Optimizing the Therapeutic Strategies in ANCA-Associated Vasculitis – Single Centre Experience with International Randomized Trials

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Abstract: ANCA-associated vasculitides, including Wegener's granulomatosis and microscopic polyangiitis, are systemic autoimmune diseases with poor prognosis in untreated patients, which can be dramatically improved by current therapeutic modalities. The aim of multi-centre randomized trials of the European Vasculitis Study Group is to optimise and standardise treatment of these diseases. From 1995–2001 our department contributed a total of 40 patients to the trials CYCAZAREM (cyclophosphamide versus azathioprine during remission for generalised vasculitis), MEPEX (plasma exchange versus methylprednisolone for severe renal vasculitis) and CYCLOPS (daily oral versus pulse cyclophosphamide during induction phase for generalised vasculitis). In this paper, we report on the preliminary results of long-term follow up of our patients included in international trials. The mean time of follow-up of the patients was 55.7 months with the patient survival rate of 72% and renal survival rate of 65%. Remission was achieved in 82% of patients, out of which 42% suffered a relapse. In generalised forms of vasculitides, treatment with cyclophosphamide is nowadays established as the standard therapy. The aim, however, is to further minimise its toxic effects by choosing the optimal therapeutic strategies. Complete results of all trials have not yet been published; nevertheless, the preliminary available data have already revealed new potential therapeutic approaches.

Introduction

Vasculitides are a heterogeneous group of clinical and pathological units characterised by inflammatory infiltration and potential destruction of blood vessels' walls. According to the so-called Chapel Hill's classification, vasculitides can be divided into large vessel, medium-sized vessel and small vessel vasculitides. Wegener's granulomatosis (WG), microscopic polyangiitis (MPA) and Churg-Strauss syndrome (CSS), mentioned in detail below, all belong to the group of small vessel vasculitides [1, 2].

They are systemic autoimmune diseases characterised by pauciimmune necrotizing vasculitis with a strong association with ANCA (AntiNeutrophil Cytoplasmic Autoantibodies), which are circulating autoantibodies directed against different target antigens of azurophilic granules of polymorphonuclear leucocytes. In WG, ANCA are usually directed against proteinase 3 (antiPR3) and have a cytoplasmic type of immunofluorescence (c-ANCA, Figure 1). In MPA the target antigen is mostly myeloperoxidase (MPO) and the type of immunofluorescence is perinuclear (p-ANCA, Figure 2). Wegener's granulomatosis, microscopic polyangiitis and Churg-Strauss syndrome, together with renal-limited vasculitis (RLV – also called idiopathic pauciimmune rapidly progressive glomerulonephritis), which is a limited form of disease not involving any other organ but kidney, more often antiMPO positive, are all ranked among ANCA-associated vasculitides (AAV). At present, it is not known whether there is any difference between them in prognosis or response to therapy [3].

Their incidence (the annual incidence of renal vasculitis in Europe may exceed 20 per million per year) is not as low as it was originally believed, most probably due to higher awareness of the disease and the possibility of routine ANCA testing. Nevertheless, still relatively low numbers of patients in single centres led to the need of international multi-centric cooperation in finding optimal therapeutic approaches (see below). Basically any organ may be afflicted by vasculitis. Apart from renal involvement (ANCA-associated vasculitis is the cause of end-stage renal disease in about 5% [4]), the disease often involves lungs (WG, MPA, CSS) or ENT organs (WG), and patients can also suffer from neurological symptoms.

Untreated, generalised WG and MPA follow a progressive course with a fatal outcome due to vital organ failure. The mean survival of untreated patients was 5 months. In the 1970s Fauci and Wolff introduced empirical therapeutic scheme of daily oral cyclophosphamide (CYC) administered for one year after remission achievement and corticosteroids (prednisolone 1 mg/kg/day on a tapered-off basis). The so-called 'Fauci-scheme' dramatically improved the prognosis of the patients, and although not supported by any controlled study, it was established as the standard treatment. Remission was induced in 80–100% of cases. However, the toxicity of the regimen caused considerable morbidity and mortality. Moreover, in a long-term follow-up it has turned out that at least 50% of the patients relapse even under continuing immunosuppression or when the therapy is reduced [5]. To maintain remission, CYC and later azathioprine or other alternative drugs (chlorambucil, cyclosporin etc.) have been tested.

European Vasculitis Study Group (EUVAS)

The European Vasculitis Study Group (EUVAS) was founded in 1993 to verify new therapeutic schemes and strategies in the treatment of AAV. Its aim is to improve the clinical, histological and serological diagnostics of vasculitides and to optimise their treatment. Therefore, EUVAS has designed and conducted several randomized trials. Our centre has closely cooperated with EUVAS and repeatedly included a considerable number of patients in the studies [6, 7].

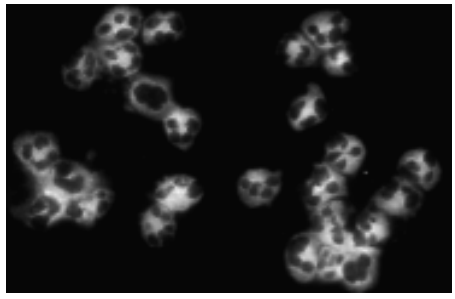

Figure 1 – cANCA.

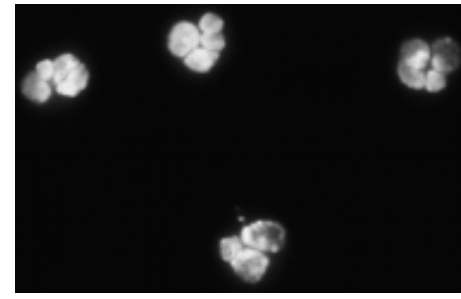

Figure 2 – pANCA.

Several semi-objective scoring tools, which enable unbiased assessment of patients in different centres, have been introduced by EUVAS. BVAS (Birmingham Vasculitis Activity Score) is a score representing the total disease activity attributable to vasculitis (BVAS.1 new or worse activity, range 0–66; BVAS.2 persisting or grumbling disease, range 0–33) [6]. VDI (Vasculitis Damage Index) is another score representing chronic (at least 3 months lasting), often non-healing organ damage caused by vasculitis (unlike BVAS, VDI score does not represent disease activity). The potential range of VDI is from 0 to 64, and one of the important aims of our treatment is to preserve this index at the lowest possible level during the long-term follow-up, which means to prevent persistent organ damage in patients with AAV [6].

As the initial severity of the disease and its extent relate to the degree of damage at long-term follow-up, treatment needs to be tailored to clinical subgroups characterised at the time of diagnosis [7], and clinical trials are therefore needed. EUVAS has determined five subgroups to cover the spectrum of severity of AAV at presentation (Table 1). Four of them were associated with any of the so-called first wave randomised clinical trials starting in the early 90's [8]. Later, several second wave trials were designed to study newer therapeutic approaches, e.g. pulsed cyclophosphamide (CYCLOPS) or long-term remission therapy with low dose

Table 1 – Classification of AAV according to the disease severity [8]

Clinical subgroup	Characterisation	Serum creatinine	ANCA	Trial
Localised	No constitutional symptoms No threat to vital organ function	< 120 μmol/l	+ or –	
Early systemic	Constitutional symptoms No threat to vital organ function	< 120 μmol/l	+ or –	NORAM
Generalised	Constitutional symptoms Imminent vital organ failure Renal involvement	< 500 μmol/l	+	CYCAZAREM
Severe renal	Constitutional symptoms Severe renal involvement	> 500 μmol/l	+	MEPEX
Refractory	Frequently relapsing or progressive Life-threatening Standard treatment of no use	Any	+ or –	SOLUTION

azathioprine and prednisolone (REMAIN). The trials in which our patients were enrolled (i.e. CYCAZAREM, MEPEX and CYCLOPS) will be mentioned in more detail.

In all three studies, patients with newly diagnosed WG, MPA or RLV with renal involvement defined as biopsy-proven necrotizing glomerulonephritis (Figure 3) and/or microhematuria and proteinuria (more than 1 g/24 hours) were included.

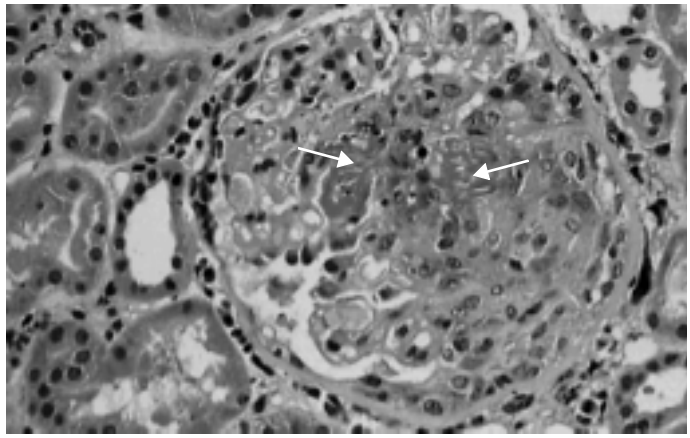

Figure 3 – Necrotizing glomerulonephritis.

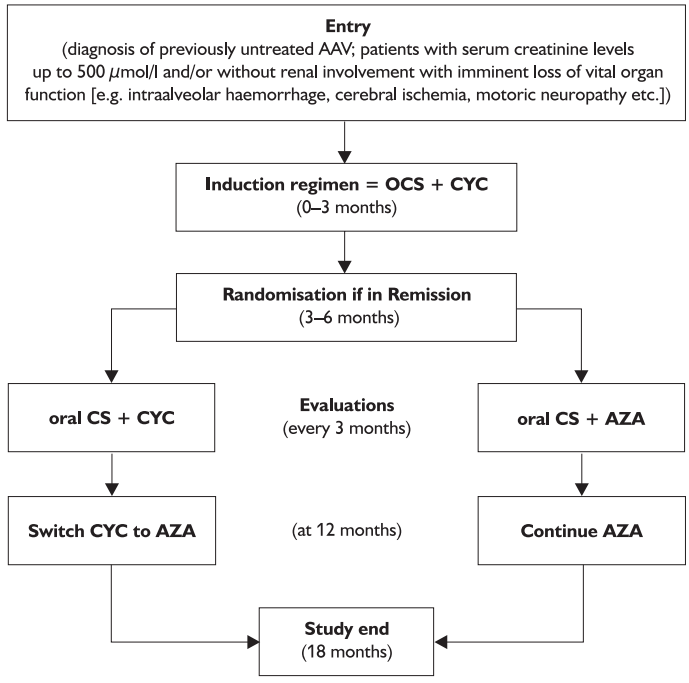

Figure 4 – CYCAZAREM – design of the study.

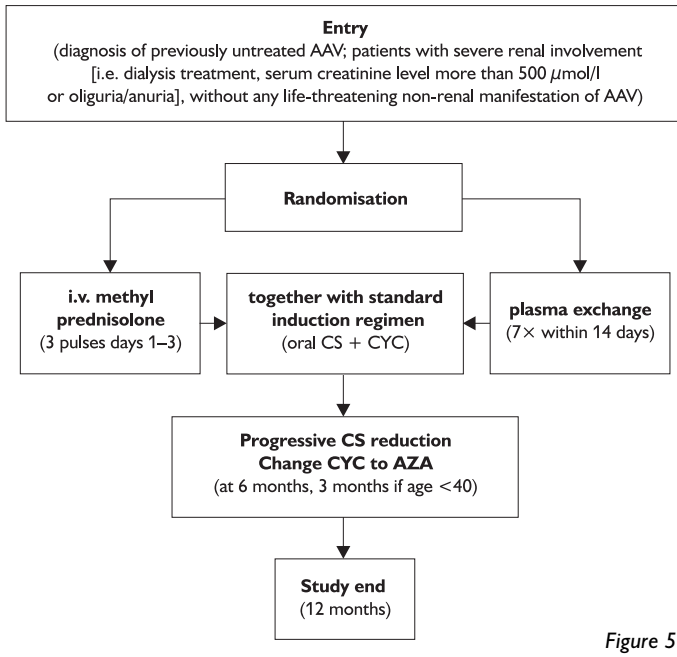

Figure 5 – MEPEX – design of the study.

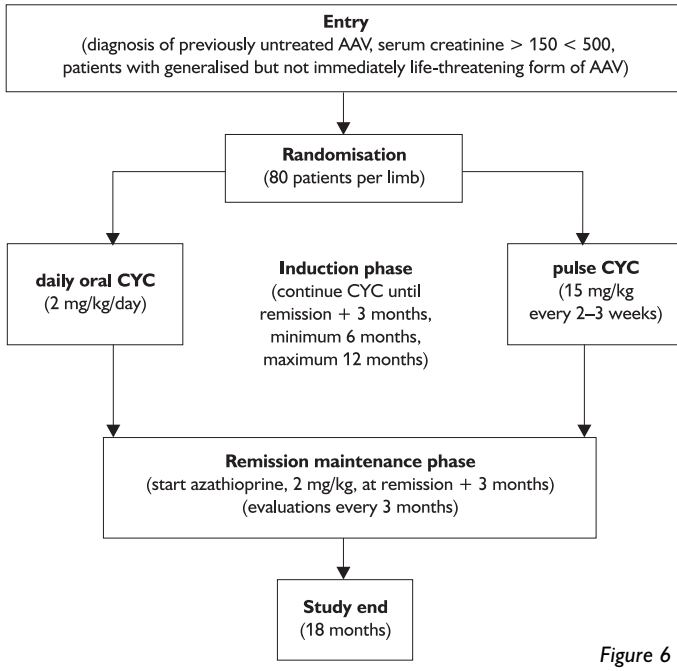

Figure 6 – CYCLOPS – design of the study.

The severity of renal involvement differed in the studies (see Figure 4–6). Other inclusion criteria were ANCA positivity and the age limit 18–80 years (75 years in CYCAZAREM, respectively). Patients who had already received immunosuppressive treatment or were anti-GBM positive, patients with other autoimmune diseases, pregnant women, women without adequate contraception and patients who had a known malignancy or were HbsAg, antiHCV or HIV positive were excluded [6, 14]. To complete the list, some of our patients have recently been recruited for the trial IMPROVE (azathioprine versus mycophenolate-mophetil in remission therapy), which has not yet finished.

CYCAZAREM

The combination of corticosteroids (CS) and cyclophosphamide (CYC) is usually effective in the therapy of generalised active vasculitis. However, it is obvious that the treatment has to continue after remission achievement to prevent a relapse [9]. The aim of this study was to optimise the treatment of the remission phase of AAV and to compare the efficiency and safety of standard CYC to alternatively used azathioprine (AZA) with presumed lower toxicity [9, 10] (for study design see Figure 4 [adapted from 6]). In total 155 patients were included in this study. The primary end-point of the study was relapse rate in both limbs of the study; secondary end-points were adverse effects and cumulative doses of administered drugs.

MEPEX

In the case of severe renal form of AAV with a rapidly progressive course to dialysis- dependent renal failure, the standard therapy with oral CS and CYC may be insufficient. The aim of this study was to compare the efficiency of adjunctive therapy with methylprednisolone with the therapy with plasma exchange (considered to be a more efficient, but potentially more risky and certainly more expensive way of treatment in previous smaller studies) in patients with severe renal involvement [6, 11] (for study design see Figure 5 [adapted from 6]). The study was designed for 150 patients. The end-points of this study were renal survival at 1 year and renal recovery to creatinine levels less than 200 μmol/l. Adverse effects were also monitored and evaluated.

CYCLOPS

Unlike the previous studies, CYCLOPS belongs to the second wave trials, in which the combined immunosuppressive treatment with CS and cytotoxic drugs has already been standardised. The aim of this study was to reduce the cumulative dose of CYC during the induction period of generalised but not life-threatening AAV by using it in an intermittent pulsed form (for study design see Figure 6 [adapted from 6]). There was an international consensus that a regimen comprising pulsed CYC would be associated with less toxicity compared to the gold standard, continuous oral CYC regimen. However, there were concerns that less toxicity may

be at the expense of reduced efficacy and especially higher relapse rate [12, 13]. The required number of patients was 160. The primary end-point of this study was the disease-free period (time from remission until relapse or study end, i.e. 18 months from the study start). The secondary end-points were adverse effects, cumulative doses of CYC and cumulative organ damage caused by vasculitis.

Patients – entry data

From November 1995 – November 2001, our department contributed a total of 40 patients to the above-mentioned international EUVAS studies (MEPEX, CYCAZAREM and CYCLOPS). Fourteen men and 26 women were included, with the mean age at the start of the study 53.9 years (range 18–73 years). Nineteen patients were diagnosed with Wegener's granulomatosis (47.5%), 16 patients with microscopic polyangiitis (40%), 4 patients with renal-limited vasculitis (10%) and 1 patient suffered from Churg-Strauss syndrome (2.5%). All our patients were ANCA positive, cANCA positivity was found in 24 patients (60%) and pANCA positivity in the remaining 16 (40%). Three of the patients were included in MEPEX (1 man, 2 women, the diagnoses were 2× WG, 1× MPA, mean age 59.3 years), 9 patients entered the trial CYCAZAREM (3 men, 6 women, the proportion of diagnoses: 5× WG, 4× MPA, mean age 51.7 years) and 28 patients participated in CYCLOPS (10 men, 18 women, 12× WG, 11× MPA, 4× RLV, 1× CSS, mean age 54 years).

Renal involvement (by definition) was present in all our patients, also biopsy-proven. The median serum creatinine level at the start of the study was 251 μmol/l (range 79–660), 3 patients (patients included in MEPEX) were initially dialysed. The mean proteinuria was 1.33 g/day. The mean entry level of CRP was 58 mg/l, the mean haemoglobin level 94 g/l. The mean entry BVAS.1 score in our patients was 20.2 (range 11–41).

Results – study end

The trial was successfully (after 12 months in MEPEX and 18 months in the other two studies) completed by 31 patients (77.5%). At the end of the study the mean CRP was 5.5 mg/l, the median serum creatinine level was 134 μmol/l, the mean proteinuria was 0.75 g/day. Only 1 patient out of those who completed the study protocol was dialysis-dependent (in trial CYCLOPS), out of 3 initially dialysed patients in study MEPEX 1 patient died, 1 patient recovered renal functions to serum creatinine level at about 160 μmol/l and the last patient was not dialysis-dependent at the study end. Nevertheless, his serum creatinine level was about 450 μmol/l and it was necessary to commence with dialysis later.

Remission during the study course was achieved in 33 patients (82.5%), 1 patient (trial CYCLOPS) remained mildly active during the whole study period. The other 6 patients who did not achieve remission died soon after inclusion in the trial (1× MEPEX, 2× CYCAZAREM, 3× CYCLOPS). In 3 patients who achieved

remission a relapse leading to a change of their treatment was observed during the study period (2× CYCLOPS, 1× CYCAZAREM). The mean VDI at the end of the study in patients who had survived was 2.0 (range 0–4). The mean last available VDI in all patients (in dead patients including their last index before death) was 2.03 (range 0–5).

As far as the adverse effects of the therapy are concerned, more or less significant leucopoenia was reported in 14 patients (35%), 15 patients (37.5%) suffered from some form of infectious complications (bacterial, mycotic, as well as viral infections, including CMV infection or active hepatitis B, were found). The infectious events were usually successfully treated with antimicrobial drugs. In some patients the immunosuppressive treatment had to be reduced or temporarily stopped. However, despite this therapy, in 4 patients the infectious complications resulted in their death. We also had to deal with dyspepsia several times, osteoporosis and amenorrhea in women was also observed. In one patient, haemorrhagic cystitis was observed during the CYC treatment.

Nine patients did not terminate the study; two of them were withdrawn by the physicians. The reasons for their withdrawal were newly diagnosed hepatitis B after the start of haemodialysis treatment in 1 patient (CYCLOPS) and intolerance of immunosuppressive treatment in the other patient (MEPEX – severe leucopoenia, severe dyspepsia), who later died in another hospital, the cause of death presumably being cardiac failure and chronic hyper-hydration in haemodialysis treatment (however, active vasculitis was not excluded). The other

Table 2 – Preliminary results of long-term follow up of the patients included in EUVAS trials

	CYCAZAREM	MEPEX	CYCLOPS	TOTAL
No. of patients included	9	3	28	40
Remission achieved	7/9	2/3	24/28	33/40
Relapses				
during the trial	1/7	0/2	2/24	3/33
after the trial	2/7	1/1	10/23	13/31
Deaths				
during the trial	2/9	1/3	5/28	8/40
after the trial	0/7	1/2	2/23	3/32
Median serum creatinine (in μmol/l)				
study start	266	all HD	218	251
study end	121	306	126	134
at present	151	240	118	128
Dialysis dependent or renal transplantation				
study start	0/9	3/3	0/28	3/40
study end	0/7	1/2	1/23	2/32
at present	1/7	0/1	2/21	3/29

7 patients died during the study period (5 patients in CYCLOPS, i.e. 18%, 2 patients in CYCAZAREM, i.e. 22%). The cause of death was a septic shock with multi-organ failure in 4 patients (3× CYCLOPS, 1× CYCAZAREM) – at the time of death these patients were not at remission. In two of them haemodialysis treatment had to be started before death. One patient died of pulmonary embolism (CYCLOPS), one of severe gastrointestinal bleeding related to chronic anticoagulation therapy with possible adverse effect of corticosteroids (CYCLOPS), while another died of a stroke soon after being diagnosed with AAV, which was not assessed as the disease activity, even though the patient did not achieve remission (CYCAZAREM).

Results – long-term follow-up

The mean time of follow-up of our patients was 55.7 months, i.e. approximately 4.5 years (range 1–119 months). In the further follow-up after the end of the study 3 other patients have died (2 patients in CYCLOPS, 1 patient in MEPEX). The cause of death was unrelated to active vasculitis – 1 patient had a rupture of abdominal aortic aneurysm (CYCLOPS) and the remaining two died of cardiac and respiratory failure; both were haemodialysis-dependent (the patient in CYCLOPS was the one who had not completed the study protocol because of hepatitis B). The mean time to death in our patients was 10.5 months.

At present, 29 patients included in the studies are alive. The overall survival rate at 4 years (as the study start was at least four years ago in all patients) is 72.5%. In patients who were included in the trial more than 5 years ago (31 patients) the overall survival at 5 years is 64.5%. The median serum creatinine level in the patients alive is 128 μmol/l. Two patients are dialysis-dependent (CYCLOPS) and 1 patient underwent a successful renal transplantation (CYCAZAREM). One of the initially dialysed patients (MEPEX) is still dialysis-independent with serum creatinine level at about 240 μmol/l. End-stage renal failure with the need of renal replacement therapy was reported in 5 other patients who had already died. Thus the renal survival rate at 4 years in our patients is 65%. In patients included in the trial more than 5 years ago (31 patients) the renal survival rate at 5 years is 61.3%.

Thirteen patients of the total 31 who finished the study have suffered from at least one relapse (42%) – 2 patients in CYCAZAREM (in one of these patients we observed a relapse already during the study period), the only patient alive in MEPEX and 10 patients in CYCLOPS (in 2 of them relapse was also reported during the study period). All three patients who relapsed during the study period have shown a tendency to repeat the relapses. Moreover, they do not respond sufficiently to standard treatment. In another patient in CYCAZAREM 2 relapses were also observed, on the contrary, in the remaining 9 patients no other relapse was reported. The mean time to relapse in our patients is 33.5 months. The immunosuppressive treatment (i.e. either combined immunosuppression or corticosteroids in the doses above 10 mg of prednisolone per day) was

administered to 4 patients at the time of the first relapse. The other patients did not receive immunosuppression at the time of relapse. The most common type of relapse was that involving kidney (9 out of 13 patients). Relapses were normally managed by standard immunosuppressive treatment (according to the severity either CS or CYC or AZA, later also MMF). In 2 patients with repeating relapses we have used some of the newer alternative ways of the treatment of AAV (infliximab, deoxyspergualin or etoposide).

Severe adverse effects and non-healing organ damage are some of the most important points in long-term follow-up of patients with vasculitides. In one patient in CYCLOPS we diagnosed a solid tumour (papilocarcinoma of urinary bladder). The diagnosis was made 31 months after the diagnosis of AAV. In another patient in CYCLOPS a myelodysplastic syndrome was reported. Malignancy was observed in a total of 5% of our patients. At the time of the diagnosis of AAV, there were 5 patients with diabetes mellitus. In other 4 patients diabetes started after the inclusion in the trial (i.e. in total 22.5% patients with diabetes mellitus). Symptoms of coronary heart disease manifested in 6 patients (15%), 3 patients underwent a stroke after the start of the study (7.5%). Deep venous thrombosis or pulmonary embolism was observed in 4 patients during the follow-up (10%). Osteoporosis caused a fracture in 4 of our patients (10%). The mean VDI has increased a little since the study ended, it is now 3.9 (range 0–8) in those patients who have survived. The last available mean VDI in all patients is 3.88 (range 0–8).

Results – respective trials

Both of our female patients in MEPEX (1× MPA, 1× WG) were randomised to receive additionally plasma exchange. The only male patient was randomised to intravenous corticosteroids. Because of the small number of our patients in this trial, the results naturally cannot be statistically assessed. Nevertheless, mortality of our patients in this trial was higher in comparison with the other studies. The only patient alive with moderate deterioration of renal functions was treated with plasma exchange. According to the published preliminary international data, the mortality in both trial limbs was the same, but the renal survival was much better in patients treated with plasma exchange [15]. In our department, we always consider adjunctive treatment with plasma exchange in patients with severe and vital organ function threatening course of AAV.

In CYCAZAREM, remission was achieved in 7 out of 9 patients who could be further randomised. In three cases they were randomised to AZA (1× MPA, 2× WG), in 4 cases to continuing CYC (2× MPA, 2× WG). In our small group of patients we observed relapses in 2 patients treated with AZA, while there was no relapse in the CYC limb. However, published results of this trial support the original hypothesis that an early substitution of AZA for CYC does not increase the relapse rate in the short-term follow-up and significantly reduces the exposure to CYC, which might be necessary in the treatment of later relapses [16]. Whether

the relapse rate in the long-term follow-up (after 18 months) is higher in the patients early switched to AZA than in the patients treated with CYC for 12 months has not yet been answered. Any increase is, however, unlikely to be large [16]. A comparison of treatments with AZA and MMF in the remission phase of AAV is one of the aims of the newest trial IMPROVE.

In CYCLOPS, 15 of our patients were randomised to the intermittent pulse administration of CYC (11 women, 4 men, 7× MPA, 3× RLV, 5× WG, mean age 48.3 years). Thirteen patients were included in the continuous oral CYC limb (7 women, 6 men, 7× WG, 1× RLV, 1× CSS, 4× MPA, mean age 55.2 years). Remission was achieved in 10 patients in the oral limb (77%) in comparison with 14 patients in the intravenous limb of the trial (93%). The number of infectious complications of immunosuppressive treatment was comparable in both limbs. However, their course in the patients in the oral limb was more severe. Out of 7 patients in this trial who died, 4 were treated with oral CYC (i.e. mortality 31% in the oral limb of the trial). In three instances, the cause of death was a septic shock with a multi-organ failure – the death was therefore directly related to the immunosuppression. In one case the cause of death was severe gastrointestinal bleeding. In the intravenous limb of CYCLOPS there were 3 deaths (i.e. mortality 20%) and the death was never directly related to immunosuppressive treatment. The causes of death were cardiac failure in one dialysed patient, pulmonary embolism in another patient and rupture of abdominal aortic aneurysm in the last patient. The renal survival rate only slightly differs in both limbs (it is 73% and 69% in patients in the intravenous limb and oral limb respectively). The relapse rate was also similar in both groups of patients. Four relapses were observed in the oral limb (i.e. 44% of the patients who completed the study) and six relapses observed in the intravenous limb (i.e. 46% of the patients who completed the trial protocol). Malignancy was diagnosed in both groups (papilocarcinoma of the urine bladder in the intravenous limb and myelodysplastic syndrome in the oral limb). We can sum up that, in accordance with published preliminary results of CYCLOPS, the intermittent pulse administration of CYC in generalised but not life-threatening forms of AAV represents as efficient therapy as pulse administration of CYC with significant reduction of cumulative dose and both short-term and long-term toxicity of CYC [17].

Conclusion

ANCA-associated vasculitides are serious systemic diseases with a poor outcome in untreated patients. Nevertheless, their prognosis has dramatically improved in the last 30 years thanks to better diagnostics and an earlier start of the treatment as well as new therapeutic approaches tailored to the disease severity. The ultimate results of all studies conducted by EUVAS have not yet been published; however, some of the preliminary published data have already revealed new promising therapeutic approaches. Our data support some the original hypotheses

of the studies. Nevertheless, due to low numbers of our patients the results cannot be generalized. Moreover, the effect of alternative therapy in long-term follow-up remains to be elucidated. In the future, there will surely be an effort to further minimalise toxic effects of standard treatment and to find new treatment possibilities for the patients with refractory or frequently relapsing forms of vasculitis.

Following current recommendations for the standard treatment of AAV stem from our long-term experience with patients with generalised form of AAV with renal involvement. It should be, however, underlined that there is a need of tailoring the treatment according to the status of every single patient. Furthermore, patients with AAV should definitely be monitored in an experienced and specialized centre.

In our hands, combined immunosuppressive treatment with CS (mostly i.v. methylprednisolone for the first three days, followed by oral administration of prednisolone) and CYC remains the standard therapy in the induction phase of generalised form of AAV and is usually effective in achieving remission. In severe and vital organ function threatening vasculitis the addition of plasma exchange to the standard therapy should always be considered. The toxicity of CYC may be reduced by the pulse administration of CYC which significantly reduces the cumulative dose and does not increase the relapse rate. Therefore, the pulse administration is usually preferred in our department. It is also proven that early substitution of AZA for CYC is as safe as long-term treatment with CYC in the short-term follow-up of the patients and further reduces the cumulative dose. In some patients, MMF can be successfully used instead of AZA. The maintenance therapy has to be long-term, with the reduction of CS on a tapered-off basis.

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