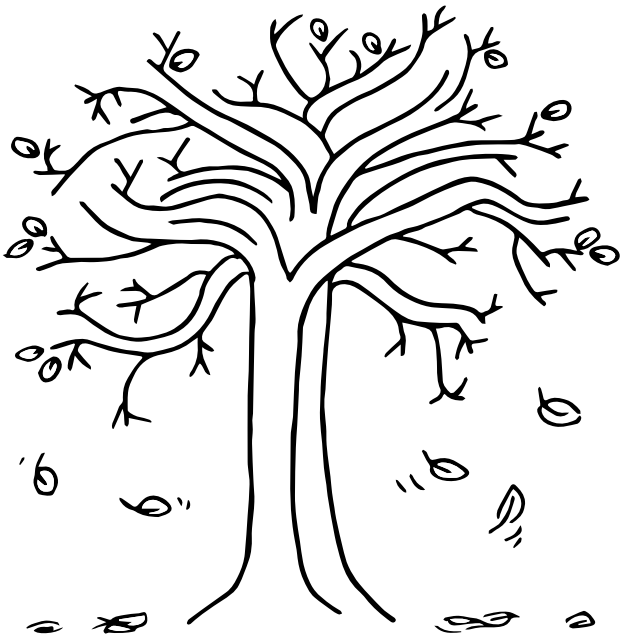
Consequences of disease and treatment in ANCA-associated vasculitis

Tuin, Janneke

Chapter three

3



BRIEF REPORT

Menopause and Primary Ovarian Insufficiency in Women Treated for Antineutrophil Cytoplasmic Antibody–Associated Vasculitides

J. Tuin BSc[1], J.S.F. Sanders MD PhD[1], A.P. van Beek MD PhD[2], A. Hoek MD PhD[3], C.A. Stegeman MD PhD[1]

University of Groningen, University Medical Center Groningen, Department of Internal Medicine, Division of [1]Nephrology. University of Groningen, University Medical Center Groningen, Department of Internal Medicine, Division of [2]Endocrinology. University of Groningen, University Medical Center Groningen, Department of Obstetrics and Gynaecology[3].

Arthritis & Rheumatology 2016;68(4):986–992

Abstract

Objective. One of the side effects of cyclophosphamide is earlier menopause and primary ovarian insufficiency. This study was undertaken to investigate the onset of menopause and the incidence of primary ovarian insufficiency in women with antineutrophil cytoplasmic antibody–associated vasculitis (AAV), especially after treatment with orally administered cyclophosphamide.

Methods. We retrospectively studied the onset of menopause and the influence of cyclophosphamide in women diagnosed as having AAV in our center between 1970 and 2012.

Results. Ninety-four premenopausal women diagnosed as having AAV were included. Sixty-seven patients received cyclophosphamide, and 27 received other, mostly immunosuppressive, medication. Forty-six cyclophosphamide-treated women developed menopause, 22 of whom were considered to have primary ovarian insufficiency. None of the patients who were not treated with cyclophosphamide developed primary ovarian insufficiency. There was a significant association between a cumulative cyclophosphamide dose of >16.6 gm, versus a cumulative dose of <16.6 gm, and menopause (χ^2=8.72, P=0.003; odds ratio [OR] 2.60 [95% confidence interval 1.38–4.90]). In addition, there was a significant association between a cumulative cyclophosphamide dose of <16.6 gm, versus no cyclophosphamide exposure, and menopause (χ^2=16.37, P < 0.001; OR 7.32 [95% confidence interval 2.79–19.20]). Both women who received cyclophosphamide and those who did not experienced involuntary childlessness.

Conclusion. Earlier menopause and primary ovarian insufficiency frequently develop after oral cyclophosphamide therapy in premenopausal women with AAV. Involuntary childlessness is common after the development of primary ovarian insufficiency, but it also occurs in women not treated with cyclophosphamide. These findings emphasize the importance of the use of drugs that are not toxic to gonadal function in women of childbearing age.

Introduction

Cyclophosphamide combined with corticosteroids has been the standard immunosuppressive therapy for antineutrophil cytoplasmic antibody (ANCA)– associated vasculitis (AAV) for many years. More than 80% of patients with granulomatosis with polyangiitis (Wegener's) (GPA) or microscopic polyangiitis (MPA) who are treated with cyclophosphamide and corticosteroids will attain remission (1). Despite its fundamental contribution to the survival of AAV patients, cyclophosphamide is associated with significant toxicity and long-term side effects, including earlier menopause and its extreme form, primary ovarian insufficiency (2).

Cyclophosphamide induces irreversible DNA cross-linking between and within DNA strands, which interferes with cell division. It is hypothesized that in the ovaries, cyclophosphamide induces proliferation of dormant primordial follicles and induces cell death in granulosa cells of larger follicles. Therefore, the recruitment of primordial follicles will be enhanced, leading to early depletion of the primordial follicle pool and finally menopause (3).

Earlier menopause, and especially primary ovarian insufficiency, have considerable implications for reproduction, bone mineral density, general and sexual well-being, and cardiovascular health (4). Limited data are available on the effect of treatment, especially orally administered cyclophosphamide, on the onset of menopause or the incidence of primary ovarian insufficiency in women with AAV (5). In addition, treatment regimens have been adjusted over the years and possible implications of these diagnoses for fertility have hardly been evaluated. Therefore, this study was undertaken to study the effect of cyclophosphamide on the onset of menopause in women treated for AAV over the last 4 decades.

Patients and methods

Patients

All premenopausal female patients diagnosed as having AAV and treated at the University Medical Center Groningen were included. Patients were classified according to criteria adopted from the Chapel Hill Consensus Conference Nomenclature and the algorithm developed by Watts et al (6,7). Patient characteristics and laboratory results were obtained from medical records. Disease activity at diagnosis was scored using the Birmingham Vasculitis Activity Score (BVAS) (8). Women were followed up until July 2014 or until they were lost to follow-up or died. This study was conducted according to the principles of the Declaration of Helsinki (October 2013 version).

Treatment protocol

Standard treatment for AAV consisted of daily oral cyclophosphamide (2 mg/kg) and prednisolone (1 mg/kg; maximum dosage of 60 mg/day). Prednisolone was first tapered by 10 mg every 2 weeks until a dosage of 30 mg was reached and thereafter by 5 mg every 2–4 weeks. Until 1996, once remission was induced, maintenance therapy consisted of oral cyclophosphamide, which was tapered by 25 mg every 3 months.

From 1996 on, cyclophosphamide therapy was switched to azathioprine maintenance therapy (1.5–2mg/kg body weight daily) after 3 months of stable remission. Azathioprine was tapered by 25 mg every 3 months beginning 1 year after diagnosis. Other therapies included the standard

prednisolone scheme combined with mycophenolate mofetil 1,000 mg twice daily, methotrexate 25 mg once a week, or rituximab. A few patients with localised GPA were treated with cotrimoxazole monotherapy (800 mg trimethoprim/160 mg sulfamethoxazole twice daily). The cumulative cyclophosphamide dose was calculated by adding the daily dosages of cyclophosphamide. In patients in whom menses ceased during the treatment period and did not resume, the cumulative dose was calculated over the complete treatment period.

Definitions

Menopause was defined as the last menstrual period followed by at least 12 months of amenorrhea. This was preferably confirmed by follicle-stimulating hormone (FSH) and luteinizing hormone (LH) measurements; however, these data were lacking in part of the cohort due to the retrospective nature of this study. Primary ovarian insufficiency was defined as the occurrence of menopause in women younger than 40 years old.

Statistical analysis

SPSS version 22.0 was used for data analysis. Data are shown as the mean ± SD or median (interquartile range [IQR]). Differences in various parameters between groups were tested using Student's t-test, Mann-Whitney U test, or Pearson's chi-square test for normally distributed, non-normally distributed, and categorical variables, respectively. Time to menopause after cyclophosphamide therapy was evaluated by Kaplan-Meier actuarial survival curves with log rank testing. Women were grouped into 2 strata, and odds ratios (ORs) were calculated. Left censoring was applied to take into account age at cyclophosphamide initiation; therefore, age appears on the x-axis in graphs. P values less than 0.05 (2-sided) were considered significant.

Results

Cohort characteristics

Ninety-four premenopausal female patients diagnosed as having AAV were included in this study. The mean ± SD age at diagnosis was 34±10 years. Sixty-seven women received cyclophosphamide for the induction of remission, and 27 women were treated with other, mostly immunosuppressive, medication. A summary of patient characteristics is shown in Table 1. Women treated with cyclophosphamide were significantly older at initiation of therapy, were more often diagnosed with GPA, were more often proteinase 3–ANCA positive, more frequently had renal involvement, and had a higher BVAS compared to women treated with other agents. The median follow-up time exceeded 13 years in both groups, with a total follow-up of 1,362 patient-years. A flow chart of eligible patients and therapies administered is shown in Figure 1.

Treatment

Thirteen of the 67 women treated with cyclophosphamide were initially treated with another agent in order to prevent cyclophosphamide exposure. Due to therapy failure or an early relapse, these patients started standard treatment with oral cyclophosphamide. Sixty-six patients received oral cyclophosphamide, and 1 woman initially received intravenous cyclophosphamide pulse therapy,

Table 1. Patient and treatment characteristics.

	All women (n= 94)	CYC treatment (n= 67)	No CYC treatment (n=27)	*p*
Age at diagnosis, mean ± SD years	34 ± 10	36 ± 11	31 ± 8	0.033
Diagnosis				
GPA	70 (74.5%)	56 (83.6%)	14 (51.9%)	0.001
MPA	15 (16.0%)	8 (11.9%)	7 (25.9%)	0.094†
EGPA	9 (9.6%)	3 (4.5%)	6 (22.2%)	0.008†
ANCA-type				
PR-3	57 (60.6%)	48 (71.6%)	9 (33.3%)	0.001
MPO	24 (25.5%)	15 (22.4%)	9 (33.3%)	0.271
No specificity	13 (13.8%)	4 (6%)	9 (33.3%)	0.001†
BVAS at diagnosis, median [IQR]	15 [9- 22]	19 [12- 25]	10 [7- 14]	<0.001
Organ involvement at diagnosis				
ENT	76 (80.9%)	57 (85.1%)	19 (70.4%)	0.101
Chest	47 (50%)	36 (53.7%)	11 (40.7%)	0.254
Renal	49 (52.1%)	41 (61.2%)	8 (29.6%)	0.006
eGFR (ml/min) diagnosis, mean ± SD	78 ± 36 (n= 69)	76 ± 38 (n= 48)	84 ± 34 (n= 21)	0.394
BMI at diagnosis, kg/m2, median [IQR]	23 [21- 26] (n= 48)	24 [21- 27] (n= 31)	22 [21-25] (n= 17)	0.262
Smoking				
Current	11 (11.7%)	5 (7.5%)	6 (22.2%)	0.044†
Former	15 (16%)	12 (17.9%)	3 (11.1%)	0.415†
Therapy received before menopause or end FU				
Cyclophosphamide	67 (71.3%)	67 (100%)	0 (0%)	
Azathioprine	42 (44.7%)	32 (47.8%)	10 (37%)	0.344
Glucocorticoids	86 (91.5%)	67 (100%)	19 (70.4%)	<0.001†
Mycophenolate mofetil	14 (14.9%)	9 (13.4%)	5 (18.5%)	0.531†
Rituximab	4 (4.3%)	1 (1.5%)	3 (11.1%)	0.037†
Methotrexate	11 (16.4 %)	7 (10.4 %)	4 (14.8 %)	0.551†
CT monotherapy	19 (20.2%)	7 (10.4%)	12 (44.4%)	<0.001
Other	11 (11.7%)	8 (11.9%)	3 (11.1%)	0.910†
eGFR (ml/min) menopause/ end follow up, mean ± SD	78 ± 28 (n= 73)	74 ± 27 (n= 51)	85 ± 29 (n= 22)	0.106
Renal replacement therapy at menopause/ end FU	5 (5.3%)	4 (6%)	1 (3.7%)	0.658†
FU after diagnosis, median [IQR] months	165 [90- 232]	168 [94- 234]	160 [75- 232]	0.561

* Except where indicated otherwise, values are the number (%). CYC- cyclophosphamide; GPA-granulomatosis with polyangiitis (Wegener's); MPA-microscopic polyangiitis; EGPA-eosinophilic granulomatosis with polyangiitis (Churg-Strauss); ANCA-antineutrophil cytoplasmic antibody; PR3-proteinase 3; MPO-myeloperoxidase; BVAS-Birmingham Vasculitis Activity Score; IQR-interquartile range; ENT-ear, nose, and throat.

† Expected count of <5 in statistical analysis.

‡ Data on estimated glomerular filtration rate (eGFR) at diagnosis were available for 69 patients (48 who were treated with cyclophosphamide and 21 who were not treated with cyclophosphamide).

§ Data on body mass index (BMI) at diagnosis were available for 48 patients (31 who were treated with cyclophosphamide and 17 who were not treated with cyclophosphamide).

¶ Data on eGFR at menopause or end of follow-up were available for 73 patients (51 who were treated with cyclophosphamide and 22 who were not treated with cyclophosphamide).

but was subsequently switched to oral cyclophosphamide due to an insufficient response. Thirty-nine patients switched to maintenance therapy with azathioprine after induction therapy with cyclophosphamide and were exposed to a significantly lower cumulative cyclophosphamide dose compared to women treated with a protocol of cyclophosphamide alone (median 16.6 gm [IQR 12.4–26] versus 47.7 gm [IQR 22.5–70]; P < 0.001). All patients received oral contraceptives in order to attenuate cyclophosphamide-induced ovarian insufficiency and prevent pregnancy during treatment.

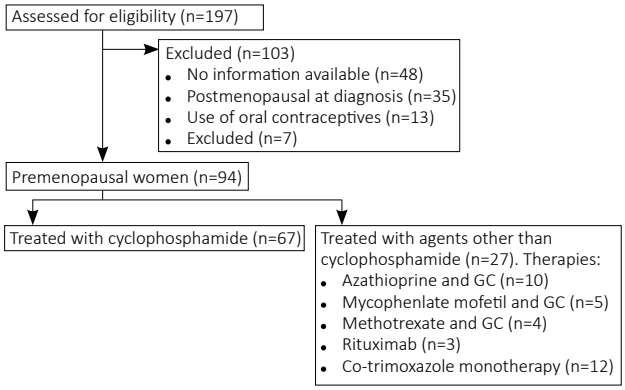


Figure 1. Flowchart of patients included in the study and therapies administered. GC=glucocorticoid.

Menses and menopause

In 33 cyclophosphamide treated patients (49%), menses did not resume within a year after treatment with cyclophosphamide. Patients without resumption of menses (n=33) were significantly older (mean ± SD 42±9 versus 30±8 years; P < 0.001) and tended to be exposed to a higher cumulative dose of cyclophosphamide (median 22.7 gm [IQR 17.4–50]) compared to patients in whom menses did resume (median 17.3 gm [IQR 11.6–45]; n=34) (P=0.055). Menses resumed more often in women who switched to azathioprine maintenance therapy (n=39) than in women who did not switch (n=28) (61.5% versus 35.7%) ($\chi^2[1]=4.35$, P=0.037).

At the end of the follow-up period, menopause had occurred in 46 (69%) of the cyclophosphamide treated women, at a mean ± SD age of 41±8 years. Menopause was confirmed by FSH and LH measurement in 18 (39%) of 46 patients. Menopause occurred after a median cumulative cyclophosphamide dose of 26.5 gm (IQR 16.6–54.7). Twenty-two women (48%) were considered to have developed primary ovarian insufficiency, since menopause occurred before the age of 40 years. The median cumulative cyclophosphamide dose in women with primary ovarian insufficiency was 48.7 gm (IQR 22.9–68.6) and was significantly higher than in patients who became menopausal but did not have primary ovarian insufficiency (median 19.6 gm [IQR 16.2–28.8]; n=24) (P=0.008). At the end of the follow-up period, menopause had occurred in 4 women who were not previously exposed to cyclophosphamide. Menopause occurred at the age of 47 years in 2 patients and at the age of 50 years in 2 patients.

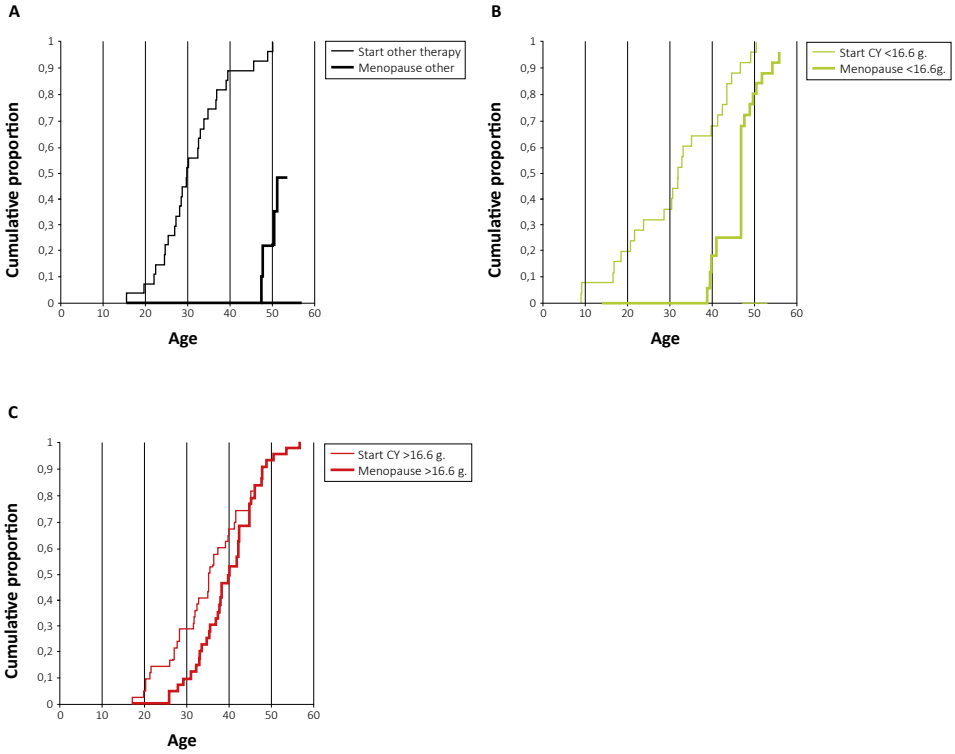


Figure 2. Cumulative proportion with menopause in women treated for antineutrophil cytoplasmic antibody–associated vasculitis. **A**, Patients who were not exposed to cyclophosphamide (n=27). **B**, Patients who were exposed to a cumulative cyclophosphamide dose of <16.6 gm (n=25). **C**, Patients who were exposed to a cumulative cyclophosphamide dose of >16.6 gm (n=42). Thin lines represent age at cyclophosphamide initiation; thick lines represent age at menopause or age at censoring. Therefore, patients are represented twice in these graphs.

A Kaplan-Meier plot with left censoring of the cumulative proportion of subjects in menopause against age and the interval between therapy initiation and menopause is shown in Figure 2. Women were stratified according to the mean cumulative cyclophosphamide dose in patients who switched to sequential azathioprine maintenance, which was 16.6 gm. At the age of 40, the cumulative proportion in menopause was 18% for women who received <16.6 gm compared to 50% for women who received higher doses. None of the women who received other agents developed menopause at age 40.

At younger ages, the effect of cyclophosphamide dose was less pronounced. The time interval between initiation of therapy and menopause was considerably longer in younger women, especially women who received <16.6 gm of cyclophosphamide. With increasing age, the interval was shortened, and women older than ~44 years became menopausal promptly after cyclophosphamide initiation, irrespective of dosage. There was a significant association between cumulative cyclophosphamide doses of >16.6 gm (n=42), versus cumulative doses of <16.6 gm

(n=25), and menopause (χ^2[1]=8.72, P=0.003; OR 2.60 [95% confidence interval 1.38–4.90]). In addition, there was a significant association between a cumulative dose of <16.6 gm, versus no cyclophosphamide exposure (n=27), and menopause (χ^2[1]516.37, P<0.001; OR 7.32 [95% confidence interval 2.79–19.20]).

Family planning

The number of children born after diagnosis was low in the entire cohort; 17 children were born to cyclophosphamide-treated women (n=62) and 9 were born to women who were not treated with cyclophosphamide (n=26). At least 6 women with primary ovarian insufficiency were documented to be involuntarily childless. In addition, at least 2 patients not treated with cyclophosphamide and 1 cyclophosphamide treated woman regarded pregnancy as undesirable due to the burden of disease and were consequently involuntarily childless as well. At the end of the follow-up period, 3 women previously treated with agents other than cyclophosphamide were still pregnant.

Discussion

This study aimed to investigate the onset of menopause and incidence of primary ovarian insufficiency following therapy for induction of remission of AAV and the role of orally administered cyclophosphamide therein. The present study indicates that cyclophosphamide is related to earlier menopause and primary ovarian insufficiency in a dose- and age-dependent manner and that current treatment protocols reduce, but by no means eliminate, ovarian toxicity.

Our findings are consistent with those of previous studies. In systemic lupus erythematosus the risk of sustained amenorrhea is estimated to be 25–59% after treatment with oral cyclophosphamide and 11–54% after treatment with intravenous cyclophosphamide (9). In our cohort, 49% of the women had no resumption of menses, and 48% of menopausal women were considered to have developed primary ovarian insufficiency. The high primary ovarian insufficiency incidence in this study might be explained by the fact that most of these women received high cumulative cyclophosphamide doses. As demonstrated in previous studies, cyclophosphamide induces menopause in a dose-dependent manner (10).

Several of our patients were treated in the 1970s and 1980s, and at that time, cyclophosphamide was continued for ~2 years with slow tapering. Over the years, treatment protocols based on an induction phase followed by sequential therapy have reduced cyclophosphamide exposure dramatically. In the present study, the odds of developing menopause were >2 times lower in women who received <16.6 gm of cyclophosphamide, a cumulative dose comparable to those reached in current treatment protocols. Our results therefore show that the introduction of induction therapy followed by sequential therapy, due to among others the Cyclophosphamide versus Azathioprine During Remission of Systemic Vasculitis (CYCAZAREM) study, led to an important improvement in the treatment of AAV by lowering cyclophosphamide exposure and in parallel reducing the risk of early menopause and primary ovarian insufficiency (11).

Besides the cumulative dose received, age at cyclophosphamide initiation is considered a risk factor for the development of menopause or primary ovarian insufficiency, in which age is considered to be a reflection of ovarian reserve. The present study clearly showed an inverse association between age at cyclophosphamide initiation and the onset of menopause. This supports the idea

that younger women have a larger follicle pool and, hence, are relatively protected against follicle depletion. However, there appeared to be a certain age threshold, of ~44 years, above which the risk of menopause was unequivocally high irrespective of the cyclophosphamide dose.

This study underscores an important dilemma in the care of women of childbearing potential with AAV. Is there still a place for cyclophosphamide in the treatment of these women? Fortunately, new treatment options such as rituximab have emerged, but cyclophosphamide is still a highly effective agent which might be indicated in patients who show insufficient response to other therapies. Despite its toxicity, insufficient treatment of severely ill patients could be more detrimental. Additionally, poor disease control, a higher relapse rate, or vital organ damage hamper patients' abilities to fulfill their desire to have a child. This is supported by our finding that several women regarded pregnancy as undesirable due to the burden of disease and were involuntarily childless, irrespective of treatment choice.

Preserving fertility is a difficult challenge, especially with the need for early treatment in AAV. In this study, all women received oral contraceptives in order to attenuate cyclophosphamide-induced menopause and prevent pregnancy during treatment. However, a meta-analysis did not show beneficial effects of oral contraceptives (12). In addition, the efficacy of gonadotropinreleasing hormone (GnRH) analogs remains uncertain, though a recent study showed a protective effect of a GnRH agonist on fertility (13,14). Established preservation methods such as oocyte or embryo cryopreservation might take too long when early treatment is warranted. The results of experimental methods, for example, in vitro maturation of immature oocytes which can be harvested quickly, ovarian cortex cryopreservation, and neoadjuvant pharmacotherapy, should be awaited (15). In addition, therapies that are not toxic to gonadal function, such as rituximab, should be considered.

Obviously, our results should be interpreted with caution. A limitation of this study was the lack of confirmation of menopause by biomarkers such as FSH or anti–müllerian hormone in part of the cohort. Menopause is diagnosed when the last menstruation is followed by 12 months of amenorrhea, but menses might resume after a longer period of time. However, it cannot be unequivocally concluded that resumption of menses reflects reproductive potential. A third potential limitation was the inclusion of all women treated over the last 40 years. Considering the changes in therapy, our results might be harder to extrapolate to the current situation. However, we stratified the patients according to a current, relevant cumulative cyclophosphamide dose. In addition, these results provide insights into the declining risk of earlier menopause and primary ovarian insufficiency over the past decades attributable to the development of new treatment protocols. A fourth limitation was the assessment of cumulative cyclophosphamide doses in women in whom menses did not resume, since the time of menopause could not be accurately determined. We therefore calculated the cumulative dose over the complete treatment period, which meant in some cases a cumulative dose over more than 1 year. This probably led to an overestimation of the cumulative cyclophosphamide dose that induced menopause. A strength of this study was the extensive follow-up consisting of 1,362 patient-years.

This study showed that earlier menopause and primary ovarian insufficiency frequently develop after oral cyclophosphamide therapy for induction of remission in women of childbearing potential with AAV. A treatment protocol based on the CYCAZAREM study successfully increased the interval between therapy and onset of menopause and reduced the incidence of primary ovarian

insufficiency in this study. We expect further improvement with newly emerging therapeutic options such as rituximab.

Women should be counseled about the risk of earlier menopause, and in women older than 44 years of age, resumption of menses is not to be expected. Appropriate therapy, for example, estrogen replacement therapy, should be considered. An issue that arises from this study is the incidence of involuntary childlessness due to disease activity or morbidity, irrespective of choice of therapy. This also emphasizes the need for better disease management, preferably without cyclophosphamide. Nevertheless, our findings suggest that when other drugs are unable to induce remission or are not well tolerated, cyclophosphamide may be used up to a cumulative dose of 16 gm, which might still give patients, especially younger women, a "window of opportunity" if a child is desired. Induction of sustained remission is still vital in these incurable and life-threatening diseases.

ACKNOWLEDGMENTS

We acknowledge Dr. C. F. M. Franssen, R. T. Gansevoort, R. R. H. Nap, A. Ozyilmaz, A. Rutgers, and W. J. Sluiter for assistance with data collection and statistical support.

AUTHOR CONTRIBUTIONS

All authors were involved in drafting the article or revising it critically for important intellectual content, and all authors approved the final version to be published.

Dr. Tuin had full access to all of the data in the study and takes responsibility for the integrity of the data and the accuracy of the data analysis.

Study conception and design. Tuin, Sanders, Stegeman.
Acquisition of data. Tuin, Sanders, Stegeman.
Analysis and interpretation of data. Tuin, Sanders, van Beek, Hoek, Stegeman.

References

1. Mukhtyar C, Flossmann O, Hellmich B, Bacon P, Cid M, Cohen-Tervaert JW, et al, on behalf of the European Vasculitis Study Group (EUVAS). Outcomes from studies of antineutrophil cytoplasm antibody associated vasculitis: a systematic review by the European League Against Rheumatism systemic vasculitis task force. Ann Rheum Dis 2008;67:1004–10.
2. Kovarsky J. Clinical pharmacology and toxicology of cyclophosphamide: emphasis on use in rheumatic diseases. Semin Arthritis Rheum 1983;12:359–72.
3. Kalich-Philosoph L, Roness H, Carmely A, Fishel-Bartal M, Ligumsky H, Paglin S, et al, Cyclophosphamide triggers follicle activation and "burnout"; AS101 prevents follicle loss and preserves fertility. Sci Transl Med 2013;5:185ra62.
4. Shuster LT, Rhodes DJ, Gostout BS, Grossardt BR, Rocca WA. Premature menopause or early menopause: long-term health consequences. Maturitas 2010;65:161–6.
5. Clowse ME, Copland SC, Hsieh TC, Chow SC, Hoffman GS, Merkel PA, et al. Ovarian reserve diminished by oral cyclophosphamide therapy for granulomatosis with polyangiitis (Wegener's). Arthritis Care Res (Hoboken) 2011;63:1777–81.
6. Jennette JC, Falk RJ, Bacon PA, Basu N, Cid MC, Ferrario F, et al. 2012 Revised International Chapel Hill Consensus Conference Nomenclature of Vasculitides. Arthritis Rheum 2013;65:1–11.
7. Watts R, Lane S, Hanslik T, Hauser T, Hellmich B, Koldingsnes W, et al. Development and validation of a consensus methodology for the classification of the ANCA-associated vasculitides and polyarteritis nodosa for epidemiological studies. Ann Rheum Dis 2007;66:222–7.
8. Luqmani RA, Bacon PA, Moots RJ, Janssen BA, Pall A, Emery P, et al. Birmingham Vasculitis Activity Score (BVAS) in systemic necrotizing vasculitis. QJM 1994;87:671–8.
9. Katsifis GE, Tzioufas AG. Ovarian failure in systemic lupus erythematosus patients treated with pulsed intravenous cyclophosphamide. Lupus 2004;13:673–8.
10. Park MC, Park YB, Jung SY, Chung IH, Choi KH, Lee SK. Risk of ovarian failure and pregnancy outcome in patients with lupus nephritis treated with intravenous cyclophosphamide pulse therapy. Lupus 2004;13:569–74.
11. Jayne D, Rasmussen N, Andrassy K, Bacon P, Tervaert JW, Dadoniene J, et al. A randomized trial of maintenance therapy for vasculitis associated with antineutrophil cytoplasmic autoantibodies. N Engl J Med 2003;349:36–44.
12. Ben-Aharon I, Gafter-Gvili A, Leibovici L, Stemmer SM. Pharmacological interventions for fertility preservation during chemotherapy: a systematic review and meta-analysis. Breast Cancer Res Treat 2010;122:803–11.
13. Turner NH, Partridge A, Sanna G, Di Leo A, Biganzoli L. Utility of gonadotropin-releasing hormone agonists for fertility preservation in young breast cancer patients: the benefit remains uncertain. Ann Oncol 2013;24:2224–35.
14. Moore HC, Unger JM, Phillips KA, Boyle F, Hitre E, Porter D, et al. Goserelin for ovarian protection during breast-cancer adjuvant chemotherapy. N Engl J Med 2015;372:923–32.
15. De Vos M, Smitz J, Woodruff TK. Fertility preservation in women with cancer. Lancet 2014;384:1302–10.