Original Article

Prolonged treatment of refractory Wegener’s granulomatosis with 15-deoxyspergualin: an open study in seven patients

Wilhelm H. Schmitt1, Rainer Birck1, Peter A. Heinzel2, Ursula Göbel3, Mira Choi3, Klaus Warnatz4, Hans H. Peter4 and Fokko J. van der Woude1

1Fifth Medical Clinic, University Hospital Mannheim, Heidelberg University, Mannheim, 2Euro Nippon Kayaku GmbH, Frankfurt, 3Department of Nephrology, University Hospital Berlin-Buch, Berlin and 4Division of Rheumatology and Clinical Immunology, Department of Medicine, Freiburg University Hospital, Freiburg, Germany

Abstract

Background. A subset of patients with Wegener’s granulomatosis does not respond to daily oral cyclophosphamide (CYC) plus corticosteroids or suffers from intolerable side effects. A 6 month course of the immunosuppressant 15-deoxyspergualin (DSG) has previously been employed successfully in these refractory cases. However, there are no reports on long-term treatment with DSG.

Methods. To document the effects of prolonged DSG treatment, this study reports on seven patients suffering refractory Wegener’s granulomatosis, who were successfully treated with DSG over an average of 26.5 months (range: 11–55.5 months).

Results. Before administration of DSG, patients had experienced an average of 6.6 relapses (range: 3–12) under an average of 5.4 (range: 2–11) different therapeutic approaches, which included CYC in all cases. All suffered active disease when DSG was initiated. Four were unresponsive to CYC and three did not tolerate it. DSG (0.5 mg/kg/day subcutaneous) was given for 2–3 weeks until the leukocyte count dropped to 3000/μl, followed by a rest until a leukocyte count of 4000/μl was reached again. No other immunosuppressants besides corticosteroids were given. All patients showed a long-lasting, favourable response to DSG with complete (n=5) or partial (n=2) remission. Only one case relapsed while being treated with DSG. Termination/interruption of DSG was followed by relapse in four of five occasions. Resumption of DSG led to complete remission. Currently, five of the seven patients are still treated with DSG and are in remission. Infections, mainly of the respiratory tract, were observed in five cases and resolved after treatment. One case developed a third-degree heart block that required pacing.

Conclusions. In patients with refractory Wegener’s granulomatosis, prolonged treatment with DSG seems safe and successful.

Keywords: 15-deoxyspergualin; immunosuppression; vasculitis; Wegener’s granulomatosis

Introduction

Wegener’s granulomatosis (WG), the most common primary systemic vasculitis associated with antineutrophil cytoplasmic antibodies (ANCA), commonly takes a lethal course or results in severe organ damage if left untreated. About 90% of cases respond to daily oral cyclophosphamide (CYC) plus glucocorticosteroids (GC), at the cost of substantial treatment-induced morbidity and mortality. Complications of this treatment, such as infections, haemorrhagic cystitis, bladder cancer, lymphoproliferative disease, myelodysplasia and infertility, are experienced by >40% of the cases [1]. Furthermore, ≤25% of patients achieve no complete remission and relapses are seen in ≤50% [1]. Thus, new treatment strategies, combining less toxicity with comparable efficacy, are warranted.

15-Deoxyspergualin (DSG; generic name: gusperimus) is a synthetic analogue of spergualin, a natural product of the bacterium Bacillus laterosporus. Spergualin was discovered in 1981 by Takeuchi et al [2] in a culture filtrate while screening for natural products that inhibit the transformation of chicken embryo fibroblasts through Rous sarcoma virus. Subsequently, in various animal models of transplantation and autoimmune diseases, such as experimental
crescentic glomerulonephritis and lupus nephritis [3], the drug was shown to possess strong immunosuppressive properties. The precise mechanisms of action are not understood clearly, but from the available data it seems that DSG inhibits both T- and B-cell differentiation, possibly by blocking the nuclear translocation of transcription factor NF-κB [4–6]. In clinical trials of recurrent kidney transplant rejections, DSG, which was generally given at 3 or 5 mg/kg/day by 3 h intravenous (IV) infusion for 5–7 days, led to remission in 79% of cases and was shown to be of similar efficacy as the monoclonal antibody OKT3 [7]. Later, Hotta et al. [8] demonstrated a therapeutic potential of DSG in four patients with crescentic proliferative glomerulonephritis using a low-dose regimen of DSG, which was administered at daily doses of 0.25 and 0.5 mg/kg by 1 h IV infusion for 28 days. From pre-clinical and clinical data it appears that DSG is a potent immunosuppressant with a favourable side-effect profile, exerting no renal and liver toxicity and only reversible bone marrow suppression [7–9]. Based on these favourable results, we previously conducted an open-label pilot trial of DSG in 20 patients with crescentic proliferative glomerulonephritis using a low-dose regimen of DSG, which was administered at daily doses of 0.25 and 0.5 mg/kg by 1 h IV infusion for 28 days. From pre-clinical and clinical data it appears that DSG is a potent immunosuppressant with a favourable side-effect profile, exerting no renal and liver toxicity and only reversible bone marrow suppression [7–9]. Based on these favourable results, we previously conducted an open-label pilot trial of DSG in 20 patients with active WG or microscopic polyangiitis refractory to standard immunosuppressants [10]. Fourteen of 19 cases of WG showed a favourable response to DSG, with six complete and eight partial remissions. Four cases continued treatment with DSG after termination of the trial on a compassionate use basis. We now report the further course of these patients and of three further cases that subsequently were treated with DSG for refractory WG.

Subjects and methods

Patients

In the setting of tertiary academic referral centres (two nephrology units and one rheumatology unit), seven patients with histologically proven active refractory WG received long-term treatment with DSG. In four of the patients, treatment with DSG had been initiated between May 1999 and July 2000 within an open-label, prospective, uncontrolled multicentre pilot trial, published previously (Table 1, patients 1–4) [10]. After termination of the pilot trial, treatment with DSG was continued in these four cases on compassionate use basis. The other 16 patients of the pilot trial were switched to other treatments [10]. Three further cases (patients 5–7) were started on DSG between February 2002 and February 2003 and were treated according to the same protocol. All patients gave written informed consent before inclusion. Ethical approval for the DSG protocol had been obtained in each participating centre.

Refractory WG

This was defined according to the consensus of the European Vasculitis Study Group (EUVAS) [11] as (a) progressive disease (persistently active or relapse) unresponsive to ≥6 weeks of standard treatment (CYC/GC), (b) progressive disease in patients intolerant of standard treatment or (c) constant grumbling disease in patients who relapse after reduction or omission of cytotoxic agents.

Disease extent

The extent of disease was assessed at entry and subsequently every 4 weeks or at relapse using the disease extent index (DEI) as described previously [12]. Organ manifestations were recorded as follows: E, upper respiratory tract; L, lung; K, kidney; Ey, eye; H, cardiac involvement; S, skin; C, central nervous system; P, peripheral nervous system; A, arthritis/arthralgia and myalgia; GI, gastrointestinal manifestation (each organ: two points); B, constitutional symptoms (fever >38°C, undesired weight loss >10% of body weight within <6 months) (one point). The maximum possible score was 21 points.

Table 1. Patient characteristics

<table>
<thead>
<tr>
<th>Case (no.)</th>
<th>Age (years)</th>
<th>Gender</th>
<th>Cumulative histological findings</th>
<th>Cumulative disease extent/DEI</th>
<th>Duration of WG at start of DSG (years)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>37</td>
<td>M</td>
<td>Nose: vasculitis, granuloma</td>
<td>E, subglottic stenosis, L, K, A, B 9</td>
<td>4</td>
</tr>
<tr>
<td>2</td>
<td>52</td>
<td>F</td>
<td>Jejunum: granuloma</td>
<td>E, K, Ey, GI, P, A, B 13</td>
<td>3</td>
</tr>
<tr>
<td>3</td>
<td>36</td>
<td>F</td>
<td>Nose: vasculitis, granuloma</td>
<td>E, subglottic stenosis, L 4</td>
<td>7</td>
</tr>
<tr>
<td>4</td>
<td>64</td>
<td>M</td>
<td>Nose: vasculitis, granuloma</td>
<td>E, L, A 6</td>
<td>5</td>
</tr>
<tr>
<td>5</td>
<td>35</td>
<td>F</td>
<td>Nose: vasculitis, granuloma</td>
<td>E, L, Ey, P, A 10</td>
<td>9</td>
</tr>
<tr>
<td>6</td>
<td>67</td>
<td>F</td>
<td>Kidney: necrotizing crescent GN</td>
<td>E, K, L, A, P, B 11</td>
<td>1.5</td>
</tr>
<tr>
<td>7</td>
<td>31</td>
<td>M</td>
<td>Kidney: necrotizing crescent GN</td>
<td>E, L, K, A, Raynaud, B 9</td>
<td>3.5</td>
</tr>
</tbody>
</table>

Classification to assess disease extent and DEI according to [12].
F, female; M, male; GN, glomerulonephritis; E, ENT region; L, lungs; K, kidneys; Ey, eye; P, polyneuropathy; A, arthritis, arthralgia, myalgia; GI, gastrointestinal tract; B, constitutional symptoms; Bx, biopsy.
Disease activity

Disease activity was assessed at the same time-points as disease extent using the Birmingham Vasculitis Activity Score (BVAS) [13]. The BVAS is calculated from new or deteriorated symptoms and signs within the previous month attributable to active vasculitis in nine separate organ systems that are weighed according to their relative contribution to mortality and morbidity of systemic vasculitis. The maximum possible score was 63.

Definition of remission

The activity of disease was evaluated according to clinical, radiological and serological data. Adopted from the criteria used within the therapeutic trials of EUVAS, complete remission was defined as the absence of pathological findings in these variables attributable to active vasculitis, irrespective of ANCA titres. The absence of clinical disease activity was indicated by a BVAS and DEI of zero. Partial remission was defined as partial regression of disease activity. Response to DSG therapy was defined as induction of partial or complete remission for ≥ 1 month [11,14].

Definition of relapse

As described previously [11], major relapse required the recurrence of active disease threatening the function of major organs (e.g. glomerulonephritis, pulmonary infiltrates or granulomata). Minor relapse required the recurrence of less-severe disease (i) in non-vital organs (e.g. arthralgia and nasal crusting) or (ii) involving vital organs without threatening organ function (e.g. pulmonary symptoms, such as cough or wheeze without radiological changes).

Treatment protocol

It was the hypothesis of the study that DSG is able to induce and then to maintain remission of refractory WG. Inclusion criteria were biopsy-proven WG with a refractory course as defined by the criteria given above. Exclusion criteria were a leukocyte count < 4000/µl. Pregnancy and lactation, the presence of uncontrolled acute or chronic infections, anticonvulsive treatment and inadequate contraception. Endpoints were induction of remission as the primary endpoint and maintenance of remission, sparing of immune suppressants and side effects of DSG as secondary endpoints.

Administration of DSG

Cytotoxic agents, IV immunoglobulins or plasma exchange had to be discontinued before and were not given during DSG treatment. Corticosteroids were allowed according to local practice. As described previously [10], all patients were given cycles of DSG in a dosage of 0.5 mg/kg daily by self administered subcutaneous (SC) injection. Patients 1 and 2 received the first week of DSG treatment by IV injection with the same dosage. Within each cycle, DSG was given for 2–3 weeks with the aim to reach a leukocyte (WBC) nadir of 3000/µl. WBC were monitored at least twice weekly. At the nadir, treatment was discontinued for ≥ 2 weeks to allow the leukocyte count to recover (WBC ≥4000/µl). Then, treatment was resumed in the same way.

Results

The clinical characteristics and demographic details of the seven patients are summarized in Tables 1–3. WG was histologically proven in all patients (Table 1) and all were C-ANCA/PR3-ANCA positive. All patients suffered from a generalized, severe course of WG with a cumulative DEI of 8.9 (range: 4–13) before the start of DSG treatment. During a disease duration of 4.7 years (range: 1.5–9 years) prior to the administration of DSG, the patients had suffered an average of 6.6 relapses (range: 3–12) (Table 2). Previous therapies consisted of 5.4 (range: 2–11) different therapeutic approaches (Table 2), which included GC and daily CYC in all. The mean cumulative dose of CYC was 58.4 g (range: 4.5–90 g). Four cases had been treated with experimental therapies (three mycophenolate mofetil, three etanercept, two antithymocyte globulin, one intravenous immunoglobulins, two cyclosporin A and one plasma exchange), without sufficient control of disease activity (Table 2).

Indication for DSG

All seven patients suffered from active, refractory generalized WG at the time of first administration of DSG and all had been treated previously with CYC (cumulative dose given in Table 2). Four patients (patients 3 and 5–7) received DSG for progressive disease under CYC [three oral daily CYC (latest dose given in Table 3); one bolus IV CYC]. In the other three, who presented with a relapse, CYC had to be avoided due to intolerance (patient 1: severe hepatotoxicity clearly associated with CYC; patient 2: haemorrhagic cystitis) or a high cumulative dose (patient 4: 90 g). No patient was treated with DSG for grumbling disease.

At the time of DSG administration, the patients showed a mean DEI of 5.1 (range: 3–9), indicating generalized active disease. Major organ involvement, threatening organ function, was present in all but one patient (Table 2). Four patients had severe pulmonary or subglottic disease (two suspected pulmonary granulomata and two subglottic stenosis, plus pulmonary infiltrates in one) and three suffered renal involvement. Immediately prior to DSG treatment, all patients were taking immunosuppressive drugs, including GC (mean dosage: 18.3 mg/day; range: 5–75 mg/day), that failed to control disease activity (Table 2).

Treatment effects

All patients showed a favourable response with complete (n = 5) or partial (n = 2) remission of disease activity (Table 3). Beneficial effects were observed both in lesions of (suspected) granulomatous nature, such as pulmonary opacities and subglottic stenosis, and in lesions thought to be of merely vasculitic origin, such as pulmonary infiltrates, episcleritis and polyneuropathy. Noteworthy, this did include
<table>
<thead>
<tr>
<th>Case (no.)</th>
<th>No. of relapses prior to DSG</th>
<th>Pre-treatment history (with cumulative CYC dosage)</th>
<th>Localization of disease activity at start of DSG</th>
<th>BVAS/DEI at start of DSG</th>
<th>Therapy at start of DSG (in mg/day p.o. unless otherwise indicated)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (1st course)</td>
<td>5</td>
<td>CYC 4.5 g, AZA, T/S, MMF, IVIG, PPS, ATG, MTX, GC</td>
<td>Rhinitis, progressive subglottic stenosis, episcleritis, arthralgia</td>
<td>6</td>
<td>MTX 15 SC weekly/PRED 20</td>
</tr>
<tr>
<td>1 (2nd course)</td>
<td>7</td>
<td>Additionally: etanercept, MMF, GC</td>
<td>Tracheitis, subglottic lesions, rhinitis, sinusitis, lacrimal duct obstruction, kidney (microhaematuria), arthralgia, fever</td>
<td>9</td>
<td>MMF 2000, etanercept 25 SC twice weekly, PRED 20</td>
</tr>
<tr>
<td>2 (1st course)</td>
<td>3</td>
<td>CYC 34 g, AZA, MMF, GC</td>
<td>Arthritis, polyneuropathy, abdominal pain, constitutional symptoms</td>
<td>7</td>
<td>MMF 2000, PRED 5</td>
</tr>
<tr>
<td>2 (2nd course)</td>
<td>5</td>
<td>Additionally: ATG, etanercept, MMF, GC</td>
<td>Arthritis, constitutional symptoms</td>
<td>3</td>
<td>MMF 1000, etanercept 25 SC twice weekly, PRED 7.5</td>
</tr>
<tr>
<td>3</td>
<td>12</td>
<td>CYC 73 g, AZA, MTX, CsA, GC</td>
<td>Rhinitis, subglottic granuloma</td>
<td>4</td>
<td>CYC 100, PRED 7.5</td>
</tr>
<tr>
<td>4</td>
<td>4</td>
<td>CYC 90 g, GC</td>
<td>Progressive subglottic granuloma, pulmonary infiltrates, kidney (microhaematuria), constitutional symptoms</td>
<td>5</td>
<td>PRED 75</td>
</tr>
<tr>
<td>5</td>
<td>4</td>
<td>CYC 88 g, MTX, MMF, etanercept, GC</td>
<td>Rhinitis, sinusitis, mastoiditis, constitutional symptoms</td>
<td>3</td>
<td>CYC 150, PRED 5</td>
</tr>
<tr>
<td>6</td>
<td>3</td>
<td>CYC 51 g, AZA, GC</td>
<td>Kidney (microhaematuria), arthritis, constitutional symptoms</td>
<td>5</td>
<td>CYC 100, PRED 15</td>
</tr>
<tr>
<td>7</td>
<td>3</td>
<td>CYC 68 g, AZA, GC</td>
<td>Rhinitis, sinusitis, pulmonary granuloma</td>
<td>4</td>
<td>CYC bolus, PRED 10</td>
</tr>
</tbody>
</table>

PRED, prednisone; AZA, azathioprine; MTX, methotrexate; MMF, mycophenolate mofetil; CsA, cyclosporin A; ATG, antithymocyte globulin; PPS, plasmapheresis; IVIG, intravenous immunoglobulin; T/S, trimethoprim/sulphamethoxazole.
complete resolution of severe disease activity in two of three cases of subglottic stenoses, two cases of pulmonary nodules/infiltrates (one case each) and two cases of glomerulonephritis.

Under the administration of DSG, the disease extent declined, as indicated by a drop of the DEI from 5.1 prior to DSG to 0.4 (range: 0–2) after 6 months and to 0.6 (range: 0–3) at the latest follow-up under DSG. Accordingly, a reduction of disease activity was demonstrated by a fall of the BVAS score from 16.1 (range: 4–25) directly before the start of DSG to 2.0 (range: 0–7) after 6 months and 1.2 (range: 0–9) at the latest follow-up under the study drug (individual data given in Table 3). Remaining disease activity in the two patients with partial remission was restricted to residual lesions of subglottic stenosis and nasal crusting (one case each). The full effect of DSG became apparent no earlier than after 8 weeks of treatment (not before the end of cycle 2), on average after 3 months, following 2.9 cycles of DSG.

<table>
<thead>
<tr>
<th>Case (no.)</th>
<th>Clinical response to DSG (0.5 mg/kg unless stated otherwise) and follow-up</th>
<th>BVAS/DEI</th>
<th>Cumulative dose of DSG (g)</th>
<th>Cumulative duration of DSG treatment (months)</th>
</tr>
</thead>
<tbody>
<tr>
<td></td>
<td></td>
<td>At 6 months</td>
<td>At latest follow-up under DSG</td>
<td>Including 1st course:</td>
</tr>
<tr>
<td></td>
<td></td>
<td></td>
<td></td>
<td>Including 1st course:</td>
</tr>
<tr>
<td>1 (1st course)</td>
<td>Complete remission after four cycles. GC stopped after 16 months. Maintenance of remission following dose reduction to 0.3 mg/kg DSG. Minor relapse (arthralgia, nasal crusting, epistaxis) while on 7.5 mg GC 2 months after discontinuation of DSG</td>
<td>0</td>
<td>0</td>
<td>11.3 16</td>
</tr>
<tr>
<td>1 (2nd course)</td>
<td>Complete remission after three cycles. Minor flare (nasal crusting, sinusitis) when DSG interrupted for 2 months due to pleuropericarditis. Maintained complete remission after therapy resumed (0.3 mg/kg DSG). GC reduced to 5 mg</td>
<td>0</td>
<td>0</td>
<td>Including 1st course: 45.5</td>
</tr>
<tr>
<td>2 (1st course)</td>
<td>Complete remission after four cycles. GC reduced to 5 mg after 6 months. Maintenance of complete remission following dose reduction to 0.3 mg/kg DSG. After 27 months, DSG replaced by MMF 2 g/day, followed by major relapse (renal, arthritis, polyneuropathy) after 1 month</td>
<td>6</td>
<td>0</td>
<td>11.9 27</td>
</tr>
<tr>
<td>2 (2nd course)</td>
<td>Complete remission achieved and maintained following two cycles of DSG and GC 25 mg. GC subsequently reduced to 5 mg</td>
<td>0</td>
<td>0</td>
<td>Including 1st course: 21.5</td>
</tr>
<tr>
<td>3</td>
<td>Complete remission achieved at end of cycle 3. DSG reduced to 0.2 mg/kg for 2 weeks after 9 months. GC finally 7.5/5 mg/day. Interruption of DSG for logistical reasons after 12 months, followed by nasal relapse and bronchial stenosis. Complete resolution of lesions 4 weeks after resumption of DSG (0.2 mg/kg). A second interruption of DSG 9 months after resumption was again followed by relapse (sinusitis) 2 months later (complete resolution after resumption plus GC pulse). DSG replaced by leflunomide after 27 months for logistical reasons (latest GC dose 7.5 mg)</td>
<td>3</td>
<td>0</td>
<td>7.5 23</td>
</tr>
<tr>
<td>4</td>
<td>Partial remission at the end of cycle 3. GC reduced to 7.5 mg. Remission maintained for 11 months, followed by relapse (haemoptysis, pulmonary nodules, B symptoms). Remission induction by CYC/GC pulse</td>
<td>7</td>
<td>9</td>
<td>7.7 11</td>
</tr>
<tr>
<td>5</td>
<td>Complete remission achieved and maintained following two cycles of DSG. GC kept at 5 mg</td>
<td>0</td>
<td>0</td>
<td>7.6 13</td>
</tr>
<tr>
<td>6</td>
<td>Complete remission achieved and maintained during third cycle of DSG and an increased dose of GC (initially bolus IV for 3 days). GC stopped after 22 months</td>
<td>0</td>
<td>0</td>
<td>18.1 24</td>
</tr>
<tr>
<td>7</td>
<td>Partial remission achieved/maintained at end of cycle 2. Residual nasal crusting. GC reduced to 5 mg</td>
<td>2</td>
<td>2</td>
<td>8.4 12</td>
</tr>
</tbody>
</table>

MMF, mycophenolate mofetil; B symptoms, constitutional symptoms.
(patients 1 and 2 during their first course of DSG) achieved complete remission only after four cycles.

Although the use of GC could be stopped in only two cases, the average dose of prednisolone could be reduced from 18.3 mg/day (range: 5–75 mg/day) directly prior to DSG to 4.3 mg/day (range: 0–7.5 mg/day) at the latest follow-up under the study drug.

Because the clinical course of most patients was stabilized under DSG, the drug was continued for a long time, on average for 26.5 months (range: 11–55.5 months). Only one of the seven patients (case 4), who had only achieved partial remission, relapsed on the drug and developed pulmonary nodules, haemoptysis and constitutional symptoms after treatment with DSG over 11 months. This major relapse responded to CYC bolus and GC bolus. In three of the remaining cases, who all went into complete remission, DSG was stopped after 16, 27 and 27 months, respectively. All received maintenance therapy with GC and/or other drugs, as specified in Table 3. Two of them (patients 1 and 2) subsequently relapsed after 1 and 2 months (one major and one minor relapse). During their further course, both of them received additional immunosuppressive treatments, including etanercept [tumour necrosis factor (TNF)-α blocker] and mycophenolate mofetil (plus antithymocyte globulin in patient 2), but no CYC, as they did not tolerate it, without long-lasting effect. Finally, treatment with DSG was resumed in both and resulted in complete remission (follow-up time: 49 and 20 months; Tables 2 and 3). In two cases, treatment was interrupted on three occasions for 2 months because of infection (patient 1, second course) or for logistical reasons (patient 3, two episodes). All these episodes were followed by a relapse that resolved after resumption of DSG (Table 3). Patients who had received CYC directly before DSG was initiated seemed to respond equally well as those who were on other agents.

**Dosage of DSG other than 0.5 mg/kg**

Eleven months (range: 9–12 months) after induction of complete remission using 0.5 mg/kg DSG, the dose of DSG was decreased to 0.3 mg/kg in two cases (patients 1 and 2, first course of DSG) and 0.2 mg/kg in one case (patient 3, details given in Table 3) in order to avoid infections. In patients 1 and 3, the duration of cycles was furthermore fixed at 2 weeks and leukopenia did not develop. The patients were kept on this regimen for 43.5, 45 and 14 months, with complete control of the disease.

**ANCA**

ANCA were positive in all patients directly before DSG treatment, as detected by indirect immunofluorescence and/or proteinase-3 enzyme-linked immunosorbent assay. Follow-up on ANCA titres was available in six cases. Under DSG, ANCA titres became negative in three, declined by more than two titre-steps in one and remained unchanged in two cases.

**Leukopenia**

Within each treatment cycle, the targeted level of leukopenia (WBC nadir of 3000/µl) was induced in all patients, after an average of 3 weeks of treatment. The duration of treatment cycles did not change significantly during the observation period. In all cases leukopenia was transient and followed a foreseeable course without any signs of increased toxicity associated with the long-term use of DSG. As described previously [10], DSG affected mainly neutrophil counts. During the treatment period, the intervals between WBC determinations were therefore extended from checks performed initially every other day [10] to a regimen where WBC determinations were started not before the second week during each cycle and were limited to two determinations per week. In contrast to neutrophil counts, lymphocyte and monocyte counts were hardly affected by DSG treatment. A slight decrease in absolute lymphocyte counts of ~20% below the normal range was observed in four of the seven patients (Table 4). In three of them, lymphopenia was associated with neutropenia. A relation between lymphopenia and remission could not be detected, as the non-lymphopenic cases equally responded to DSG.

**Table 4.** Adverse events observed under treatment with DSG

<table>
<thead>
<tr>
<th>Case (no.)</th>
<th>Side effects under DSG</th>
</tr>
</thead>
<tbody>
<tr>
<td>1 (1st course)</td>
<td>Two episodes of bronchitis in presence of subglottic stenosis</td>
</tr>
<tr>
<td>1 (2nd course)</td>
<td>Recurrent bronchitis and dacryopyosis in presence of subglottic and lachrymal duct stenosis</td>
</tr>
<tr>
<td>2 (1st course)</td>
<td>One pharyngitis (probably viral), lymphopenia</td>
</tr>
<tr>
<td>2 (2nd course)</td>
<td>Aggravation of renal anaemia, requiring use of erythropoietin; lymphopenia</td>
</tr>
<tr>
<td>3</td>
<td>Three urinary tract, one hair follicle infection at labia majora (drainage), one erysipelas</td>
</tr>
<tr>
<td>4</td>
<td>Acne</td>
</tr>
<tr>
<td>5</td>
<td>Pain/exanthema at injection site, three respiratory tract infections</td>
</tr>
<tr>
<td>6</td>
<td>Five urinary tract infections, two respiratory tract infections, lymphopenia</td>
</tr>
<tr>
<td>7</td>
<td>Three episodes of respiratory tract infection, lymphopenia</td>
</tr>
</tbody>
</table>
Other side-effects

No fatalities occurred. Side effects necessitating treatment are listed in Table 4. Infections, mostly of the upper respiratory tract, were observed most often and usually resolved quickly with the use of antibiotics, without further consequences. However, one case, who suffered from recurrent bronchitis and dacryocystitis in the presence of subglottic stenosis and lachrymal duct stenosis (patient 1, second course of DSG), experienced two episodes of pleuropericarditis that required inpatient treatment. During the first episode, he developed a third-degree heart block in the context of suspected myocarditis. The condition required the permanent placement of a pacemaker. Despite an intensive search, no micro-organisms could be identified. On both occasions, the condition of the patient improved under antibiotics and an interruption of DSG treatment, which was resumed subsequently. All other infections could be managed on an outpatient basis. No cases of typical opportunistic infections, such as herpes zoster, cytomegalovirus, *Pneumocystis carinii* or fungal infections, were observed in this group of patients. The infections were not related to the neutropenic phases during the cycles.

Besides the above-mentioned side effects, local and systemic tolerability of DSG was excellent. Only one patient experienced a painful exanthema following each injection, which resolved always after 1–2 h. Treatment with DSG was not interrupted.

Slight decreases in haemoglobin levels and thrombocytes were noted in two of the seven cases, but were clinically insignificant with the exception of patient 2, who additionally suffered from concomitant renal insufficiency due to renal scarring. During her second course of DSG, anaemia aggravated under DSG in conjunction with worsening renal function (active glomerulonephritis excluded by biopsy) and required the use of erythropoietin.

Discussion

This study shows that the prolonged use of DSG for an average of 26.5 months (range: 11–55.5 months) was a safe treatment in WG patients who had all been pre-treated with several immunosuppressive agents, including CYC. All patients had multiple relapses and active refractory disease prior to the administration of the study drug. Nevertheless, DSG induced long-lasting remissions (five complete and two partial) in all patients and only one of the cases relapsed after 11 months under treatment with DSG. In contrast, termination/interruption of DSG treatment was followed by relapse in four of five occasions and resumption of DSG again led to complete remission. The good clinical response corresponded with a fall in the DEI from a mean of 5.1 prior to DSG to 0.6 (range: 0–3) at the latest follow up. Accordingly, a reduction of disease activity was demonstrated by a fall of the BVAS score from 16.1 (range: 4–25) to 1.2 (range: 0–9).

One might argue that concomitant treatment with GC added significantly to the favourable response of DSG. However, it is known that treatment with GC alone is not capable of inducing or maintaining remission in WG [1]. Also, all patients had received GC before the administration of DSG, without control of disease activity. Finally, the dose of GC could be reduced from an average of 18.3 mg/day (range: 5–75 mg/day) directly prior to DSG to 4.3 mg/day (range: 0–7.5 mg/day) at the latest follow-up under the drug and two cases were able to completely stop GC treatment.

It cannot be excluded fully that a postponed effect of the other immunosuppressants given prior to study entry contributed to the favourable course of the patients under DSG. However, it seems unlikely that the effect of these drugs continued for several months or years under sole treatment with the study drug. Furthermore, the patients were selected for DSG treatment on the assumption that the previous immunosuppressive medications were either not sufficiently effective or were contraindicated due to severe side effects. In fact, four of the seven patients showed progressive disease under the standard therapy with CYC and GC and clearly improved following the employment of DSG. However, it should be stressed that — as in other immunosuppressive drugs — the full effect of DSG treatment became apparent not earlier than after 8 weeks of treatment, on average after 3 months, following 2.9 cycles of DSG.

It is noteworthy that preliminary experience with anispermus (LF 15-0195), a derivative of DSG, showed a similar response rate in patients with refractory systemic vasculitis, mostly ANCA-associated vasculitis (induction of remission in seven out of nine cases), thereby confirming this novel therapeutic approach [15].

A selection bias may have contributed to the favourable results reported here, as only those four patients of the pilot trial were included in the long-term study who were kept on DSG treatment because they had shown a beneficial response. It should be mentioned, however, that only three of the 19 WG cases reported in the initial pilot trial relapsed during treatment with DSG and only one patient had not responded at all. In two further patients, DSG had to be stopped due to side effects. In contrast, the majority of cases (13/19) did successfully complete the pilot trial and could have been kept on DSG further. For unclear reasons, however, only four of these 13 were selected for continuous treatment with DSG by their local physicians and are reported here. The other nine patients were switched to different regimens of maintenance therapy. Two of them relapsed during the following 6 months [10].

The present study extends the previous observation that treatment with DSG, given in a cycled manner at daily dosages of 0.5 mg/kg SC until the target leucocyte count of 3000 cells/μl is reached, was feasible and safe [10]. This regimen is in contrast to the treatment schemes applied in acute kidney rejection, where DSG...
is normally used in doses ranging from 3 to
5 mg/kg/day for 5–7 days [7,9]. However, data from
animal models and the initial clinical experience from
patients with various forms of proliferative glomerulo-
nephritides with or without crescent formation [8]
suggest that the efficacy of DSG may be dependent on
the duration of application rather than on the absolute
amount given. This impression is now further sub-
stantiated by the experience in three patients who were
treated with even smaller doses of 0.3 (two cases) and
0.2 mg/kg, without a loss of efficacy. In two of these
patients, the duration of treatment cycles was restricted
to 2 weeks, independent of whether the leukocyte
nadir of 3000 cells/µl was reached or not. In fact, both
of these patients retained normal leukocyte counts
during their treatment cycles, without any apparent
loss of efficacy. Monitoring of treatment was, therefore,
much easier in these cases. However, the number of
patients needs to be extended before definite conclu-
sions can be drawn.

Despite the prolonged administration of DSG
reported here, long-term tolerability was good and
no increase in the incidence of side effects over
time could be observed. Importantly, the duration of
treatment cycles was stable over time, with an average
of 3 weeks per cycle, until the target leukocyte count
3000 cells/µl was reached. Thus, no signs of cumulative
bone marrow toxicity of the drug after repeated cycles
were observed.

Despite the significant reduction in neutrophil counts
induced by a dosage of 0.5 mg/kg DSG within each
cycle, most infections observed under the study drug
were of mild to moderate intensity and resolved after
adequate treatment. The rate of infections was in the
range that could be expected from previous trials
in ANCA-associated vasculitis. A good tolerability
of DSG was also observed in other studies, including
phase I studies that involved patients suffering from
renal allograft rejection, crescentic glomerulonephritis
and various advanced cancers. These treatment regi-
mens included short-term doses of ≤75 mg/kg/day,
given mostly as 3 h infusions for ≤7 days. The main
adverse effects, listed in Table 5, were transient and
included leukopenia, anaemia and thrombocytopenia,
hypotension, oral numbness and infections, which
were mostly mild with the exception of one case
of septicemia. A randomized, double-blind study of
DSG in multiple sclerosis, which employed placebo or
2 or 6 mg/kg DSG given IV in five 4 day courses at
4-week intervals did not demonstrate any differences
in the number and severity of adverse effects during
a 2 year follow-up period, beside a mild and transient
leukopenia and anaemia that was noted in association
with the treatment phases. No long-term toxicities were
reported [3]. Furthermore, in all clinical and pre-clinical
studies, DSG revealed no mutagenicity, teratogenicity
or carcinogenicity [3].

It is noteworthy that one of the patients reported
here developed a third-degree heart block after a
total of 29 months of treatment with DSG. This
adverse event, which required permanent placement
of a pacemaker, occurred while the patient showed
signs and symptoms of infective pleuropericarditis,
which responded to antibiotic treatment. It seems
unlikely that a cardiotoxic effect of DSG was the
cause of this conduction deficit and no further
arrhythmias or other signs of cardiotoxicity were
observed despite resumption of treatment after
2 months. However, concerns over potential cardio-
toxicity have been raised with a DSG analogue,
anisperimus, in two vasculitis patients who developed
changes in left ventricular function (personal commu-
nication, Dr David Jayne, Cambridge, UK). As for
DSG, cardiotoxicity has not been observed in previous

<table>
<thead>
<tr>
<th>Table 5. Adverse effects reported in clinical experimental use of DSG</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Haematological</strong></td>
</tr>
<tr>
<td>Transient leukopenia observed in all patients, which resolved after interruption of therapy</td>
</tr>
<tr>
<td>Transient anaemia, leukopenia (neutropenia), thrombocytopenia in ≤30% of cases</td>
</tr>
<tr>
<td><strong>Cardiovascular</strong></td>
</tr>
<tr>
<td>Reversible hypotension and hypertension, myocardial infarction (one case)</td>
</tr>
<tr>
<td>Oedema, hot flushes in ≤25% of cases</td>
</tr>
<tr>
<td><strong>Gastrointestinal tract</strong></td>
</tr>
<tr>
<td>Loss of appetite, nausea, vomiting, heart burn</td>
</tr>
<tr>
<td><strong>Neurological</strong></td>
</tr>
<tr>
<td>Transient perioral numbness, dose-dependent in ≤33% of cases</td>
</tr>
<tr>
<td><strong>Infections</strong></td>
</tr>
<tr>
<td>Sinusitis, oral, upper respiratory tract, urinary tract, skin, septicaemia (one case), herpes zoster</td>
</tr>
</tbody>
</table>
stimulated T and B cells after activation and the production of interferon-γ by Th1 effector cells [4]. It also leads to an arrest of T-lymphocyte maturation in the thymus during CD4 CD8- to CD4+CD8- transition [6]. B-cell maturation and κ light-chain expression is reduced as well, possibly by blocking the nuclear translocation of transcription factor NF-κB [5]. Finally, also the antigen presentation by macrophages was shown to be altered under DSG, including a diminished expression of MHC class II antigens in conjunction with a reduced production of TNF-α and interleukin-1β [16].

Finally, the availability of alternative experimental therapies for patients with refractory WG needs to be mentioned. Although most of the work has been published only in abstract form, B-cell depletion by a monoclonal anti-CD 20 antibody (rituximab) was successfully used, with few side effects [17], and may be a therapeutic option especially in patients who developed infections under conventional therapeutic agents. Furthermore, anti-TNF-α blockade [18] and anti-T cell-directed treatment [11] may be therapeutic options in patients who do not respond to conventional means.

In conclusion, prolonged treatment with DSG appeared effective and safe in seven patients with refractory WG who showed active, progressive disease despite standard treatment with GC and CYC or who did not tolerate CYC. Although the study was open and uncontrolled and the number of patients was small, the standardized regimen employed here seems promising for both the induction and the maintenance of remission in WG. Prolonged administration of the drug did not lead to increased toxicity and the main side effects were infections of mild to moderate intensity, all of which could be controlled with appropriate therapy. However, the treatment regimen employed here necessitates determination of blood cell counts in regular intervals to monitor the myelosuppressive effects of the drug. It remains an open question whether a treatment regimen with lower doses of the drug, which may avoid myelosuppression, would be equally effective. In view of the good tolerability of the drug, randomized studies are warranted to investigate the efficacy of DSG in comparison with CYC as a secondary or even primary agent in patients with active WG.

Acknowledgements. This study was supported by Euro Nippon Kayaku GmbH, Staufenstrasse 4, D-60323 Germany.

Conflict of interest statement. W.H.S., R.B. and F.J.v.d.W. have received research support from Euro Nippon Kayaku GmbH.

References
7. Okubo M, Tamura K, Kamata K et al. 15-Deoxyspergualin ‘rescue therapy’ for methylprednisolone-resistant rejection of renal transplants as compared with anti-T cell monoclonal antibody (OKT3). Transplantation 1993; 55: 505–508

Received for publication: 10.7.04
Accepted in revised form: 2.2.05