Pure red cell aplasia associated with large granular lymphocytic leukemia: a rare association in Western countries

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The coexistence of large granular lymphocytic leukemia (LGLL) and pure red cell aplasia (PRCA) has been previously described, but is rare in Western countries (7%) in a recent series of LGLL cases). We present the clinical features, hematological parameters and immunophenotype of two patients with PRCA associated with CD3+ LGLL.

Large granular lymphocyte leukemia (LGLL) is a clinical and pathologic heterogeneous lymphoproliferative disorder involving large granular lymphocytes (LGL).[1] Recent immunophenotypic and molecular analyses have shown that LGL can be divided in two major lineages: a) cytotoxic $CD3^+$ T cells with NK cell properties (T-LGLL), and b) NK cell lineage with $CD3^-CD7^+$ and variable expression of CD16, CD56 and CD57 (NK-LGLL).[2] It is known that patients with LGLL can have pure red cell aplasia (PRCA).[3] This complication is rare in Western patients (only 7%),[4] but is more frequent in Japanese patients (occurring at a high frequency of 64%).[5]

We present two Caucasian patients with PRCA associated to T-LGLL. Anemia and low reticulocyte count was found in both patients. Clinical features are summarized in Table 1. Lymphocyte count was 11 and 8×10^9/L with 15 and 35% of LGL, respectively. Serologies for Parvovirus B19, HIV, CMV and HTLV-1 were negative. Both cases showed an absence of erythroid precursors in the bone marrow (BM) with an interstitial pattern of LGL infiltration, ranging from 17 to 21%, respectively. Immunophenotyping of lymphocytes in peripheral blood (PB) by flow cytometry with a large panel of monoclonal antibodies (MoAbs) directed against T and NK antigens showed a common pattern of T-LGLL with expression of CD3 (Table 2). Case #2 expressed the TCRδγ receptor (Cγ-1). Case #1 showed a normal karyotype.

Table 1. Clinical features of PRCA associated to LGLL.

Case	Sex/Age (yr)	Liver (cm)	Spleen (cm)	LN	Survival (months)
1	F/52	–	–	–	60
2	M/62	4	6	–	72

LN: lymph nodes.

Table 2. Immunophenotypic markers.

Case	$CD3^+CD8^+$ (%)	$CD3^+CD16^+$ (%)	$CD3^+CD56^+$ (%)	$CD3^+CD57^+$ (%)	$CD3^+DR^+$ (%)
#1	74	69	2	60	55
#2	76	70	3.5	70	39

Percentages are referred to the lymphoid population.

Case #1 was treated with steroids and cyclophosphamide, with no response. She achieved a complete remission with ATG that lasted for 9 months. At present after a follow-up of 72 months she has active disease unresponsive to cyclosporin A (CyA). Case #2 was diagnosed as having PRCA associated with thymoma. He was refractory to thymectomy, steroids, cyclophosphamide and intravenous gammaglobulin. He responded to treatment with CyA.

Five years later, the patient relapsed and did not respond to CyA. At that time the morphology and immunophenotype of lymphocytes in PB and BM were consistent with LGLL. Nine months later the patient died with unresponsive PRCA and persistent $CD3^+CD57^+$ TCRγδ$^+$ lymphocytes after a follow-up of 72 months.

T-LGLL is a lymphoproliferative disease characterized by the chronic proliferation of $CD3^+$ lymphocytes with clonal rearrangement of TCR genes.[3] The evidence of a LGL lymphocytosis greater than 2×10^9/L lasting for more than 6 months is currently regarded as a clinical criterion to define the disease.[3,6] Although TCR gene rearrangement could not be studied in our patients, the associated hematologic and immunologic abnormalities with a follow-up longer than six months are compatible with T-LGLL. In case #2, TCRγδ was expressed in LGL. These TCRγδ$^+$ cases seem to have a similar clinical presentation as TCRαβ$^+$ cases, although only a few patients with this phenotype has been described[2] and γδ-T-cell lymphoma should be excluded.[7] PRCA complicating LGLL is very rare in Western populations[2] but in a large Japanese serie, the frequency of PRCA in LGLL was quoted to be as high as 64%.[5] In these cases PRCA could be considered a feature of LGLL and not an entity by itself. In our case #2, PRCA associated with thymoma preceded the development of $CD3^+$ LGLL. There are several reports that thymoma is associated with peripheral T-cell lymphocytosis.[8,9] In our case at the moment of diagnosis of PRCA the patient had a normal lymphocyte count without expansion of $CD3^+CD8^+$ cells.[10] However, T-cell disorders must be excluded at diagnosis or in the follow-up of patients with thymoma.

Key words

Aplasia, large granular lymphocyte, leukemia, immunophenotype

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Effect of cyclosporin-A on anemia in idiopathic myelofibrosis

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The recent report of Pietrasanta *et al.*,[1] described an improvement of anemia in a case of idiopathic myelofibrosis (IMF) treated with cyclosporin-A (CyA) for psoriatic skin lesions.
We have also used CyA in four IMF patients, all with transfusion dependent anemia. We report briefly the clinical aspects, hematological parameters and responses to the treatment for every patient.

Case #1. A 61-year-old female with diagnosis of IMF, pathologic stage III, low risk according to the LILLE scoring system, became transfusion dependent 62 months after diagnosis. Before use of CyA blood test results were: median levels Hb 7.9 g/dL (range 7.1-8.7), WBC 15×10^9/L, Plt 624×10^9/L, reticulocytes 3.7%, sEPO 8.2 mIU/mL (vn 4-25), Coombs D/I negative, ratio CD4/CD8 1.1. The patient was treated with hydroxyurea (HU) and had received splenic irradiation 6 months previously. A median of 2 packaged red blood cell units was needed monthly. The treatment with CyA (200 mg/day) was performed for 4 weeks: no response was observed.

Case #2. A 65-year-old male with IMF, pathologic stage III, intermediate risk, needed 7 packaged red blood cell units monthly, he became transfusion dependent 29 months after diagnosis. Median levels Hb 5.4 g/dL (range 4.6-6.7), WBC 13×10^9/L, Plts 26×10^9/L, reticulocytes 4.7%, sEPO 13.4 mIU/mL, ratio CD4/CD8 0.6. The patient received hydroxyurea (HU). The treatment with CyA (200 mg/day) was performed for 12 weeks, but the hemoglobin level and the transfusional support remained invariable.

Case #3. A 57-year-old female afflicted by IMF, pathologic stage II, intermediate risk, was treated with recombinant human erythropoietin (4000 U/three times weekly) and HU for 2 years, with improvement of Hb levels (Hb > 10 g/dL). After the loss of response to EPO, the Hb was 6.7 g/dL, reticulocytes 1.9%, sEPO 1496 mIU/mL, and transfusional support was need. CyA (200 mg/day) was administered for 6 weeks, without any effect.

Case #4. A 66-year-old male, 2 months after diagnosis of IMF (pathologic stage I) developed a pure red cell aplasia (PRCA). Blood test results were Hb 4.5 g/dL, WBC 21×10^9/L, Plt 342×10^9/L, reticulocytes 0.5%, sEPO 15 mIU/mL, Coombs D/I negative, ratio CD4/CD8 0.9. The treatment with CyA was performed for 12 weeks, without response. Considering the level of sEPO inadequate for the degree of anemia, r-EPO (10,000 U/three times weekly) was associated to CyA for another 10 weeks. Again the treatment failed.

In all our cases, the use of CyA was ineffective. The report by Centenara *et al.*[2] described a reduction of transfusional support in 4 out of the 7 IMF patients available for evaluation. No other reports on the efficacy of CyA in IMF have been published. Immunologic abnormalities described in IMF[3,4] and the positive action of CyA on certain types of anemia (PRCA during lymphoproliferative disorders) could justify the use of CyA. Nevertheless, the need to clarify the role of immune mechanisms in the pathogenesis of IMF and then perform further investigations about the use of immunosuppressive agents in selected patients are essential.

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Splenic peliosis with spontaneous splenic rupture in a patient with immune thrombocytopenia treated with danazol

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We present a 79-year-old man diagnosed with immune thrombocytopenia (ITP), treated with danazol, who died as a result of a spontaneously ruptured spleen. The histopathological diagnosis was splenic peliosis. This patient presents a chronological association between the treatment with danazol and the development of peliosis, which suggests a clear cause-effect relationship. Facing an individual patient with ITP, clinicians should weigh the potential benefits of danazol with the possible development of serious complications, such as hepatic failure or splenic rupture due to peliosis.

Peliosis is an ectasial vascular process, characterized by the presence of blood-filled cavities in the parenchyma of the liver or spleen.[1,2] Isolated splenic peliosis is rare.[3] The liver is the most frequently affected organ, and most of the reported cases have been incidentally discovered in autopsies[3] relating to tuberculosis[4] and hematological malignancies.[3,5]. Recently, cases of patients treated with anabolic corticosteroids, with or without steroids, as well as Rickettsia-like organisms in HIV-infected patients have been reported. Peliosis may be asymptomatic, or may be responsible for hepatic failure or life-threatening intraperitoneal bleeding.[4,5]

Danazol is a synthetic androgen used in the treatment of ITP.[6] The response rate of ITP to danazol is variable.[7] It may exhibit synergic action with steroids, which are considered standard initial treatment of ITP, reduces the need for steroids and may even replace them once remission has set in. Danazol is a well-tolerated drug. However, some unfavorable effects have been described.[6,7] More significantly, danazol is regarded as a potential cause of hepatic injury, including cholestatic hepatitis, peliosis and neoplasia.[4-6,8.]

We present a 79-year-old male patient diagnosed with ITP in July of 1994 (8×10^9 platelets/L). Initially, he was treated with non-specific gammaglobulins (25 g/day, 5 days) with a favorable response (239×10^9 platelets/L). Later he developed a new episode of severe thrombocytopenia(19×10^9 platelets/L), and treatment with prednisone (1 mg/kg/day) was started, with response after three weeks of treatment (160×10^9 platelets/L). Steroid-related effects developed thereafter (diffuse osteoporosis, vertebral collapse, myopathy and behavior disorders) and the dose was therefore reduced (0.1 mg/Kg/day). In January, 1995, danazol was added to the treatment (400 mg/day) with gradual normalization of platelet count. This allowed the gradual tapering of steroids, which were discontinued in July, 1995 (214×10^9 platelets/L). Two months later the patient was admitted to hospital with acute abdomen. Ultrasonography showed evidence of hemoperitoneum and spleen rupture. An emergency splenectomy was performed. The patient developed multi-organic failure, dying 11 days after surgery. The histopathologic findings showed a ruptured spleen with extensive splenic peliosis. No liver biopsies were obtained.

Peliosis appears to be the general histopathologic expression of a wide range of agents capable of damaging viscera, particularly the liver and spleen. How peliosis develops is unclear. One hypothesis relates its appearance to sinusoidal barrier damage.[4] In HIV patients with peliosis, treatment of the rickettsial infection resolved both the hepatosplenomegaly and also liver function test abnormalities, which suggests the possible reversibility of the injury.[9,10]

Danazol has been said to play an etiologic role in peliosis development. Nesher[5] and Makdisi[4] have published two cases of patients with hepatosplenic peliosis who received danazol as treatment for ITP. Because the exposure to danazol in both patients was brief; these reports did not clearly demonstrate that danazol was the causal agent. However, it is probable that danazol could have had an additive or synergistic effect with other potential causes of peliosis like steroid therapy. The patient we present developed splenic rupture while being exposed to this drug, steroid treatment having been stopped three months before.

We feel that this report strongly suggests that danazol plays a main casual role in the development of splenic peliosis. It is tempting to speculate that the effects of steroids and danazol on endothelial function could converge not only in their therapeutic effect but also in the development of peliosis.

We think that the clinician must weigh the potential benefits of danazol with the possible development of serious complications, such as hepatic failure or

splenic rupture due to peliosis, specially in patients with myeloproliferative disorders and/or receiving glucocorticoids. In these patients, prompt detection of enlargement of the liver or spleen and careful monitoring of liver function tests may be rewarding, as peliosis can be reversible after stopping these drugs.

Key words

Peliosis, splenic, immune thrombopenia, danazol

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Organization of an umbilical cord blood transplant program

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The development of human umbilical cord blood transplants with hematopoietic repopulating cells has enabled some problems associated with bone marrow transplants to be solved. Frozen umbilical cord blood banks should facilitate the finding of suitable stem cell donors. However, further experience is necessary to develop the optimal method for collection, separation, storage and cryopreservation of umbilical cord blood. We report our experience in the organization of a Cord Blood Bank.

Bone marrow transplants from related donors are the only alternative for some genetic, neoplastic, and non-neoplastic diseases, but they require an HLA identical donor, or with one mismatched antigen at most. Human umbilical cord blood (UCB) contains hematopoietic stem/progenitor cells and might be a clinically useful source of transplantable hematopoietic repopulating cells.[1]

In 1993 the *New York Blood Center* created the first cord blood bank for hematopoietic stem cell transplantation. In Spain there are two banks authorized by the National Transplant Organization, one in Barcelona and the other in Malaga. It is important to define the problems involved in organizing Cord Blood Banks.[2] The different international centers should be associated in an *International Cord Group*, such as *Eurocord*. The legislation should be similar to that applied to the transplantation of other tissues.[3]

We report our experience in the organization of a cord blood bank. UCB was collected from mothers of children who were candidates for a bone marrow transplant and from others for the constitution of an unrelated donor cord blood bank. Cord blood was not collected if there were obstetric complications. We had previously studied the organizational structure of the obstetrics unit which was co-ordinated with the cord blood bank. Subjects were recruited from the *Materno-Infantil Hospital* at the time of admission to labour and delivery. Human UCB samples were obtained from normal full-term vaginal deliveries. The mothers were from families with no known genetic disease and they gave written informed consent prior to delivery. Women with a history of a sexually transmitted disease, hepatitis, or other infectious disease were excluded from the study, even if an analysis was negative.

The method used by us for blood cord collection was the blood bag sterilized by betadine, to obtain the maximum volume for each collection in order to separate the cord blood mononuclear cell population. This is the method most commonly used.[4] It may be important to reduce the cryopreserved cord blood volume from 100 mL (±50) to 50 mL or less.

We used conventional 350 mL blood bags with CPD-A, reducing the anticoagulant volume to 25 mL in a sterile laminar flow hood. The modified bag and the samples for immunohematological controls, bacteriological and fungal cultures, flow cytometry and hematopoietic progenitor cell count were sterilized

and packed. The obstetrics staff kept the sterile packs awaiting collection by the UCB.

Immediately after delivery the umbilical cord was double clamped and transected 5 cm from the navel. Within 30 seconds the umbilical vein was catheterized, and the blood was collected by gravity into a 300 ml blood bag containing 25 mL of CPD-A, which was carefully shaken during collection to prevent blood clots. The doubly clamped blood bag was taken to the cord blood bank with two samples of maternal blood for standard controls. The umbilical cord blood was kept at 22°C in continuous agitation until processing within 24h to 48h.

HLA typing studies were made first for HLA class I antigens by serology, defining the split antigens DRB and DQB by generic DNA typing. Later, DNA typing for DRB, DQA, DQB and DPB HLA antigens was performed.

The personnel involved in this program is shown in Table 1. A total of 400 samples of umbilical cord blood were collected from full-term infants in 19 months, from January 1996 to July 1997. We examined the volume collected, the total mononuclear cells, the number of $CD34^+$ cells, and checked for the presence of infectious diseases or bacteriological and fungal contamination.

The mean volume of the 400 samples of umbilical cord blood collected over 19 months was 71 mL (range: 31-128), the mean number of total cells was 8.46×10^8 (range: 1.60-29.10), the mean number of mononuclear cells was 5.44×10^6 (2.00-22.5), and the mean perccetage of $CD34^+$ cells was 0.26% (0.10-0.90). Microbiological contamination was found in 35 samples (8.7%) (Table 2). Maternal blood tests for AgHBs, antiHCV, antiHIV or syphilis RPR were positive in 1.25% of the cases (2 HIV were initially reactive but Western-Blot negative, and 3 antiHCV were ELISA reactive).

Table 1. Umbilical cord blood program: human resources.

Department	*Obstetrics*	*CBB**	*Immunol*	*Serologic*	*Bacteriol*	*Hematol*	*Total*
Physician	2	1	1	–	–	1	5
Midwife	25	–	–	–	–	–	25
Technician	–	1	1	1	1	1	5
Trainer	–	2	–	–	–	–	2
Total	27	4	2	1	1	2	37

CBB: Cord Blood Bank.*

Umbilical cord blood from a single birth contains a number of stem/progenitor cells within the range required for autologous and HLA compatible allogeneic transplantation in both the infant and the adult. The establishment of banks has been proposed in order to store frozen cord blood samples.[5] Umbilical cord blood can be used fresh, with minimal periods of cryopreservation, or after a long period of frozen storage. Several studies have attempted to determine the optimal collection, separation and cryopreservation techniques for cord blood banking and transplantation.[6,7]

An important question is the number of litres of nitrogen necessary to store a determinate number of UCB units. Many studies have been aimed at reducing the volume of the cord blood per unit to 24.1 mL, using a closed system and centrifuging the buffy coat, thereby reducing the storage requirements and resulting in a good recovery of MNC, lymphocytes, $CD34^+$ cells and CFU.[8]

Table 2. Microbiological contamination in samples of cord blood.

Bacillus sp.	1
Citrobacter freundi	1
Candida albicans	1
Clostridium sp.	1
Corynebacterium sp.	4
Escherichia coli	6
Enterococcus durans	1
Escherichia faecalis	1
Staphylococcus	7 (1 *staphylococcus aureus*, 1 *staphylococcus warnieri*, 2 *staphylococcus hominis*, 2 *staphylococcus epidermidis*, 1 *staphylococcus haemolyticus*)
Streptococcus	8 (5 *streptococcus agalactiae*, 1 *streptococcus bovis*, 1 *streptococcus microaerofilico sp*, 1 *streptococcus lactobacillus sp*)
Proteus mirabilis	2

The staff involved in the program, belonging to two hospitals and six departments to a total of 37 persons including physicians, midwives, technicians and assistant staff, create an important organizational challenge, similar to that of the co-ordination of organ and tissue transplants.

The *National System of Public Health* pays the costs, which are distributed between each of the integrating centres of the program. The greatest workloads are in the *Umbilical Cord Blood Bank*, which requires a specific full-time technician for every 4 cryopreserved UCB units/day, and in the histocompatibility laboratory which carries out the typing study.

It is necessary to make a decision about the pre-cryopreservation period, that is whether the samples should be stored at 22°C or 4°C during periods of 24 or 48 hours. Results so far on pre-cryopreservation storage at 22°C show a higher significant post-thaw viability than storage at 4°C. No significant differences in cell viability or nuclear cell count were noted post-thaw when storage for 24 hours was compared with storage for 48h.[7]

The minimum acceptable volume was 30 mL. It was difficult to obtain a volume superior to 120 mL, though a mean volume around 70 mL is considered

acceptable for use in one blood bag. Our mean nuclear cellularity was 5.44×10^6, but this could possibly be increased by raising the collected blood volume. Checking for the existence of transmissible diseases must be performed in the maternal blood, the IgG antibodies levels are very low or undetectable in the newborn. Positive serological results invalidate the UCB for transplant. The serological tests are repeated in the mothers 3-6 months after the birth.

In conclusion, the establishment of Cord Blood Centers/Banks could open new fields in the procurement of donors for transplantation.

Key words

Program umbilical cord blood, cord blood bank, umbilical cord blood, blood donors

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Incidence and prognostic significance of idiopathic thrombocytopenic purpura in patients with Hodgkin's disease in complete hematological remission

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Idiopathic thrombocytopenic purpura (ITP) is a frequent and well recognized complication of lymphoproliferative diseases (especially chronic lymphatic leukemia),[1] but it is an unusual and poorly documented disease in the acute phase (1-2%).[2] We report on two female patients out of 812 patients with Hodgkin's disease (HD) followed between 1970 and 1995 at the Institute of Hematology and Medical Oncology "Seràgnoli" in Bologna, who developed ITP unrelated to bone marrow failure, 26 and 15 months after achievement of complete hematologic remission from HD.

Case #1. A 22-year-old woman presented in May 1991 with fatigue, fever and adenopathy in the right supraclavicular region. A chest radiograph defined the presence of intrathoracic adenopathy. Supraclavicular lymph nodes' biopsy revealed HD of nodular sclerosing type. A full blood count revealed hematocrit 44%, leukocyte count 0.54×10^9/L and a normal platelet count, described as being normal from the smear. The patient was placed in stage IIB, and chemotherapy was begun in June 1991. She received 3 cycles of MOPP protocol and 3 cycles of ABVD regimen.[3] At the end of chemotherapy, she began radiotherapy, receiving a total of 36 Gy to the mediastinal area.[4] The patient achieved complete remission (CR) in February 1992. Over the next 24 months she remained well and numerous complete blood counts were normal. In June 1994 she suddenly noted extensive purpura and easy bruising; examination revealed only ecchymoses and petechiae. The spleen was not palpable and there was no clinical evidence of recurring HD. The hematocrit was 44.8%, leukocyte count was 4.8×10^9/L with a normal differential count and the platelet count 4.0×10^9/L. Posterior iliac spine bone marrow biopsy revealed marrow in which erythroid and myeloid maturation appeared normal. No granulomas, tumor cells or any increase in fibrous tissue were seen. Roentgenograms of the chest and abdomen were negative. The diagnosis of ITP was suspected, and a trial of corticosteroids was begun. After 30 days, the platelet count was normal and corticosteroids were stopped. Over the next 14 months the full blood count remained normal. In September 1995 she again noted ecchymoses and petechiae. The blood platelet count was 5.0×10^9/L and an ITP relapse was diagnosed. After steroid therapy for two months without evidence of increased platelets, splenectomy was performed.[5] The platelet count rose immediately after splenectomy and remained normal. After two months, an autoimmune hemolytic anemia (AHA) appeared and the patient achieved a partial remission after prednisolone therapy. To date, there is no evidence of HD or ITP relapse.

Case #2. In December 1993, a 29-year-old woman noted enlarged lymph nodes in her right supraclavicular region. Lymph nodes biopsy revealed nodular sclerosing type HD. CT scanning of the chest, abdomen and pelvis revealed lymph node involvement of the anterior mediastinum without involvement of bone marrow. The patient was placed in stage IIIA and chemotherapy was started in January 1994. She received 6 cycles of ABVD regimen.[3] After completing chemotherapy she received the scheduled

radiation therapy to the mediastinum (36 Gy). The patient achieved CR in February 1995. Over the next 14 months she remained well and in complete hematologic remission. In May 1996 she noted extensive ecchymoses and petechiae. Her hematocrit was 43.2%, leukocyte count 0.64×10^9/L and the platelet count 1.0×10^9/L. The spleen was not palpable and there was no evidence of HD relapse. A bone marrow biopsy revealed normal erythroid and myeloid maturation. The diagnosis of ITP was suspected and a trial of corticosteroids was begun. After 30 days the platelet count was normal (224×10^9/L) and corticosteroids were stopped. To date, the patient remains in complete hematologic (ITP) and HD remission. Although some studies have reported that the presence of ITP in HD patients could signify active lymphoma, especially with splenic involvement,[2] other reports have shown that ITP can occur in the absence of active HD,[6] and that the HD status is independent of the onset of ITP.

Our two patients did not have active HD when they developed ITP, and the development of ITP did not indicate subsequent relapse, supporting the hypothesis of two independent diseases with different onsets. Furthermore, as for primary ITP occurring in association with HD, so thrombocytopenic purpura also seems to respond to conventional therapy.[2] Both patients responded with a single course of oral prednisolone, but one relapsed after 15 months and splenectomy was necessary for CR.[5]

The recognition of a picture of ITP as a complication of HD could have important implications, as described by Doan and Bouroncle.[7] We suggest, therefore, that an occult HD should be considered in any patient who presents with thrombocytopenic purpura of the ITP type, and particularly in a patient known to have been previously treated for HD, where the ITP picture may signal the recurrence of lymphoma. It should be noted that we have also observed thrombocytopenic purpura in chronic myeloid leukemia (CML) during treatment with interferon-α.[8]

In conclusion, ITP associated with HD responds to therapy in a similar manner to that of primary ITP. Its presence seems to confer no prognostic significance,[9] although the possibility of localization of splenic HD disease[10] has to be considered.

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Bacteremia caused by CDC Group IV c-2 in a patient with acute leukemia

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Human infections due to CDC group IV c-2, a gram-negative bacillus, are rare. We describe a case of nosocomial bacteremia caused by this organism in a neutropenic patient with acute lymphoblastic leukemia and include a literature review of CDC group IV c-2 infection in patients with hematologic malignancies.

Human infections due to CDC group IV c-2, a gram-negative bacillus, have been recently reported.[1-8] Bacteremia caused by this organism is rare in patients with hematologic malignancies.[2,5,6,8]

We report here a case of septicemia caused by CDC group IV c-2 in a neutropenic patient with acute leukemia and review the literature on this entity (MEDLINE, National Library of Medicine, Bethesda, MD covering the years 1965-97).

A 71-year-old previously healthy woman was admitted to our hospital with a temperature of 37.4°C, dyspnea and pleuritic chest pain. The patient had a history of fatigue, weakness and weight loss of 10 kg over the three previous months. Physical examination revealed cutaneous pallor, shortness of

breath and a few crackles at the left lung base. White blood cell count was 188×10^9/L with 95% blastic forms, the hemoglobin level was 6.8 g/dL and platelet count 44×10^9/L. A chest radiograph showed a left lower lobe infiltrate and a small left pleural effusion. Empirical therapy with amoxicillin/clavulanate was administered intravenously. On the second day of admission the patient was afebrile. Sputum and blood cultures were negative.

Microscopical examination of a bone marrow aspirate revealed acute lymphoblastic leukemia, L-2 according to the criteria of the FAB classification. Immunophenotype was consistent with ALL-preB. An induction chemotherapy regimen consisting of daunorubicin, vincristine, L-asparaginase and prednisone was initiated.

After two weeks of antibiotic treatment, the patient's condition improved and a new chest radiograph was normal. Amoxicillin/clavulanate was stopped and oral prophylaxis with norfloxacin was then initiated. Twelve days later, while neutropenic, the patient's temperature rose to 38.5°C and her blood pressure fell to 80/50 mm Hg. After acquisition of two specimens for blood culturing, fluid replacement was initiated and, ceftazidime and amikacin were given intravenously. Twenty-four hours after beginning empirical antibiotic therapy she became afebrile with no clinical signs of infection. A gram-negative rod was isolated from all the blood cultures. Cultures from urine and indwelling line specimens were negative.

The chemotherapy course was complicated by several episodes of intestinal bleeding due to severe thrombocytopenia. Several days later, the patient became agitated and severe hypotension and oligoanuria developed despite fluid replacement and inotropic drug therapy. The patient died because of a new bleeding episode. At that time blood cultures were negative.

The isolate was identified as CDC group IV c-2 by conventional methods from the blood cultures after 72 hours. Its microbiological characteristics were consistent with those previously defined for the bacillus.[9,10] With standard disk diffusion testing, the organism was susceptible to trimethoprim-sulfamethoxazole, ciprofloxacin and ceftazidime, but was resistant to ampicillin, amoxicillin plus clavulanate, piperacillin, ticarcillin, ticarcillin plus clavulanate, cephalothin, cefoxitin, cefotaxime, ceftriaxone, imipenem, aztreonam, gentamicin, tobramycin, amikacin and tetracycline.

CDC group IV c-2 is a gram-negative environmental bacillus rarely isolated from clinical specimens. However, several cases of well-documented infections caused by this organism have been reported, with eight cases of septicemia (five of them occurring in a hospital outbreak),[2,5,6,8] two cases of peritonitis related to peritoneal catheter infection,[1,4] one case of plantar abscess complicated with bacteremia[3] and one case of tenosynovitis.[7] All but one of these patients had underlying disease. We found five previously reported cases of infection caused by CDC IV c-2 in patients with hematologic malignancies in our review of the literature.[2,5,6,8] These 5 cases with the addition of our case are summarized in Table 1. Four patients had acute leukemia and two non-Hodgkin's lymphoma. Our review shows that most of the infections were nosocomial-acquired and presented with bacteremia, mainly catheter-related, occurring during chemotherapy-induced neutropenia.

In spite of the well-known severity of gram-negative bacteremia in patients with cancer, all reported patients with bacteremia caused by CDC group IV c-2 were cured from infection following appropriate antimicrobial therapy, probably indicating a low level of pathogenicity of this organism. It should be noted that catheter removal was only necessary in one of these cases.

It has been shown that CDC group IV c-2 is susceptible to a wide range of antimicrobial agents, including aminopenicillins, antipseudomonal penicillins, cephalosporins, carbapenems and fluoroquinolones. However, this organism is usually resistant to aminoglycosides.[1-8,10] In our case, the organism showed an unusual pattern of antibiotic-susceptibility, showing resistance to a broad spectrum of antimicrobial agents. To our knowledge, this is the

Table 1. CDC group IV c-2 infection in patients with hematologic malignancies.

Reference	*Age/Sex*	*Underlying condition*	*Neutropenia*	*Specimen cultured*	*Source of infection*	*Acquisition*	*Outcome*
Dan *et al.*[2]	37/M	Plasma cell leukemia	No	Blood	Unknown	Community	Recovery
Arduino *et al.*[5]	77/M	Non Hodgkin's lymphoma	No?	Blood	Catheter	Nosocomial	Recovery
Ramos *et al.*[6]	10/F	Acute leukemia	Yes	Blood	Catheter	Nosocomial	Recovery
Moissenet *et al.*[8]	11/NR	Non Hodgkin's lymphoma*	Yes	Blood	Catheter	Nosocomial	Recovery
Moissenet *et al.*[8]	14/NR	Acute leukemia*	Yes	Blood	Catheter	Nosocomial	Recovery
Salar *et al.*[PR]	77/F	Acute leukemia	Yes	Blood	Unknown	Nosocomial	Recovery

*PR, present report; NR, not reported; *cases reported in a hospital outbreak of CDC group IV c-2 infection.*

first report in which resistance of this bacillus to imipenem is noted.

In summary, CDC group IV c-2 bacillus is an opportunistic pathogen that rarely causes bacteremia in patients with hematologic malignancies and indwelling intravenous lines.

Key words

CDC group IV c-2, bacteremia, acute leukemia, neutropenia.

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