Isolated hepatic tuberculoma after orthotopic liver transplantation: a case report

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Tuberculosis (TB) is an unusual infection in liver transplant recipients, its incidence ranges from 0.9-2.3% in developed countries¹. In these patients, TB is frequently seen as disseminated disease or as pulmonary disease, and liver involvement has been described only in cases of disseminated disease. Here we describe the uncommon case of a liver transplanted patient who developed an isolated liver abscess due to Mycobacterium tuberculosis (BK).

A 58-year-old man was referred to our unit 8 months after orthotopic liver transplantation (OLT) for cryptogenic cirrhosis. He complained of episodes of high fever and night sweats for over 1 month. On admission he had intermittent fevers up to 39°C, and no clear focus of infection determined by a physical examination. He did not complain of any other symptoms. The patient was on chronic immunosuppressive treatment with tacrolimus (1.5 mg bid) and steroids (prednisone 10 mg/day) due to the presence of lupus anticoagulant phenomenon.

Results of blood analyses were as follows: white blood cell count 6.95 × 10³ cells/l, with 84% neutrophils, 9.1% lymphocytes, 4.6% monocytes; platelets 151 × 10⁹ cells/l; haemoglobin 13.7 g/dl; erythrocyte sedimentation rate 36 mm/h (normal value < 15 mm/h); C-reactive protein 132 mg/l (normal value < 80 mg/l); fibrinogen 6190 mg/l (normal value 2000-4000 mg/l); gamma-glutamyl transferase was 2.1 times the upper normal values, alkaline phosphatase was 1.2 times the upper normal value. Serum transaminase, bilirubine, creatinine, lactate dehydrogenase and serum electrolytes were normal. Serology for parvovirus, cytomegalovirus and Epstein-Barr virus was negative. Blood cultures excluded bacterial infection. Radiological examination of the chest was normal.

An abdominal ultrasonography revealed a 35 mm solid hypoechoic mass with poorly defined walls in the fourth segment (Fig. 1) without dilatation of the biliary tree. The mass was not seen on a previous ultrasound examination done by the same operator 3 months before.

A Doppler study did not show arterial or venous blood flow inside the mass, whereas the hepatic artery flow was normal.

A contrast-enhanced computed tomography of the thorax and abdomen was then performed, showing a normal pattern of lungs, and confirming a 50 mm lesion in the fourth hepatic segment, with the pattern of a partly solid and only in small part liquefied abscess. No enlarged lymph nodes were detected. During this procedure, by inserting a 20G needle in the liquid part of the mass, an aspiration biopsy was obtained. The small quantity of collected material presented as a dense, purulent, slightly haematic fluid.

The microbiological examination of the specimen showed a large amount of acid-fast bacilli. The patient was treated on an empirical basis with clarithromycin (500 mg bid), ethambutol (400 mg tid) and levofloxacin (500 mg bid), and rapidly improved, with fever resolution on the second day of treatment. The erythrocyte sedimentation rate and C-reactive protein normalised 8 days later. Blood levels of tacrolimus progressively increased, and its dosage was reduced to 0.5 mg every 3 days. The patient was discharged from the hospital on clarithromycin, ethambutol and levofloxacin. After 1 month, the bacterial strain was identified as Mycobacterium tuberculosis (sensitive to all tested drugs).
In consideration of this finding the treatment was modified and ethambutol 400 mg bid, levofloxacin 500 mg/day, isoniazide 200 mg/day and pyrazinamide 500 mg tid were added. Three months later, an ultrasound re-evaluation showed that the liver was free from lesions. Now the patient is in the tenth month of therapy, in good health. In his case antituberculotic therapy did not cause any adverse event.

In solid organ transplant recipients TB has a significant morbidity and mortality, reaching 25-40% despite modern chemotherapy. In liver transplanted patients, TB causes infection in 0.5-11%, the higher prevalence is present in developing countries, and ranges 0.9-2.3% in Europe and United States\(^1\)-\(^3\). The time of onset of the disease after OLT is variable, ranging 1-60 months, with a median time of 9 months. In our patient TB infection occurred 8 months after OLT. While the type of immunosuppressive therapy does not seem to influence the probability of developing TB infection, it has been reported that it is one of the predictors of early onset of the disease; it is interesting to note that in the 11 patients with TB receiving tacrolimus, the infection developed within 12 months of transplantation in 100% of cases\(^1\), and that our case is in agreement with these previous experiences.

Our patient had been found positive to lupus anticoagulant phenomenon several months before the clinical onset of TB; lupus anticoagulant has been reported in 53% of patients with TB\(^4\), and this laboratory finding might have been the first manifestation of the presence of BK in our patient.

In solid organ transplantation recipients, TB is frequently disseminated to extrapulmonary sites\(^1\)-\(^5\), and fever is most likely to occur in disseminated TB\(^1\). In our patient, even if the disease was exclusively intrahepatic, the clinical presentation was dominated by intermittent fever and constitutional symptoms (night sweats). Whereas hepatic involvement is often observed in the setting of disseminated disease, the finding of an isolated liver abscess due to BK (or hepatic tuberculosis) is very unusual even in immunocompromised hosts\(^1\)-\(^5\); Alothman et al.\(^6\) described a case of isolated liver abscess due to BK in an OLT recipient, who, similarly to our patient, was TB-negative prior to OLT.

Re-activation of dormant TB is the usual mode of acquisition\(^3\)-\(^5\), so we investigated the immunological status pre-transplantation of our patient, finding that he had no signs of previous contact with BK (negative tuberculin skin test, standard radiological and computed tomography examination of the chest, negative blood, urine and stool microbiological tests for BK). In the absence of a previous positive test for BK, the finding of a TB infection in a liver transplant recipient may be due to a recently acquired infection (usually a nosocomial transmission) or to transmission from the graft\(^7\), which was the proposed source in 4% of recent series\(^1\)-\(^3\).

In newly acquired infection the most common clinical manifestation is pulmonary or disseminated TB, and no case of localisation in hepatic parenchyma has been confirmed in this setting up to date. Therefore we searched for a possible donor’s infection. We found out that the donor was a 65-year-old woman of Croatian origin. TB is highly endemic in Croatia, and it used to be particularly frequent in the decades prior to the second World War\(^8\). Even if we cannot provide clear evidence of transmission from the graft, in this case the donor had a high probability of having been exposed to BK.

As for the ultrasonographic aspect, liver BK abscesses usually appear either as hypoechoic lesions without a distinct wall, formed by coalescence of small tubercles, or as a hypoechoic lesion with hyperechoic rims; very rarely do they look like hyperechoic masses\(^9\). Our case presented the most typical ultrasound aspect, being hypoechoic without a well-defined wall (Fig. 1).

The microbiological finding of acid-fast bacilli in the biopsy obtained from the mass can be considered diagnostic for BK\(^10\), as it was in our case. Currently, the diagnosis can be confirmed by rapid tests such as automatic liquid culture or by amplification of genetic material. In the absence of direct observation, the specimen must be processed and inoculated at 37°C for 8 weeks to exclude TB.

As for treatment, in our patient the infection was rapidly controlled by an empirical therapy including clarithromycin, ethambutol and levofloxacin without significant side effects; the observed increased plasma levels of tacrolimus was probably due to competitive antagonism for the cytochrome oxidase between anti-TB agents and the immunosuppressive agent. The initial decision to treat the patient with an unconventional regimen of antibacterial therapy was due to the frequent reported toxicity of recommended agents\(^11\), and to the good clinical response that we observed. At BK infection confirmation the patient was switched to a more conventional regimen of therapy, which did not induce adverse events in his case.

In conclusion, in liver transplanted patients, BK infection can be restricted in the hepatic tissue as an isolated abscess, and thus should be included in the differential diagnosis of liver masses in OLT recipients.

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Weight loss, articular pain and bone fractures: neoplastic disease or malabsorption syndrome?

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The body systems that control the growth and maintenance of the skeleton can be disrupted in different ways that result in a variety of bone diseases and disorders. Primary osteoporosis is mainly a disease of the elderly, the result of the cumulative impact of bone loss and deterioration of bone structure that occurs as people age, and is quite rare in young adults1-3. In addition to conditions that affect bone directly, there are many other disorders that indirectly affect bone by interfering with mineral metabolism. Young adults and even older individuals who get osteoporosis, often do so as a by-product of another condition or medication use. Diseases that reduce intestinal absorption of calcium and phosphorus, or impair the availability of vitamin D, can also cause bone disease. Bone loss is seen after gastric bypass surgery even in morbidly obese women who do not have low bone mass initially4. Fractures are also seen in patients with Crohn’s disease and ulcerative colitis5. Similarly, diseases that impair liver function (primary biliary cirrhosis, chronic active hepatitis, cirrhosis due to hepatitis B and C, and alcoholic cirrhosis) may result in disturbances in vitamin D metabolism. Coeliac disease is an important cause of secondary osteoporosis6. The active absorption of calcium takes place in the duodenum and in the proximal jejunum7, which are known to be the most damaged intestinal segments in patients with coeliac disease. Coeliac disease is considered to be one of the most frequent predisposing conditions to metabolic osteopathy. Studies have shown that more than 75% of untreated adults with coeliac disease suffer from osteopaenia or osteoporosis. Patients on a gluten-free diet show a lower prevalence of bone loss, suggesting that dietary therapy may improve bone mineral density8-10.

We report the case of a 30-year-old woman; she was 163 cm tall and 86 kg weight. She had two miscarriages during the previous 10 years, and had given birth to her only son 4 years beforehand. During nursing, rachidian (spinal) pain had appeared for the first time. During the previous 12 months, axial pain had become more severe, and she had felt “unwell” and had a progressive weight loss (she was 97 kg). She had also abdominal bloating after dinner, with some episodes of abdominal pain and diarrhoea. She had been seen by her family physician. The practitioner had ordered general blood tests that had revealed an increased serum level of alkaline phosphatase as well as iron deficiency anaemia. Therefore, he suggested the admission to hospital suspecting an occult neoplastic pathology.

The general history did not reveal a reduction in food intake during the prior months. The body mass index was 32.36 kg/m² in contrast to the past year when it had been 36.5 kg/m². General examination revealed paleness and asthenia. The pain was localised in the upper thorax and in the pelvis involving shoulders and hips. Vital signs, cardiopulmonary function, body temperature and blood oxygenation were normal, as well as initial laboratory testing of the kidney and liver. The phlogosis (inflammatory) blood test was normal, and the complete blood count revealed an iron deficiency anaemia; the alkaline phosphatase, and parathyroid hormone were
raised, and there was a little hypocalcemia and hypocalciuria (Table 1). No gastrointestinal bleeding or neoplastic markers dosed in the serum were found. A radiographic study explained the pain in the upper thorax and in pelvis, showing fractures of the right scapula, of both first ribs and the pubis. Bone scintigrams revealed an increased osteogenesis in many ribs and in the right fibula. An echographic study of the thyroid gland showed bilateral oval masses compatible with parathyroid gland hypertrophy.

The clinical aspects were suggestive for a malabsorption syndrome with secondary hyperparathyroidism. The endomysial antibody and duodenal biopsy were positive and confirmed the diagnosis of coeliac disease (mucosa characterised by villus flattening and infiltration of the epithelium with lymphocytes).

We discharged the patient with a strict gluten-free diet, and with a dietetic supplementation with calcium plus vitamin D (the blood concentration was 14 pg/ml; normal values 15-60 pg/ml). A re-evaluation was performed after 40 days: she felt well, with no dyspeptic symptoms or bloating, and the axial pain was reduced. There were radiologic signs of re-ossification to both first ribs and pubic branches. She weighted 86.5 kg. She refused a repeat duodenal biopsy.

This 30-year-old woman was obese (with a body mass index of 32.36 kg/m²) at the time of coeliac disease diagnosis, despite weight loss of 11 kg during the past year. Few clinicians would expect obesity to be part of coeliac disease presentation; in fact, initially her family physician and us thought the patient had an occult neoplasm.

The excess flatulence and the low-grade persistent abdominal pain led us to consider the possibility of irritable bowel syndrome, or a case of inflammatory bowel disease such as Crohn’s disease. The hypothesis of a neoplasm or of a bowel pathology were reinforced by an iron deficiency anaemia, and after the finding of the pathologic fractures on the radiographic studies. To understand the nature and the extent of the fractures in the body, we performed an examination of neoplastic markers, parathyroid hormone levels, calciuria, bone scintigrams and an echographic study of the neck.

The bone metabolism abnormalities in patients affected with coeliac disease were first described in 1929. The mechanism of development of bone pathology in patients with untreated coeliac disease is not fully defined, but chronic negative calcium balance caused by loss of villous surface area, binding of calcium to fatty acids in the intestinal lumen, and impairment of active intestinal calcium transport mechanisms all thought to be operative. Increased calcium intake could potentially compensate for the reduced fractional calcium absorption in treated adult coeliac patients, but may not normalise the bone mineral density. In addition, the inverse correlation between parathyroid hormone and time following treatment is suggestive of a continuing long-term benefit of gluten withdrawal on bone metabolism in coeliac patients.

Other factors also playing an important role for the development of a negative calcium balance in coeliac disease are the low calcium intake with food, probably mediated by the contemporary secondary lactase deficiency, the higher fecal excretion of the endogenous calcium, which is likely to be due to an increased intestinal secretion or to a decreased reabsorption. Bone loss in coeliac disease might also be caused by a cytokine imbalance directly affecting osteoclastogenesis and osteoblast activity. Secondary hyperparathyroidism may develop as a compensatory response to hypocalcaemia, and this will cause increased bone resorption. Furthermore, the tendency of parathyroid hormone to reduce serum phosphate will decrease the calcium:phosphate product, further diminishing one of the important driving forces for mineralisation of bone. The role of vitamin D deficiency in the metabolic bone disease of coeliac disease is also debated; it is not certain whether hypovitaminosis D contributes to the intestinal calcium malabsorption. It appears that mild deficiency can lead to so-called pre-osteomalacia, in which increased bone turnover caused by secondary hyperparathyroidism is associated with accelerated bone loss.

Coeliac disease is also associated with anaemia, which is the most common laboratory manifestation of coeliac disease. Iron is absorbed in the proximal small intestine, where coeliac manifestations are most prominent. In addition, hidden blood loss related to intense small-bowel inflammation may occur in 50% of the patients with gluten-sensitive enteropathy. The risk of fractures in coeliac disease is still an unsolved problem: as our case report reveals, many authors point out an increase in both the absolute and relative risk of fracture in people with coeliac disease. The risk seems to be slightly lower in those with a more recent diagnosis of coeliac disease. Diagnostic and therapeutic strategies to prevent bone loss and fracture should be preferentially

### Table 1. Haematological data of the patient.

<table>
<thead>
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<th>Parameter</th>
<th>Value</th>
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<tr>
<td>Red cell count (µl)</td>
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<tr>
<td>White blood cell count (x 10⁹ cells/l)</td>
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</tr>
<tr>
<td>Haemoglobin (mmol/l)</td>
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</tr>
<tr>
<td>Haematocrit (%)</td>
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<tr>
<td>Mean cell volume (fl)</td>
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</tr>
<tr>
<td>Platelet count (x 10⁹ cells/l)</td>
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</tr>
<tr>
<td>Siderin (µg/dl)</td>
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</tr>
<tr>
<td>Ferritin (µg/dl)</td>
<td>8</td>
</tr>
<tr>
<td>Calcium (mg/dl)</td>
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</tr>
<tr>
<td>Calciuria (mEq/day)</td>
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<tr>
<td>Alkaline phosphatase (IU/l)</td>
<td>998</td>
</tr>
<tr>
<td>Vitamin D3 (µg/ml)</td>
<td>14</td>
</tr>
<tr>
<td>Parathyroid hormone (pmol/l)</td>
<td>466</td>
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</tbody>
</table>
used in the subgroup of patients with classic clinical disease19, and any spontaneous (pathologic) fracture deserves a careful search for metabolic bone disease20. We believe this case report to be of interest because it underlines some unusual aspects of coeliac disease, such as the obesity of the patient in spite of her progressive weight loss, and the pain involving upper thorax and pelvis that initially suggested pathological bone fractures of neoplastic disease.

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A novel nonsense mutation in exon 2 of the factor IX gene resulting in severe haemophilia B

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Haemophilia B (Christmas disease) is a recessive X-linked coagulation disorder caused by the plasma deficiency of coagulation factor IX (FIX)1,2. An approximate incidence of 1 in 25 000 males is registered worldwide3. It is clinically defined by low factor IX coagulant activity (FIX:C) and, as with haemophilia A, it is classified as mild (FIX:C between 5-30%), moderate (FIX:C between 1-5%) and severe (FIX:C < 1%)3. Molecular testing of FIX identifies disease-causing mutations in 99% of cases4. The FIX gene is localised on the X chromosome (q27.1) and is characterised by 8 exons (33 kb)5. Human FIX, a vitamin K-dependent single-chain glycoprotein, is synthesised by the hepatocyte, first as a precursor protein; then, it undergoes extensive post-translational modifications to become the fully γ-carboxylated mature zymogen secreted into the blood6,7. The mature protein consists of 415 amino acids proceeded by a pre-peptide (28 amino acids) and by a pro-peptide (18 amino acids)8. The pre-peptide

Received 20 April 2006; revised 27 June 2006; accepted 31 August 2006.

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acts as a signal peptide, and it is coded by exon 1. The propeptide acts as a signal for the vitamin K-dependent γ-carboxylation, and it is coded by a portion of exon 2. The rest of exon 2 codes for a γ-carboxyglutamate (GLA) domain in which some residues are γ-carboxylated. The process of γ-carboxylation is essential for the production of a biologically active protein. Exon 3 is very short, and codes for the last part of GLA domain. Exon 4 and 5 code for the epidermal growth factor-like domain, and exon 6 codes for an activation peptide. Exon 7 and 8 code for a catalytic domain in which there is a catalytic triad: His221, Asp269, Ser365. The cleavage of a small peptide (from 145 to 180 amino acids) acts as activation mechanism. This paper is the description of a new nonsense mutation in the FIX gene causing severe haemophilia B.

A Sicilian male patient, 1 year old, had severe haemophilia B with FIX:C < 1 IU/dl. He had large haematomas, haemarthroses and spontaneous mucocutaneous bleeding. He was treated only with 500 U/ml of FIX recombinant for 1 year. He was also followed up for activated partial thromboplastin time activity, FIX:C and for FIX inhibitor (Table 1). His parents are unrelated, and his mother showed no clinical symptom of bleeding with FIX:C 81 IU/dl. Blood samples were taken from both the mother and the propositus after obtaining informed consent. Genomic DNA was extracted from whole blood using a commercial DNA extraction kit (QiaAmp, Qiagen, Hilden, Germany). Genotyping was performed by direct sequencing FIX gene. The FIX coding region (8 exons) and the promoter region were amplified as described by Wulff et al. and Yoshitake et al. using sets of oligonucleotide primers, which were positioned within the intronic sequence to ensure inclusion of intron-exon boundaries. For polymerase chain reaction (PCR), uniform conditions were applied to all exons: 5 min of initial denaturation at 94°C; 30 cycles were carried out at 94°C for 20 s, 54°C for 30 s, 68°C for 45 s; a final extension at 68°C for 6 min. PCR products were electrophoresed in 2% agarose gel and visualised by ethidium bromide staining. All the 8 exons, step by step, were amplified in each region, and PCR products purified with the Qiagroup PCR kit (Qiagen) that underwent automated direct sequencing by using Applied Biosystem Prism BigDye Terminator Cycle Sequencing Ready Reaction Kit. Sequencing analysis was performed using ABI Prism 3100 Genetic Analysers. FIX gene sequences of the patient and his mother were compared with the native sequence obtained from the Haemophilia B Mutation Database, version 12, and the Human Gene Mutation Database (http://www.hgmd.cf.ac.uk/ac/index.php). Genetic analysis revealed the same new point mutation in both cases: a nonsense A→T transversion in exon 2 at nucleotide position 6421 corresponding to codon +62 of coding sequence (Fig. 1) generating a change at position +16 of the mature FIX protein. The codon AGA (coding for Arg in normal conditions) is changed into TGA coding for a stop signal. Severe haemophilia B is caused by a variety of gene alterations, frameshift, nonsense or missense mutations and also promoter, splice site and cryptic splice site mutations. These alterations produce abnormal gene products or the total absence of FIX. This novel nonsense mutation in exon 2 of FIX gene found in a Sicilian patient with severe haemophilia B consists in a transversion A→T at nucleotide position 6421 (Fig. 1). The transversion leads to a stop codon, so that translation is interrupted at the level of GLA domain of the mature protein. This novel nonsense mutation has never been described in the Haemophilia B Mutation Database and is compatible with the severe phenotype of the patient. He presented with undetectable FIX coagulant activity while his mother with no clinical symptoms, is carrier.

An important consequence of our molecular analysis is that we are now able to use this mutation for prenatal diagnosis in the family.

Table 1. Patient follow-up during therapy.

<table>
<thead>
<tr>
<th></th>
<th>Pre-treatment</th>
<th>Post-treatment</th>
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<tbody>
<tr>
<td>FIX:C (%)</td>
<td>0.9</td>
<td>41.5</td>
</tr>
<tr>
<td>aPTT (s)</td>
<td>64</td>
<td>58</td>
</tr>
<tr>
<td>INR</td>
<td>1.2</td>
<td>1.1</td>
</tr>
<tr>
<td>FIX inhibitor</td>
<td>Absent</td>
<td>Absent</td>
</tr>
<tr>
<td>aPTT, activated partial thromboplastin time; C, coagulant activity; FIX, factor IX; INR, international normalised ratio.</td>
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Figure 1. Representation of exon 2 of factor IX gene and comparison between electropherogram of propositus, his mother and negative control.

References


Calciphylaxis: evolving concepts

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Calciphylaxis (often referred to as calcific uraemic arteriopathy) is a vascular disease characterised by medial calcification and intimal hyperplasia of the small arteries that lead to ischaemia and necrosis of the skin, subcutaneous fat, visceral organs and skeletal muscle. It predominantly occurs in patients with end-stage renal disease (ESRD) on dialysis or after kidney transplantation1,2. The pathogenesis of calciphylaxis remains unclear, but an association with high serum phosphate, calcium and parathyroid hormone (PTH) levels has been reported in many affected patients. Its largely empirical treatment is based on correcting hyperphosphataemia, avoiding high calcium-phosphate products, local wound care, and parathyroidectomy. The prognosis of patients with calciphylaxis is poor, with a mortality rate of 60-80%, which is mainly due to sepsis and ischaemic events3.

We here describe a patient on chronic haemodialysis who developed fatal calciphylaxis, and summarise recent advances in our understanding of the pathogenesis, diagnosis and treatment of this devastating clinical syndrome. In June 2004, a 41-year-old non-smoking woman on maintenance haemodialysis was referred to our department because of multiple cutaneous ulcers in both legs. She had a history of chronic kidney disease secondary to chronic obstructive uropathy requiring haemodialysis in March 2000. A significant degree of hyperparathyroidism had developed as a consequence of inadequate hyperphosphataemia control due to poor compliance with dietary phosphorus restriction and oral phosphorus binders. In November 2003, she had undergone a cadaveric renal transplantation, but the transplant had failed because of urological complications, and haemodialysis had been re-established. Five months before admission, she had developed a painful livedo reticularis-type rash on both thighs, with the gradual emergence of ecchymosis and necrotic skin ulcerations. On admission, the temperature was 38°C, the pulse was 100/min and the arterial blood pressure 140/85 mmHg. Physical examination showed multiple large necrotic ulcers (Fig. 1) on both thighs. The results of cardiovascular and chest examinations were unremarkable. The pulses in both legs were palpable. Predialysis laboratory tests revealed a white blood cell count of 12,000/mm3 with left shift.

Figure 1. Skin ulceration with overlying black eschars on the left thigh.
The patient presented with ESRD, secondary hyperparathyroidism and painful cutaneous ulcerations; these findings suggested the diagnosis of calciphylaxis, but other diseases were considered in the differential diagnosis of the cutaneous ulcers. Peripheral vascular disease may cause ischaemic ulcers but, in our case, the proximal distribution of the lesions and the preserved peripheral pulses favoured the diagnosis of calciphylaxis. Cutaneous ulcers may occur in a variety of processes: small-vessel vasculitis, systemic autoimmune disorders, inflammatory bowel disease, malignant tumours, diabetes mellitus and infections, but our patient had no history of any of these conditions. The possibility of septic embolism was considered, but the blood cultures before antibiotic administration were always negative; in addition, no valvular vegetations were found on cardiac ultrasonography. Cholesterol emboli can lead to cutaneous ulcerations, but these develop mainly on the lower extremities and almost exclusively in elderly patients with widespread atheromatosis. The clinical diagnosis of calciphylaxis was supported by the skin biopsy specimens, which ruled out other causes of ulceration, such as small-vessel vasculitis (no presence of leukocytoclasis, fibrinoid necrosis and granulomas) and pyoderma gangrenosum (no cellular inflammation with leukocytoclasia, fibrinoid necrosis and granulomas) and pyoderma gangrenosum (no cellular inflammation with leukocytoclasia, fibrinoid necrosis and granulomas) and pyoderma gangrenosum (no cellular inflammation with leukocytoclasia, fibrinoid necrosis and granulomas) and pyoderma gangrenosum (no cellular inflammation with leukocytoclasia, fibrinoid necrosis and granulomas) and pyoderma gangrenosum (no cellular inflammation with leukocytoclasia, fibrinoid necrosis and granulomas).

The pathogenesis of calciphylaxis is unclear. Obesity, female gender, diabetes mellitus, hypoalbuminaemia, an excess of vitamin D, and hypercoagulability syndromes (such as protein C or protein S deficiency) have all been reported to be important risk factors. Although many patients have an abnormal Ca × P product or high PTH levels, the fact that the deranged calcium-phosphate metabolism is usually no more severe than in dialysis patients without calciphylaxis strongly suggests the involvement of other aetiopathogenetic factors. One of these may be vascular calcification (VC) that is highly prevalent in ESRD patients. VC is not a passive, uncontrolled precipitation of calcium phosphate, but a tightly regulated process in which vascular smooth muscle cells (VSMCs) are induced to transform into osteoblast-like cells, capable of mineralisation and production of bone matrix proteins. A number of in vitro studies indicate that hyperphosphataemia may directly stimulate VSMCs to undergo a phenotypic transition characterised by the loss of smooth muscle cell markers (SM α-actin and SM22α) and the gain of osteogenic markers (osteopontin, Cbfa-1, alkaline phosphatase and osteocalcin). Interestingly, almost identical changes in smooth muscle gene expression are observed in biopsy specimens from ESRD patients with calciphylaxis. However, hyperphosphataemia is not the only factor involved in vascular cell calcification. As blood vessels normally have inhibitory mechanisms preventing mineralisation, it is likely that VC occurs as a result of failure of these mechanisms. A number of local and systemic calcification inhibitors have been identified, including matrix GLA protein, osteoprotegerin and fetuin-A. The last one, which is also known as alpha-
Heremans-Schmid glycoprotein, is an inhibitor of basic calcium-phosphate mineral precipitation, and accounts for about 50% of the mineral inhibitory activity of blood. Low serum fetuin-A levels have been found in dialysis patients, especially in those with raised C-reactive protein concentrations. Although chronic inflammation and uraemia may both contribute to exhausting fetuin-A release, variations in the gene encoding fetuin may have an additional effect on its low circulating levels. It can be hypothesised that patients with a genetic propensity for low fetuin-A levels are at higher risk of VC when they are exposed to factors promoting calcification, such as hyperphosphataemia, hypercalcaemia and uraemia. There is no diagnostic laboratory test for calciphylaxis. Hyperphosphatemia, a high Ca × P product, high plasma PTH levels and a slight increase in serum calcium remain essential components of any evaluation of calciphylaxis, but do not necessarily correlate with its development. As low fetuin-A levels are associated with inflammation and VC in patients on dialysis, measuring its serum concentrations may help identify individuals at high risk of calciphylaxis. Bon scan, roentgenography and xeroradiography are not diagnostic procedures, but may be helpful in detecting extraosseous calcification. Given the high mortality rate associated with calciphylaxis, early diagnosis and treatment are essential. In ESRD patients, strict control of phosphate and calcium balance should avoid the metabolic milieu that predispose to calciphylaxis. Secondary hyperparathyroidism should be prevented by means of a low-phosphorus diet, the use of Ca-free (and aluminium-free) phosphate binders (such as sevelamer), and the careful use of vitamin D analogues. Calcimimetics agents provide a novel drug therapy to suppress PTH release and synthesis and prevent parathyroid cell proliferation. The therapeutic role of parathyroidectomy is still debated. Most authors recommend surgery only in patients with severe hyperparathyroidism (PTH > 500 pg/ml) and an increased Ca × P product (> 70 mg²/dl²). Dialysis with low-calcium dialysate solutions (e.g. 1.5 or 1.0 mEq/l), bisphosphonates and sodium thiosulfate may lower serum calcium and phosphorous levels and mobilise calcium salts from calcified tissues, thus promoting healing of skin ulcerations. However, the positive results of using bisphosphonates reported in one paper were not confirmed by our case.

As sepsis is the main cause of death, an aggressive programme of wound care with careful use of antibiotics is essential. Success of surgical debridement is poor, as observed in our patient. Hyperbaric oxygen therapy may improve tissue oxygenation and the cutaneous ulcers. Pain is a frequent and devastating complication of calciphylaxis, and so large doses of analgesics are often needed to provide relief. Some authors report that a neurolytic lumbar sympathetic block is beneficial in patients with intractable pain, but this was not the case for our patient.

Despite recent diagnostic and therapeutic advances, calciphylaxis-related mortality remains unacceptably high. A better understanding of the factors contributing to the development of VC in ESRD may lead to the identification of more effective therapeutic approaches.

References