LETTERS

Cladribine in the treatment of systemic lupus erythematosus nephritis

Systemic lupus erythematosus (SLE) nephritis often requires treatment with cyclophosphamide, which carries the risk of major side effects including infection, ovarian failure and bladder malignancy. Therapeutic strategies that would specifically target lymphocytes are appealing. Following the first report of the use of the purine nucleoside analogue cladribine (2-chloro-2'-deoxyadenosine), a selective lymphocyte depleting agent, in the treatment of lupus nephritis,¹ we report our experience in two patients with severe renal involvement.

CASE 1

A 32 year old woman was diagnosed with SLE at age 28, with polyarthritis, photosensitive rash, subcutaneous nodules, fatigue and lymphopenia. ANA, anti-dsDNA, anti-Sm and anti-RNP antibodies were present. Various immunosuppressants and corticosteroids failed to maintain a sustained remission. Two and a half years after presentation, she developed haematuria and proteinuria and renal biopsy revealed WHO Class III lupus nephritis. Treatment with pulsed intravenous cyclophosphamide and methylprednisolone had to be stopped after four months and a total cyclophosphamide dose of 9 g because of an anaphylactic reaction during an infusion. Despite azathioprine, 150 mg/day, and prednisolone, up to 20 mg/day, she developed severe hypertension (210/120 mm Hg) and biopsy confirmed lymphocytic cutaneous vasculitis. Cladribine (0.05 mg/kg/day for seven days as continuous intravenous infusion) and prednisolone 60 mg/day were started. Cutaneous vasculitis resolved within five days and serum creatinine fell from 190 to 120 µmol/l in five weeks. Cladribine was well tolerated apart from a herpes simplex infection in the natal cleft that responded to acyclovir. She relapsed three months later, with a new rise in creatinine (154 µmol/l) and recurrence of cutaneous vasculitis.

A further infusion of cladribine was given, keeping prednisolone at 5 mg/day. Although the vasculitic rash again resolved, renal function and proteinuria continued to deteriorate.

She has subsequently been maintained with mycophenolate mofetil 1 g twice daily and oral prednisolone. Serum creatinine has

returned to 98 µmol/l and proteinuria to 5.3 g/24 h and remained stable despite gradual reduction of prednisolone dose to 15 mg daily.

CASE 2

A 35 year old woman was diagnosed with SLE at age 31, with fever, pancytopenia, and nephrotic syndrome (proteinuria 6.65 g/24 h). ANA and anti-dsDNA antibodies were present. Renal biopsy revealed WHO Class III lupus nephritis. In the next four years she required three treatment cycles of intravenous cyclophosphamide (total dose per six month cycle: 9-10 g). Azathioprine, methotrexate, cyclosporin A and prednisolone 5-40 mg/day in the interim had failed to control her disease. Cyclophosphamide, additionally, had resulted in premature ovarian failure. Repeat renal biopsy showed progression to Class IV nephritis with focal necrosis and crescents. Cladribine (continuous IV infusion of 0.05 mg/kg/day for seven days) and prednisolone 40 mg/day proved ineffective as creatinine rose from 149 to 243 µmol/l in two months. She also developed a perineal herpes simplex infection but drug was otherwise well tolerated. Pulse intravenous cyclophosphamide and methylprednisolone were subsequently reintroduced and creatinine has again fallen to 118 µmol/l.

Table 1 shows the results of investigations before and after cladribine infusions for both cases.

In the initial study by Davis *et al*,¹ three of seven patients treated with continuous cladribine infusion for a week responded completely and renal function did not deteriorate in any of the seven patients. Our limited experience suggests that cladribine may be effective in other manifestations of SLE (that is, cutaneous vasculitis), but it does not seem to have a consistent effect in severe nephritis. Good tolerability of the drug was confirmed and although herpes simplex infections occurred in both patients the role of corticosteroids cannot be ignored.

Further studies are required to establish the position of cladribine in the treatment of SLE especially in the presence of other lymphocyte depleting agents such as mycophenolate mofetil, which is reported to be effective in lupus nephritis,²⁻⁴ even in cases refractory to cyclophosphamide.⁵

VLASSIS KONTOGIANNIS

PETER C LANYON

RICHARD J POWELL

Clinical Immunology Unit, University Hospital,
Queens Medical Centre,
Nottingham NG7 2UH

Correspondence to: Dr V Kontogiannis.

- 1 Davis CJ, Austin H, Boumpas D, Fleisher TA, Yarboro C, Larson A, *et al*. A pilot study of 2-chloro-2'-deoxyadenosine in the treatment of systemic lupus erythematosus-associated glomerulonephritis. *Arthritis Rheum* 1998;41:335-43.
- 2 Briggs W, Choi M, Scheel PJ. Successful mycophenolate mofetil treatment of glomerular disease. *Am J Kidney Dis* 1998;31:213-17.
- 3 Ho A, Magder L, Petri M. The effect of azathioprine/mycophenolate on systemic lupus erythematosus activity. [Abstract]. *Arthritis Rheum* 1998;41(suppl.):S281.
- 4 Pashinian N, Wallace D, Klinenberg J. Mycophenolate mofetil for systemic lupus erythematosus (abstract). *Arthritis Rheum* 1998;41(suppl.):S110.
- 5 Glicklich D, Acharya A. Mycophenolate mofetil therapy for lupus nephritis refractory to intravenous cyclophosphamide. *Am J Kidney Dis* 1998;32:318-22.

Leg bone pain syndrome in a kidney transplant patient treated with tacrolimus (FK506)

Patients with chronic renal failure often develop musculoskeletal problems such as renal osteodystrophy and amyloid arthropathy,¹ and in successful renal transplantation other complications may ensue, particularly avascular necrosis.² Since the availability of immunosuppressive agents for rejection, there has been a decrease in musculoskeletal problems, however, new complications have been described such as a symmetrical bone pain syndrome and reflex sympathetic dystrophy syndrome (RSDS), some of them related to cyclosporin.³⁻⁶

Tacrolimus is a novel macrolide with potent immunosuppressive effects and with a very similar mechanism of action to cyclosporine A—that is, calcineurin phosphatase inhibition.^{7,8} We report on a patient treated with tacrolimus, who developed a leg bone pain syndrome, two months after kidney transplantation.

The patient was a 50 year old woman with severe hypertension, treated with atenolol (100 mg/day), verapamil (240 mg/day) and clonidine (0.150 mg/day). She developed chronic renal failure and was treated with peritoneal dialysis in 1995. In 1997 she underwent a kidney transplant from a cadaver and immunosuppressive treatment with tacrolimus (4 mg/day) and prednisone (15 mg/day) was started. Two months after transplantation she reported progressive bilateral symmetric pain in the knees. Because of pain and difficulty in walking she was readmitted to our unit. At this time, the patient was receiving tacrolimus (4 mg/day) and prednisone (5 mg/day). Clinical examination revealed pain on movement and tenderness over the bone and joint line, without swelling

Table 1 Results of investigations before and after cladribine infusions

Patient 1				Patient 2	
First infusion		Second infusion			
Before	After	Before	After	Before	After
Proteinuria	12.25 g/24 h	4.2 g/24 h	5.2 g/24 h	4 g/24 h	7.2 g/24 h
Serum creatinine	190 µmol/l	120 µmol/l	154 µmol/l	149 µmol/l	243 µmol/l
Anti-ds DNA	132 IU/ml	58 IU/ml	292 IU/ml	171 IU/ml	49 IU/ml
C3	0.51 g/l	0.67 g/l	0.72 g/l	0.39 g/l	0.60 g/l
C4	0.13 g/l	0.14 g/l	0.15 g/l	0.12 g/l	0.16 g/l
C3d	22 units/ml	10 units/ml	23 units/ml	13 units/ml	12 units/ml
Urine analysis	red cells, hyalogramular, cellular casts			red cells, a few casts	

Reference ranges: serum creatinine 50-100 µmol/l, anti-dsDNA: 50-300 IU/ml positive, >300 IU/ml strongly positive, C3: 0.63-1.19 g/l, C4: 0.11-0.43 g/l, C3d: up to 12 units/ml.

Figure 1 Bone scintigraphy, showing increased uptake in both knees.

or increased temperature. She had no signs of autonomic vasomotor disturbances and articular mobility was normal. Examination of the remaining peripheral and axial joints was normal.

Blood tests showed creatinine levels of 1.3 mg/dl, calcium of 10.1 mg/dl, phosphate of 3.5 mg/dl and urate of 7.2 mg/dl. Other laboratory findings were normal. Patchy osteoporosis in the knees was seen radiographically. Bone scintigraphy showed intense uptake in both the osseous and vascular phases in the knees (fig 1). Calcitonin treatment was begun (three monthly cycles of 100 intramuscular units/day during 20 days) without clinical improvement. Because of the high serum concentrations of tacrolimus (15 µg/ml) and the ineffective calcitonin treatment, tacrolimus was reduced to 2 mg/day. Nine months after transplantation, she was free of symptoms and radiographs and tacrolimus concentration (5.1 µg/ml) were normal. Changes in plasma tacrolimus concentrations subsequent to the resolution of symptoms did not occur and the patient continued asymptomatic.

We describe a complication in a patient treated with tacrolimus after kidney transplantation that is similar to that described by other authors in transplanted patients treated with cyclosporin.⁴⁻⁶ Although the radiographic and bone scintigraphy findings suggested RSDS, the symptoms of this patient were not the classic features of this entity. The efficacy of corticosteroids in the treatment of uncomplicated RSDS has been demonstrated,⁴⁻⁵ so it is possible that corticosteroids might have a protective role against a full RSDS development, as she was treated with high doses of prednisone after the renal transplantation.

The early onset of symptoms after the administration of the drug and the clinical improvement after the reduction of the immunosuppressant dose, are features that support a possible relation between tacrolimus and the leg bone pain syndrome. The patient had high plasma tacrolimus concentrations at the onset of the clinical symptoms and the improvement appeared only when the drug doses went down. Although recurrence of knee symptoms with an increase in tacrolimus dose would be much stronger proof of this association, it is not ethically justifiable. Furthermore, she was treated with verapamil in addition to other drugs for controlling hypertension. Verapamil might have played a part in a possible increased risk for this clinical complication, because it decreases tacrolimus clearance.¹⁰ However, there are reports that calcium channel block-

ers (albeit of the dihydropyridine type) can improve the bone pain syndrome.¹¹

Although leg bone pain syndrome in kidney transplant patients who have received cyclosporin A is very rare, there are case reports described in the literature.⁴⁻⁶ To our knowledge, this is the first case of a renal transplant patient with pain in the lower limbs, related to tacrolimus treatment. Additional case reports are needed to support this association.

VIRGINIA VILLAVERDE
MIGUEL CANTALEJO
ALEJANDRO Balsa
EMILIO MARTIN MOLA
Department of Rheumatology, Hospital
Universitario La Paz, Madrid, Spain

AURELIO SANZ
Department of Nephrology, Hospital
Universitario La Paz, Madrid, Spain

Correspondence to: Dr V Villaverde, Rheumatology Unit, Hospital Universitario La Paz. Paseo de la Castellana 265, 28046 Madrid, Spain.

- Muñoz-Gómez J, Bergada E, Gómez R, Llopert E, Subías R, Sole M. Amyloid arthropathy in patients undergoing periodical haemodialysis for chronic renal failure. *Ann Rheum Dis* 1985;44:729-33.
- Bonomini V, Bortolotti GC, Feletti C, Scolari MP. Serial histomorphometric and histochemical bone biopsy studies in dialysis and transplantation. *J Urol Nephrol* 1975;12:941-50.
- Barbosa LM, Gauthier VJ. Bone pain in transplant patients. *Arthritis Rheum* 1993;36 (suppl):91.
- Muñoz-Gómez J, Collado A, Gratacos J, Campistol JM, Lomeña F, Llena J, et al. Reflex sympathetic dystrophy syndrome of the lower limbs in renal transplant patients treated with cyclosporin A. *Arthritis Rheum* 1991;34:625-30.
- Naredo E, Balsa A, Sanz A, Pantoja L, Martín Mola E, Gijón Baños J. Leg bone pain syndrome due to cyclosporin A in a renal transplant patient. *Clin Exp Rheumatol* 1994; 12:653-6.
- Dubost JJ, Fourcade J, Soubrier M, Ristori JM, Sauvezie B, Deteix P. Epiphyseal pain from cyclosporine in renal transplants. *Nephrologie* 1997;18:17-22.
- Sugiyama E, Suzuki H, Tunru I, Yamashita N, Hori T, Kobayashi M. FK506, an immunosuppressant partially inhibits interleukin 6 production by adherent rheumatoid synovial cells. *J Rheumatol* 1994;21:1597-601.
- Suzuki N, Kaneko S, Ichino M, Mihara S, Wakisaka S, Sakane T. In vivo mechanisms for the inhibition of T lymphocyte activation by long-term therapy with tacrolimus (FK506). *Arthritis Rheum* 1997;40:1157-67.
- Kozin F, Ryan L, Carrera G, Soin J, Wortmann R. The reflex sympathetic dystrophy syndrome (RSDS): scintigraphic studies, further evidence for the therapeutic efficacy of systemic corticosteroid and proposed diagnostic criteria. *Am J Med* 1981;70:23-30.
- Christians U, Schmidt G, Bader A, Lampen A, Schottmann R, Linck A, et al. Identification of drugs inhibiting the in vitro metabolism of tacrolimus by human liver microsomes. *Br J Clin Pharmacol* 1996;41:187-90.
- Redlich K, Pietschmann P, Stule T, Peterlik M. Comparative study on the effect of calcium channel blockers on basal and parathyroid hormone-induced bone resorption in vitro. *Pharmacol Toxicol* 1997;80:262-5.

Spleen haemorrhagic infarction and hazards of anticoagulation in Wegener's granulomatosis

In the largest cohort published to date, no splenic involvement is described in Wegener's granulomatosis (WG).¹ We report on two patients who required splenectomy for symptomatic splenic infarction in the course of WG.

CASE 1

A 42 year old man was admitted with an eight month history of arthritis and lower limb dysesthesia. Examination showed an acutely ill patient with a 39°C fever, oral ulcers, haemorrhagic gingival hyperplasia, bilateral haemorrhagic nasal discharge with crusts, diffuse necrotic purpura, neuritis, and black discoloration of some fingers and toes. The spleen was not palpable. Silent anterior myocardial infarction was diagnosed because of raised MB-CK levels and ST-segment increase with loss of R waves in leads V1,V2,V3 on electrocardiogram.² Antineutrophil cytoplasmic antibodies (c-ANCA) were disclosed in serum and necrotising vasculitis was shown on skin biopsy specimen.³ No antiphospholipid antibody or coagulation protein abnormality could be disclosed. Treatment consisted of intravenous administration of prednisolone, cyclophosphamide, sodium heparinate, diltiazem, dinitrosorbide and enalapril. His short-term course was uneventful. At day 14, the patient suddenly developed a severe haemorrhagic shock. Echotomography of the abdomen showed a splenic mass. At laparotomy, the spleen was almost disrupted by voluminous haematoma. Histological analysis of the spleen showed widespread necrotising vasculitis with haemorrhagic infarction. After five years of follow up, the patient is in complete remission with oral cotrimoxazole treatment.

CASE 2

A 23 year old young man was admitted in August 1996 because of repeated otitis media, sinusitis, epistaxis, headache, arthralgia with fever and weight loss. Despite a short course of oral corticosteroids and antibiotics, his general condition worsened. Antiproteinase 3 c-ANCA were disclosed in serum. Chest computed tomography showed pulmonary nodules. Intranasal endoscopic biopsies demonstrated necrotising vasculitis with epithelioid and giant cells. Treatment included oral prednisone and intravenous cyclophosphamide pulses. After a few days, serum creatinine concentrations abruptly increased to 198 µmol/l and urine analysis showed microscopic haematuria and proteinuria. High dose methylprednisolone pulses were then given, intravenous cyclophosphamide was changed to a 100 mg oral daily regimen and the patient eventually achieved remission. In October 1996, abdomen computed tomography showed an intrasplenic lesion that was consistent either with a splenic infarct or haematoma (fig 1). The later course was marked by a WG flare in January 1997, which was complicated with massive thrombosis of the left iliofemoral vein and the inferior vena cava. No thrombophilic disorder could be found. Intravenous heparin then oral anticoagulation with acenocoumarol were given. Because of persistent left hypocondrium tenderness, splenectomy was performed in September 1997. Histological examination showed splenic infarction with organised haematoma and sequelae of vasculitis (fig 2).

COMMENT

Because they are vessels without collateral flow, occlusion of distal parenchymal splenic arteries leads invariably to splenic infarction. Of note, two of the three patients described by Wegener in 1936 had spleen involvement.⁴ The frequency of spleen involvement ranges from 50% to 100% of WG cases at necropsy.⁵⁻⁷ Histological data frequently

Figure 1 Contrast enhanced upper abdominal computed tomography: large hypodense area with a peripheral rim of normally enhancing tissue.

showed massive or multiple areas of splenic necrosis, associated to a variable extent with central arteritis, splenic trabeculitis, follicular arteriolitis, disseminated parenchymatous granulomata and capsulitis (fig 2). Patients with splenic infarction in WG usually remain asymptomatic. Prominent splenomegaly is rare.⁹ With computed tomography, focal splenic infarction appears as well defined, peripheral wedge shaped areas of low attenuation. In WG, the diffuse vasculitis process often results in massive vasculature lesions involving the spleen parenchyma (fig 1).⁸⁻¹⁰ A peripheral rim of enhancement may be seen, as for spleen abscess, haematoma and lymphoma.¹¹ Splenectomy has been performed successfully in some patients.^{10 12 13} Spleen lesions may also appear to heal on repeated computed tomography under medical treatment only, consisting of prednisone and cyclophosphamide.¹⁴

Few recent reports of splenic involvement in WG provide histological analysis from live patients.^{10 12 13} Inaugural spontaneous rupture of a normal sized spleen with only subcapsular neutrophil infiltrate has been described in a patient who subsequently developed full blown WG.¹⁵ In another case, a spontaneous splenic haemorrhage was ascribed to vasculitis in a patient who had severe WG that required haemodialysis.¹³ In our two patients, microscopical study of the spleen also showed haemorrhagic infarction caused by specific WG related vasculitis

Figure 2 Cut gross pathological section of spleen shows changes corresponding to those seen by computed tomography. Large firm yellow (white) area consistent with infarction was surrounded by dark peripheral zone of splenic parenchyma.

process. A severe splenic haemorrhage occurred in patient 1, which was clearly related to both necrotising vasculitis and hypocoagulable state. Anticoagulation was indicated for inaugural myocardial infarction in case 1 and deep venous thrombosis in case 2, in both cases during active WG flare. Splenectomy was required in both our cases.

Our data suggest that antithrombotic treatment entails a specific risk of bleeding complications in patients with WG vasculitis. When anticoagulation is necessary in WG patients, computed tomography of the abdomen should be systematically performed and, if splenic infarction is disclosed, splenectomy should be considered.

THOMAS PAPO
DU LE THI HUONG
JEAN-CHARLES PIETTE

Internal Medicine, Hôpital Pitié-Salpêtrière,
Paris, France

MARC ANDRE
OLIVIER AUMAITRE
Internal Medicine, Hôpital Cébazat,
Clermont-Ferrand, France

FRÉDÉRIC CHARLOTTE
Histopathology, Hôpital Pitié-Salpêtrière, Paris,
France

JEAN-LOUIS KEMENY
Histopathology, Hôpital Cébazat,
Clermont-Ferrand, France

Correspondence to: Dr T Papo, Internal Medicine Unit, Hôpital Pitié-Salpêtrière, 83 Boulevard de l'Hôpital 75651 Paris cedex 13, France.

- Hoffman GS, Kerr GS, Leavitt RY, Hallahan CW, Lebovics RS, Travis WD, *et al.* Wegener granulomatosis: an analysis of 158 patients. *Ann Intern Med* 1992;116:488-98.
- Papo T, Piette JC, Laraki R, Bletty O, Le Thi Huong Du, Godeau P. Silent myocardial infarction in Wegener's granulomatosis. *Ann Rheum Dis* 1995;54:233-4.
- Leavitt RY, Fauci AS, Bloch DA, Michel BA, Hunder GG, Arend WP, *et al.* The American College of Rheumatology 1990 criteria for the classification of Wegener's granulomatosis. *Arthritis Rheum* 1990;33:1101-7.
- Wegener F. Über eine eigenartige rhinogene granulomatose mit besonderer beteiligung des arteriensystems und der nieren. *Beitr Pathol Anat* 1939;102:36-68.
- Fahey JL, Leonard E, Chung J, Godman G. Wegener's granulomatosis. *Am J Med* 1954;17:168-78.
- Pinching AJ, Lockwood CM, Pussell BA, Rees AJ, Sweny P, Evans DJ, *et al.* Wegener's granulomatosis: observations on 18 patients with severe renal disease. *Q J Med* 1983;208:435-60.
- Walton EW. Giant-cell granuloma of the respiratory tract (Wegener's granulomatosis). *BMJ* 1958;2:265-70.
- McHugh K, Manson D, Eberhard BA, Laxer RM, Shore A. Splenic necrosis in Wegener's granulomatosis. *Pediatr Radiol* 1991;21:588-9.
- Fonner BT, Nemcek AA, Boschman C. CT appearance of splenic infarction in Wegener's granulomatosis. *AJR* 1995;164:353-4.
- Ruel M, Bobrie G, Jarrousse B, Henry-Biabaud E. Iconographie splénique au cours de la granulomatose de Wegener. *Presse Med* 1989;18:725.
- Kalaitzoglou I, Drevelengas A, Paladas P, Asimaki A. MRI appearance of pulmonary Wegener's granulomatosis with concomitant splenic infarction. *Eur J Radiol* 1998;8:367-70.
- Morayati SJ, Fink-Bennet D. Indium-111 leukocyte scintigraphy in Wegener's granulomatosis involving the spleen. *J Nucl Med* 1986;27:1864-6.
- Kettritz R, Anders S, Kettritz U, Schneider W, Göbel U, Luft FC. Spontaneous splenic hemorrhage in a patient with Wegener's granulomatosis. *Am J Kidney Dis* 1998;5:860-2.
- Gregorini G, Campanini M, Tira P, Lancini L, Tincani A, Majorca R. Spleen involvement in Wegener's granulomatosis: two case reports. *APMIS* 1990;S19:23.

- Franssen CFM, Ter Maaten JC, Hoorntje SJ. Spontaneous splenic rupture in Wegener's vasculitis. *Ann Rheum Dis* 1993;52:314.

Amiodarone induced lupus

Lupus related to amiodarone has not previously been described. We report on a patient who developed drug induced lupus (DIL) in association with amiodarone treatment. To our knowledge, this is the first report of amiodarone induced lupus (CD ROM: Medline, USA National Library: 1966-98).

A 71 year old white woman was admitted because of two weeks of pleuritic chest pain, dyspnea on exertion, and non-productive cough. She had malaise, intermittent fever, arthralgia, and weight loss for more than six months. There was no history of Raynaud's phenomenon, oral ulcers or photosensitivity. She had a six year history of arterial hypertension and atrial fibrillation treated with amiloride, digital and amiodarone (200 mg two times daily) for the past two years.

Physical examination disclosed malar rash, an aortic systolic murmur (grade II/VI), and hypoventilation in both pulmonary bases. Laboratory studies showed an erythrocyte sedimentation rate of 90 mm 1st h. Peripheral blood examination revealed a mild normochromic and normocytic anaemia (10 g/dl), normal white blood cells count (4000/μl), with lymphopenia (20 per cent), and normal platelets count (180 000/μl). Coagulation tests were normal. All serum chemistries, including thyroid function tests, creatinine phosphokinase, immunoglobulins, complement levels, and urine analysis were within normal limits. Coombs's tests were negative. Rheumatoid factor was 1:320. Circulating immune complexes (IgG-C1q) were positive. Antinuclear antibodies (ANAs) were positive at 1:640; anti-Ro, anti-La, anti-dsDNA, anti-Sm, anti-histone antibodies, antiphospholipid antibodies, cryoglobulins, C reactive protein, VDRL and Mantoux test were negative. Blood and urine cultures were negative. Electrocardiogram was within normal limits, and the two dimensional echocardiogram showed mild aortic stenosis. Chest radiography revealed bilateral pleural effusions, without fibrosis or cardiomegaly. Pleural fluid was exudative, with lymphocytic predominance, without cytological features for malignancy. Cultures of pleural fluid for bacteria, including for *Mycobacterium tuberculosis*, were negative. Bone scan with technetium-99m showed increase uptake in hands, elbows, and knees. The histopathological examination of biopsy specimens of the skin, including indirect immunofluorescence stain, muscle and temporal artery did not show abnormal features.

The amiodarone was stopped and the patient progressively improved. No corticosteroids were given. On the third week she developed a transient relapse, with fever, malaise and with evidence of unilateral pleural effusion. One year after no clinical, analytical or radiological findings were present, and three years later she still remained free of symptoms, and the ESR, complete blood count, and radiological data were normal. The titre of ANA decreased but remained weakly positive at 1:40.

It is estimated that 3-7% of all patients with systemic lupus erythematosus (SLE) might have DIL.¹ Clinical features of SLE and DIL are similar but there are certain distinguishing characteristics between them: the

patients with DIL are usually older; the prevalence of men and women is similar; and the presenting symptoms are usually mild, with the patient usually complaining of malaise, fever and arthralgia, with or without arthritis, while skin, central nervous system or renal involvement is rare. Pleuropericardial disease is frequent and, as in classic SLE, anaemia and leucopenia may be present. Serum complement components are usually normal, ANAs are positive but anti-dsDNA and anti-Sm are negative, while anti-histones antibodies can be detected in most of patients.²

The pathogenic mechanisms proposed for DIL include: cross reactivity between drug and the nucleic acid; hapten complex formation between drug and nucleic acid, or structural damage to the chromosomal DNA; action of drug as an adjuvant or immunostimulant, which, in concert with appropriate immune response genes, triggers polyclonal B/T cell activation; and interference with the complement pathway.¹

The incidence of side effects associated with amiodarone ranges from 40% to 93% and, in most of cases, these side effects are consequence of its potential to be directly toxic to several organ systems.^{3,4} However, there is also some evidence of immunologically mediated phenomena related to amiodarone. A positive skin and basophil degranulation tests with amiodarone, secretion of leucocyte inhibitory factor, positive lymphoblastic transformation and circulation of a specific antibody of the IgG class have been described.^{5,6} Moreover, several studies suggest that various biological and immunological markers of "systemic" disease activity are present in patients taking this drug. Circulating immune complexes, ANAs, and non-specific increase in ESR and white blood cell count, sometimes with eosinophilia, are common findings.^{7,8}

Low ANA titre is not uncommon in an elderly patient. However, spontaneous SLE in elderly people is not usual and DIL must always be considered in the differential diagnosis. This case, presenting with malaise, fever, arthralgia, circulating immune complexes, and autoantibodies strongly suggests an immunological underlying condition. Moreover, this patient meets four SLE criteria: malar rash, serositis, haematological disorder (lymphopenia), and positive ANAs test. Imputability criteria of amiodarone induced lupus are present on a semiological basis with classic features of DIL and on a chronological basis with disappearance of most of the symptoms after amiodarone withdrawal. The relapse could be explained because of the long elimination half time of the drug⁹ and, in consequence, the immune response might progress despite discontinuation of the treatment.

RUI SUSANO

LUIS CAMINAL

DAVID RAMOS

BERNARDINO DÍAZ

Department of Medicine, Hospital Central de Asturias (University Centre), Oviedo, Spain

Correspondence to:

Dr R Susano, Internal Medicine, Hospital Central de Asturias, C/ Julian Claveria s/n, 33006, Oviedo, Spain.

1 Faubert PF, Porush JG, Venkatesh VS, Grishman E. Lupus-like syndromes. In: Grishman E, Churg J, Needle MA, Venkatesh VS, eds. *The kidney in collagen-vascular diseases*. New York: Raven Press, 1993:87–119.

- 2 Dubois EL, Wallace DJ. Drugs that exacerbate and induce systemic lupus erythematosus. In: Wallace DJ, Dubois EL, eds. *Dubois's lupus erythematosus*. Philadelphia: Lea and Febiger, 1987:450–60.
- 3 Raeder EA, Podrid PJ, Lown B. Side effects and complications of amiodarone therapy. *Am Heart J* 1985;109:975–83.
- 4 Martin WJ, Rosenow EC. Amiodarone pulmonary toxicity. Recognition and pathogenesis (Part I). *Chest* 1988;93:1067–74.
- 5 Akoun GM, Gauthier-Rahman S, Milleron BJ, Perrot JY, Mayaud CM. Amiodarone-induced hypersensitivity pneumonitis. Evidence of an immunological cell-mediated mechanism. *Chest* 1984;84:133–5.
- 6 Fan K, Bell R, Eudy S, Fullenwider J. Amiodarone-associated pulmonary fibrosis. Evidence of an immunologically mediated mechanism. *Chest* 1987;94:625–30.
- 7 Manzano L, Yebra M, Merino F, Lladós G, Diego F, Durántez A. Afección pulmonar por la amiodarona y anticuerpos antinucleares. *Rev Clin Esp* 1986;179:80–1.
- 8 Harris L, McKenna WJ, Rowland E, Holt DW, Storey GC, Krikler DM. Side effects of long term amiodarone therapy. *Circulation* 1983; 67:45–51.
- 9 Andreasen P, Agerback H, Bjerregard P, Gotzsche H. Pharmacokinetics of amiodarone after intravenous and oral administration. *Eur J Clin Pharmacol* 1981;19:293–9.

Antinuclear antibodies in relapsing polychondritis

The prevalence of antinuclear antibodies (ANA) in relapsing polychondritis (RP) has been recently reported by Zeuner *et al* to be as high as 66%,¹ usually in a low titre with a speckled pattern. We report here on our experience of ANA testing in patients with RP.

The charts of 180 patients followed up in our institution fulfilling the criteria for RP proposed by Michet *et al*² have been recently retrospectively reviewed, with special focus on dermatological manifestations and their relation with myelodysplasia.^{3,4} This aim led us to exclude 36 patients because the association of RP with potentially confounding diseases,⁵ such as systemic lupus erythematosus (SLE) present in nine, mixed connective tissue disease (MCTD) in five, rheumatoid arthritis in three, Takayasu arteritis in three, mesenteric panniculitis in three, spondyloarthropathy in two, Crohn's disease in two, psoriasis in two, or Lichen planus in two. Among the 144 patients remaining, 111 have been tested for ANA by using either Hep-2 cells or liver sections as substrate, or both. Most patients had repeated ANA determinations, including initial testing before onset of corticosteroid treatment, and the higher titres were considered for analysis. Figure 1 shows the histogram of ANA positivity according to ANA titre.

Figure 1 Prevalence of ANA in "pure" RP according to ANA titre.

ANA were either absent or present in low titres in a majority of patients (73% and 18%, respectively). "Significant" titres—that is, titre > 1/100 ANA—were demonstrated in only 10 of 111 patients (9%). The pattern of fluorescence was as follows: homogeneous in five, homogeneous and speckled in two, speckled and nucleolar in one, perinuclear and nucleolar in one, and not provided in one. Among those 10 patients, five had clinical or ophthalmological features, or both, suggestive of an associated Sjögren's syndrome including two who had antibodies to both SS-A and SS-B, and two others had a myelodysplastic syndrome.⁴ None of these 10 patients had antibodies to ds-DNA. Using the same methods, ANA > 1/100 were found in 15 of the 36 patients initially excluded (42%). Among the nine with SLE, ANA > 1/100 and antibodies to ds-DNA (by Farr assay or Crithidia fluorescence) were found in eight patients. All five patients with MCTD had ANA > 1/1000 (in a speckled pattern in four), with positive antibodies to RNP and negative tests for ds-DNA. The two remainders had either rheumatoid arthritis, or Lichen planus associated with multinodular goitre; both of them also had features suggestive of sicca syndrome. Therefore, within these 36 patients, strong ANA positivity was mainly restricted to SLE, MCTD or Sjögren's syndrome, or all three.

Beside the recent article by Zeuner *et al*,¹ the prevalence of ANA has rarely been reported in RP. McAdam *et al* found positive ANA in four of their 18 patients tested (22%), and noted 3 of 23 (13%) ANA positivity in a literature review.⁵ Data regarding ANA were not provided in the large series of patients with RP followed up at the Mayo Clinic.^{2,6} The low prevalence of ANA observed in our cases with "pure" RP—that is, RP not associated with another connective tissue disorder except for a possible Sjögren's syndrome—agrees with the negative results of tests for IgG antinucleosome antibodies recently reported by our group in this condition.⁷

We conclude that: (a) the prevalence of ANA observed in RP is low and, (b) as suggested by other authors^{6,8} the finding of a significant titre of ANA in a patient with RP strongly suggests the presence of an associated disorder, such as SLE, MCTD, Sjögren's syndrome or acquired myelodysplasia.

JEAN-CHARLES PIETTE

ROULA EL-RASSI

ZAHIR AMOURA

Internal Medicine Unit, Hôpital Pitié-Salpêtrière, 83 Boulevard de l'Hôpital, 75651 Paris Cedex 13 France

Correspondence to: Dr J-C Piette.

- Zeuner M, Straub RH, Rauh G, Albert ED, Schölmerich J, Lang B. Relapsing polychondritis: clinical and immunogenetic analysis of 62 patients. *J Rheumatol* 1997;24:96–101.
- Michet CJ Jr, McKenna CH, Luthra HS, O'Fallon WM. Relapsing polychondritis. Survival and predictive role of early disease manifestations. *Ann Intern Med* 1986;104:74–8.
- Piette JC, El-Rassi R, Wechsler B, Laporte JL, Rybojad M, Francès C. Dermatological manifestations in relapsing polychondritis. Association with myelodysplasia. [Abstract]. *Arthritis Rheum* 1996;39 (suppl 9):S230.
- Piette JC, Papo T, Chavanon P, Du LTH, Francès C, Godeau P. Myelodysplasia and relapsing polychondritis. *J Rheumatol* 1995;22:1208–9.
- McAdam LP, O'Hanlan MA, Bluestone R, Pearson CM. Relapsing polychondritis. Prospective study of 23 patients and a review of the literature. *Medicine (Baltimore)* 1976;55:193–215.
- Isaak BL, Liesegang TJ, Michet CJ Jr. Ocular and systemic findings in relapsing polychondritis. *Ophthalmology* 1986;93:681–9.
- Amoura Z, Cacoub P, Chabre H, Amoura I, Wechsler B, Musset L, et al. Prevalence of antinucleosome IgG in connective tissue diseases. [Abstract]. *Lupus* 1998;7 (suppl 1):82.
- Trentham DE, Le CH. Relapsing polychondritis. *Ann Intern Med* 1998;129:114–22.

There is no association between polymyalgia rheumatica and acute parvovirus B19 infection

Parvovirus B19 has been associated with a growing number of diseases. Besides the frequent manifestations such as erythema infectiosum, aplastic crisis in persons with underlying haemolytic anaemia, hydrops fetalis in pregnant women and acute or chronic arthritis a range of rather rare diseases have been described in recent reports.¹ Among them are case reports on persistent parvovirus B19 infection in immune incompetent people, encephalitis, myocarditis, systemic lupus erythematosus (reviewed by Anderson and Young²) and rheumatoid arthritis.³ Furthermore, parvovirus B19 has been suspected to play a part in the aetiology of polymyalgia rheumatica (PMR).^{3,4} Because of the acute onset of PMR and its systemic symptoms an infectious cause may be a relevant factor. Additionally, autoimmune processes have been demonstrated in both, PMR and parvovirus B19 infection.^{5,6} As the receptor for parvovirus B19, the P-blood group antigen (globoside), is also present on endothelial cells an interrelation between parvovirus B19 and giant cell arteritis or PMR may be possible.

Parvovirus B19 can only replicate in erythroid precursor cells in human bone marrow, but it is known that infection of cells non-permissive for viral replication leads to an excess production of the viral non-structural protein (NS1) without production of capsid proteins.⁷ As the NS1 protein is cytotoxic and able to induce apoptosis, it probably plays a part in the pathogenetic process of the parvovirus B19 induced tissue damage. This is confirmed by the fact that antibodies against NS1 of parvovirus B19 are preferentially produced during chronic or persistent parvovirus B19 infections, for example in parvovirus B19 associated chronic arthritis.^{8,9}

To test the hypothesis whether PMR is associated with acute parvovirus B19 infection, we tested the seroprevalence of IgG antibodies against the two structural proteins VP1 and VP2 and against the non-structural protein NS1 in 110 PMR patients (patients

Figure 1 Seroprevalence of IgG antibodies against VP1/VP2 (black bars) and NS1 (open bars) in control subjects and patients with polymyalgia rheumatica (PMR). The number of healthy controls in each subgroup of normal subjects was at least 20. * $p=0.057$ v seroprevalence of IgG NS1 in age matched healthy controls (groups 56–65 and 66–75 with 42 controls v 110 patients with PMR).

with giant cell arteritis excluded; mean (SEM) age 67.0 (0.8) years, range: 48–77) and, for comparison, in 135 healthy controls of different ages. At the time point of blood sampling (median disease duration at the time point of blood sampling: 0.6 years, range: 0–7.3, mean (SEM): 1.4 (0.2) years), 35 patients had no corticosteroids and 75 patients received on an average 15.2 (1.8) mg prednisolone/day. Furthermore, we investigated the interrelation between age, symptoms¹⁰ or laboratory parameters and the presence of NS1 specific antibodies in healthy controls and patients with PMR. Non-parametric Kruskal-Wallis one way analysis was used to compare means of different subgroups. The significance level was $p<0.05$.

Subjects in the control group had various ages between 18 to 75 years. Overall seroprevalence of IgG against the capsid proteins VP1 and VP2 was 78% (fig 1). Overall IgG seroprevalence against VP1 and VP2 was 88% in patients with PMR (not significantly different versus the age matched control group). With respect to the NS1 IgG antibody, overall seroprevalence in the control group was 22% (fig 1) and in patients with PMR 20% ($p=0.057$ versus the age matched control group: 10%; fig 1).

Furthermore, we investigated the association between the presence of NS1 IgG antibodies and PMR related symptoms¹⁰ or laboratory parameters (patients with NS1 as compared with patients without NS1 IgG antibodies were not different in age, sex, and medication). The symptoms were assessed using standard record forms from the medical histories (at the time serum was collected).¹⁰ We asked the patients for muscular pain in the left/right shoulder, left/right upper arms, left/right neck, left/right gluteal muscle, and left/right thigh. If one muscle group was painful, the corresponding item was scored with one point (the sum of the item points was the overall muscle score).¹⁰ In PMR patients with NS1 IgG as compared with patients without NS1 IgG, arthralgia was more frequent (with versus without: 73% v 40%, $p=0.024$). However, the overall muscle score was lower in NS1 positive than in NS1 negative patients (0.5 (0.2) SEM v 1.6 (0.3) SEM score points; $p=0.021$). With respect to other PMR related symptoms, no

significant differences were found. In patients with a positive NS1 IgG antibody, interleukin 6 (4.6 (0.9) SEM v 11.3 (2.2) SEM; $p=0.037$) and soluble ELAM (48.2 (4.8) SEM v 71.4 (5.2) SEM; $p=0.024$) were significantly lower as compared with patients without NS1 IgG. No significant differences were found with respect to erythrocyte sedimentation rate, C reactive protein, tumour necrosis factor, interleukin 2, and interleukin 1 β .

In view of these data, there was a positive association between NS1 and arthralgia. However, other symptoms and parameters of inflammation such as erythrocyte sedimentation rate were not associated with the presence of NS1 IgG. Furthermore, disease related immune mediators such as interleukin 6 or soluble ELAM were lower in patients with as compared with patients without NS1 IgG. As a positive NS1 IgG titre indicates an active infection, an acute parvovirus B19 infection does not seem to be a pathogenetic factor in our patients with PMR.

ANDREA HEMAUER
SUSANNE MODROW

Institute for Medical Microbiology and Hygiene,
University Medical Centre Regensburg,
Germany

JOACHIM GEORGI

Department of Internal Medicine, Ostseelinklinik,
Damp, Germany

KLAUS HELMKE

Department of Internal Medicine IV, Hospital
Munich-Bogenhausen, Germany

PETER VAITH

Department of Internal Medicine, University
Medical Centre Freiburg, Germany

BERNHARD LANG

Rheumazentrum Baden-Baden, Germany

JÜRGEN SCHÖLMERICH

RAINER H STRAUB
Department of Internal Medicine I, University
Medical Centre Regensburg, Germany

Correspondence to: Dr R H Straub, Department of
Internal Medicine I, University Medical Centre,
D-93042 Regensburg, Germany.

- Anderson LJ, Young NS. Human parvovirus B19. In: Parks WP, ed. *Monographs in virology*. Vol 20. New York: Karger, 1997.
- Murai C, Munakata Y, Takahashi Y, Ishii T, Shibata S, Muryoi T, et al. Rheumatoid arthritis after human parvovirus B19 infection. *Ann Rheum Dis* 1999;58:130–2.
- Cimmino MA. Genetic and environmental factors in polymyalgia rheumatica. *Ann Rheum Dis* 1997;56:576–7.
- Cimmino MA, Negrari A, Grazi G, Franconi G, Accardo S. Parvovirus B19 infection in polymyalgia rheumatica (PMR). [Abstract]. *Arthritis Rheum* 1997;40:238.
- Kerr JR, Boyd N. Autoantibodies following parvovirus B19 infection. *J Infect* 1996;32:41–7.
- Soloninka CA, Anderson MJ, Laskin CA. Anti-DNA and antilymphocyte antibodies during acute infection with human parvovirus B19. *J Rheumatol* 1989;16:777–81.
- Liu JM, Green SW, Shimada T, Young NS. A block in full length transcript maturation in cells nonpermissive for B19 parvovirus. *J Virol* 1992;66:4686–92.
- von Pöblitzki A, Gigler A, Lang B, Wolf H, Modrow S. Antibodies to parvovirus B19 NS-1 protein in infected individuals. *J Gen Virol* 1995;76:519–27.
- von Pöblitzki A, Hemauer A, Gigler A, Puchhammer-Stöckl E, Heinz FX, Pont J, et al. Antibodies to the nonstructural protein of parvovirus B19 in persistently infected patients: Implications for pathogenesis. *J Infect Dis* 1995;172:1356–9.
- Straub RH, Herfarth HH, Rinkes B, Konecna L, Glück T, von Landenberg P, et al. Favorable role of interleukin 10 in patients with polymyalgia rheumatica. *J Rheumatol* 1999;26:1318–25.

Neutrophil chemotaxis in Behçet's syndrome

It has been suggested that the marked cellular inflammatory response, which characterises Behçet's syndrome (BS), may be attributable to increased neutrophil locomotion.¹⁻⁴ However, others disagree.^{5,6} We have re-evaluated chemotaxis of polymorphonuclear leucocytes (PMNs) in BS among a greater number of patients in a controlled setting.

Fifty four male BS patients, nine male patients with ankylosing spondylitis, eight with psoriasis and 37 male healthy controls were studied with 28 female patients with BS and 16 healthy female controls. Behçet patients with severe disease were those with active major vessel and/or eye involvement.

We measured chemotaxis with the "under the agarose method".^{7,8} The measurements were masked with the assessors not knowing the diagnoses. An inverted microscope fitted with an ocular micrometre disc to measure the migration of neutrophils from middle wells to outer (chemotaxis) and inner wells (chemokinesis) was used. Zymosan activated sera (patients or controls) were used as a source of C5a. Results were expressed as micrometre square (1 mm=8 squares). Additionally the plates were evaluated macroscopically for observation of the migration between neutrophil wells.

Tables 1 and 2 show the results. There were no significant differences between the chemotactic indices of the various groups of patients and controls studied of either sex. Maximal chemotaxis rates in the groups varied from 67% to 100%.

The Boyden millipore filter system has extensively been used for chemotaxis experiments.^{9,10} The agarose method is simple and cheap. This method can preferentially be used to differentiate chemokinesis from chemotactic migration.⁷

There is marked heterogeneity in disease expression in men and women in BS¹¹ and we reasoned that some of the confusion in the literature about neutrophil activity might be related to this. Thus we analysed our data separately for either sex. Although there was a tendency for male patients with severe disease to have higher chemotactic indices this was not statistically significant (p=0.62). We did not study any diseased controls for female patients with BS.

Abdulla and Lehner⁶ observed decreased chemotaxis in BS. Fordham *et al.*² on the other hand reported increased chemotaxis, but normal random migration. While Wilkinson,⁵ similar to our experience, observed normal chemotaxis in BS, more recently Carletto *et al*³ reported augmented chemotaxis especially in the active phases of the disease. Finally, Ben Ezra *et al.*, among a group of Behçet patients with uveitis could demonstrate increased chemotactic activity only among a few of these patients, compared with that observed among patients with other forms of uveitis. They concluded that increased chemotactic activity was

Table 2 Chemotactic indices in women*

Groups	Number	Mean (SD)
Behçet's	28	7.2 (8.2)
Healthy controls	16	10.1 (10.2)

*t=1.032, p>0.05.

not a regular feature of ocular BS (personal communication).

In vivo assays do not differentiate chemotaxis from chemokinesis. In the Carletto study clinically active Behçet patients demonstrated increased chemotaxis to sera by Senn's modified in vivo assay.³ Others had found hyperchemotaxis to neutrophil cytoplasmic fractions again by using an in vivo assay.⁴ Although it is difficult to compare the results of in vitro and in vivo assays, we thought these reported increases might have resulted from increased chemokinesis. In our experiments we observed maximal chemotaxis (3 mm) frequently, however we did not find any significant differences in chemotactic indices between diseased and healthy subjects.

An interesting aspect of our study was the migration between neutrophil wells that was observed in many of the Petri dishes. This was observed even though we had not used cellular materials as chemotactic agents. Presumably the gravity of the cellular materials overcame the chemical gradient of zymosan activated sera in some Petri dishes. Because of the observed migration between neutrophil wells, we suggest that there should be only one "triple well rank" in a Petri dish. On the other hand our method of preincubation of the whole blood for 45 minutes at 37°C before harvesting the PMNs (intended for better viability) might have been responsible for this phenomenon by increasing the chemotactic activity in all groups studied. Further studies are needed to clarify these issues.

BINNUR TÜZÜN
Department of Dermatology, Trakya University
Medical Faculty, Edirne, Turkey

YALÇIN TÜZÜN
CEM MAT
Department of Dermatology, Istanbul University
Cerrahpasa Medical Faculty, Istanbul, Turkey

SEBAHATTİN YURDAKUL
VEDAT HAMURYUDAN
HASAN YAZICI
Department of Rheumatology, Istanbul
University Cerrahpasa Medical Faculty,
Istanbul, Turkey

YILMAZ ÖZYAZGAN
Department of Ophthalmology, Istanbul
University Cerrahpasa Medical Faculty,
Istanbul, Turkey

Correspondence to: Dr B Tüzün, Büyükciftlik Sokak Belde Apt 24/3, Nisantasi, Istanbul 80200, Turkey.

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1 Efthimiou J, Addison IE, Johnson BV. In vivo leukocyte migration in Behçet's syndrome. *Ann Rheum Dis* 1989;48:206-10.
2 Fordham JN, Davies PG, Kirk A, Currey HL. Polymorphonuclear function in Behçet's syndrome. *Ann Rheum Dis* 1982;41:421-5.
3 Carletto A, Pacor ML, Biasi D, Caramaschi P, Zeminians S, Bellavite P, *et al.* Changes of neutrophil migration without modification of in vitro metabolism and adhesion in Behçet's disease. *J Rheumatol* 1997;24:1332-6.
4 Takeuchi A, Kobayashi K, Mori M, Mizushima Y. The mechanism of hyperchemotaxis in Behçet's disease. *J Rheumatol* 1981;8:40-4.
5 Wilkinson PC. Comments on enhanced leukocyte locomotion in Behçet's disease. In: Lehner

T, Barnes CG, eds. *Recent advances in Behçet's disease*. London: Royal Society of Medicine Services, International Congress and Symposium Series, no 103, 1986:97-8.
6 Abdulla YH, Lehner T. The effect of immune complexes on chemotaxis in Behçet's syndrome and recurrent oral ulcers. In: Lehner T, Barnes CG, eds. *Behçet's syndrome*. London: Academic Press, 1979:55-75.
7 Nelson RD, Bauman MP, Gracyk JL, Fiegel VD, Herron MJ. Leukocyte chemotaxis: migration under agarose method. In: Douglas SD, Quie PG, eds. *Investigation of phagocytes in disease*. Edinburgh: Churchill-Livingstone, 1981:20-31.
8 Nelson RD, Quie PG, Simmons RL. Chemotaxis under agarose: A new and simple method for measuring chemotaxis and spontaneous migration of human polymorphonuclear leucocytes and monocytes. *J Immunol* 1975;115:1650-6.
9 Fernandes AC, Anderson R, Ras GJ. An objective filter-based, enzymatic method for the in vivo measurement of the migration of human polymorphonuclear leukocytes. *J Immunol Methods* 1985;83:259-71.
10 Fernandez HN, Henson PM, Otani A, Hugli TE. Chemotactic response to human C3a and C5a anaphylatoxins. *J Immunol* 1978;120:109-15.
11 Yazici H, Tüzün Y, Pazarli H, Yurdakul S, Özyazgan Y, Özdoğan H, *et al.* Influence of age of onset and patient's sex on the prevalence and severity of manifestations of Behçet's syndrome. *Ann Rheum Dis* 1984;43:783-7.

MATTERS ARISING

Neuropsychiatric systemic lupus erythematosus

The considerable difficulties in making sense of the literature on patients with lupus involving the central nervous system are re-emphasised in the paper by Rood *et al.*¹ The authors, who to be fair take a sensibly cautious approach to their results, nevertheless seek to persuade us that the IL10 locus is associated with neuropsychiatric lupus on the basis of a historical case notes review of 42 lupus patients with neuropsychiatric disease, compared with 50 who lack such involvement.

Their conclusion needs to be treated with caution. Does it make sense to lump together 42 highly diverse patients and make the kind of claim they have made? The authors suggest that CNS lupus is attributable to either antiphospholipid antibody related thrombotic events, or "immune mediated" disease. This division is artificial. There is a considerable literature on CNS lupus that proposes that a wide variety of immunopathogenic mechanisms may be responsible in individual cases. These mechanisms include thrombotic effects, which may be linked to antiphospholipid antibodies, a true vasculitis, a cross reaction between antibodies that recognise the lymphocyte surface targets and neurological antigens, and antibodies to a wide variety of neurological targets. A considerably larger number of patients will have to be studied before any claims of links to an IL10 promoter haplotype can be truly convincing.

We agree with the authors that patients with SLE have a higher innate production of IL10 than controls. However, as there is no significant difference in the frequency of the IL10 promoter single nucleotide polymorphisms (SNP)s in SLE patients when

Table 1 Chemotactic indices in men*

Groups	Number	Mean (SD)
Behçet's	54	6.4 (8.0)
Severe Behçet's	9	11.3 (12.1)
Ankylosing spondylitis	9	5.9 (7.1)
Psoriasis	8	10.8 (11.0)
Healthy controls	37	10.0 (10.3)

*Kruskal-Wallis one way analysis of variance, corrected for ties: $\chi^2=2.1381$; DF=4; p>0.05.

compared with controls in their study, we suspect that the difference in IL10 production is not attributable to functional difference between patients with SLE and controls in terms of the IL10 SNP alleles frequencies.²⁻⁴ Differences have been described with respect to microsatellites⁵ and one awaits confirmation from other populations or family studies. To our knowledge, a difference in IL10 production between patients with neuropsychiatric disease SLE and non-neuropsychiatric disease SLE has not been described. The described associations would be biologically meaningless if IL10 production is similar between these two groups.

The authors suggest that the -1082A allele is associated with a higher innate IL10 production, however, they appear to ignore the only published study to date that showed that the A allele was associated with lower IL10 production.⁶ In addition we have confirmed that the A allele is associated with lower IL10 production in transient transfection studies and the ATA/ATA genotype is associated with lower IL10 production in whole blood culture.⁴ The increase in the A allele is mainly accounted for by an increase in the ATA haplotype in their neuropsychiatric disease patients and therefore they are describing an association with a low IL10 producing haplotype, not a high IL10 producing haplotype. One interpretation of this would be that patients with neuropsychiatric disease symptoms are unable to adequately control inflammation from a variety of different pathological mechanisms because of low IL10 production.

DAVID ISENBERG

Centre for Rheumatology, UCLH, London

ESTHER CRAWLEY

Department of Molecular Pathology, UCLMS

PAT WOO

Department of Molecular Pathology, UCLMS

- 1 Rood MJ, Keijsers V, van der Linden MW, Tong TQT, Borggreve SE, Verweij CL, *et al.* Neuropsychiatric systemic lupus erythematosus is associated with imbalance in interleukin 10 promoter haplotypes. *Ann Rheum Dis* 1999;58:85-9.
- 2 Mok CC, Lanchbury JS, Wai Chan D, Sing Lau C. Interleukin 10 promoter polymorphisms in southern Chinese patients with systemic lupus erythematosus. *Arthritis Rheum* 1998;41:1090-5.
- 3 Lazarus M, Hajeer AH, Turner D, Sinnott P, Worthington J, Ollier WER, *et al.* Genetic variation in the interleukin 10 gene promoter and systemic lupus erythematosus. *J Rheumatol* 1997;24:2314-17.
- 4 Crawley E, Kay R, Sillibourne J, Patel P, Hutchinson I, Woo P. Polymorphic haplotypes of the interleukin-10 5' flanking region determine variable interleukin-10 transcription and are associated with particular phenotypes of juvenile rheumatoid arthritis. *Arthritis Rheum* (in press).
- 5 Mehrian R, Quismorio FP, Strassmann G, Stimmler MM, Horwitz DA, Kitridou RC, *et al.* Synergistic effect between IL-10 and bcl-2 genotypes in determining susceptibility to systemic lupus erythematosus. *Arthritis Rheum* 1998;41:596-602.
- 6 Turner DM, Williams DM, Sankaran D, Lazarus M, Sinnott PJ, Hutchinson IV. An investigation of polymorphism in the interleukin-10 gene promoter. *Eur J Immunogenet* 1997;24:1-8.

Authors' reply

We thank Drs Isenberg, Crawley and Woo for their interest in our paper.¹

They argued that the dichotomy of the pathogenesis of CNS lupus in "immune

mediated" and thromboembolic disease is too rigid, because a wide variety of immunopathogenetic mechanisms can be deemed responsible for CNS lupus. As the hallmark of SLE is the production of autoantibodies, it seems to be justified to assume that the pathogenesis underlying CNS lupus is B cell mediated. Based upon this assumption we clustered the individual neuropsychiatric disease SLE patients and tested the hypothesis that a genetic marker in the promoter of the IL10 gene is associated with the phenotype of CNS-SLE.

In general, a positive result in a genetic association study is only possible after a correct definition of the phenotype. After all, if the phenotype is inadequately defined, the magnitude and statistical significance of the association will be less or lost because of the random distribution of the genetic marker in the misclassified patients. If misclassification occurred in the sense that CNS lupus patients were attributed to the non-neuropsychiatric disease SLE population, the fact that we still found a positive result strengthens our conclusions instead of weakens it.

It might be argued that thromboembolic events do not fit in the pathogenetic model of B cell mediated CNS lupus. But, as stated clearly in the article, even after exclusion of these ambiguous patients, the distribution of the frequencies in the neuropsychiatric disease SLE and non-neuropsychiatric disease SLE patients remains the same.

Of course we agree with the notion that our findings must be repeated in another group of patients. Interestingly, the increased prevalence of ATA in neuropsychiatric disease SLE patients has already been reported by Mok in a group of Chinese SLE patients.² Currently we are investigating the distribution of the IL10 promoter haplotypes of neuropsychiatric disease SLE patients in an ethnically different population.

In our article we have elaborated on two possible explanations of our findings. Firstly, the increased frequency of the ATA haplotype might be associated with an increased production of IL10. We made this assumption in the light of previous studies stating that SLE as a whole is characterised by an increased innate IL10 production.^{3,4} It is wrong to extrapolate these conclusions to our population. Because of the retrospective character of our study, we were not able to measure IL10 production in our populations and therefore cannot say whether or not IL10 production in our SLE patients as a whole was similar to or different from the control population. It might well be that differences in IL10 production would only emerge after stratifying into neuropsychiatric disease SLE and non-neuropsychiatric disease SLE patients. Furthermore, it might be that in the populations mentioned before, there was an excess of patients with neuropsychiatric disease SLE.

The second explanation for the skewing found in IL10 promoter polymorphisms might be that the increased susceptibility to neuropsychiatric disease SLE in the ATA patients is not conferred via an increased IL10 production at all, but that it is merely a marker for the real neuropsychiatric disease SLE susceptibility allele.

It is not clear whether or not IL10 promoter SNPs are associated with low or high IL10 production, because of the ambiguous reports in the literature. In our laboratory, the -1082 A allele has been found to

be associated with high IL10 production.⁵ In this light we have speculated about the possible link of high IL10 production and the pathogenesis of neuropsychiatric disease SLE. Isenberg *et al* have referred to another group stating that -1082 A is associated with a low ex vivo IL10 production and they interpret our results with this finding in mind.⁶ In conclusion, we do not know the relevance of the IL10 promoter in the in vivo regulation of IL10 production and therefore both explanations are equally speculative.

M J ROOD

T W J HUIZINGA

Leiden University Medical Centre, Department of Rheumatology, Building 1, C4-R, PO Box 9600, 2300 RC Leiden, the Netherlands

- 1 Rood MJ, Keijsers V, Van der Linden MW, Tong TQT, Borggreve S, Verweij CL, *et al.* Neuropsychiatric systemic lupus erythematosus is associated with imbalance in interleukin10 promoter haplotypes. *Ann Rheum Dis* 1999;58:85-9.
- 2 Mok CC, Lanchbury JS, Wai Chan D, Sing Lau C. Interleukin-10 promoter polymorphisms in southern Chinese patients with systemic lupus erythematosus. *Arthritis Rheum* 1998;41:1090-5.
- 3 Llorente L, Richaud-Patin Y, Fior R, Alcocer-Varela J, Wijdenes J, Morel-Fourrier B, *et al.* In vivo production of interleukin-10 by non-T cells in rheumatoid arthritis, Sjogren's syndrome, and systemic lupus erythematosus. *Arthritis Rheum* 1994;37:1647-55.
- 4 Hagiwara E, Gourley MF, Lee S, Klinman DM. Disease severity in patients with systemic lupus erythematosus correlates with an increased ratio of interleukin-10: interferon gamma secreting cells in the peripheral blood. *Arthritis Rheum* 1996;39:379-85.
- 5 Keijsers V, Verweij CL, Hazes JMW, Westendorp RGJ, Breedveld FC, Huizinga TWJ. Innate differences in IL-10 production are present at the level of transcription and associated with haplotypes: association of IL-10 haplotypes and rheumatoid arthritis. [Abstract]. *Clin Exp Rheumatol* 1998;16:200.
- 6 Turner DM, Williams DM, Sankaran D, Lazarus M, Sinnott PJ, Hutchinson IV. An investigation of polymorphisms in the interleukin-10 gene promoter. *Eur J Immunogenet* 1997;24:1-8.

A man with intermittent fever and arthralgia

Knight and Symmons report a very interesting case of a man with Whipple's disease and provide us with a timely update on this rare condition.¹ They describe how six months after initial presentation the diagnosis of adult onset Still's disease (AOSD) was made and despite regular review at several centres, this diagnosis was upheld for a further five years. Multiple investigations were performed adding little to the original diagnosis. It is not mentioned whether a serum ferritin was taken. This may have been useful given the initial diagnosis of AOSD as it might have resulted in questioning this diagnosis, permitting an earlier diagnosis of Whipple's disease.

It is widely reported in the literature, as far back as 1975,² that increased serum ferritin may be of use in both diagnosis³⁻⁵ and monitoring of AOSD.^{6,7} Hyperferritinaemia is not however exclusive to AOSD, as various malignancies, hepatic necrosis and haemochromatosis can all cause it. However, values up to 25 000 µg/l have been recorded in AOSD and reportedly, values rarely exceed 3-5000 µg/l in the above conditions. The pathogenesis is not clearly understood, but it has been hypothesised that in AOSD, cytokine upregulation of ferritin mRNA

translation may occur.⁸ This compares with the iron regulated pathway of ferritin synthesis in haemochromatosis and iron overload syndromes.

A comparative study of diagnostic criteria in AOSD by Mason *et al*⁹ suggest the Yamaguchi criteria are superior to the others tested, including Cush *et al* quoted by Knight and Symmons. However, none of the criteria to aid diagnosis make use of serum ferritin measurement despite the claims for its use in the literature and acceptance in clinical practice. Although undoubtedly useful if very high, it is not clear what the relevance of a normal value in AOSD is, in a case satisfying clinical diagnostic criteria (although we have never seen such a case). In rare diseases such as AOSD, it is difficult to assess and evaluate diagnostic criteria and calculate sensitivity and specificity of possible disease markers. If serum were stored on this patient it would be interesting to know the serum ferritin measurement and how, if at all, it would have affected this patient's management.

MARK QUINN

ANDREW GOUGH

Department of Rheumatology, Old Home, Leeds General Infirmary, Great George Street, Leeds, West Yorkshire, LS1 3EX.

Correspondence to: Dr M A Quinn.

- 1 Knight SM, Symmons PM. A man with intermittent fever and arthralgia. *Ann Rheum Dis* 1998;57:711–14.
- 2 Jacobs A, Worwood M. Ferritin in serum; clinical and biochemical implications. *N Engl J Med* 1975;292:951–6.
- 3 Gonzalez-Hernandez T, Martin-Mola E, Fernandez-Zamirano A, Balso-Criado A, Miguel-Mendieta E. Serum ferritin can be useful for diagnosis in adult onset Still's disease. *J Rheumatol* 1989;16:412–13.
- 4 Ota T, Higashi S, Suzuki H, Eto S. Increased serum ferritin in adult Still's disease. *Lancet* 1987;i:562–3.
- 5 Schiller D, Mittermayer H. Hyperferritinaemia as a marker of Still's disease. *Clin Infect Dis* 1998;26:534–5.
- 6 Akritidis N, Giannakakis I, Giougli T. Ferritin levels and response to treatment in patients with adult onset Still's disease. *J Rheumatol* 1996;23:201–2.
- 7 Schwarz-Eywill M, Heilig B, Bauer H, Breitbart A, Pezzutto A. Evaluation of serum ferritin as a marker for adult onset Still's disease activity. *Ann Rheum Dis* 1992;51:683–5.
- 8 Rogers JT, Bridges KR, Durmowicz GP, Glass J, Auron PE, Munro HN. Translational Control during the acute phase response. Ferritin synthesis in response to interleukin-1. *J Biol Chem* 1990;265:14572–8.

- 9 Masson C, Loet X, Liote F, Dubast JJ, Boissier M-C, Perroux-Goumy L. Comparative study of 6 types of criteria in adult Still's disease. *J Rheumatol* 1996;23:495–7.

Authors' reply

We thank Drs Quinn and Gough for their interest in our paper. Our patient did have his serum ferritin measured in 1992. It was 197 µg/l (normal range 15–200). This was therefore a situation in which the patient satisfied clinical diagnostic criteria for adult onset Still's disease (AOSD) but had a normal ferritin concentration. As the authors point out, had the ferritin concentration been high, this would have helped to confirm the diagnosis but given that it was in the normal range, it could not actually be used to refute the diagnosis. It was always felt that this patient's disease was not typical of AOSD and the various physicians who looked after the patient were always willing to consider alternatives. However, it is difficult, even with the benefit of hindsight, to conclude that Whipple's disease could have been diagnosed earlier. Although the normal serum ferritin was not in keeping with the diagnosis of AOSD it did not point towards any other diagnosis in particular.

DEBORAH SYMMONS

S M KNIGHT

Rheumatology, Macclesfield District General Hospital, Victoria Road, Macclesfield, Cheshire SK10 3BL

Ear, ear, what's going on in Norfolk?

Having recently started work in the rheumatology department of the Norfolk and Norwich Hospital I read with great interest the article on Hug(h)e(s') ears: an unusual presentation.¹ Amazingly we have recently seen an almost identical, but less catastrophic case.

A 27 year old white man presented with a 24 hour history of ears so swollen and painful, that he could not lie in bed with them touching the pillow. One year previously he was diagnosed with primary antiphospholipid syndrome (APLS) after recurrent deep vein thromboses and a raised IgG anticardiolipin

antibody at 92 iu/ml. He was subsequently given warfarin.

On admission this time, his INR, while taking warfarin 7 mg per day, was subtherapeutic at 1.6. Biopsy from his left ear lobe showed numerous fibrin thrombi with no associated vasculitis, consistent with thrombosis secondary to APLS.

His warfarin dose was increased to obtain an INR between 3 and 4. Within a few days he had recovered and was discharged home well.

Perhaps Hughes' ears should be renamed Norfolk ears?

EMMA CLARK

KARL GAFFNEY

PETER MERRY

Department of Rheumatology, Norfolk and Norwich Hospital, Brunswick Road, Norwich NR1 3SR

- 1 O'Gradaigh D, Scott DG, Levell N. Hug(h)e(s') ears: an unusual presentation of antiphospholipid syndrome. *Ann Rheum Dis* 1999;58:65–6.

Authors' reply

We note with interest the report from Dr Gaffney. While the coincidence is indeed curious, these cases do suggest possible mechanisms for activation of thrombosis. The external ear is characterised particularly by a lower average temperature than core body temperature, and by its susceptibility to trauma and pressure effects. In our case, cryoglobulins were not identified, and no comment in this regard is made by the authors. There is no specific reference to any aural trauma, though presumably, as in our case, it is difficult to assess what pressure was exerted on the external ear during sleep. It is plausible that such pressure causes a degree of blood stasis, which together with inadequate anticoagulation, resulted in thrombosis. Such speculation may be interesting, but it is this latter point that deserves emphasis—patients with antiphospholipid syndrome who have had thrombi will do so again, potentially with serious consequences, if the INR is not scrupulously maintained above 3.0, a message that must be spread widely: "Friends, Norfolk countrymen, lend me your ears!"

DONNCHA O'GRADAIGH

DAVID SCOTT

Department of Rheumatology, Norfolk and Norwich Hospital, Brunswick Road, Norwich NR1 3SR