occurs far more frequently in northern areas (Europe and United States), whereas MPO-ANCA-associated MPA is more frequent in southern areas and in East Asia and Japan. In the latter countries the clinical presentation of GPA (Wegener's) is more associated with MPO-ANCA than with PR3-ANCA [10]. Whether this is based on genetic factors or explained environmentally is not clear.

The exogenous factors involved in disease induction are largely unknown. Toxic substances, such as chronic silica exposure, as well as microbial agents, have been incriminated. Silica could play a role but the widespread exposition to silica and the rarity of AAV indicates that it probably plays only a minor role. The role of infections is intriguing and will be explored further in a large European Community (EC)-funded European–US project called INTRICATE (infectious triggers of autoimmunity). Until now at least three microbial factors have been proposed. The first relates to the observation that nasal colonization with *Staphylococcus aureus* is frequently present in GPA (Wegener's) and is a strong risk factor for relapse. *S. aureus* infection could create an environment for autoantigen presentation and immune activation [11]. More mechanistic data are needed to explain the role of *S. aureus* colonization and to establish its significance as a therapeutic target. The second relates to the concept of complementary proteins; that is, a protein that results from transcription of the anti-sense strand of the gene encoding for the original protein, which could induce an immune response that leads, via idiotypic–anti-idiotypic interaction, to an immune response against the original protein. Antibodies to complementary PR3 (cPR3) have been demonstrated in patients with PR3-ANCA disease, and immunization of mice with cPR3 led to anti-PR3 antibodies [12]. The immunodominant part of cPR3 showed homology with various microbial peptides including peptides from *S. aureus* which, thus, could potentially induce anti-cPR3 and anti-PR3. Although being an attractive theory, the presence of anti-cPR3 in patients with AAV has not been confirmed as yet by others. The third proposed mechanism of induction of AAV has already been mentioned, and concerns the development of anti-hLAMP2 antibodies [7]. Again, the presence of anti-hLAMP2 antibodies in AAV patients should be confirmed by other groups.

Less attention has been given to the age of presentation of the AAV which, generally, present at a relatively old age. Whether ageing of the immune system, so-called immunosenescence, plays a role in disease development has not yet been addressed.

As mentioned, experimental studies have taught us much about the pathophysiological events resulting in the clinical expression of AAV. This is particularly true for MPO-ANCA-associated MPA, although the clinical association of changes in levels of MPO-ANCA with variation in disease activity is limited. The pathophysiological significance of PR3-ANCA is less clear. The *in vitro* data showing that PR3-ANCA, comparable to MPO-ANCA, are able to activate primed neutrophils to the production of reactive oxygen species and the release of lytic enzymes, could explain the potential of the antibodies to induce endothelial damage. However, the distinct clinical differences between PR3-ANCA disease and MPO-ANCA disease, in particular the granulomatous inflammation and relapsing course in the former, are not explained. The (additional) involvement of cell-mediated

immunity in the pathogenesis of PR3-ANCA disease has been suggested by various studies [13], but full understanding of the pathogenic routes leading to granuloma formation requires an animal model. Unfortunately, such a model is currently not available. Any model for PR3-ANCA disease should include granulomatous inflammation in conjunction with necrotizing vasculitis. Furthermore, such a model requires not only a PR3-directed immune response but also an *in vivo* expression of the target antigen identical to that which occurs in patients with PR3-ANCA disease.

Finally, a better understanding of the relapsing course of PR3-ANCA disease is necessary. Although risk factors for relapse have been reported, including persistence of ANCA after induction of remission, chronic nasal carriage of *S. aureus* and the $CD8^+$ T cell transcription signature, elucidation of the events leading to relapse could provide clues to prevent relapses and/or to select those patients who need long-term maintenance treatment.

Treatment

As mentioned, great advances have been made in the treatment of AAV, due largely to the unique collaborations in EUVAS and VCRC. The results from large RCTs on patients with AAV have enabled an evidence-based approach in treatment. New trials are under way, including an evaluation of the role of plasma exchange in AAV (PEXIVAS) and the role of intermittent B cell depletion for maintenance of remission.

What are other unmet needs in the treatment of AAV? First, targeted treatment in the acute initial phase of the diseases could be helpful to reduce the amounts of corticosteroids needed to control the inflammatory process. Here, targeting complement factor C5, its chemotactic split product C5a and/or the C5a-receptor seems promising based on *in vivo* experimental data [14]. Rapid control of inflammation could reduce the accrual of damage. Secondly, although immediate mortality has been strongly reduced, late mortality, reflected in 5 years' survival data, and morbidity is still considerable. As discussed before, better insight in the mechanisms of relapse is essential to reduce this late mortality and morbidity. Also, slowly ongoing decline in renal function, particularly in patients with MPO-ANCA disease, should be recognized and explained. Better markers are needed to detect ongoing subacute active renal disease. Possibly, the presence of effector T cells in urine specimens could be a sensitive biomarker for renal activity in the AAV, better than the urinary sediment [15]. Long-term follow-up data from RCTs are essential to appreciate the efficacy of therapeutic regimens on outcome in terms of late mortality and damage.

Conclusion

Enormous progress has been made in our understanding of the aetiopathogenesis of the AAV. This has contributed to improvements in early diagnosis, treatment and even secondary prevention of these severe diseases. Although many unmet needs are still apparent, as discussed in this paper, the combined efforts of the international community in the field of ANCA-associated vasculitides set the stage for further improvements in the near future, of benefit for patients suffering from AAV.

Disclosure

None.

References

1 Chen M, Kallenberg CGM. ANCA-associated vasculitides – advances in pathgenesis and treatment. Nat Rev Rheumatol 2010; **6**:653–64.

2 Falk RJ, Jennette JC. ANCA disease: where is this field heading? J Am Soc Nephrol 2010; **21**:745–52.

3 Hunder GG, Arend WP, Bloch DA *et al.* The American College of Rheumatology 1990 criteria for the classification of vasculitis. Introduction. Arthritis Rheum 1990; **33**:1065–7.

4 Jennette JC, Falk RJ, Andrassy K *et al.* Nomenclature of systemic vasculitides: the proposal of an international consensus conference. Arthritis Rheum 1994; **37**:187–92.

5 Kallenberg CGM. Autoantibodies as biomarkers for ANCA-associated vasculitides. Curr Immunol Rev 2011; in press.

6 Cartin-Ceba R, Golbin J, Keogh KA *et al.* Rituximab for remission induction and maintenance in ANCA-associated vasculitis: a single-center ten-year experience in 108 patients. Arthritis Rheum 2010; **62**:S283–4.

7 Kain R, Exner M, Brandes R *et al.* Molecular mimicry in pauci-immune focal necrotizing glomerulonephritis. Nat Med 2008; **14**:1088–96.

8 McKinney EF, Lyons PA, Carr EJ *et al.* A CD8+ T cell transcription signature predicts prognosis in autoimmune disease. Nat Med 2010; **16**:586–91.

9 Ciavatta DJ, Yang J, Preston GA *et al.* Epigenetic basis for aberrant upregulation of autoantigen genes in humans with ANCA vasculitis. J Clin Invest 2010; **120**:3209–19.

10 Chen M, Yu F, Zhang Y *et al.* Characteristics of Chinese patients with Wegener's granulomatosis with anti-myeloperoxidase autoantibodies. Kidney Int 2005; **68**:2225–9.

11 Popa ER, Stegeman CA, Kallenberg CGM, Tervaert JW. *Staphylococcus aureus* and Wegener's granulomatosis. Arthritis Res 2002; **4**:77–9.

12 Pendergraft WF, Preston GA, Shah RR *et al.* Autoimmunity is triggered by cPR- (105-201), a protein complementary to human autoantigen proteinase-3. Nat Med 2004; **10**:72–9.

13 Berden AE, Kallenberg CGM, Savage COS *et al.* Cellular immunity in Wegener's granulomatosis. Characterizing T lymphocytes. Arthritis Rheum 2009; **60**:1578–87.

14 Huugen D, van Esch A, Xiao H *et al.* Inhibition of complement factor C5 protects against anti-myeloperoxidase antibody-mediated glomerulonephritis in mice. Kidney Int 2007; **71**:646–54.

15 Abdulahad WH, Kallenberg CGM, Limburg PC, Stegeman CA. Urinary CD4+ effector memory T cells reflect renal disease activity in antineutrophil cytoplasmic antibody-associated vasculitis. Arthritis Rheum 2009; **60**:2830–8.