The Journal of Laryngology and Otology
May 1994, Vol. **108,** pp. 420–422

Negative ANCA in Wegener's granulomatosis

S. CARRIE, F.R.C.S., K. B. HUGHES, F.R.C.S., D.L.O., M. G. WATSON, F.R.C.S.

Abstract
Wegener's granulomatosis is a systemic vasculitis characterized by necrotizing granulomata and without treatment is associated with a poor prognosis. Antineutrophil cytoplasmic antibodies (ANCA) have proved to be a useful serological marker allowing for early diagnosis and treatment of this condition. We report two cases of patients who were ANCA-negative on presentation despite clinical suspicion of Wegener's granulomatosis. In both cases the patients developed a positive ANCA titre as the disease progressed.

Key words: Antibodies, antineutrophil cytoplasmic (ANCA); Wegener's granulomatosis

Introduction

Wegener's granulomatosis is a specific vasculitis first defined by Friedrich Wegener (1939). Fauci and Wolff (1973) described the condition as a specific disease having necrotizing granuloma with vasculitis of the upper and lower respiratory tracts, systemic vasculitis and focal necrotizing glomerulitis.

The disease may be difficult to diagnose, especially in the early stage due to the multiplicity of symptoms (Van der Woude *et al.*, 1978) and the lack of specific histological findings on biopsy specimens (McCluskey and Fienberg, 1983).

Approximately one-third of the patients present with (limited) locoregional forms of the disease which can last from months to years (Gross *et al.*, 1991). Progression occurs to the classic generalized form which is associated with a rapidly fatal course unless treated aggressively. The locoregional type is the more difficult to diagnose for the reasons explained earlier, but the recent development of the serological marker antineutrophil cytoplasmic antibody (ANCA) has enabled Wegener's granulomatosis to be diagnosed much earlier in its clinical course.

Van der Woude *et al.* (1985) reported IgG antibodies directed against intracytoplasmic antigens of granulocytes and monocytes in patients with Wegener's granulomatosis. They suggested this anticytoplasmic antibody, later termed ANCA, was specific for Wegener's granulomatosis and that the antibody titre correlated with disease activity. Since then ANCA has been subtyped into a cytoplasmic pattern (c-ANCA) and a perinuclear pattern (p-ANCA) based on the findings of immunofluorescence techniques. c-ANCA directed against proteinase 3 is more specific for Wegener's granulomatosis than p-ANCA which is found in other types of vasculitis and idiopathic glomerulitis. c-ANCA has been used in the early diagnosis of otological Wegener's granulomatosis prior to the disease becoming systemic (Macias *et al.*, 1993).

We report on two patients who presented with locoregional (limited) forms of Wegener's granulomatosis and who did not develop raised ANCA levels until their disease had progressed. This underlines the importance of interpreting antibody levels in the context of the clinical picture and other diagnostic investigations.

Case reports

Case 1

A previously healthy 30-year-old man was admitted with a 24 h history of the sudden onset of vertigo. Associated with this he had a two-month history of increasing bilateral deafness, otalgia and intermittent otorrhoea, for which a presumptive diagnosis of acute otitis media was made prior to his admission.

On examination he had second degree nystagmus and was extremely unsteady. He was apyrexial and had no demonstrable cerebellar signs. His external auditory canals were grossly oedematous and the tympanic membranes were inflamed and bulging. Fistula test was negative bilaterally.

Haemoglobin and white cell count were not elevated. Plasma viscosity, c-reactive protein and alpha-globulins were raised and albumin levels were reduced. c-ANCA titres carried out shortly after admission were negative. Urinalysis was positive for red blood cells and free light chains. Chest X-ray, posterior fossa CT scan and CSF analysis were normal. Syphilis serology and Mantoux test were negative.

Pure tone audiometry revealed a profound hearing loss on the right side and a severe mixed deafness on the left. Caloric tests confirmed bilateral absence of vestibular function.

He was treated with intravenous antibiotics and topical antibiotic/steroid ear drops on the presumptive diagnosis of acute otitis media with secondary labyrinthitis. Twenty-four hours after commencing treatment the hearing deteriorated in his left ear. Examination under anaesthesia revealed thickened inflamed tympanic membranes and a serous otitis media. Culture of this produced no growth.

His hearing continued to deteriorate and as there was no response to antibiotics it was felt his otitis media was immunologically mediated. He was commenced on prednisolone 60 mg per day combined with a high dose of intravenous penicillin. Within 48 h there was a marked improvement in the appearances of the tympanic membranes with complete resolution of the inflammation after one week. Two weeks following the introduction of steroids there was a marked improvement in his hearing thresholds amounting to 20–30 dB bilaterally (Figures 1 and 2). He was rehabilitated with binaural hearing aids and postural physiotherapy prior to discharge. Upon review two weeks later his pure tone thresholds were unchanged and he appeared to be adjusting to his disability.

Four weeks after review he was admitted under the care of a general physician with a two-week history of polyarthralgia and a recent haemoptysis. Renal function had deteriorated and c-ANCA had become positive at a titre of 1 in 50. Chest X-ray

From the Department of Otolaryngology, Doncaster Royal Infirmary, Doncaster.
Accepted for publication: 30 October 1993.

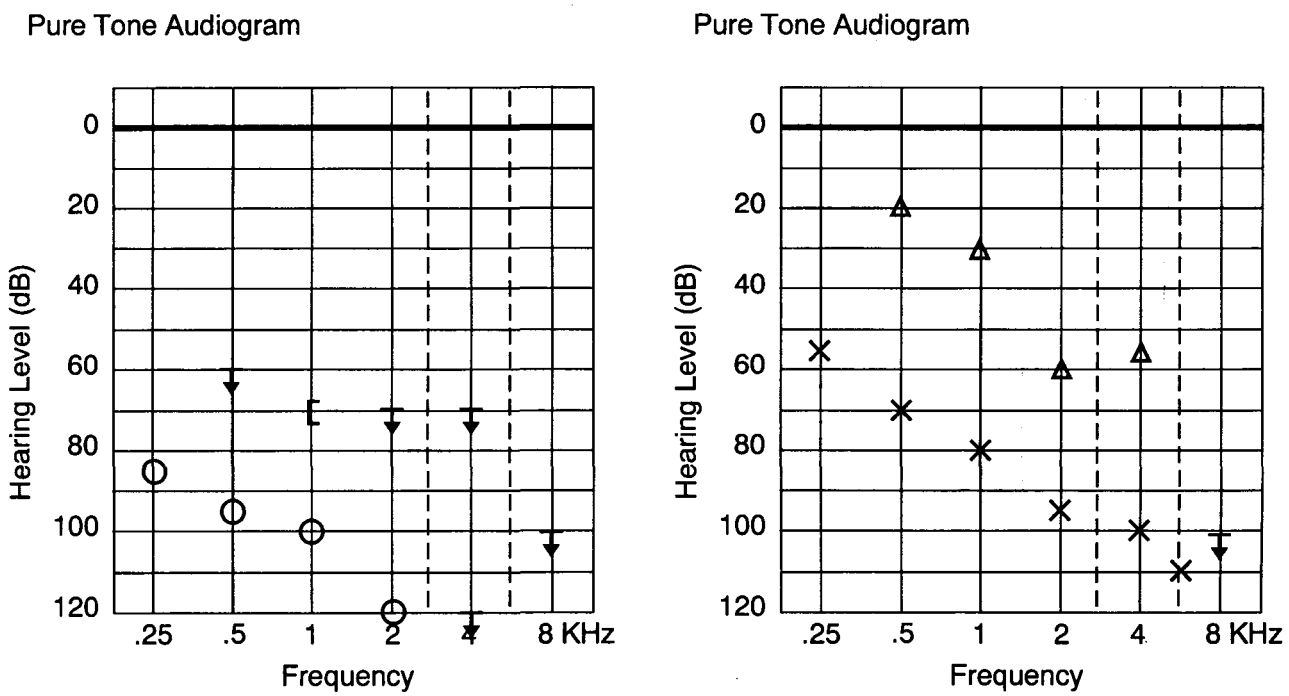

Fig. 1
Pre-treatment audiogram.

and CT scan were highly suggestive of pulmonary infarct and a renal biopsy revealed crescentic formation in the glomeruli. A diagnosis of Wegener's granulomatosis was made and he was commenced on prednisolone and cyclophosphamide resulting in a marked improvement in his general condition and renal function.

Case 2

A 41-year-old butcher, previously well, was admitted as an emergency with increasing facial pain, a painful red eye and malaise. He had been treated as an outpatient for acute sinusitis one week previously, when he had presented with facial pain, nasal obstruction and epistaxes. Examination of his nose had revealed extensive mucopus and mucosal oedema. A nasal swab grew *Staphylococcus aureus* and he was commenced on intravenous flucloxacillin plus metronidazole to cover the possibility of anaerobic infection. An ophthalmology opinion confirmed simple conjunctivitis.

His temperature spiked repeatedly over the next few days and to rule out a septic focus in his maxillary sinuses he had bilateral antral washouts. The return from these was haemorrhagic fluid with no pus. Grossly abnormal nasal mucosa was biopsied from the left antrum and inferior turbinate, the histology of which was

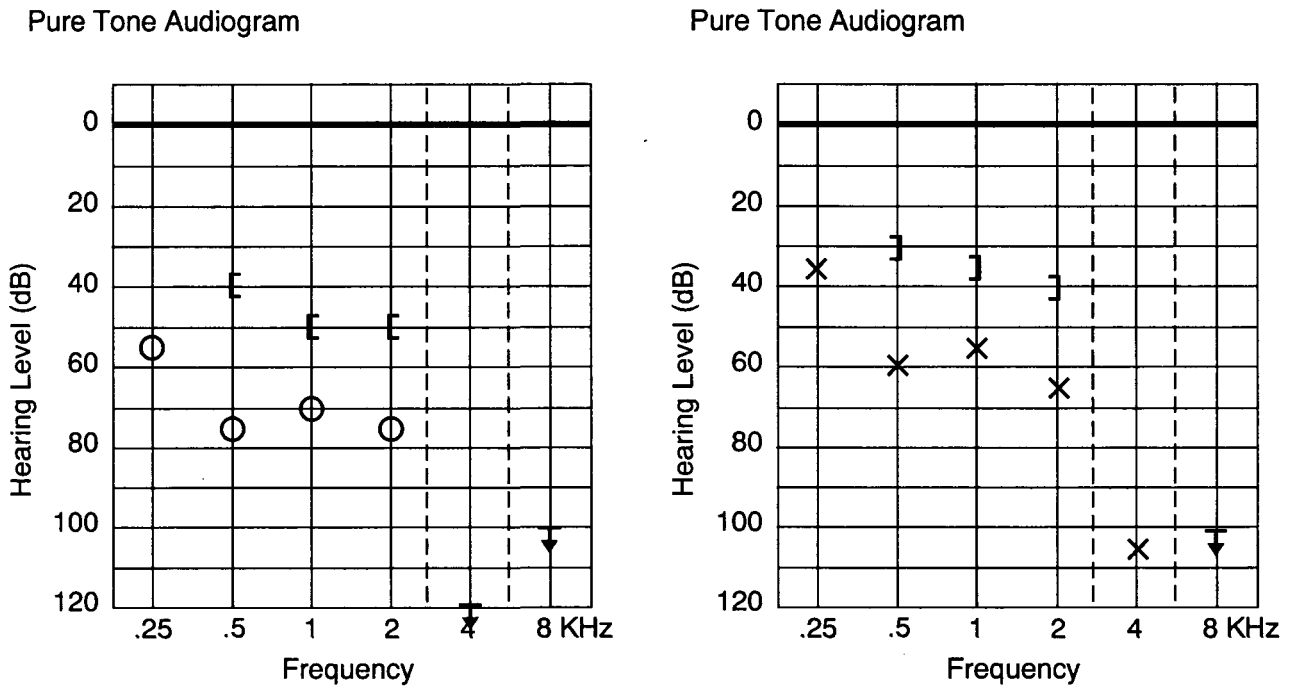

Fig. 2
Post-treatment audiogram.

https://doi.org/10.1017/S0022215100126969 Published online by Cambridge University Press

chronic inflammation with a granuloma and giant cells, compatible with Wegener's granulomatosis. c-ANCA was negative but plasma viscosity was markedly elevated at 2.24 mPas. Autoantibody screen and renal function were normal. He had a microcytic anaemia with a Hb of 10.0 g/dl and his white cell count and platelets were not elevated. A chest X-ray revealed a suspicious lung lesion in the right midzone. Bronchoscopy and biopsy of this were performed, the histology of which was nonspecific chronic inflammation and fibrosis. Bronchial brushings were negative for malignant cells.

Despite the negative ANCA the most likely diagnosis was Wegener's granulomatosis. This was supported by the clinical picture of generalized illness, joint pains, rhinosinusitis and a lung lesion, plus a raised plasma viscosity, a microcytic anaemia and suggestive nasal biopsy. He was commenced on daily prednisolone and cyclophosphamide as well as ferrous sulphate and made a marked clinical improvement. His plasma viscosity returned to normal range and his c-ANCA became positive at a titre of 1 in 25 eight weeks after presentation.

Discussion

In both cases the patients were previously well with no history of immunological disease. The diagnosis of Wegener's granulomatosis was considered in each case and c-ANCA titres were measured early after presentation using the indirect immunofluorescence technique. A positive titre was taken at level of 1 in 10. ANCA has been demonstrated to be highly specific for Wegener's granulomatosis with several studies suggesting more than 97 per cent positivity (Van der Woude *et al.*, 1985; Nolle *et al.*, 1989; Tervaert *et al.*, 1989). The sensitivity of ANCA depends on the disease activity and extent. Using enzyme-linked immunoadsorbent assay (ELISA) techniques in active locoregional (limited) disease there is 60 per cent sensitivity and in the active generalized form there is 93 per cent sensitivity (Nolle *et al.*, 1989). These figures rise to 67 and 96 per cent respectively if indirect immunofluorescent antibody detection is used.

The two cases discussed illustrate how an initially negative titre can become positive as the disease progresses to the more generalized form. This would confirm recent work suggesting that ANCA levels parallel the extent of disease activity. However, in *Case 2*, although the ANCA titre was negative on presentation, the disease was clinically generalized. This may have resulted from an initial delay in antibody production or because antibody levels at this stage were too low to be detected by conventional means. Tervaert *et al.* (1989) demonstrated that antibody titres fall in patients during treatment and tend to be negative in patients without disease activity. A rising ANCA titre is also a sensitive indicator of a relapse following treatment of Wegener's granulomatosis.

The significance of a negative ANCA test must be taken in the context of the overall clinical picture. A single negative ANCA does not rule out the diagnosis of Wegener's granulomatosis, although serial titres may demonstrate a rise in antibody levels as the disease progresses. In the presence of strong clinical and histological evidence, with a negative ANCA titre, appropriate treatment should be commenced while the disease is at an early stage. It is, however, important to remember that false positive ANCA cases do occur and have been demonstrated in patients with glomerulonephritis (Davies *et al.*, 1982) and systemic vasculitis (Hall *et al.*, 1984), and also in a case of nasal septal perforation (Mains, 1989) with no evidence of vasculitis.

References

Davies, D. J., Moran, J. E., Niall, J. F., Ryan, G. B. (1982) Segmental necrotizing, glomerulonephritis with antineutrophilic antibody: possible arbovirus aetiology? *British Medical Journal* **2:** 606.

Fauci, A. S., Wolff, S. M. (1973) Wegener's granulomatosis: studies in 18 patients and a review of the literature. *Medicine* **52:** 535–561.

Gross, W. L., Schmitt, W. H., Csernok, E. (1991) Antineutrophil cytoplasmic antibody-associated diseases: a rheumatologist's perspective. *American Journal of Kidney Diseases* **18:** 175–179.

Hall, J. B., Wadham, B. M., Wood, C. J., Ashton, V., Adam, W. R. (1984) Vasculitis and glomerulonephritis: a subgroup with an antineutrophil cytoplasmic antibody. *Australian and New Zealand Journal of Medicine* **14:** 277–278.

Macias, J. D., Wackym, P. A., McCabe, B. F. (1993) Early diagnosis of otological Wegener's granulomatosis using serological marker c-ANCA. *Annals of Otology Rhinology and Laryngology* **102:** 337–341.

Mains, B. (1989) Wegener's granulomatosis: false positive antineutrophilic cytoplasmic antibody test. *Journal of Laryngology and Otology* **103:** 524–525.

McCluskey, R. T., Fienberg, R. (1983) Vasculitis in primary vasculitides, granulomatoses and connective tissue diseases. *Human Pathology* **14:** 305–313.

Nolle, B., Specks, U., Ludemann, J. (1989) Anticytoplasmic autoantibodies: their immunodiagnostic value in Wegener's granulomatosis. *Annals of Internal Medicine* **111:** 28–40.

Tervaert, J. W. C., Van der Woude, F. J., Fauci, A. S., Ambrus, J. L., Velosa, J., Keane, W. F., Meijer, S., Van der Giessen, M., The, T. H., Van der Hem, G. K., Kallenberg, C. G. M. (1989) Association between Wegener's granulomatosis and anticytoplasmic autoantibodies. *Archives of Internal Medicine* **149:** 2461–2465.

Van der Woude, F. J., Arisz, L., Meijer, S., Donker, A. J. M., Hoedemaeker, P. H. J., Van Overbeek, J. J. M. (1978) Wegener's granulomatosis: a clinicopathological study in twelve patients. *Netherlands Journal of Medicine* **21:** 205–220.

Van der Woude, F. J., Rasmussen, N., Lobatto, S., Wikk, A., Permin, H., Van Es, L. A., Van der Giessen, M., Van der Hem, G. K., The, T. H. (1985) Autoantibodies to neutrophils and monocytes: a new tool for diagnosis and a marker of disease activity in Wegener's granulomatosis. *Lancet* **1:** 425–429.

Wegener, F. (1939) Uber eine eigenartige rhinogene Granulomatose mit besonderer Beteiligung des Arteriensystems und der Nieren. *Beitrage zur Pathalogischen Anatomie und zur Allgemeinen Pathologie* **102:** 36–68.

Address for correspondence:
Mr Sean Carrie,
Department of Otolaryngology,
Doncaster Royal Infirmary,
Armthorpe Road,
Doncaster DN2 5LT.

Fax: (0302) 761205.