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Short-term effects of two continuous combined oestrogen–progestogen therapies on several cardiovascular risk markers in healthy postmenopausal women: A randomised controlled trial

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ABSTRACT

Objective: To compare the short-term effects of two oral continuous combined oestrogen–progestogen treatment regimens on blood concentrations of several cardiovascular risk markers in healthy postmenopausal women.

Study design: In a 12-week randomised controlled study, 48 healthy non-hysterectomised postmenopausal women, aged 41–58 years, received either no treatment (control group; $n = 16$), or daily oral continuous combined treatment with 1 mg micronised 17β-oestradiol plus 5 mg dydrogesterone (E/D group; $n = 18$) or 0.625 mg conjugated equine oestrogens plus 5 mg medroxyprogesterone acetate (CEE/MPA group; $n = 14$).

Fasting blood sampling was performed at baseline and after 12 weeks of follow-up.

Results: Compared with the control group, 12-week treatment with E/D or CEE/MPA reduced fibrinogen (−7.7%, $p = 0.004$ and −3.3%, $p = 0.083$, respectively), factor VII-act (−8.7%, $p = 0.14$ and −9.7%, $p = 0.06$, respectively), homocysteine (−20.5%, $p = 0.02$ and −26.7%, $p = 0.005$, respectively), and IGF-1 (−27.9%, $p < 0.001$ and −18.1%, $p = 0.002$, respectively), but increased factor VII-ag (+10.1%, $p = 0.03$ and +4.4%, $p = 0.46$, respectively), endothelin-1 (+15.2%, $p = 0.12$ and +20.0%, $p = 0.13$, respectively) and C-reactive protein (+88.8%, $p = 0.18$ and +71.0%, $p = 0.44$, respectively). Fibrinolytic factors were not affected by either hormone therapy (HT).

Conclusions: Short-term oral continuous combined therapy with oestradiol/dydrogesterone and conjugated equine oestrogens/medroxyprogesterone acetate had comparable effects on the investigated cardiovascular risk markers.

1. Introduction

Coronary heart disease (CHD) is a major cause of morbidity and mortality in postmenopausal women. The increase of CHD after menopause is related to the state of oestrogen loss. In observational studies treatment with postmenopausal hormone therapy (HT) showed a 50–70% decreased risk in coronary events in healthy postmenopausal women [1].

Contradictions exist between the results of these observational studies and those of large randomised controlled trials as the Heart and Estrogen/progestin Replacement Study (HERS) [2] and the Women's Health Initiative (WHI) trials [3,4]. These studies showed either no change or an early increase in CHD in healthy women [3,4] and in women with established CHD [2]. In these trials patients were treated with oral conjugated equine oestrogens plus medroxyprogesterone acetate (CEE/MPA), the most commonly prescribed HT in the United States. Pro-coagulatory and inflammatory influences of especially oral HT have been suggested to explain these effects [5,6], despite the observed beneficial changes in lipids and lipoproteins [7]. It is unclear whether these findings may be generalized to all forms of HT. In Europe oestradiol containing hormone combinations are prescribed more frequently. Oestradiol plus dydrogesterone is an oral HT with beneficial effects on various cardiovascular risk markers [8,9]. The dosage of MPA used in this study is higher than in the HERS and WHI study, because we treated younger early postmenopausal women.

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Clinical studies on the effect of HT on non-lipid cardiovascular risk markers might give important information about the mechanisms involved in the development of CHD. In an earlier study we tested the effect of conjugated equine estrogens plus medroxyprogesteroneacetate versus oestradiol plus dydrogesterone on various lipid cardiovascular risk markers [10].

The objective of this study was to compare in healthy postmenopausal women the effect on several non-lipid cardiovascular risk markers of oral 17β-oestradiol plus dydrogesterone with non-treatment and of conjugated equine oestrogens plus medroxyprogesterone acetate with non-treatment. The secondary objective is to compare both treatment groups with each other.

We evaluated the levels of coagulation factors: fibrinogen, factor VII activity (factor VII-act) and factor VII antigen (factor VII-ag); the fibrinolytic factors: tissue-type plasminogen activator (t-PA), plasminogen activator inhibitor type-1 (PAI-1), total fibrin degradation products (TDP) and plasmin-α2-antiplasmin (PAP) complex, and other cardiovascular risk markers: C-reactive protein (CRP), homocysteine and endothelin-1 (ET-1). Furthermore insulin-like growth factor-1 (IGF-1), associated with several cancers, was measured.

2. Materials and methods

This 12-week study was performed at the outpatient clinic of the Department of Obstetrics and Gynaecology of the Vrije Universiteit University Medical Center, Amsterdam. The study conformed to the principles outlined in the declaration of Helsinki and was approved by the Institutional Review Board. Informed consent was obtained from each participant before study entry.

This study is a single-center extension of the multicenter study published earlier [10]. The participants were recruited by advertisements in newspapers, they were postmenopausal, 41–58 years old, had an intact uterus, a blood pressure of 170/105 mmHg or lower, had a body mass index between 17 and 31 kg/m^2 and smoked no more than 20 cigarettes per day. They have at least 12 months amenorrhoea before screening, their follicle-stimulating hormone (FSH) concentration was within the postmenopausal range (>30 U/L) and they had not received any oestrogen or progestogen containing treatment in the 4 weeks proceeding randomisation. Exclusion criteria included a history of metabolic, endocrinological, cardiovascular or (pre-)malignant disease, alcohol or drug abuse, as well as clinically relevant abnormalities in laboratory tests of haematological, renal and hepatic function.

Subjects were screened within 4 weeks before randomisation. A medical history was obtained and a complete physical and pelvic examination, including a transvaginal ultrasound. Cervical cytology and a mammogram were performed.

After randomisation to the treatment or the non-treatment arm the women in the treatment arm were randomly assigned in a double-blind fashion to either oral continuous combined 1 mg micronised 17β-oestradiol plus 5 mg dydrogesterone (E/D group; Femoston® continu 1/5, Solvay Pharmaceuticals, Weesp, The Netherlands) or to oral continuous combined 0.625 mg conjugated equine oestrogens plus 5 mg medroxyprogesterone acetate (CEE/MPA group; Premique®, Wyeth Lederle/AHP, Berks, United Kingdom) given every day. The packaging of the CEE/MPA tablets into capsules to match the E/D capsules was carried out by the Department of Pharmaceutical Development at Solvay Pharmaceuticals (United Kingdom) in compliance with Good Manufacturing Practice.

At baseline and after 12 weeks, venous blood samples were taken in the morning after at least 10 h of fasting. In case of study discontinuation, participants were invited to come for a last visit to perform blood sampling. Blood was collected with a Vacutainer® system into plain tubes and tubes containing etylenediaminetetraacetic acid (EDTA).

The blood samples were centrifuged (30 min 3000 × g) and was frozen at 80 °C until analysis.

All samples for a given parameter were assayed in a single run at the end of the study. The laboratory was blinded to the treatment code. We measured the following plasma and serum levels:

- Fibrinogen, quantified by the Claus method [11].
- Factor VII-act by a chromogenic assay; Chromogenix AB, Mölndal, Sweden.
- Factor VII-ag by ELISA; Asserachrom VII: AG; Diagnostica Stago, Asnières-sur-Seine, France.
- t-PA by Imulyse (Biopool International, Umea, Sweden).
- PAI-1 by Innotest (Innogenetics, Gent, Belgium).
- PAP complex antigen by ELISA; Enzygnost PAP, Behringwerke AG, Marburg, Germany.
- TDP by ELISA; Thrombonostika TDP, Organon Teknika, Turnhout, Belgium.
- CRP by a homemade highly sensitive ELISA.
- Hcy defined as the sum of all homocysteine subfractions in plasma including free and protein-bond forms determined by high-performance liquid chromatography with fluorescence detection.
- ET-1 by a high-sensitive ELISA (Amersham, Little Chalfont, UK).
- IGF-1 by Immunoradiometrische assay (DSL, Webster, TX, USA).
- Serum FSH with a specific immunoassay (Bayer Immuno-1 microparticle enzyme immunoassay).

Statistical analysis was performed using the Statistical Package for the Social Sciences PC +10.0 (SPSS Inc., Chicago, IL). Values are given as mean ± standard deviation (S.D.) or as median and range in case of a skewed distribution. We compared baseline measurements from baseline at 12 weeks between groups using standard parametric tests. Analyses of covariance (ANCOVA) for repeated measurements, with the baseline value of the variable under consideration as a constant covariate, were used for comparisons between the groups. Because there was a statistically significant difference in baseline blood pressures between the groups, we also used the systolic and diastolic blood pressure as a constant covariate. We give p-levels and confidence intervals of the absolute between-group difference in change from baseline. A two-tailed $p < 0.05$ was accepted as the level of statistical significance. Because of a skewed distribution of Factor VII-ag, PAI-1, PAP, TDP and CRP these analyses were conducted on log-transformed data.

3. Results

After screening, 48 women were randomised (Fig. 1). Baseline descriptive characteristics of the three groups are shown in Table 1. At baseline there was no relevant difference among the groups except for the systolic and diastolic blood pressures (p = 0.001 and 0.036, respectively).

There were no dropouts. The measurements of PAI-1 at baseline and week 12 were missing in the control group for one patient. In the E/D group of one patient the baseline and week 12 measurement of t-PA was missing and of one patient in the CEE/MPA group the baseline and week 12 values of ET-1.

Table 2 shows the levels of the markers of coagulation (Fig. 2) and fibrinolysis (Fig. 3). It shows the levels at baseline and week 12, the percentage change from baseline and the ANCOVA (overall, between treatment and control group and between treatment groups). The ANCOVA indicated that there was a statistically significant between-group difference for fibrinogen after 12 weeks