The objective of this study was the evaluation of clinical, demographic and treatment-associated mortality factors in patients with diffuse alveolar haemorrhage (DAH) associated with systemic lupus erythematosus (SLE). Clinical, laboratory test, SLEDAI-2K, predictors of mortality (APACHE II) and different treatments including cyclophosphamide, methylprednisolone and rituximab were evaluated in SLE patients who were diagnosed with DAH, to determine potential association with mortality. Twenty-nine episodes of DAH in 22 SLE patients were included (one patient with four episodes, four patients with two episodes (seven recurrences)), 15 died. Mean age was 25.1 years and 1.5 years of SLE evolution with haemoglobin drop 3.4 g/dl. In 4 of 22 patients, the DAH diagnosis was confirmed by autopsy. Six episodes were in patients under 18 years of age (2 patients with recurrence). DAH was the initial manifestation of SLE in 10 patients. Of the 22 patients, 17 were women and 22/29 had DAH episodes. Dyspnoea and nephritis occurred in all patients, less common were arthritis (75.9%) and fever (65.5%); haemoptysis was present only in 44.8%. Through evaluation of all included factors, only thrombocytopenia, renal failure, requirement for mechanical ventilation and high APACHE II were associated with higher mortality. Cyclophosphamide use was associated with less mortality (not statistically significant).

Key words: diffuse alveolar haemorrhage; systemic lupus erythematosus

Introduction

Diffuse alveolar haemorrhage (DAH) associated with systemic lupus erythematosus (SLE) has been found in 0.5% of SLE patients admitted to a hospital in Taiwan, 5.4% of a cohort of 630 Mexican patients, 18.4% of Mexican hospital autopsies, and 29 of 120 autopsies at The Johns Hopkins Hospital. Alveolar haemorrhage occurs more frequently in association with diseases other than SLE. The low prevalence and difficulties in achieving specific diagnosis have probably contributed to the slow progress related to this pathology. Even though the first report of DAH associated with SLE was more than 100 years ago from a description by Osler in 1904, the progress has been very poor compared for example with the outcome for lupus nephritis. Mortality of DAH associated with SLE is high, although some series are encouraging with survival greater than 80%. Few publications have evaluated the prognostic factors; Zamora et al. reported mechanical ventilation, the presence of infection and the use of cyclophosphamide as mortality related factors; Chang et al. reported eight cases in which there were differences in APACHE II (Acute Physiology, Age and Chronic Health Evaluation) and organ system failure (OSF) scores. Barile et al. reported that the ‘massive treatment’ with 3 g or more of methylprednisolone was associated with benefit. Badsha et al. found differences in creatinine and requirement of mechanical ventilation. Recently Shen et al. found differences in infection and mechanical ventilation when comparing survivors and non-survivors. Few case reports have described the use of new biologic drugs such as rituximab. In our study, the main objective was...
to evaluate the clinical, demographic and treatment-associated mortality factors in patients with DAH associated with SLE.

Materials and methods

Study design
We performed a retrospective cohort study of case series.

Patient selection
Since January 2004 all our patients with SLE, according to the American College of Rheumatology (ACR) criteria of classification, and DAH were treated under the same management protocol and were evaluated.

Management protocol
All patients received antibiotics (covering Gram-negative bacteria) and 1 g of methylprednisolone for 24 hours or its equivalent in paediatric patients for at least 3 days after admission or at DAH diagnosis; the dose was continued until there was no evidence of bleeding. Cyclophosphamide was administered 500 mg/m² of body surface area if within 24 hours the patient had no infection evidence. Rituximab was administered to patients financially solvent (main limitation in our hospital) and who had evidence of ongoing bleeding after 3 days of treatment.

DAH definition
The presence of new lung infiltrates in the chest radiograph suggestive of pulmonary haemorrhage, abrupt drop in haemoglobin of at least 2 g/dl without evidence of bleeding elsewhere, with or without the presence of the following symptoms and signs: dyspnoea, haemoptysis and haemosiderophages in bronchoalveolar lavage.

Recurrent DAH
Recurrent DAH was defined when a new episode occurred after complete resolution of previous DAH with asymptomatic period and normal chest radiograph or computed tomography.

Clinical measurements at admission

Demographic factors
The demographic factors were sex, age and smoking.

SLE evolution and treatment
SLE evolution time, previous use of steroids or immunosuppressive treatment were measured.

Clinical manifestations
The clinical manifestations were dyspnoea, haemoptysis, fever, chest pain, mucositis, arthritis, serositis, pericarditis, haemolytic anaemia, neuropsychiatric and cutaneous manifestations.

Laboratory tests
Haemoglobin, haemoglobin drop, leukocytes, lymphocytes, platelets, serum creatinine, C-reactive protein (CRP), glomerular filtration rate (GFR) calculated by the CKD-EPI equation, proteinuria, presence of dysmorphic red cells and serum levels of complement were recorded. Cultures of blood, sputum and urine were taken at admission.

Treatment and management factors
We recorded mechanical ventilation, days of mechanical ventilation, haemodialysis and medications used to treat DAH (antibiotics, methylprednisolone, cyclophosphamide, rituximab, intravenous immunoglobulin).

Prognostic factors described previously
Diagnosis of infections, Systemic Lupus Erythematosus Disease Activity Index 2000 (SLEDAI-2K) and APACHE II scores had been described previously. Renal failure was defined as estimated GFR <60 ml/min/1.73 m², nephritis was defined as the presence of dysmorphic red cells and/or casts on urinalysis and thrombocytopenia as platelets <150,000/mm³.

Haematology test, haemoglobin (Hb), white blood cell (WBC) count, differential count of WBC, platelets and nucleated red blood cell were counted using an XE-2100 automated haematology analyser; creatinine measurement was made with kinetic enzymatic method, CRP was evaluated by immunoturbidimetric method, urinalysis was evaluated for proteins, cells and casts with high power field.

Statistical analysis
As the episodes were at different times, with diverse clinical features, received different treatment, and results obtained were different (outcome from resolution to death), each episode (in the same patient in case of recurrence) was considered for analysis as a ‘different episode’ even though DAH recurrence had developed in the same patient, to compare patients who died and who lived.
Continuous data were reported as means and standard deviations (SD) for normally distributed data. Non-normally distributed data are expressed as median (upper–lower limits) and percentages for categorical variables. Means were compared using Student’s *t*-test, medians with Mann–Whitney *U*-test; categorical data were compared using chi-square test or Fisher’s exact test. Statistical analysis was made using SPSS 15 (SPSS, Chicago, IL).

Results

From January 2004 until March 2010, 321 SLE patients were admitted; 29 episodes of DAH presented in 22 SLE patients (29 DAH associated with SLE in 22 patients) were evaluated (one patient with four episodes, four with two episodes (seven recurrences)), 51.7% (15/29) died and in 4 of these patients the diagnosis was confirmed by autopsy. The mean age was 25.1 ± 10.4 years, 6 episodes were in patients under 18 years (2 patients with recurrence, another 2 patients with one episode). Of the 22 patients, 17 were women and had 22 of the 29 episodes (Table 1). Of 22 patients only 3 (one with 4 episodes of DAH) manifested speckled pattern antinuclear antibodies, the other 19 had homogeneous and peripheral patterns. Anti-DNA and complement levels were evaluated in 11 of our 22 patients, all of them had high anti-DNA levels and 9 had hypocomplementemia.

From the 29 episodes, 10 (34.5%) had DAH as the first manifestation of SLE and in 17 (58.6%) this massive haemorrhage occurred during the first year of SLE diagnosis. The mean duration of disease at DAH diagnosis was 1.5 ± 2.2 years; 24.1% of the episodes were in autumn, 31% in winter, 24.1% in spring and 20.7% in summer. In 14 patients the type of nephropathy was known: 9 were type IV, 2 were type V, 2 of them were type IV–V and one type III–V of the World Health Organization (WHO) classification. During the previous month of the DAH in 15/29 episodes (51.7%), patients had received treatment with steroids, on average 35.3 mg of prednisone per day or its equivalent; 13 (44.8%) were treated with one or more additional immunosuppressive therapy (disease-modifying anti-rheumatic drugs [DMARDs]); 11 with methotrextate (37.9%) as the most frequent, followed by 8 with azathioprine (27.6%), 3 with chloroquine (10.3%), 3 with cyclophenolate mofetil (10.3%), 2 with cyclophosphamide (6.9%) and only one patient (3.4%) received rituximab. One patient had hypothyroidism, one type 2 diabetes mellitus, and three had secondary antiphospholipid syndrome (only one was under anticoagulant therapy). Dyspnoea and nephritis were the most common symptoms (all patients), followed by arthritis (75.9%), fever (65.5%) and haemoptysis (44.8%); 31% had chest pain, 31% mucositis and mucocutaneous manifestations, 24.1% neuropsychiatric manifestations and 6.9% pericarditis (Figure 1). Of 13 patients without haemoptysis, 9 of them had haemosiderophages (broncho-alveolar lavage) and 4 were not pursued.

The mean haemoglobin drop was 3.4 g/dl. Of all episodes, 21 (72.4%) required mechanical ventilation (6.8 ± 5.4 days). Table 2 shows laboratory test and prognostic factors measured when patients were admitted.

In 10 patients, bacterial infections (1 polymicrobial) were documented in culture processed at admission (Staphylococcus aureus in 5 patients, Citrobacter freundii in 1 patient, Rothia dentocariosa in 1 patient, Pseudomonas aeruginosa in 2 patients and Gram-negative bacilli in 2 patients), in 3 patients fungal infection was documented with biopsy or autopsy (cryptococcosis, coccidioidomycosis and systemic mucormycosis + aspergillosis), and 1 patient had disseminated cytomegalovirus infection. Inpatient time was 10.7 ± 8.8 days, and of patients who died, 60% were in first 7 days. Median dose of methylprednisolone was 3 g (2–5 g). In 58.6% of cases (17 episodes) the patients received cyclophosphamide, in 2 (6.8%) cases they received rituximab (both died) and in 1 additional patient was treated with intravenous immunoglobulin (also died).

Through evaluation of all included factors, only thrombocytopenia, renal failure, requirement for mechanical ventilation and high APACHE II were associated with increased mortality (Tables 2 and 3).

Risk factors found in our study were re-evaluated for cyclophosphamide, and we did not find

<table>
<thead>
<tr>
<th>Table 1 Main demographic characteristics</th>
<th>All</th>
<th>Deceased</th>
<th>Alive</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>Patients (n)</td>
<td>22</td>
<td>15</td>
<td>7</td>
<td>NA</td>
</tr>
<tr>
<td>Episodes (n)</td>
<td>29</td>
<td>15</td>
<td>14</td>
<td>NA</td>
</tr>
<tr>
<td>Female (%)</td>
<td>75.9</td>
<td>41.4</td>
<td>34.5</td>
<td>0.682</td>
</tr>
<tr>
<td>Age (mean ± SD)</td>
<td>25.1</td>
<td>27.7</td>
<td>22.4</td>
<td>0.172</td>
</tr>
<tr>
<td>SLE evolution (years)</td>
<td>1.5</td>
<td>1.3</td>
<td>1.6</td>
<td>0.613</td>
</tr>
<tr>
<td>Previous steroids (%)</td>
<td>51.7</td>
<td>27.6</td>
<td>24.1</td>
<td>0.858</td>
</tr>
<tr>
<td>Previous DMARD (%)</td>
<td>44.8</td>
<td>20.7</td>
<td>24.1</td>
<td>0.588</td>
</tr>
</tbody>
</table>

NA: Not applicable; DMARD: Disease-modifying anti-rheumatic drug.
statistically significant differences, but in general, the patients who received cyclophosphamide had more of the risk factors found (associated with mortality) than non-cyclophosphamide patients: 7/12 patients with thrombocytopenia received cyclophosphamide, 6/12 with renal failure, in addition, 13/21 patients with mechanical ventilation received this drug.

Discussion

Our series highlights the clinical characteristics of a good number of patients, the use of newly introduced drugs (such as rituximab), and the likely benefit of cyclophosphamide in addition to highlighting the factors associated with mortality.

To our knowledge, our series is one of the largest reported,1–3,8–12,19–25 assessing both clinical data as well as factors associated with mortality, including six episodes in paediatric-age patients (Table 4), a rare eventuality with only a few cases reported in the literature.26–28 Moreover, we have described multiple patients with recurrent DAH.8–10,23,25,28–31 Even though Santos-Ocampo et al.8 describe survival of 100%, the mortality of DAH associated with SLE continues to be very high; the clinician must analyse these data because Santos-Ocampo et al. included patients with a haemoglobin drop of only 0.5 g/dl.8

Table 2

<table>
<thead>
<tr>
<th></th>
<th>All (mean ± SD)</th>
<th>Alive (mean)</th>
<th>Deceased (mean)</th>
<th>p-value</th>
</tr>
</thead>
<tbody>
<tr>
<td>CRP (mg/dl)</td>
<td>9.9 ± 8.1</td>
<td>7.3</td>
<td>12.4</td>
<td>0.137</td>
</tr>
<tr>
<td>Leukocytes (×10³/mm³)</td>
<td>10.8 ± 8.9</td>
<td>10.4</td>
<td>11.1</td>
<td>0.832</td>
</tr>
<tr>
<td>Lymphocytes (×10³/mm³)</td>
<td>0.72 (0.1–4.6)</td>
<td>0.98</td>
<td>0.54</td>
<td>0.252</td>
</tr>
<tr>
<td>Haemoglobin (g/dl)</td>
<td>6.9 ± 1.7</td>
<td>7.0</td>
<td>6.8</td>
<td>0.772</td>
</tr>
<tr>
<td>Platelets (×10³/mm³)³</td>
<td>162 (4–496)</td>
<td>224</td>
<td>144</td>
<td>0.055</td>
</tr>
<tr>
<td>Creatinine (mg/dl)³</td>
<td>1.2 (0.3–31.9)</td>
<td>0.79</td>
<td>2.54</td>
<td>0.016</td>
</tr>
<tr>
<td>GFR (ml/min/1.73 m²)</td>
<td>73.4 ± 51.9</td>
<td>104.1</td>
<td>42.9</td>
<td>0.001</td>
</tr>
<tr>
<td>SLEDAI-2K</td>
<td>17.1 ± 7.5</td>
<td>15.9</td>
<td>18.3</td>
<td>0.416</td>
</tr>
<tr>
<td>APACHE II</td>
<td>18.4 ± 6.1</td>
<td>15.7</td>
<td>21.1</td>
<td>0.015</td>
</tr>
</tbody>
</table>

³Median (minimum–maximum). SD: standard deviation; CRP: C-reactive protein; GFR: glomerular filtration rate; SLEDAI-2K: Systemic Lupus Erythematosus Disease Activity Index 2000; APACHE II: Acute Physiology And Chronic Health Evaluation.

Table 3

<table>
<thead>
<tr>
<th></th>
<th>All</th>
<th>Alive</th>
<th>Deceased</th>
<th>p-value</th>
<th>RR</th>
<th>95% CI</th>
</tr>
</thead>
<tbody>
<tr>
<td>Thrombocytopenia (%)</td>
<td>41.4</td>
<td>10.3</td>
<td>31.0</td>
<td>0.015</td>
<td>5.5</td>
<td>1.1–28.4</td>
</tr>
<tr>
<td>Renal failure (%)</td>
<td>41.4</td>
<td>6.9</td>
<td>34.5</td>
<td>0.004</td>
<td>12.0</td>
<td>1.9–75.7</td>
</tr>
<tr>
<td>Haemodialysis (%)</td>
<td>27.6</td>
<td>6.9</td>
<td>20.7</td>
<td>0.215</td>
<td>NA</td>
<td>NA</td>
</tr>
<tr>
<td>MV (%)</td>
<td>72.4</td>
<td>24.1</td>
<td>48.3</td>
<td>0.014</td>
<td>14.0</td>
<td>1.4–137.3</td>
</tr>
<tr>
<td>Cyclophosphamide (%)</td>
<td>58.6</td>
<td>34.5</td>
<td>24.1</td>
<td>0.176</td>
<td>NA</td>
<td>NA</td>
</tr>
</tbody>
</table>

MV: Mechanical ventilation; RR: Relative risk; 95% CI: 95% confidence interval for RR, NA: Not applicable.
Predictors of mortality in diffuse alveolar haemorrhage associated with systemic lupus erythematosus

MG Martinez-Martinez and C Abud-Mendoza

Table 4 Main characteristics of paediatric patients

<table>
<thead>
<tr>
<th></th>
<th>n</th>
</tr>
</thead>
<tbody>
<tr>
<td>Episodes</td>
<td>6</td>
</tr>
<tr>
<td>Female</td>
<td>2</td>
</tr>
<tr>
<td>Recurrences</td>
<td>2</td>
</tr>
<tr>
<td>Deceased</td>
<td>2</td>
</tr>
<tr>
<td>Age (years)</td>
<td>12 (6–13)</td>
</tr>
<tr>
<td>Thrombocytopenia</td>
<td>1</td>
</tr>
<tr>
<td>Renal failure</td>
<td>0</td>
</tr>
<tr>
<td>Haemodialysis</td>
<td>0</td>
</tr>
<tr>
<td>MV</td>
<td>3</td>
</tr>
<tr>
<td>Cyclophosphamide</td>
<td>2</td>
</tr>
</tbody>
</table>

As Badsha et al. reported previously in their cases, DAH was the initial manifestation of SLE in a third of our patients. Multiple series have reported previously the association of DAH and glomerulonephritis in SLE patients as occurred in our current series; it perhaps could be secondary to the pathogenic relationship between the lung and kidney deposits of antibodies and complement fractions. It is evident that patients with DAH associated with SLE may not have clinical manifestations of respiratory haemorrhage with haemoptysis, a sign that was present in only 45% of our patients, as reported previously. Haemosiderophages were found in our series in all patients even without haemoptysis (in whom were looked for it); even though these cells that support the diagnosis could not be found until after the second day of bleeding onset, haemosiderophages should be a good marker for DAH diagnosis.

As mortality associated factors we observed those described by Chang et al. such as APACHE II and mechanical ventilation, Zamora et al. reported the use of cyclophosphamide as a bad prognosis factor; in contrast, our study shows the potential benefit of cyclophosphamide (although it was not statistically significant, perhaps because of our small sample size), agreeing with Shen et al. Cyclophosphamide is perhaps used in patients with more severe disease, as we mention in the final analysis; it is difficult to give advice without analysing the risk factors described for our study; and we understand that is necessary to study more patients with multivariate analysis to evaluate this drug.

The use of rituximab has been reported with potential benefit in this pathology, and our two patients who received the drug died, both required mechanical ventilation and had deep thrombocytopenia and end-stage renal failure, all of which are recognized as bad prognostic factors in this and other reported series. Rituximab should be a drug to treat and prevent relapses because rituximab action mechanism is not limited to diminishing autoantibodies and it is also associated with cell immune regulation and modifying other protagonists involved in SLE damage such as regulatory T cells. One of our patients who died received immunoglobulin and another one mycophenolate mofetil, which are also drugs that have been used with potential benefit to achieve a better clinical response and increase survival in the treatment for DAH associated with SLE, as has been recently reported.

Among the mortality-associated factors, the infection seems to have an important role, and lower mortality has been associated when SLE with DAH patients received antibiotics as has been reported previously, and is evidently the opportune approach for infectious processes; Rojas-Serrano et al. demonstrated infection in 57% of patients with DAH associated with SLE at admission (see also Zamora et al.). All our patients received empirical antibiotics and we found after cultures that 34.5% of episodes were associated with bacterial infections (with high frequencies of S. aureus and P. aeruginosa); another of our findings, besides bacterial infection, is the relatively high frequency of deep fungal infections, a very grave process directly associated with high mortality; all of our patients with mycosis received previous immunosuppressive therapy with steroids and other drugs potentially useful in severe SLE.

One of the limitations of this work is the non-randomization of our study, but it included a large number of DAH associated with SLE patients ratifying the severity of this pathology associated with high morbidity and mortality. Another limitation of our study is the sensitivity of the cultures (blood, urine and sputum) mainly for fungal infections. In three of our patients, fungal infection was not found until necropsy study. A risk–benefit assessment of lung biopsy is necessary in selected patients with DAH associated with SLE, as it would be very useful to discard or confirm opportunistic infections, mainly in patients with immunosuppressive therapy, as well as to ratify disease activity. It is difficult to separate infection manifestations from SLE activity mainly in DAH associated with SLE, for these reasons we always decide to use antibiotic therapy and select the patients who were potential receptors to receive cyclophosphamide, but for most patients with DAH associated with SLE we should perhaps consider both factors, infection and disease activity, to explain the severe expression of pulmonary damage.
Three of our patients had antiphospholipid syndrome, a disease which is associated with DAH, particularly when deep thrombocytopenia is present. Our three patients with antiphospholipid syndrome had initial thrombocytopenia, in addition to the other mentioned factors.

We require a larger group of DAH patients associated with SLE in order to evaluate prognostic factors and define potential benefits of the different drugs used or described as of potential benefit. We must have in mind some drugs that have recently been described as potentially useful such as abatacept and tocilizumab, biologics which could have some role in this serious and potentially fatal condition.41–43

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Conflict of interest statement

All authors declare that they have no financial interests to disclose.

References

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MU Martínez-Martínez and C Abud-Mendoza

574


