Case Report

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# MPO-C-ANCA-associated necrotising and crescentic glomerulonephritis

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### Lesson

The patterns of ANCA staining usually relate closely to antibodies against myeloperoxidase and proteinase-3. C-ANCA is mainly antibodies to proteinase-3 and P-ANCA is antibodies to myeloperoxidase. C-ANCA with antibodies to MPO with clinical sequelae is unusual.

### Keywords

myeloperoxidase, cytoplasmic anti-neutrophil cytoplasmic antibodies, glomerulonephritis

## Introduction

Anti-neutrophil cytoplasmic antibodies (ANCA) are autoantibodies directed against antigens in the cytoplasm of neutrophils and are often associated with systemic vasculitis. ANCAs are classified based on their patterns of staining: cytoplasmic (C-ANCA) and perinuclear (P-ANCA). C-ANCA is mainly antibodies to proteinase-3 (PR-3) and P-ANCA is antibodies to myeloperoxidase (MPO). C-ANCA with antibodies to MPO is unusual and is deemed interpretation error during visualisation with immunofluorescence. Therefore, our case of MPO positive C-ANCA (MPO-C-ANCA) associated with necrotising and crescentic glomerulonephritis induces diagnostic and pathogenic dilemmas.

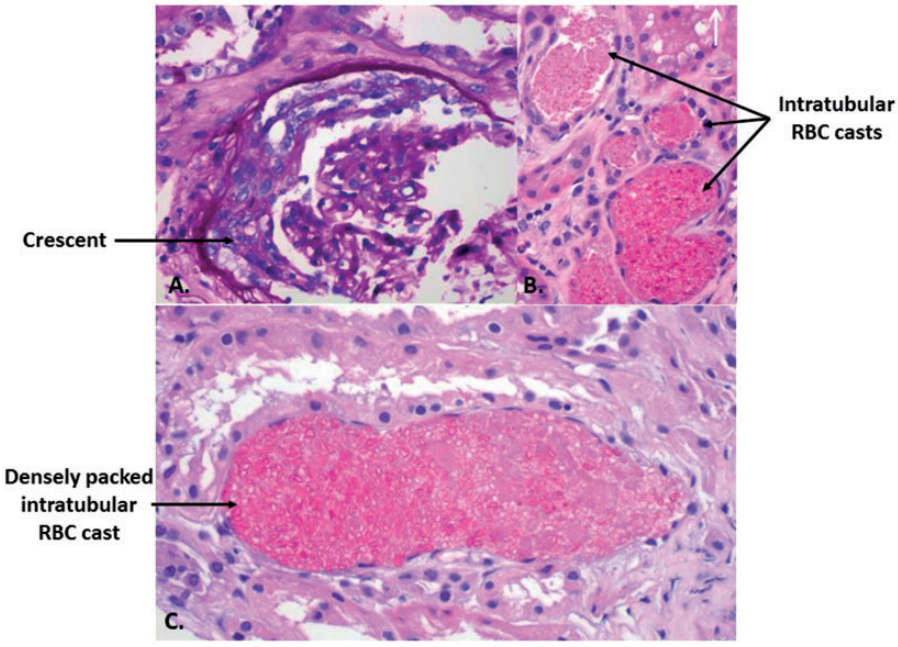
## Case report

A 73-year-old Caucasian male smoker was admitted to the hospital with progressive dyspnoea due to multifocal pneumonia superimposed on underlying pulmonary fibrosis. He rapidly developed acute respiratory distress syndrome and was intubated and put on mechanical ventilation. Bronchoscopy revealed diffuse alveolar hemorrhage. He also developed non-oliguric acute kidney injury. Laboratory tests demonstrated urea nitrogen 97 mg/dL, creatinine 2.35 mg/dL (baseline 0.8 mg/dL) and potassium 4.4 meq/L. Urinalysis revealed 288 erythrocytes/high power field (hpf), dysmorphic erythrocytes, 2 white blood cells/hpf, dense, coarse granular casts and 1–2 erythrocyte cast/hpf. C-ANCA was positive but anti-PR-3 antibody was negative (optical density 0.032). Anti-MPO antibody was positive (optical density 111.0; cutoff 0.244). Renal biopsy revealed necrotising and crescentic glomerulonephritis with 50% glomerular involvement with necrosis and crescents (Figure 1(a)), arterial and arteriolar nephrosclerosis, intratubular casts with densely packed red blood cells (Figure 1(b) and (c)). Immunofluorescence showed nonspecific staining of IgG, IgM, IgA, C3, C1Q, albumin, kappa and lambda. Treatment with steroids, cyclophosphamide and plasmapheresis resulted in improvement in renal function. Patient did not require renal replacement therapy. His total hospital length of stay was 30 days and time on mechanical ventilation was 12 days.

## Discussion

Commercial laboratories perform ANCA screen that is cell-based, indirect immunofluorescence using ethanol-fixed as well as formalin fixed human neutrophils. Results are reported as positive or negative. Positive results are reflexed to titre of the relevant pattern, e.g. C-ANCA to anti-PR3 and P-ANCA to anti-MPO. The anti-MPO and anti-PR-3 antibodies are measured using semi-quantitative multiplex immunoassays with an analytic sensitivity of 0.2 antibody index. Discrepancy in ANCA patterns has raised concerns regarding the basic standards required for ANCA testing, the value of formalin fixation of neutrophils in differentiating P-ANCA from antinuclear antibodies and use of alternative methods such as image analysis and/or multiplex immunoassays.[1] The European Vasculitis Study Group tested for the presence of cytoplasmic pattern/perinuclear pattern and atypical ANCA (A-ANCA) by indirect immunofluorescence and for the presence of PR3-ANCA and MPO-ANCA by different immunoassays and reported a large variability between the immunofluorescence methods tested and a high diagnostic performance of PR3-ANCA and MPO-ANCA by

**Figure 1.** (a) Fibrocellular crescent. (b and c) Intra-tubular erythrocyte casts highlighting the relationship between structure and function.




immunoassay to discriminate ANCA-associated vasculitis from disease controls.[2] However, false positive MPO-C-ANCA does occur[3,4] and the explanations include ANCA directed against other constituents of neutrophil granules,[5] DNA contained within the antigen binding site of anti-DNA antibodies that could bind to the highly cationic MPO used as substrate antigen in immunoassays, resulting in a false-positive MPO test[6] and cross-reaction of MPO-C-ANCA with PR-3, unspecific anti-MPO reaction or that the reactivity of the MPO-C-ANCA positive sera was directed against a contaminant in the MPO preparation. However, these assumptions have previously been disproved by Segelmark et al.[7] who have shown that sera from MPO-C-ANCA patients did not react with PR-3, that the reaction could be inhibited by a small amount of MPO in solution and that the reactivity is against MPO itself. Recently, Simon et al.[8] have reported autoantibodies directed against preformed Pentraxin 3, a class of soluble pattern recognition receptor, that can be found in the neutrophil granules. The fluorescence pattern in their study was remarkable for staining of small cytoplasmic granules that was distinctly different from the classic C- and P-ANCA patterns. The presence of anti-Pentraxin 3 antibodies appear to be limited to a few autoimmune disorders such as lupus and vasculitis. Therefore, the significance of ANCA patterns relative to target antigens may be of renewed interest.

Our patient with MPO-C-ANCA-associated necrotising and crescentic glomerulonephritis was treated per Kidney Disease/Improving Global Outcomes guidelines that recommend that cyclophosphamide and corticosteroids be used as initial treatment and the addition of plasmapheresis for patients with diffuse pulmonary hemorrhage.[9] The patient did not require dialysis and serum creatinine at discharge had improved to 1.58 mg/dL. Treatment with rituximab was considered as it has been shown not to be inferior to daily cyclophosphamide treatment for induction of remission in severe ANCA-associated vasculitis per the Rituximab versus cyclophosphamide in ANCA-associated renal vasculitis (RITUXVAS) and Rituximab versus cyclophosphamide for ANCA-associated vasculitis (RAVE) studies.[10,11] However, both the studies excluded patients with alveolar hemorrhage severe enough to require mechanical ventilation and serum creatinine level greater than 4.0 mg/dl attributed to renal failure from the current episode of renal disease activity. Our patient with diffuse alveolar hemorrhage was therefore not a candidate for rituximab therapy. Little is known about the prognosis of MPO-C-ANCA-associated necrotising and crescentic glomerulonephritis, but data from a small series ($N = 21$) report that progression to chronic kidney disease and end stage renal disease may occur despite an initial favorable response to treatment.[12] Our patient died at home,

two months after discharge from the hospital. He was not on dialysis.

In conclusion, we present an unusual case of MPO-C-ANCA-associated glomerulonephritis supported by clinical, serological and histological data.

## Declarations

**Competing Interests:** None declared.

**Funding:** None declared.

**Ethical approval:** Written informed consent for publication was obtained from the patient's spouse as the patient was critically ill and intubated.

**Guarantor:** AK

**Contributorship:** All authors were involved in the care for the patient (AK, DNW, KFA and AAE). AK drafted the initial version of manuscript, performed literature search and approved the final manuscript. KFA drafted the manuscript and approved the final manuscript. DNW has provided pathology images, reviewed and revised the manuscript. AAE reviewed and revised the manuscript, supervised and approved the final manuscript for submission.

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## References

1. Csernok E and Holle JU. Twenty-eight years with antineutrophil cytoplasmic antibodies (ANCA): how to test for ANCA – evidence-based immunology? *Auto Immun Highlights* 2010; 1: 39–43.
2. Damoiseaux J, Csernok E, Rasmussen N, Moosig F, et al. Detection of antineutrophil cytoplasmic antibodies (ANCAs): a multicentre European Vasculitis Study Group (EUVAS) evaluation of the value of indirect immunofluorescence (IIF) versus antigen-specific immunoassays. *Ann Rheum Dis*. Epub ahead of print 2016. DOI: 10.1136/annrheumdis-2016-209507.
3. Seema C, Ranjana WM, Lekha G and Nidhi S. Myeloperoxidase-antineutrophil cytoplasmic antibodies with cytoplasmic fluorescence pattern. *J Lab Phys* 2010; 2: 42–43.
4. Savige JA, Paspaliaris B, Silvestrini R and Davies D. A review of immunofluorescent patterns associated with antineutrophil cytoplasmic antibodies (ANCA) and their differentiation from other antibodies. *J Clin Pathol* 1998; 51: 568–575.
5. Stroncek DF, Egging MS, Eiber GA and Clay ME. Neutrophil alloantibodies react with cytoplasmic antigens: a possible cause of false-positive indirect immunofluorescence assays for antibodies to neutrophil cytoplasmic antigens. *Am J Kidney Dis* 1993; 21: 368–373.
6. Hirai Y, Iyoda M, Shibata T and Ashikaga E. Lupus nephritis associated with positive MPO-ANCA in a patient with underlying autoimmune hemolytic anemia. *Clin Exp Nephrol* 2008; 12: 393–397.
7. Segelmark M, Baslund B and Wieslander J. Some patients with anti-myeloperoxidase autoantibodies have a C-ANCA pattern. *Clin Exp Immunol* 1994; 96: 458–465.
8. Simon A, Subra JF, Guilpain P and Jeannin P. Detection of anti-pentraxin-3 autoantibodies in ANCA-associated vasculitis. *PLoS One* 2016; 11: e0147091.
9. KDIGO. Clinical practice guideline for glomerulonephritis. www.kdigo.org/clinical_practice_guidelines/pdf/KDIGO-GN-Guideline.pdf (2012, accessed 6 September 2016).
10. Jones RB, Tervaert JW, Hauser T and Luqmani R; European Vasculitis Study Group. Rituximab versus cyclophosphamide in ANCA-associated renal vasculitis. *N Engl J Med* 2010; 363: 211–220.
11. Stone JH, Merkel PA, Spiera R and Seo P; for the RAVE−ITN Research Group. Rituximab versus cyclophosphamide for ANCA-associated vasculitis. *N Engl J Med* 2010; 363: 221–232.
12. Franssen CF, Stegeman CA, Oost-Kort WW and Kallenberg CG. Determinants of renal outcome in anti-myeloperoxidase-associated necrotizing crescentic glomerulonephritis. *J Am Soc Nephrol* 1998; 9: 1915–1923.