ISSN 2472-1972

The Use of Resveratrol as an Adjuvant Treatment of Pain in Endometriosis: A Randomized Clinical Trial

Daniel Mendes da Silva,[1] Luiza Azevedo Gross,[2] Ernesto de Paula Guedes Neto,[3] Bruce A. Lessey,[4] and Ricardo Francalacci Savaris[1,2]

[1]Postgraduate Program in Health Science: Gynecology and Obstetrics, Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil, 90035-903; [2]Department of Obstetrics and Gynecology, Universidade Federal do Rio Grande do Sul, Porto Alegre, Rio Grande do Sul, Brazil, 90035-903; [3]Clínica Mulher, Porto Alegre, Rio Grande do Sul, Brazil, 90570-060; and [4]Department of Obstetrics and Gynecology, Greenville Health System, Greenville, South Carolina

Context: Resveratrol has been used for the treatment of endometriosis.

Objective: To compare resveratrol (40 mg/d) with monophasic contraceptive pill (COC) to COC with placebo for the reduction of pain scores.

Design: A randomized clinical trial.

Setting: University Hospital.

Patients: Women (ages 20 to 50) with laparoscopic diagnosis of endometriosis were eligible for the study. Exclusion criteria: pregnancy, allergy to resveratrol, or contraindications to COC, use of agonists of gonadotropin release hormone or danazol in the last month, or had used depot medroxyprogesterone acetate or Mirena®.

Intervention: Subjects were randomized using a computer-generated randomization list to receive COC for 42 days to be taken with identical capsules containing 40 mg of resveratrol or placebo in coded bottles (1:1 ratio). Allocation was concealed in coded, sequenced, opaque-sealed envelopes.

Main Outcome: Median pain scores measured with a visual analog scale on day 42.

Results: Between 18 June and 6 November 2015, 44 subjects were enrolled. Mean [95% confidence interval (CI)] pain scores on day 0 were 5.4 (4.2 to 6.6) in the placebo group and 5.7 (4.8 to 6.6) in resveratrol groups. After treatment, pain values were [3.9 (2.2 to 5); n = 22] and [3.2 (2.1 to 4.3); n = 22] in the placebo and resveratrol groups, respectively (P = 0.7; Mann-Whitney U test). Median (95% CI) difference between groups was 0.75 (–1.6 to 2.3).

Conclusion: Resveratrol is not superior to placebo for treatment of pain in endometriosis.

Freeform/Key Words: endometriosis, pain, resveratrol

Endometriosis, defined as the presence of endometrial glands and stroma outside the uterine cavity, affects 176 million women worldwide [1], and it is present in up to 60% of adolescent women with chronic pelvic pain or dysmenorrhea [2]. Increased endometrial proliferation and

Abbreviations: CI, confidence interval; COC, monophasic oral contraceptive; GEE, generalized estimating equation; ITT, intention to treat; PP, per protocol; RM-2 Way ANOVA, repeated-measures, two-way analysis of variance; VAS, visual analog scale.

Received 13 January 2017
Accepted 10 March 2017
First Published Online 15 March 2017

estrogen responsiveness are hallmarks of endometriosis [2–4]. The eutopic and ectopic endometrium of women with endometriosis have an increased expression of estrogen receptors (ESR1) [1, 5], thought to be due to a defect in the action of progesterone, (progesterone resistance) [4, 6], or by an excessive production of estrogen [7–9]. Estrogen receptor beta expression (ESR2) is also increased the endometrium of women with endometriosis [10, 11]. Given these changes, endometriosis is considered a predominantly estrogen-dependent disease, and therefore hormonal suppression might be an attractive medical approach to treat this disease and its symptoms [12].

Resveratrol (3,5,4'-trihydroxystilbene) is a natural phytoestrogen synthesized by plants, such as dark grapes and blueberries, after exposure to UV radiation. Side effects with the use of resveratrol are mild, mainly related to headache and somnolence [13, 14]; the use of resveratrol has been suggested to treat endometriosis, due to its antiproliferative action on endometriotic implants in *in vitro* and animal models [15–17].

The mechanism of action for resveratrol in the treatment of endometriosis is not obvious but likely reflects changes in estrogen responsiveness. Resveratrol binds to the estrogen receptors (ESR1 and ESR2) [18] and can act as an agonist or antagonist, depending on the clinical context [19]. In breast cancer cells, for example, resveratrol acts an agonist by binding to ESR1 [19]. We have previously shown that resveratrol acts as an antagonist in an animal model of endometriosis [20]. As resveratrol has a much higher affinity for ESR2, compared with ESR1 [21], and because ESR2 is dramatically overexpressed in endometriosis [22], this preferential binding of resveratrol to ESR2 may explain the observed antagonist to ESR1 by resveratrol, blocking the ability of estrogen to induce its own receptor. We previously showed that ESR1 is reduced by resveratrol in the mouse xenograft model [20].

In women with endometriosis, resveratrol, with drosperinone 3 mg and ethinyl estradiol 30 μg, was previously investigated in a small nonrandomized, open-label trial [23]. These researchers demonstrated that pain scores were significantly reduced by resveratrol compared with drosperinone 3 mg and ethinyl estradiol 30 μg alone [23]. The ESHRE guidelines do not support the use of nutritional supplements in the treatment of endometriosis, because the potential benefits and/or harms have not been demonstrated [12]. Therefore, the primary objective of this study was to verify whether the use of a monophasic contraceptive pill (COC) plus 40 mg per day of resveratrol was superior to placebo to reduce pain values in patients with endometriosis using a randomized, double-blind, placebo-controlled study design. As a secondary objective, we compared: (1) the use of medication for pain, (2) plasma levels of carcinoembryonic antigen (CA-125) and prolactin before and after treatment as biomarkers for follow-up of endometriosis treatment [6, 24, 25], (3) side effects observed in the trial, and (4) reduction of pain levels in each treatment arm compared with pain level at the initial visit.

1. Material and Methods

A. Trial Design

This study was designed as a parallel, double-blind, randomized, placebo-controlled trial with a 1:1 ratio.

B. Participants

Women between the ages of 20 and 50 with laparoscopic-proven diagnosis of endometriosis were eligible for the study. Subjects were excluded if they were pregnant, had known allergy to resveratrol or contraindications to monophasic contraceptive pills, according to World Health Organization eligibility criteria [26], or used GnRH agonists or danazol in the last month. The use of medroxiprogesterone acetate depot in the last 12 weeks and current use of Mirena® were added as ineligibility criteria after the trial started because both could

introduce bias in the reporting of pain scores. Pregnancy was also an exclusion criteria because it is a contraindication for the use of COCs.

The trial took place at the Clinical Research Center of the Hospital de Clínicas de Porto Alegre, Brazil, between 18 June and 6 November 2015.

C. Intervention

At the first consultation (day 1), pregnancy was ruled out with a negative urinary pregnancy test. After a standard questionnaire, subjects were randomized to receive the first part of the treatment: one pack of COC (levonorgestrel 0.15 mg/ethinyl estradiol 0.03 mg) to be taken daily for 21 days without pause and a coded bottle that contained 21 identical capsules of either 40 mg of resveratrol or placebo. Capsules were prepared by a local pharmacy. Patients started both medications (contraceptive pill and a capsule) on day 1. They were instructed to record any side effects and to use analgesics as needed. Specific analgesics were prescribed, and subjects were instructed to use them in the following order: dipyrone 500 mg up to 6/6 hours, ibuprofen 600 mg up to 8/8 hours, and codeine 30 mg up to 6/6 hours. Subjects were also instructed to report the use of any other medication and to return on day 21 and again on day 42 of treatment. Seven days after the first consultation, patients were contacted by telephone to determine if they had any early complications of therapy and to provide initial pain scores. On day 21, patients returned to receive the second installment of the treatment. On day 42, patients returned for the final visit. Compliance was verified by the presence of empty packs of contraceptive pills and the capsules in the coded bottle on days 21 and 42.

D. Outcomes: Primary Outcome Measures

Pain was assessed using a visual analog scale (VAS) scale as previously described [27] and was the primary outcome. VAS is the most commonly used instrument for the assessment of pain in endometriosis [28]. Briefly, a standard question was read by one of the investigators, and a 10-cm slide ruler, marked at regular intervals in millimeters, was presented to the patient. A scale of 0 to 10 was printed on the back of the ruler, facing the investigator, and a pain intensity drawing without numbers was facing the patient. Subjects were asked to move the slide somewhere along the scale to described their average pain in the last 7 days. Pain scale varied from 0 (no pain) to 10 (worst imaginable pain). The pain scores were evaluated on days 1, 7, 21, and 42, using the same standard question. The follow-up period of 42 days was based on two consecutives cycles of COCs. Reduction of levels of pain caused by endometriosis has been observed after the first month of treatment by many authors using different hormonal treatments [29–34].

E. Secondary Outcome Measures

Serum levels of CA-125 and prolactin were measured by enzyme-linked immunosorbent assay at a local endocrine laboratory using electrochemiluminescence immunoassay (Elecsys 2010, Modular Analytics E170, Roche Diagnostic GmbH, D-68305, Indianapolis, IN), with a sensitivity of 0.6 U/mL and 0.047 ng/mL for CA-125 and prolactin, respectively. Repeatability and intermedia precision in both methods were <2 for CA-125 and prolactin, respectively. Serum levels were measured on days 1 and 42.

The use of analgesics and side effects of COCs plus placebo or resveratrol were compared in both groups after entering the trial.

Reduction of pain after 7, 21, and 42 days was compared with baseline pain scores in each group.

F. Sample Size

A sample size was calculated for a superiority trial of resveratrol over placebo for a final difference of pain score between groups, based on data derived from Maia *et al.* [23]. A sample size of at least 21 patients per arm would be necessary to have a 90% chance of detecting, as significant

at the 1% level, a decrease in the median pain levels from 5 in the placebo group to a median pain level of 2 in the resveratrol group, after 42 days of treatment. A standard deviation of 2.5 was obtained from a pilot study with 10 cases. Due to the nature of resveratrol, a nutritional supplement available as an over-the-counter drug, no stopping guidelines were established.

G. Randomization Sequence Generation

Participants were randomly assigned to resveratrol (treatment A) or placebo (treatment B) according to a computer-generated randomization list (www.randomization.com). Subjects were allocated in blocks of four, as suggested in the literature [35].

H. Allocation Concealment Mechanism

Allocation sequence was concealed using coded, numerically sequenced, opaque-sealed envelopes, which were opened after patients were included in the study. Two investigators, blinded to treatments, were responsible for assignment, enrollment, and follow-up of the subjects.

I. Statistical Methods

Normal distribution of data was verified with a Kolmogorov-Smirnov normality test. If normal distribution was not present, data were transformed into a logarithmic scale for CA-125 and prolactin levels. Direct comparison between pain scores in both groups at day 1 and day 42 was performed using a Mann-Whitney U test and unpaired t test with Welch's correction, according to data distribution. Data on CA-125 and prolactin were analyzed using repeated-measures, two-way analysis of variance (RM-2 Way ANOVA), because we compared the effect of resveratrol and placebo on the plasma concentration of CA-125 and prolactin on days 1 and 42.

Generalized estimating equations (GEEs) were used for further statistical analysis, having the patients as the subject variable, time (visits), as within-subject variable, pain scores (VAS) as the dependent variable, treatment type (placebo or resveratrol), use of medication for pain (yes/no), and time were the factors analyzed (visits on days 0, 7, 21, and 42). The analysis used a linear-scale response for pain score and a factorial model analysis for the analyzed factors. A maximum-likelihood estimate, where estimates and standard errors are based on the likelihood function given the observed data, was used in the analysis, as suggested in the literature [36]; correction for multiple comparisons was made using a Bonferroni test. GEE is recommended for missing data in clinical trials, because calculation with missing data is considered in the GEE algorithm.

Sensitivity analysis was performed on data with missing values, *i.e.*, intention to treat (ITT), and compared with data without missing values [per protocol (PP)]. In PP analysis, only patients who complied with trial protocol were considered in the analysis. In ITT analysis, patients who were lost on follow-up had their last registry on pain values or plasma level measurements repeated in the follow-up consultation. If no systematic differences were observed between participants with complete data and those with missing information, data were considered to be missing at random. Data were analyzed using GraphPad Prism 6.0 for Mac (GraphPad Software, La Jolla, CA) and SPSS version 23 for Mac (IBM Software Group, Chicago, IL).

J. Ethical Issues

This study was approved by the Ethics Committee of the Hospital de Clínicas de Porto Alegre (no. 14-626) and was registered at ClinicalTrials.gov (no. NCT02475564).

2. Results

A. Participants

Two hundred and forty-three women with diagnosis of endometriosis were screened for acceptance into the study between 18 June and 6 November 2015. Of these, 199 were excluded

for not meeting the inclusion criteria. Forty-four subjects were randomly assigned to one of the two treatment regimens. Forty-three women received treatment as allocated. Five women were removed from the PP analysis. One discontinued the study after signing the informed consent, one was removed from the study for intense abnormal uterine bleeding that stopped after medical intervention, and three were lost to follow-up (Fig. 1).

For each group, 22 subjects were analyzed as ITT. Outcomes PP were analyzed in 21 and 18 subjects in the resveratrol and placebo arms, respectively.

Baseline data of the studied population are depicted in Table 1.

B. Primary Outcome: Pain Score

After 7 days of treatment, pain scores were significantly reduced in the resveratrol group, compared with the placebo group. After 42 days of treatment, however, no difference was observed in pain scores between treatment groups. The median difference between groups was 0.75 [95% confidence interval (CI): –1.6 to 2.3; Table 2; ITT results; $P = 0.7$, Mann-Whitney

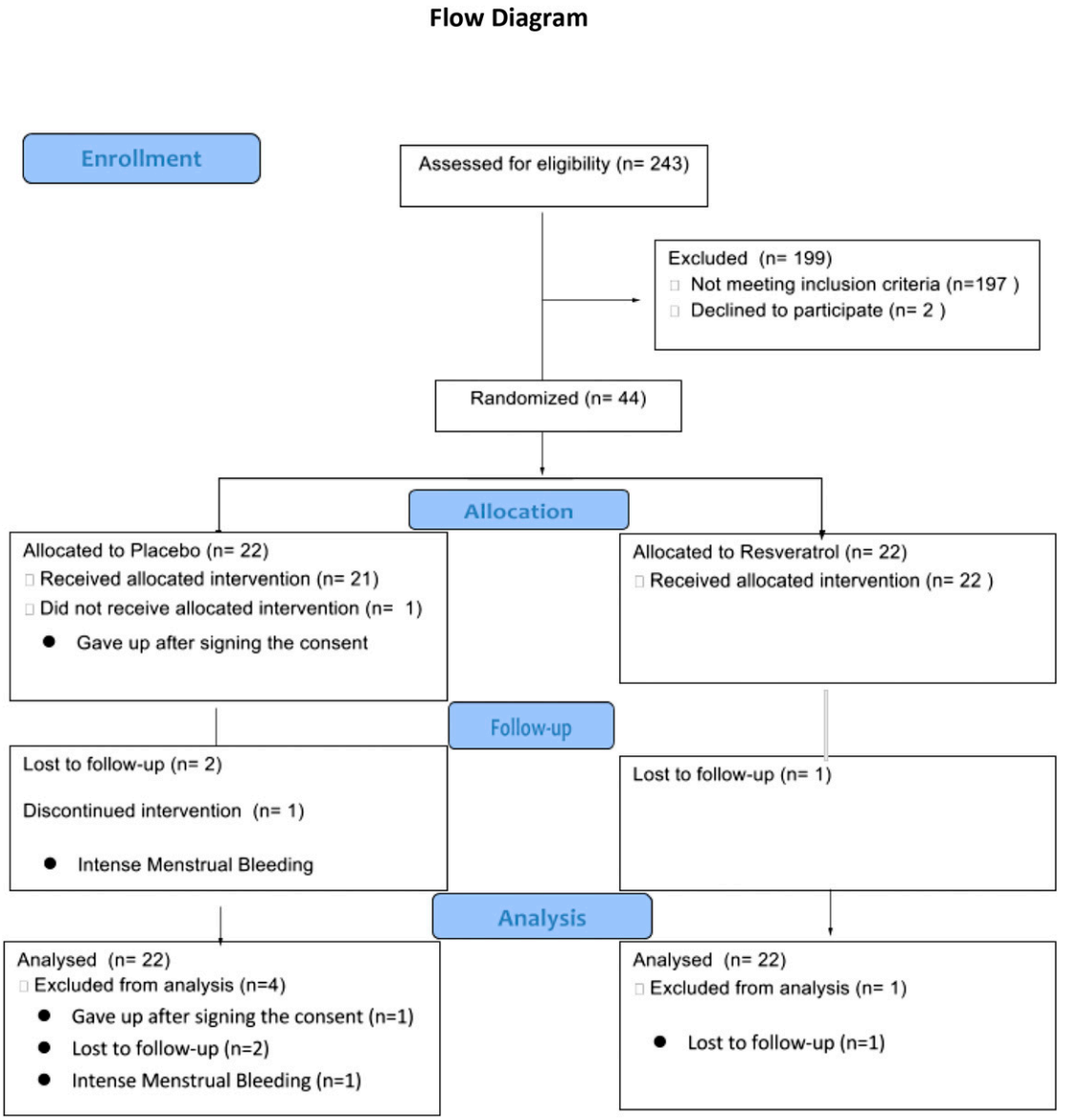

Figure 1. Flow diagram of study population randomized to two treatment groups.

Table 1. Demographic and Clinical Characteristic of the Studied Population

Characteristic	Resveratrol (n = 220)	Placebo (n = 22)	*P* Value
Mean age ± standard deviation (y)	35.4 ± 7.1	32.4 ± 7	0.1[c]
Ethnic group (n [%])			1[d]
White	16 (72)	15 (68)	
Black	6 (28)	7 (32)	
Baseline levels (mean ± standard deviation)			
Pain score (VAS)	5.4 ± 2.6	5.7 ± 2	0.6[e]
CA-125 (U/mL)[a,b]	14.1 (3.3 to 49.5)	18.7 (5.7 to 123.6)	0.2[c]
Prolactin (ng/mL)[b]	12.65 ± 5.9	14.27 ± 6.6	0.4[c]

[a]Raw values are median (range). Analysis after transformation using $Y = \log(Y)$ was performed, and no difference was found using Student *t* test. Transformed data are shown in Figure 2.
[b]n = 21 for serum levels; one case of the placebo group dropped out before collecting blood.
[c]Mann-Whitney *U* test.
[d]Fisher's exact test.
[e]Student *t* test.

U test]. Subgroup analysis was performed on subjects that followed protocol and did not use any medication for pain. Final pain scores were not significantly different between groups by PP analysis or by use or not of analgesics (Table 2). Similar results were found using a Student *t* test after data normalization (data not shown).

Further analysis using GEEs and the possible confounding factor (use of medication, time to follow-up) revealed no difference between groups [Fig. 2(a); all analyses were made using GEEs]. Further details are provided in Supplemental Table 1.

Table 2. Outcomes Measured in Both Groups

Characteristic	Resveratrol (n = 22)	Placebo (n = 22)	*P* Value
Subjects that used pain medication (n)	7	8	
Dipyrone, mg: mean (standard deviation) [n]	1944 (1793) [5]	625 (250) [5]	0.09[c]
Ibuprofen, mg: mean (standard deviation) [n]	1200 (848) [5]	5640 (328) [5]	0.09[c]
Codeine, mg (n)	0	30 (1)	
Reported side effects (n)			
Diplopia	1	0	
Headache	6	7	
Reduced libido	1	0	
Nausea	1	2	
Breast tenderness	1	0	
Hot flushes	1	0	
Increased uterine bleeding	1	0	
Candidiasis	1	0	
Dyspareunia	0	1	
Pain levels at day 42			
ITT[a]	3.2 (0 to 8)	3.9 (0 to 8.9)	0.7[b]
Difference between medians (95% CI)	0.75 (–1.6 to 2.3)		
PP[a]	3 (0 to 8) [n = 21]	2.65 (0 to 8.9) [n = 18]	0.9[b]
Difference between medians (95% CI)	0.35 (–2.3 to 2)		
PP no use of pain medication[a]	3 (0 to 7.7) (n = 15)	2.65 (0 to 8.9) (n = 14)	0.8[b]
Difference between medians (95% CI)	0.35 (–2.3 to 2.8)		
CA-125 (U/mL) at day 42[a]	11.7 (4.9 to 29.9)	13.7 (5 to 61)	0.1[b]
Prolactin (ng/mL) at day 42[a]	12 (4.3 to 32.1)	11.1 (4.2 to 31.6)	0.8[b]

[a]Values are median (range).
[b]Mann-Whitney *U* test.
[c]Unpaired *t* test with Welch's correction.

C. Secondary Outcome

C-1. CA-125 and prolactin

CA-125 levels were reduced after 42 days of treatment in placebo (P = 0.01; RM-2 Way ANOVA) and the resveratrol group (P = 0.02; RM-2 Way ANOVA). Prolactin levels did not vary by treatment type or over time [Fig. 2(b) and 2(c)].

C-2. Use of analgesics

In both arms, subjects used a similar amount of pain medication. Reported side effects were comparable in both groups and were mild (Table 2).

3. Discussion

The use of complementary and alternative medicine has been widely studied for the treatment of endometriosis [37]. Plant-based products including genistein, green tea, and resveratrol have each been reported to exhibit efficacy against endometriosis in animal studies [20, 38–41]; however, no proper clinical trials have been reported for these compounds for the treatment of endometriosis in humans.

This study investigated whether 40 mg/d of resveratrol with a COC would reduce pain scores after 42 days of use in women with endometriosis, compared with a placebo arm with a COC alone. No difference was found between treatments when pain scores were compared between groups after treatment. These results were the same when data were analyzed using ITT or PP analysis (Table 2). Likewise, no difference was found between groups when the use of analgesics and differences in pain score over time were considered (Supplemental Table 1).

This is a randomized, double-blind, controlled comparison of resveratrol vs placebo for the treatment of endometriosis-related pain. A smaller nonrandomized, open-label study by Maia *et al.* [23] reported reduced pain in response to resveratrol. Possible explanations for these differences in reported outcomes could be related to methodology, including length of treatment, pain score scales, or statistical analysis. For practical reasons, we used a more commonly available birth control pill with 21 days of active medication for two cycles (extending the treatment to 42 days) compared with their 56 days of treatment. Because *in vivo* and *in vitro* studies have shown greater benefit at higher doses [20, 41], we used a higher dose of resveratrol compared with the prior study.

This double-blinded, controlled, randomized clinical trial followed the CONSORT guidelines [42]; Maia *et al.* used a before-and-after treatment without randomization. We used a more commonly used linear pain scale (VAS) [28], whereas Maia *et al.* used a three-point scale [23]. This study used levonorgestrel as progestin, whereas drosperinone was used in the previous study; however, a difference in progestins is unlikely to be responsible for the discrepant results [43, 44].

Secondary outcomes were also examined in our study. CA-125 and prolactin levels have been shown to decrease after treatment with resveratrol [17, 45]. CA-125, but not prolactin plasma levels, were significantly reduced after treatment in both groups in our study (Fig. 2). CA-125 results are similar to those reported by de Sá Rosa e Silva *et al.* [46]. They reported that intrauterine devices with levonorgestrel reduce CA-125 plasma levels after 6 months of treatment. Although we see a reduction in CA-125, we did not find the same changes in prolactin as previously reported.

This trial has several strengths, including the controlled, randomized, double-blind study design. Less than 20% of the study population was lost to follow-up (only five cases out of 44). PP analysis was done with 39 subjects (88%). A more robust statistical analysis (GEE) was used, and similar results were observed in the PP, ITT, and sensitivity analysis (Table 2). In addition to the subjective measure of pain, nonsubjective results, including CA-125 and prolactin, were measured.

We documented activity of the treatment in both groups, with a reduction found in CA-125 levels, in both the placebo and resveratrol group (Fig. 2). In addition, a final median difference

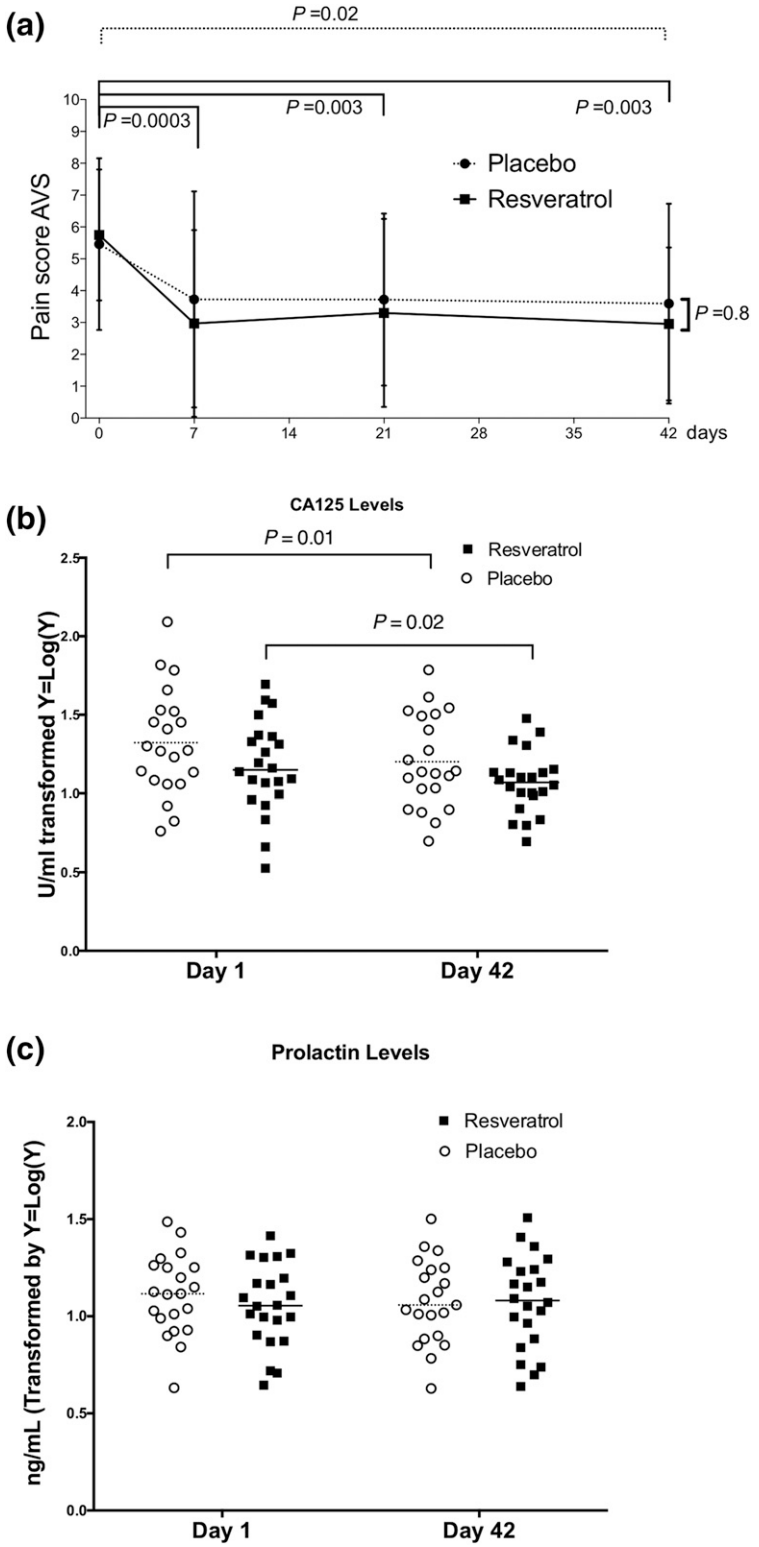

Figure 2. (a) Variation of pain scores between groups and compared with baseline; data were analyzed using GEEs. (b) CA-125 and (c) prolactin levels before and after treatment with monophasic contraceptive pill with or without resveratrol. Data were transformed into $Y = \log(Y)$ for achieving normal distribution and analyzed using RM-2 Way ANOVA. Bars represent means for each variable studied.

of 0.75 (95% CI: –1.6 to 2.3; Table 2; ITT results) was noted between groups. Based on this small median difference in outcome, we estimate that 3285 subjects would be required in each arm to detect a significant difference.

There are few limitations to this study. We treated subjects for 42 days. A longer course of treatment might give different results. However, a plateau was reached between 7 and 42 days of treatment, suggesting that longer treatment would be unlikely to change the final outcome (Fig. 2). Many other authors using different hormonal treatments have observed significant reduction of pain scores, compared with baseline, in treatments that lasted less than 6 months [29–34]. Petta *et al*. [34] found that pain scores were significantly reduced after only 30 days of treatment with the intrauterine device containing levonorgestrel [34].

Pain scores are prone to recall bias. Although we used a standard sentence for every subject to recall the mean levels of pain in the preceding week, it is possible that subjects graded their pain score according to the highest pain that they experienced; the randomization, however, should have balanced this potential bias. The registration of use of pain medications (dipyrone, ibuprofen, and codeine) may also be prone to bias. However, the randomization and the similarity of the amount of pain medication used between groups make this unlikely. A higher mean use of dipyrone was observed in the resveratrol group, but it did not reach statistical significance. Although prior laparoscopy was required to confirm a history of endometriosis, we did not examine the stage of disease between groups, as suggested by the American Society of Reproductive Medicine [47]. It is well established, however, that stage of endometriosis does not correlate well with the experience of pain [48].

Our studied population is likely to have external validity; women with pelvic pain were recruited from social media and not from a single outpatient clinic.

In conclusion, daily use of 40 mg of resveratrol, combined with continuous use of a COC, was not superior to a COC alone for the treatment of pain in women with endometriosis.

Acknowledgments

Address all correspondence to: Ricardo Francalacci Savaris, MD, MSc, PhD, Serviço de Ginecologia e Obstetrícia, Hospital de Clínicas de Porto Alegre, Rua Ramiro Barcelos, 2350/1124, Porto Alegre, Rio Grande do Sul, Brazil, 90035-903. E-mail: rsavaris@hcpa.edu.br.

This work was supported by Fundação de Incentivo à Pesquisa e Ensino, Hospital de Clínicas de Porto Alegre, Grant 14-626, which provided financial support for medications used in this study and for laboratory analysis, and Coordenação de Aperfeiçoamento de Pessoal de Nível Superior Grant 99999.003035/2015-08 (to B.A.L.).

Clinical trial registry: ClinicalTrials.gov no. NCT02475564 (registered 11 June 2015).

Disclosure Summary: The authors have nothing to disclose.

References and Notes

1. Backonja U, Buck Louis GM, Lauver DR. Overall adiposity, adipose tissue distribution, and endometriosis: a systematic review. *Nurs Res*. 2016;**65**(2):151–166.
2. Janssen EB, Rijkers ACM, Hoppenbrouwers K, Meuleman C, D'Hooghe TM. Prevalence of endometriosis diagnosed by laparoscopy in adolescents with dysmenorrhea or chronic pelvic pain: a systematic review. *Hum Reprod Update*. 2013;**19**(5):570–582.
3. Burney RO, Talbi S, Hamilton AE, Vo KC, Nyegaard M, Nezhat CR, Lessey BA, Giudice LC. Gene expression analysis of endometrium reveals progesterone resistance and candidate susceptibility genes in women with endometriosis. *Endocrinology*. 2007;**148**(8):3814–3826.
4. Young SL, Lessey BA. Progesterone function in human endometrium: clinical perspectives. *Semin Reprod Med*. 2010;**28**(1):5–16.
5. Lessey BA, Palomino WA, Apparao KB, Young SL, Lininger RA. Estrogen receptor-alpha (ER-alpha) and defects in uterine receptivity in women. *Reprod Biol Endocrinol*. 2006;**4**(Suppl 1):S9.
6. Bilibio JP, Souza CAB, Rodini GP, Andreoli CG, Genro VK, de Conto E, Cunha-Filho JS. Serum prolactin and CA-125 levels as biomarkers of peritoneal endometriosis. *Gynecol Obstet Invest*. 2014; **78**(1):45–52.

7. Igarashi TM, Bruner-Tran KL, Yeaman GR, Lessey BA, Edwards DP, Eisenberg E, Osteen KG. Reduced expression of progesterone receptor-B in the endometrium of women with endometriosis and in cocultures of endometrial cells exposed to 2,3,7,8-tetrachlorodibenzo-p-dioxin. *Fertil Steril.* 2005;**84**(1):67–74.
8. Bulun SE, Cheng Y-H, Yin P, Imir G, Utsunomiya H, Attar E, Innes J, Julie Kim J. Progesterone resistance in endometriosis: link to failure to metabolize estradiol. *Mol Cell Endocrinol.* 2006;**248**(1-2):94–103.
9. Bulun SE, Yang S, Fang Z, Gurates B, Tamura M, Sebastian S. Estrogen production and metabolism in endometriosis. *Ann N Y Acad Sci.* 2002;**955**:75–85.
10. Fazleabas AT, Brudney A, Chai D, Langoi D, Bulun SE. Steroid receptor and aromatase expression in baboon endometriotic lesions. *Fertil Steril.* 2003;**80**(Suppl 2):820–827.
11. Bulun SE, Cheng Y-H, Pavone ME, Xue Q, Attar E, Trukhacheva E, Tokunaga H, Utsunomiya H, Yin P, Luo X, Lin Z, Imir G, Thung S, Su EJ, Kim JJ. Estrogen receptor-beta, estrogen receptor-alpha, and progesterone resistance in endometriosis. *Semin Reprod Med.* 2010;**28**(1):36–43.
12. Dunselman GAJ, Vermeulen N, Becker C, Calhaz-Jorge C, D'Hooghe T, De Bie B, Heikinheimo O, Horne AW, Kiesel L, Nap A, Prentice A, Saridogan E, Soriano D, Nelen W; European Society of Human Reproduction and Embryology. ESHRE guideline: management of women with endometriosis. *Hum Reprod.* 2014;**29**(3):400–412.
13. Singh M, Parent S, Leblanc V, Asselin E. Resveratrol modulates the expression of PTGS2 and cellular proliferation in the normal rat endometrium in an AKT-dependent manner. *Biol Reprod.* 2011;**84**(5): 1045–1052.
14. Almeida L, Vaz-da-Silva M, Falcão A, Soares E, Costa R, Loureiro AI, Fernandes-Lopes C, Rocha JF, Nunes T, Wright L, Soares-da-Silva P. Pharmacokinetic and safety profile of trans-resveratrol in a rising multiple-dose study in healthy volunteers. *Mol Nutr Food Res.* 2009;**53**(Suppl 1):S7–S15.
15. Bruner-Tran KL, Osteen KG, Taylor HS, Sokalska A, Haines K, Duleba AJ. Resveratrol inhibits development of experimental endometriosis in vivo and reduces endometrial stromal cell invasiveness in vitro. *Biol Reprod.* 2011;**84**(1):106–112.
16. Ricci AG, Olivares CN, Bilotas MA, Bastón JI, Singla JJ, Meresman GF, Barañao RI. Natural therapies assessment for the treatment of endometriosis. *Hum Reprod.* 2013;**28**(1):178–188.
17. Rudzitis-Auth J, Menger MD, Laschke MW. Resveratrol is a potent inhibitor of vascularization and cell proliferation in experimental endometriosis. *Hum Reprod.* 2013;**28**(5):1339–1347.
18. Saleh MC, Connell BJ, Saleh TM. Resveratrol induced neuroprotection is mediated via both estrogen receptor subtypes, ER(α) and ER(β). *Neurosci Lett.* 2013;**548**:217–221.
19. Gehm BD, McAndrews JM, Chien PY, Jameson JL. Resveratrol, a polyphenolic compound found in grapes and wine, is an agonist for the estrogen receptor. *Proc Natl Acad Sci USA.* 1997;**94**(25): 14138–14143.
20. Amaya SC, Savaris RF, Filipovic CJ, Wise JD, Hestermann E, Young SL, Lessey BA. Resveratrol and endometrium: a closer look at an active ingredient of red wine using in vivo and in vitro models. *Reprod Sci.* 2014;**21**(11):1362–1369.
21. Bowers JL, Tyulmenkov VV, Jernigan SC, Klinge CM. Resveratrol acts as a mixed agonist/antagonist for estrogen receptors alpha and beta. *Endocrinology.* 2000;**141**:3657–3667.
22. Bulun SE, Monsavais D, Pavone ME, Dyson M, Xue Q, Attar E, Tokunaga H, Su EJ. Role of estrogen receptor-β in endometriosis. *Semin Reprod Med.* 2012;**30**(1):39–45.
23. Maia H, Jr, Haddad C, Pinheiro N, Casoy J. Advantages of the association of resveratrol with oral contraceptives for management of endometriosis-related pain. *Int J Womens Health.* 2012;**4**:543–549.
24. Mol BW, Bayram N, Lijmer JG, Wiegerinck MA, Bongers MY, van der Veen F, Bossuyt PM. The performance of CA-125 measurement in the detection of endometriosis: a meta-analysis. *Fertil Steril.* 1998;**70**(6):1101–1108.
25. Chen FP, Soong YK, Lee N, Lo SK. The use of serum CA-125 as a marker for endometriosis in patients with dysmenorrhea for monitoring therapy and for recurrence of endometriosis. *Acta Obstet Gynecol Scand.* 1998;**77**(6):665–670.
26. WHO. *Medical Eligibility Criteria for Contraceptive Use.* 5th ed. Geneva, Switzerland: World Health Organization; 2015: 267.
27. Savaris RF, Teixeira LM, Torres TG, Edelweiss MIA, Moncada J, Schachter J. Comparing ceftriaxone plus azithromycin or doxycycline for pelvic inflammatory disease: a randomized controlled trial. *Obstet Gynecol.* 2007;**110**(1):53–60.
28. Bourdel N, Alves J, Pickering G, Ramilo I, Roman H, Canis M. Systematic review of endometriosis pain assessment: how to choose a scale? *Hum Reprod Update.* 2015;**21**(1):136–152.
29. Shokeir T, Mousa S. A randomized, placebo-controlled, double-blind study of hysteroscopic-guided pertubal diluted bupivacaine infusion for endometriosis-associated chronic pelvic pain. *Int J Gynaecol Obstet.* 2015;**130**(3):219–222.

30. Schwertner A, Conceição Dos Santos CC, Costa GD, Deitos A, de Souza A, de Souza IC, Torres IL, da Cunha Filho JS, Caumo W. Efficacy of melatonin in the treatment of endometriosis: a phase II, randomized, double-blind, placebo-controlled trial. *Pain*. 2013;**154**(6):874–881.
31. Strowitzki T, Marr J, Gerlinger C, Faustmann T, Seitz C. Detailed analysis of a randomized, multicenter, comparative trial of dienogest versus leuprolide acetate in endometriosis. *Int J Gynaecol Obstet*. 2012;**117**(3):228–233.
32. Petraglia F, Hornung D, Seitz C, Faustmann T, Gerlinger C, Luisi S, Lazzeri L, Strowitzki T. Reduced pelvic pain in women with endometriosis: efficacy of long-term dienogest treatment. *Arch Gynecol Obstet*. 2012;**285**(1):167–173.
33. Guzick DS, Huang L-S, Broadman BA, Nealon M, Hornstein MD. Randomized trial of leuprolide versus continuous oral contraceptives in the treatment of endometriosis-associated pelvic pain. *Fertil Steril*. 2011;**95**(5):1568–1573.
34. Petta CA, Ferriani RA, Abrao MS, Hassan D, Rosa E Silva JC, Podgaec S, Bahamondes L. Randomized clinical trial of a levonorgestrel-releasing intrauterine system and a depot GnRH analogue for the treatment of chronic pelvic pain in women with endometriosis. *Hum Reprod*. 2005;**20**(7):1993–1998.
35. Altman DG, Bland JM. Statistics notes: How to randomise. *BMJ*. 1999;**319**:703–704.
36. Little RJ, D'Agostino R, Cohen ML, Dickersin K, Emerson SS, Farrar JT, Frangakis C, Hogan JW, Molenberghs G, Murphy SA, Neaton JD, Rotnitzky A, Scharfstein D, Shih WJ, Siegel JP, Stern H. The prevention and treatment of missing data in clinical trials. *N Engl J Med*. 2012;**367**(14):1355–1360.
37. Flower A, Andrew F, Liu JP, George L, Paul L, Qing L. *Chinese herbal medicine for endometriosis*. *Cochrane Database Syst Rev*. 2012;**5**:CD006568.
38. Xu H, Lui WT, Chu CY, Ng PS, Wang CC, Rogers MS. Anti-angiogenic effects of green tea catechin on an experimental endometriosis mouse model. *Hum Reprod*. 2009;**24**(3):608–618.
39. Yavuz S, Aydin NE, Celik O, Yilmaz E, Ozerol E, Tanbek K. Resveratrol successfully treats experimental endometriosis through modulation of oxidative stress and lipid peroxidation. *J Cancer Res Ther*. 2014;**10**(2):324–329.
40. Ozcan Cenksoy P, Oktem M, Erdem O, Karakaya C, Cenksoy C, Erdem A, Guner H, Karabacak O. A potential novel treatment strategy: inhibition of angiogenesis and inflammation by resveratrol for regression of endometriosis in an experimental rat model. *Gynecol Endocrinol*. 2015;**31**(3):219–224.
41. Lu R, Serrero G. Resveratrol, a natural product derived from grape, exhibits antiestrogenic activity and inhibits the growth of human breast cancer cells. *J Cell Physiol*. 1999;**179**(3):297–304.
42. Boers M. Updated Consolidated Standards of Reporting Trials (CONSORT): it just gets better. *J Clin Epidemiol*. 2010;**63**(8):813–814.
43. Vlahos N, Vlachos A, Triantafyllidou O, Vitoratos N, Creatsas G. Continuous versus cyclic use of oral contraceptives after surgery for symptomatic endometriosis: a prospective cohort study. *Fertil Steril*. 2013;**100**(5):1337–1342.
44. Davis L, Kennedy SS, Moore J, Prentice A. Oral contraceptives for pain associated with endometriosis. *Cochrane Database Syst Rev*. 2007; (3):CD001019.
45. Raj MHG, Abd Elmageed ZY, Zhou J, Gaur RL, Nguyen L, Azam GA, Braley P, Rao PN, Fathi IM, Ouhtit A. Synergistic action of dietary phyto-antioxidants on survival and proliferation of ovarian cancer cells. *Gynecol Oncol*. 2008;**110**(3):432–438.
46. de Sá Rosa e Silva AC, Rosa e Silva JC, Nogueira AA, Petta CA, Abrão MS, Ferriani RA. The levonorgestrel-releasing intrauterine device reduces CA-125 serum levels in patients with endometriosis. *Fertil Steril*. 2006;**86**(3):742–744.
47. American Society for Reproductive. Revised American Society for Reproductive Medicine classification of endometriosis: 1996. *Fertil Steril*. 1997;**67**(5):817–821.
48. Vercellini P, Trespidi L, De Giorgi O, Cortesi I, Parazzini F, Crosignani PG. Endometriosis and pelvic pain: relation to disease stage and localization. *Fertil Steril*. 1996;**65**(2):299–304.