

Umeå University Medical Dissertations  
New series No. 888 ISSN 0346-6612 ISBN 91-7305-635-9

---

From the Department of Clinical Sciences, Obstetrics & Gynecology,  
Umeå University, Umeå, Sweden

**HORMONE REPLACEMENT THERAPY: BENEFITS AND  
ADVERSE EFFECTS**

**Inga-Stina Ödmark**



**Umeå 2004**

**Cover picture:**

Daily rating scores of sweats by women starting conjugated estrogen/medroxyprogesterone acetate or 17 $\beta$ -estradiol/norethisterone acetate. The figure, in its whole, is presented in the article *Wellbeing at onset of hormone replacement therapy: comparison between two continuous combined regimens*, (Figure 1).

ISBN 91-7305-635-9

Copyright © Inga-Stina Ödmark

Printed in Sweden by Umeå University, Print & Media, 2004

Department of Clinical Sciences, Obstetrics & Gynecology,

Umeå University,

S-901 85 Umeå, Sweden

*To*

*the most important women in my life  
grandmothers **Hanna** and **Ester**  
former mother-in-law **Kerstin**  
my wonderful mother **Stina**  
and my daughters  
**Hanna** and **Karolina***

## Abstract

---

### HORMONE REPLACEMENT THERAPY: BENEFITS AND ADVERSE EFFECTS

Inga-Stina Ödmark

Background: Numerous studies have shown that estrogen replacement therapy (ERT) is an effective treatment for vasomotor symptoms, insomnia and vaginal dryness. Beneficial effects have also been shown on lipid patterns and on the incidence of osteoporotic fractures. As ERT increases the risk of endometrial adenocarcinoma, combinations with various progestogens have been developed in order to protect the endometrium. However, the addition of progestogens tends to reduce the beneficial effects of estrogens on mood, cognition and lipid metabolism. The added progestogen often causes side effects such as irritability and depression. There is evidence that the effect on wellbeing varies between women and with the type of progestogen used. Women who prefer to avoid withdrawal bleedings can be given continuous combined hormone replacement therapy (HRT). Unfortunately, irregular bleedings are common at the beginning of treatment and reduces compliance. Recently, several studies have reported an increased risk of breast cancer and venous thrombosis, and therefore long-term treatment with HRT for women without climacteric symptoms is no longer recommended. The ongoing debate has, for the time being, resulted in a recommendation that improving quality of life (QoL) by treatment of climacteric symptoms should be the only indication for prescribing HRT.

Aims and methods: The aims of the study were to investigate bleeding patterns, changes in wellbeing at onset and during long-term treatment, and lipid and lipoprotein profiles with two different types of continuous combined HRT. In addition, women starting, and women switching from mainly sequential HRT were compared. The design was a randomised, double-blind, one year, prospective, multicentre study including 249 healthy postmenopausal women who were given continuous daily oral treatment with either combined 0.625mg conjugated estrogen (CE) and 5mg medroxyprogesterone acetate (MPA) or combined 2mg 17 $\beta$ -estradiol (E2) and 1mg norethisterone acetate (NETA). Bleedings, if any, were recorded daily throughout the study. The main outcome measures (changes in wellbeing and climacteric symptoms) consisted of daily ratings of 12 items on a validated symptom scale. Serum concentrations of

lipids and lipoproteins were measured at baseline and after one year of treatment.

Results and conclusions: The majority of drop-outs were confined to the first three months, and the main reasons were bleedings and/or decreased wellbeing. Drop-outs were three times more common in the E2/NETA group. During the first month, 67% of the women reported irregular bleedings. The number of bleeding days decreased on both treatments during the first four months. Treatment with CE/MPA resulted in less irregular bleedings and a shorter time to amenorrhoea compared to E2/NETA.

As expected, "starters" experienced more sweats than "switchers" at the onset of treatment, but both groups improved significantly. Side effects such as breast tenderness, swelling, depression and irritability appeared during the first treatment week in both groups. The side effects of HRT appeared much more quickly than the benefits and were more frequent in women with a history of premenstrual syndrome (PMS). Breast tenderness was more common in the E2/NETA group throughout the whole study period. Apart from that, there were no differences between the two treatment regimens as regards effects on well-being at the end of the study.

Lipoprotein(a) levels, an important risk factor for cardiovascular disease, decreased in both treatment groups. Triglyceride levels increased in women treated with CE/MPA, and levels of total cholesterol, high density lipoprotein and low density lipoprotein fell in the E2/NETA group.

In conclusion, treatment with E2/NETA caused more bleeding problems than treatment with CE/MPA. CE/MPA was better tolerated than E2/NETA at the beginning of the study, but among the women remaining in the study there was no difference in QoL between the two treatment groups. HRT counselling should take into account that a history of PMS increases the likelihood of side effects and that these may precede any beneficial effects. Both treatments produced beneficial effects on lipid and lipoprotein levels, and neither of the regimens was superior in this respect.

Key words: hormone replacement therapy (HRT), bleeding pattern, progestogen, wellbeing, side effects, lipoprotein(a)

## CONTENTS

---

ABSTRACT .....	4
CONTENTS .....	6
ORIGINAL PAPERS .....	8
LIST OF ABBREVIATIONS .....	9
INTRODUCTION .....	10
Context and background .....	10
Pre-study expectations .....	13
Hot flushes and HRT .....	14
HRT and the endometrium .....	15
Wellbeing and HRT .....	16
Compliance with HRT .....	18
Premenstrual syndrome and HRT .....	19
Cognition and depression .....	20
HRT and cardiovascular disease .....	20
Estrogens .....	22
Progestogens .....	23
Alternative therapy .....	24
Present study .....	25
AIMS .....	26
SUBJECTS AND METHOD .....	27
SUBJECTS .....	27
<i>Ethical considerations</i> .....	29
<i>Multicentre study sites</i> .....	29
<i>Demographic data</i> .....	30
METHOD .....	31
<i>Bleeding diary and medication</i> .....	32
<i>Daily symptom ratings</i> .....	32
<i>The diagnosis of premenstrual syndrome</i> .....	33
<i>Lipid and lipoprotein assays</i> .....	33
STATISTICS .....	34

RESULTS	35
Baseline characteristics	35
Serious adverse events	35
Discontinuation	35
Bleeding patterns	37
Wellbeing at onset of HRT	39
Wellbeing during long-term treatment with HRT	42
Lipids and lipoproteins	45
DISCUSSION	47
Methodological strengths and weaknesses	47
Reasons for dropout	49
Effects on bleeding patterns	50
Measuring wellbeing	52
Effects on wellbeing	54
Side effects	56
HRT and cardiovascular risk	58
Conclusions and some thoughts for the future	60
SUMMARY AND GENERAL CONCLUSIONS	62
ACKNOWLEDGEMENTS	63
REFERENCES	66
APPENDIX 1	85
APPENDIX 2	86
APPENDIX 3	90

## ORIGINAL PAPERS

---

This thesis is based on the following papers, which will be referred to in the text by their Roman numerals.

- I. Ödmark I-S, Jonsson B, Bäckström T. Bleeding patterns in postmenopausal women using continuous combined hormone replacement therapy with conjugated estrogen and medroxyprogesterone acetate or 17 $\beta$ -estradiol and norethindrone acetate.  
Am J Obstet Gynecol 2001;184: 1131-8.
- II. Ödmark I-S, Bäckström T, Jonsson B, Bixo M. Wellbeing at onset of hormone replacement therapy: comparison between two continuous combined regimens.  
Climacteric, *in press*.
- III. Ödmark I-S, Bäckström T, Jonsson B, Bixo M. Long-term effects of two different continuous combined regimens of hormone replacement therapy on wellbeing.  
Gynecological Endocrinology, *in press*.
- IV. Ödmark I-S, Bäckström T, Haeger M, Jonsson B, Bixo M. Effects of continuous combined conjugated estrogen/medroxyprogesterone acetate and 17 $\beta$ -estradiol/norethisterone acetate on lipids and lipoproteins.  
Maturitas, *in press*.

These papers are reproduced with the permission of the copyright holders.



## LIST OF ABBREVIATIONS

---

AD	Alzheimer's dementia
BMI	body mass index
BMD	bone mineral density
CD	cyclicity diagnoser
CE	conjugated estrogen
CGRP	calcitonin gene-related peptide
CI	confidence interval
CVD	cardiovascular disease
E1	estrone
E2	estradiol
E3	estriol
ERT	estrogen replacement therapy
GnRH	gonadotropin releasing hormone
HDL	high-density lipoprotein
HERS	the Heart and Estrogen/progestin Replacement Study
HRT	hormone replacement therapy
ITT	intention to treat
LDL	low density lipoprotein
Lp(a)	lipoprotein(a)
MPA	medroxyprogesterone acetate
MWS	the Million Women Study
NETA	norethisterone acetate
OC	oral contraceptive
OR	odds ratio
PEPI	the Postmenopausal Estrogen/Progestin Interventions trial
PMDD	premenstrual dysphoric disorder
PMS	premenstrual syndrome
QoL	quality of life
RR	relative risk
SEM	standard error of the mean
SERM	selective estrogen receptor modulator
TC	total cholesterol
TG	triglyceride
VAS	visual analogue scale
VTE	venous thromboembolism
WHI	the Women's Health Initiative trial
WHIMS	the Women's Health Initiative Memory Study
WISDOM	the Women's International Study of long Duration Oestrogen after Menopause

## INTRODUCTION

---

### Context and background

The discontinuance of menstruation indicates a loss of ovarian follicular function and is called the menopause. Most women live long enough to make the transition from fertile to postmenopausal life. In the Western World, menopause occurs at 51.3 years of age (McKinley et al 1992), and women are postmenopausal for a third of their lives. It is expected that by the year 2025, more than one in five women will be elderly. This is a worldwide development, not limited to affluent societies (Diczfalusy 1986).

The significance of the climacteric syndrome has been debated for decades. The menopause has been defined either as a hormone deficiency (Utian 1987, Wiklund et al 1993) or as a normal physiological phase in a woman's life (Dennerstein 1996). Both arguments are difficult to accept if carried to their extremes. The first definition may lead to a medicalisation of otherwise healthy, middle-aged women (Bell 1990), whereas the second may withhold effective treatment from women suffering severe climacteric symptoms. Population studies show that hot flushes and climacteric symptoms may differ between cultures. For example, Caucasian women seem to have more vasomotor symptoms than Asian women (Ho et al 1999, Fuh et al 2001), whereas in the USA, African-American women are more likely to report symptoms than Caucasian women (Avis et al 2001). Authors have tried to attribute such differences in menopausal symptom patterns to a variety of socio-cultural factors (George 1988, Lock 1991). Asian food with its high fibre content, green tea and, in particular, soy products rich in phytoestrogens might also have some significance in reducing climacteric complaints (Knight & Eden 1996).

Three out of four women experience climacteric symptoms. Vasomotor symptoms associated with the menopause have a significant impact on women's lives and on their ability to function. Women have reported a great variety of symptoms associated with the menopause, e.g. depression, insomnia, fatigue, swelling, headache, sickness/nausea, lack of enjoyment, irritability, anxiety, palpitations, loss of libido, and poor memory. Attempts have been made to identify the symptoms attributable to the menopausal state (McKinley et al 1992, Holte 1992, Oldenhave et al 1993). Today, only hot flushes, night sweats, and vaginal atrophy are

defined as menopausal symptoms and these symptoms are effectively alleviated by hormone replacement therapy (HRT).

In fact, hot flushes are one of the chief menopausal complaints that lead women in western societies to seek medical treatment (Kronenberg 1990). In addition, there is substantial evidence that HRT increases bone mineral density (BMD) and significantly reduces the risk of fractures (Torgerson & Bell-Syer 2001, Rossouw et al 2002). Also, estrogen treatment improves neuromuscular function and balance, reducing the risk of injury (Hammar et al 1996, Naessen et al 1997). However, HRT is no longer recommended as first line treatment for osteoporosis in European countries since an increased risk of breast cancer and cardiovascular disease has been reported recently (Rossouw et al 2002, Banks et al 2003). The nature of these risks is described at the end of this chapter. Other health benefits of estrogen therapy include relief from urogenital symptoms such as vaginal dryness, dyspareunia, dysuria, urgency and recurrent urinary tract infections, (Cardozo et al 1998) and reduced risk of colon cancer (Rossouw et al 2002).

Postmenopausal women using HRT report improved quality of life, e.g. better emotional wellbeing, compared to non-users (Wiklund et al 1993). Studies suggest that there are certain predisposing factors for women starting HRT. Such factors include level of education, type of employment, earlier use of hormones, lifestyle, smoking, alcohol use, somatic health, attitude to the climacteric, and type of personality (Hunter 1990, Shelley et al 1995). A Finnish study has reported that women are more willing to start HRT if they are highly educated and live in large cities (Topo et al 1995). HRT users tend to be more physically active, smoke less, have a higher social and educational level, be leaner, and have more frequent medical examinations than non-users (Derby et al 1995, Matthews et al 1996). However, another study has shown that women who consulted their doctor because of climacteric symptoms had more negative life experiences, worse general health and higher rate of unemployment (Morse et al 1994). A Swedish study reported that women working in a stressful environment, performing demanding mental tasks and suffering from hot flushes were predisposed to starting HRT (Collins & Landgren 1997).

Thus, women using HRT are healthier and more interested in maintaining their general health than non-users. For many years there were great hopes that HRT could prevent cardiovascular disease (CVD) in women but recent studies have been a disappointment, see below/chapter HRT and cardiovascular disease. Although there is substantial evidence that HRT increases the risk of breast cancer, the debate over the last two

years has been centred upon whether short term treatment is risky and whether treatment increases the severity of the disease. The Million Women Study (MWS) a huge unselected population study of 1 084 000 British women aged 50-64 years, estimated that 32 new cases of breast cancer would occur in 1 000 women aged 50-65 years not using HRT, that five years of estrogen therapy would result in two further cases, and ten years would add another five cases. Estrogen given in combination with progesterone for five or ten years would result in six and 19 new cases of breast cancer respectively (Banks et al 2003). The first report from the Women's Health Initiative Trial (WHI) indicated increased risk after only four to five years of treatment. However, subgroup analyses have revealed that this significant increase in risk within 5 years was largely confined to women that had used HRT prior to study inclusion (Chlebowski et al 2003). A logical conclusion might be that long term use increases breast cancer risk, and treatment over a limited number of years could be considered safe in this respect.

It has been generally held that mortality in women treated with HRT is not raised because breast cancers in this group appear less aggressive (Delgado & Lopez 2001). However, contradictory results turned up in the WHI study which found that breast cancer in treated women appeared to be at a more advanced stage, and yet this was not reflected in increased mortality (Chlebowski et al 2003).

Several earlier published epidemiological studies, the Million Women Study and the WHI trial have indeed confirmed the association between HRT and breast cancer. In addition, the MWS has provided more detailed information about factors influencing this risk. At recommended doses, the CE and E2 compounds used in HRT increase the risk of breast cancer to the same degree. The risk increases with duration of use and returns to baseline level within a few (at most five) years after cessation of treatment. The relative risk is greater when the estrogen has been combined with a progestogen, given either sequentially or continuously, regardless of type. There is no evidence of a difference in risk between different routes of administration (Banks et al 2003).

Combined estrogen-progestogen treatment causes an increased density of the mammographic image, reducing the sensitivity and/or specificity of mammography, possibly resulting in delayed diagnosis and unnecessary investigations (Chlebowski et al 2003, Lundström et al 2002). The addition of progestogen to HRT and its effect on breast cancer is one of the most important questions regarding the risks of HRT.

HRT increases the risk of venous thromboembolism (VTE). Use after the menopause seems to increase the risk of VTE by 2 to 3 times, particularly

during the first year of treatment. This is based on a large randomised controlled study The Heart and Estrogen/progestin Replacement Study (HERS) which included 2 763 postmenopausal women treated with continuous combined HRT or placebo and the relative risk (RR) was 2.7 (CI 95% 1.4-5.0) for the treated women (Grady et al 2000). The WHI reported a similar (RR) of 2.13 (CI 95% 1.39-3.25) for pulmonary embolism (Rossouw et al 2002). In fact, the risk of VTE associated with HRT is about the same as the risk associated with use of oral contraceptives (OC).

The prescribing of estrogen and estrogen/progestogen combinations in Sweden reached its zenith towards the end of the 1990's. A dramatic change in attitudes has been seen since the publication of the large randomised trials WHI, and MWS. The prescribing of these hormones measured as defined daily doses (DDD) for the categories G03C (estrogens) and G03F (estrogen-progestogen combinations) decreased by 11% and 20% during the years 2002 and 2003 respectively, (information from Apoteksbolaget, Sweden 2004).

HRT is still the most effective treatment for climacteric symptoms. More recently, low-dose and limited duration treatment has been recommended. HRT should still be prescribed in situations where the benefits clearly outweigh the risks.

## Pre-study expectations

The following issues were discussed before the start of the study. Why do some women experience mood changes on HRT whilst others do not? Women reported spontaneously that they experienced negative mood changes on 17 $\beta$ -estradiol in combination with norethisterone acetate (E2/NETA), and when they were given conjugated estrogen in combination with medroxyprogesterone acetate (CE/MPA) instead, they reported an improvement in these symptoms. Clinical experience and the spontaneous comments of women prompted a desire to discover if differences between the treatments really did exist. Prescribers had differing opinions and experiences as regards the prevalence of irregular vaginal bleeding and amenorrhoea with the two different combinations. Separate studies of the two treatment options had been done, but they had never been compared in a randomised and controlled fashion as far as we knew. An evaluation of any differences in wellbeing and quality of life (QoL) was thought to be particularly desirable, in the light of feedback received from women on HRT, and in response to a general desire among

proponents of women's health for better research into the issues of wellbeing and QoL on HRT.

## Hot flushes and HRT

Estrogen concentration declines gradually during the climacteric, and women with low amounts of circulating estrogen are more likely to have symptoms. A rapid fall of estrogen levels has been thought to be the most significant triggering event for hot flushes (Sturdee & Brincat 1988). This could not be confirmed in a study of postmenopausal women without previous or current vasomotor symptoms who abruptly abandoned estrogen replacement therapy (Hammar et al 1999). Women with a surgically induced menopause often have more severe symptoms than women undergoing a natural menopause (Dennerstein et al 1978, Sherwin & Gelfand 1985). In an epidemiological study involving 5 213 women, 85% of perimenopausal women (6 months before to 12 months after the menopause) experienced vasomotor symptoms (Oldenhave et al 1993). 50-60% of postmenopausal women have reported vasomotor symptom (Oldenhave et al 1993, Lindgren et al 1993). Moreover, 15-20% of women still have symptoms more than 15 years after the menopause (Lindgren et al 1993). Estrogen replacement therapy (ERT) reduces or abolishes vasomotor symptoms associated with the climacteric (Campbell & Whitehead 1977, Kronenberg 1990). In large randomised clinical trials, HRT reduced hot flushes by 50-100% in postmenopausal women (McNaghy 1999, Mac Lennan et al 2001).

The expression "hot flushes" is commonly used synonymously with vasomotor, menopausal or climacteric symptoms. Hot flushes could be described as a subjective sensation of heat that is associated with objective signs of cutaneous vasodilatation and a subsequent drop in core temperature. The exact mechanism behind the changes in hypothalamic thermoregulation during the menopause is not known. Nevertheless, an increase in body surface temperature precedes most hot flushes (Freeman 1998). Calcitonin gene-related peptide (CGRP), which is a potent vasodilator, has been suggested as the mediator of hot flushes. It has been shown that women who have vasomotor symptoms have significantly higher total excretion of CGRP in urine compared to women who do not suffer from these symptoms (Wyon et al 1998). The average hot flush lasts about 4 minutes but its duration may range from a few seconds to as long as ten minutes. Hot flushes may be accompanied by sweats, flushing, palpitations, anxiety, irritability and even panic (Kronenberg 1994). Night

sweats are common. Some women have hot flushes a few times a week, whilst others have them hourly. Hot flushes are more common in women who do little or no exercise, women who smoke, (Gold et al 2000), women with low estradiol or estrone concentrations, and those of low bodyweight (Erlik et al 1982). There are contradicting results concerning the effect of body mass index (BMI) on hot flushes. High and low BMI may cause hot flushes (Gold et al 2000, Erlik et al 1982). It is important to recognize that hot flushes can also be related to systemic diseases, neurological disorders, alcohol and drug abuse, eating habits and food additives (Mohyi et al 1997). Moreover, the following symptoms have also been associated with the menopause: depression, nervousness, agitation and inability to concentrate (Pearce et al 1995, Montgomery & Studd 1991). It is unclear whether these symptoms trigger hot flushes, make them worse or are actually caused by the flushes themselves.

## HRT and the endometrium

One disadvantage of sequential hormone replacement therapy is that women continue to menstruate. Continuous treatment with combined estrogen and progestogen was developed in order to avoid this. Even so, there are usually some initial spottings, and some women experience troublesome irregular bleedings which makes this therapy unsuitable for them (Staland 1985, Archer et al 1994). The exact cause of irregular bleedings during continuous HRT remains unknown. Some of the theories will be discussed further on in this thesis.

High endometrial safety is important during hormone replacement therapy. There is substantial evidence that the use of unopposed estrogen is associated with an increased incidence of endometrial carcinoma (Ziel 1982, Voigt et al 1991). Estrogens stimulate proliferation of the endometrium during ERT, and uncontrolled proliferation may lead to hyperplasia and endometrial carcinoma (Deligdisch & Holinka 1987). Nowadays, combined regimens with the addition of a progestogen to antagonise the effects of estrogen and prevent the development of endometrial hyperplasia (Persson et al 1989, Woodruff & Pickar 1994, Whitehead et al 1982) is standard treatment for women with a preserved uterus. Various regimens with sequential addition of various doses of progestogen for various lengths of time have been tried. It has been shown that progestogens added cyclically reduce the relative risk of endometrial cancer in a duration-dependent manner (Weiderpass et al 1999). Furthermore, continuous combined treatment with estrogen and

progestogen actually reduces the incidence of endometrial adenocarcinoma compared to untreated women (Weiderpass et al 1999, Hill et al 2000).

## Wellbeing and HRT

One third of a woman's life will be spent in the postmenopausal state, and quality of life during these years is an important issue. QoL is a useful and appropriate way of evaluating the way women feel and function, but it has many interpretations and definitions. Walker and Rosser, offered the following definition: 'A concept encompassing a broad range of physical and psychological characteristics and limitations which describe an individual's ability to function and to derive satisfaction from doing so' (Walker & Rosser 1988). Improvement of QoL is a primary purpose of health promotion. In recent years, assessment of QoL (looking at a number of physical, psychological and social parameters) has been increasingly used as a measure of HRT outcome. A 12-week randomised placebo-controlled study of transdermal estradiol treatment reported an improvement in QoL as regards wellbeing, sleep disturbances, other somatic symptoms, cognitive function and sexual functioning. (Karlberg et al 1995). The effects of estrogen on various parts of the body, including the central nervous system, are complex. Hot flushes are probably the result of an altered thermoregulation in the brain. Estrogens may also be involved in mechanisms regulating sleep, mood and cognition (Stahl 2001). Menopausal symptoms are often associated with insomnia (Shaver & Zenk 2000) and hot flushes are strongly associated with altered sleeping patterns (Shaver et al 1988). However, sleep disturbances could also be associated with changes in estrogen concentrations *per se*. Estrogen may also lengthen the sleeping phase, facilitate sleep onset and generally improve sleep in women without hot flushes (Leibenluft 1993).

Several studies have reported an improvement in sleeping patterns after estrogen substitution (Campbell & Whitehead 1977, Padwick et al 1986). In general, QoL, wellbeing and depressed mood improve when women with menopausal symptoms are treated with estrogen (Derman et al 1995, Rebar et al 2000, Schmidt et al 2000).

Measuring wellbeing and QoL in postmenopausal women has proved to be a difficult task. A number of validated psychological measuring instruments have been tried. Other problems such as study design and differences between populations have led to inconsistent findings (Adler 2002). It has been argued that subjective outcomes, such as QoL, are



generally open to substantial bias and are often measured with less quantitative rigour. The measurement and evaluation of wellbeing requires a scale with a wide range of response options that is sensitive to small treatment-induced changes over time (Guyatt et al 1986).

The Heart and Estrogen/progestin Replacement Study compared the effects of estrogen plus progestogen with placebo on health-related quality of life and concluded that the estrogen effect was dependent on the presence or absence of menopausal symptoms at entry into the study (Hlatky et al 2002). However, this was not confirmed in a large randomised placebo controlled study published recently, (WHI), using CE 0,625mg/MPA 2,5mg, and including 16 608 postmenopausal women aged 50-79 years (Hays et al 2003). Combined estrogen/progestogen treatment gave only a small beneficial effect on sleep disturbances and no improvement in any other quality of life items (Hays et al 2003). Furthermore, the results of another study, the Postmenopausal Estrogen/Progestin Interventions Trial (PEPI), in which women were randomly assigned to treatment with placebo, unopposed estrogen, estrogen with cyclical progestogen or continuous combined estrogen/progestogen showed no significant differences in measures of psychosocial and cognitive functions (Greendale et al 1998). However, negative side effects occur with all types of progestogens in combination with estrogen (Sherwin 1991, Magos et al 1986, Björn et al 2000, Björn et al, 2002).

Progestogen only treatment does not seem to produce side effects when estrogen levels are low. A placebo-controlled, double-blind crossover study during two consecutive months compared 10mg MPA and placebo in 11 postmenopausal women. No differences between placebo and MPA were seen in progestogen side-effects or vasomotor symptom scores (Prior et al 1994). As MPA given alone does not seem to cause symptoms, it seems that it is the combination of estrogen and progestogen that lies at the root of the side effects seen when using combined regimens. Studies have also shown that vaginal progesterone suppositories in combination with oral estradiol caused depression, irritability, tension, fatigue and physical symptoms such as breast tenderness and swelling. These side effects are inversely related to dose, as a lower dose of vaginal progesterone (400mg/day) produced more negative effects than a higher dose (800mg/day) (Andréen et al 2003). This was also supported by a study by Björn et al (2002) where combinations with MPA 10mg produced more negative symptoms than those with 20mg. Estrogen dose dependent effects has also been reported. 3mg estradiol tend to produce more negative side effects than 2mg in combination with

MPA (Björn et al 2003). However, a one year study where 1.25mg CE and placebo was compared to 0.625mg CE and 5mg MPA given sequentially showed more negative mood and physical symptoms with the latter, indicating that high doses of estrogen could be tolerated as long as progestogens are not added (Sherwin 1991). There are also women who are not sensitive to progestogen at all and don't experience side effects with HRT.

## Compliance with HRT

HRT may improve QoL but fear of side effects has led many women to refrain from, or discontinue treatment prematurely. In one study in which women were given HRT because of low bone mass, as many as 39% discontinued treatment within 8 months despite the increased fracture risk (Ryan et al 1992). Improved compliance with HRT requires better information at the outset and continuous monitoring and adjustment of the treatment. This has been stressed in several review articles (Corson 1995, Dören & Schneider 1996, Mattsson 1996). It has been shown that compliance with HRT can be improved if the physician explains the purpose, risks and side effects of the treatment (Nachtigall 1990). In a longitudinal Swedish study, meticulous efforts to meet the women's consultation need resulted in high compliance. The main reasons given for discontinuing HRT were negative side effects (35%), desire to find out if climacteric symptoms had ended (26%), fear of cancer and thrombosis (25%) and disenchantment with bleedings (19%) (Björn & Bäckström 1999). Obviously, side effects are a common reason for discontinuing treatment.

The most common side effects are breast tenderness, headache, nausea and negative mood e.g. irritability, depression, anxiety, and fatigue. Other reported side effects include oedema, abdominal bloating, leg cramps, dysmenorrhoea, reduced libido and greasy skin with acne (Dennerstein et al 1979, Holst et al 1989, Bäckström 1995). Some of these symptoms have been shown to be related to progestogen type and dose (Magos et al 1986, Björn et al 2002). During sequential treatment women reported positive effects of estrogen on mood during the first part of the cycle, but negative effects of progestogen on mood during the rest of the cycle (Hammarbäck et al 1985). This pattern has many similarities with the premenstrual syndrome (PMS), and women who experience PMS during their fertile years have more of these side effects on HRT (Björn et al 2000).

## Premenstrual syndrome and HRT

There are many similarities between progestogen and progesterone, and their negative effects on mood might be mediated by the same mechanisms in the brain. Neurosteroids are metabolites of progesterone, and they have profound effects on various aspects of brain function including mood and anxiety (Sundström et al 1999). It is conceivable that synthetic progestogens follow the same metabolic pathways as endogenous progesterone and consequently produce neurosteroid-like substances.

The terms *premenstrual syndrome* (PMS) and *premenstrual dysphoric disorder* (PMDD) are sometimes used synonymously (Sundström et al 1997). However, PMS has been characterised as a milder condition with more physical symptoms and moderate psychological symptoms whereas PMDD is, according to Diagnostic and Statistical Manual of Mental disorders 4<sup>th</sup> edition (DSM-IV) (American Psychiatric Association 1994), defined as irritability, depression, tension or mood changes with negative effects on daily life (Steiner 1996). It is typical of both these conditions that symptoms appear after ovulation and gradually worsen until the start of menstruation (Bäckström et al 1983). Women with moderate and severe PMS/PMDD have reported psychological side effects when using oral contraception (Cullberg 1972). Furthermore, this phenomenon has been observed in postmenopausal women with a history of PMS who are given HRT. Unopposed estrogen is always associated with improved wellbeing (Hammarbäck et al 1985, Bäckström et al 1983) whereas combinations with progestogen tend to provoke negative mood (Hammarbäck et al 1985, Björn et al 2000). Women with prior PMS might therefore recognise these cyclical symptoms when starting sequential HRT (Björn et al 2000). The doses of both progestogen and estrogen seem to be of importance, as high doses of estrogens provoke more severe symptoms than lower doses in combined treatment (Björn et al 2002, Björn et al 2003). In addition, Klaiber et al (1997) reported that women with high plasma concentrations of 17 $\beta$ -estradiol reported a worsening in symptoms when progesterone was added. Furthermore, in a study of 30 PMS women, symptoms were more severe during cycles with high endogenous levels of estradiol during the luteal phase (Seippel & Bäckström 1998).

## Cognition and depression

A number of descriptive studies have reported that estrogen improves cognitive function in healthy women with low levels of estrogens (Matthews et al 1999). Another study of young women treated with gonadotropin releasing hormone (GnRH) and with impairment of verbal memory showed an improvement after estrogen treatment (Sherwin & Tulandi 1996). However, a recently published large controlled randomised study has reported no improvement in cognitive function with HRT. A total of 4 381 women aged 65 years or older in the Women's Health Initiative Memory Study (WHIMS), a randomized, double blind, placebo controlled clinical study, were tested with the Modified Mini-Mental State Examination after approximately four years of treatment with continuous combined HRT or placebo (Rapp et al 2003). Data from the estrogen only treatment group is still not available.

A meta-analysis of studies into the possible protective effects of estrogen therapy against Alzheimer's dementia (AD) concludes that it may have a part to play (Yaffe et al 1998). However, the epidemiological studies included in the analysis have significant methodological deficiencies, making it impossible to draw safe conclusions. Estrogen treatment of women with mild-moderate AD does not seem to improve cognitive performance, and a recently published randomised trial (WHIMS) reported increased risk of dementia with continuous combined HRT in women aged 65 years or older after approximately four years of treatment (Shumaker et al 2003).

Although estrogen treatment could improve mood in postmenopausal women, there is little evidence that estrogen could prove useful as a treatment for depression. Only one study of perimenopausal women has shown estrogen treatment to have a positive effect on clinical depression (Soares et al 2001), otherwise results have been negative.

## HRT and cardiovascular disease

Heart disease is the leading cause of death in women over 60 years of age. The risk factors are essentially the same in men and women and they include hypertension, smoking, diabetes mellitus and obesity. The risk of developing CVD increases with age, and in women this process coincides with the decline in estrogen levels. The fact that women develop coronary heart disease 10-15 years later than men suggests that estrogens protect against arteriosclerosis. It is generally accepted that lipids and lipoproteins

play an important role in the development of coronary atherosclerosis, which is the main cause of myocardial infarction. Lipoproteins influence the reuptake of cholesterol from the systemic circulation, and high levels have been associated with increased risk of coronary heart disease (Godsland 2001). Estrogen and combined estrogen/progestogen therapy in postmenopausal women lower plasma lipoprotein(a) (Lp(a)) and may, as a consequence, exert a beneficial effect on the cardiovascular system (Sacks et al 1994, Soma et al 1991).

The menopause is associated with a more atherogenic lipid profile, characterised by an increase in total cholesterol (TC) and low density lipoprotein (LDL) cholesterol with a concomitant reduction in high density lipoprotein (HDL) and cholesterol (Stevenson et al 1993). Triglyceride (TG) levels may increase after the menopause, although these data are less consistent. There are insufficient data about the relationship between the menopause and levels of Lp(a). High blood pressure is a well-known risk factor for CVD. However, estrogen alone, or in combination with progestogen has not been found to have any adverse effect on blood pressure (The Writing Group for the PEPI Trial 1995, Lip et al 1994).

Apart from the effects of estrogen on serum lipids and lipoproteins, there are some indications that it may produce beneficial effects by acting directly on the arterial wall (Diaz et al 1997, Collins et al 1994). In addition, studies have assessed estrogenic effects on the vessel walls and found a decrease in flow resistance and a subsequent increase in blood flow velocity in postmenopausal women (Bourne et al 1990, Ganger et al 1991). It is conceivable that other mechanisms influenced by HRT are equally important. For example, the thrombophilic effect of HRT might contribute to the cardiovascular risks.

To a great extent, the cardioprotection theory has been based on results of studies showing beneficial effects on lipid profiles. Countless studies have documented changes in blood lipid patterns during HRT. Results regarding effects of HRT on lipids are somewhat inconsistent as different regimens tend to produce different effects. A study by Munk-Jensen et al (1994) showed some decrease of HDL with 17 $\beta$ -estradiol/norethisterone acetate. Another study showed almost no counteracting effects of medroxyprogesterone acetate in combination with conjugated estrogen on serum lipids (Lobo et al 1994).

Epidemiological studies have reported a reduction in CVD in women using HRT compared to non users (Grodstein et al 2000). However, several large-scale randomised clinical trials, have recently demonstrated that HRT does not in fact protect women from CVD when the treatment is introduced after the age of 60 years. The HERS study was the first

randomised clinical trial of combined hormone therapy in secondary prevention of coronary events (Hulley et al 1998). It showed an unexpected increase in risk of myocardial infarction during the first year, followed by a decrease during the fourth and fifth year. An extension of this 5 year study, by a further 2.7 years (HERS II) yielded no further (positive or negative) effects (Grady et al 2002). The data on time trend are consistent with results from a large prospective cohort study where the risk of coronary events seemed to increase in short-term hormone users with previous coronary disease but to decrease with longer-term HRT use (20 years of follow up) (Grodstein et al 2001). To evaluate the effect of HRT on CVD in healthy women, two large trials were started during the nineties. In the Women's Health Initiative, 16 608 women were randomised to either continuous combined HRT or to placebo, and the main outcome was coronary events. The study was terminated early due to excess risk of breast cancer, thrombosis, stroke and cardiovascular disease. The relative risks were estimated at 1.29 for coronary heart disease (95% CI 1.02-1.63) and 1.41 for stroke (95% CI 1.07-1.85) after 5.2 years (Rossouw et al 2002). The second large trial, the Women's International Study of Long Duration Oestrogen after Menopause (WISDOM) was planned to include 22 300 postmenopausal women in the United Kingdom, Australia and New Zealand who were to be treated long-term with ERT, HRT or placebo (Vickers et al 2002). However, this study was terminated prematurely because of the negative results of the WHI. The previously assumed positive effects of HRT on CVD have been brought into question, resulting in more restrictive attitudes to the use of HRT.

## Estrogens

Estrogens are produced in several tissues including the ovary and adrenal gland. In postmenopausal women, most is derived from adipose tissue and the adrenals, whereas in fertile women, the ovaries are the largest source.  $17\beta$ -estradiol (E2), synthesised via estrone (E1) or testosterone, is the main estrogen secreted by the ovary. The metabolic pathway of the endogenous estrogens is  $E2 \rightarrow E1 \rightarrow E3$  (estriol) and the most potent is E2.

The estrogens used in the present study are the two most frequently used in Europe and North America, E2 and CE respectively. Conjugated estrogen has been used in ERT and HRT since the 1950s. CE is prepared from the urine of pregnant mares and consists of 50-60% estrone sulphate with equine estrogens such as equilin and  $17\alpha$ -dihydroequilin. They are

considered “natural” as they tend to behave in a similar way to purely human estrogens. CE has been prescribed worldwide and used more than any other preparation. As a result, many clinical studies of the effects of HRT on climacteric symptoms, endometrial and breast cancer, osteoporosis and cardiovascular disease are based on this treatment. Synthetic compounds of micronised/valerate E2 are commonly used in ERT/HRT in most of Europe including Scandinavia. As a consequence, a variety of estrogens with different properties are nowadays used in HRT, table 1.

The dose of estrogen required for amelioration of climacteric symptoms varies due to individual differences such as physical build, dietary habits and smoking. In recent times, the generally accepted strategy has been to prescribe the lowest effective dose of estrogen.

## Progestogens

In fertile women, synthesis of progesterone occurs in the corpus luteum during the second part of the menstrual cycle and during pregnancy. Progesterone counteracts the proliferative effects of estrogen in the endometrium and "prepares" the endometrium for a possible pregnancy. Natural progesterone cannot be administered orally, and for endometrial safety synthetic progestogens are used in HRT. The duration of progestogen treatment is more important than the dose and type used as regards the suppression of endometrial hyperplasia and cancer, see “HRT and the endometrium” above (Whitehead et al 1982).

The possibility that progestogens might reduce the beneficial effects of ERT on wellbeing is a hotly debated issue. Some progestogens are derivatives of 19-nortestosterone (19 nor-progestogens) e.g. norethisterone acetate and levonorgestrel. These steroids are characterised by high androgenicity which counteracts the effect of estrogen on lipid metabolism and coagulation. In addition, they may produce typically androgenic side effects, such as greasy skin and acne. Another commonly used progestogen, medroxyprogesterone acetate, is a 17 $\alpha$ -hydroxyprogesterone closely related to natural progesterone and characterised by low androgenicity. This has fewer androgenic side effects. Today, a number of different progestogens with slightly different properties are used in HRT, table 1. Equipotent doses of different progestogens as regards effect on the endometrium are shown in table 2.

The progestogens used in the present study are those used most frequently in Europe and North America, NETA and MPA, respectively.

**Table 1. Synthetic progestogens and estrogens**

19-nortestosterones	17 $\alpha$ -hydroxyprogesterones	Estrogens
norethisterone (NETA)	medroxyprogesterone (MPA)	conjugated equine estrogen
lynestrenol	cyproterone acetate	17 $\beta$ -estradiol (valerate/micronized)
levonorgestrel (LNG)	megestrol acetate	
norgestimate	dydrogesterone	
gestodene		
desogestrel		

**Table 2. Equipotent doses of synthetic progestogens.**

medroxyprogesterone acetate	10 mg
norethisterone acetate	0.7-1 mg
levonorgestrel	0.075 mg
micronized progesterone	200-300 mg
dydrogesterone	20 mg

## Alternative therapy

For those women who are unwilling or unable to use HRT, alternative treatments such as acupuncture, physical exercise, relaxation programmes, fans, air conditioners or cold showers could relieve climacteric symptoms (Stevenson & Delprato 1983, Wyon et al 1995). Phytoestrogens are often suggested as an alternative to HRT and are widely available. Results of studies into the effects of phytoestrogens on climacteric symptoms are, however, conflicting. A recently published placebo-controlled study of climacteric symptoms showed no difference between placebo and isoflavonoids (Nikander et al 2003).



Selective estrogen receptor modulators (SERM) are now available, and there is substantial evidence that they increase BMD and reduce the incidence of vertebral fractures. However, no positive effect on QoL has been shown with SERM. On the contrary, an increase in hot flushes was detected in postmenopausal women treated with raloxifen (Ettinger et al 1999).

Tibolone is a tissue-specific synthetic steroid hormone with estrogenic, progestogenic and androgenic properties. It can alleviate hot flushes and sweats (Hammar et al 1998) and there is evidence from clinical trials that it increases BMD in healthy women and women with osteoporosis (Studd et al 1998). However, an association between breast cancer and current use of tibolone was shown in the Million Women Study (Banks et al 2003).

## Present study

At the time the present study was carried out, there were two continuous combined regimens available in Sweden - Premelle® and Kliogest®. They contain different doses and types of estrogens, 0,625mg conjugated estrogen and 2mg 17 $\beta$ -estradiol respectively. The serum levels of estradiol and estrone obtained with standard doses of CE and E2 are similar (Andersson et al 1978). Both CE and E2 have been shown to effectively reduce climacteric symptoms and increase bone mineral density. The two preparations also contain different progestogens with 5mg medroxy-progesterone acetate in Premelle® and 1mg norethisterone acetate in Kliogest®. It is not known if the two preparations have equal effects on the brain, the cardiovascular system and the breast.

Placebo-controlled or open studies of the two combinations separately have reported effects on bleeding pattern, wellbeing and lipids (The Writing Group for PEPI Trial 1995, The Writing Group for PEPI Trial 1996, Staland 1985, Munk-Jensen et al 1994, Stadberg et al 1996<sup>1</sup>, Ulrich et al 1997). As far as we knew, a directly comparative study between the two combinations CE/MPA and E2/NETA as regards bleeding patterns, wellbeing and lipids had not been done before.

## AIMS

---

To investigate the effects of hormone replacement therapy on quality of life issues, especially bleeding patterns and wellbeing, and lipid profiles. The aim was also to compare two commonly prescribed continuous combined hormone replacement therapies - conjugated estrogens/medroxyprogesterone acetate and 17 $\beta$ -estradiol/norethisterone acetate.

The specific aims of the study can be summarised as follows:

1. To investigate and compare troublesome irregular bleedings in postmenopausal women given continuous combined hormone replacement therapy of two different types.
2. To describe changes in wellbeing at onset (first month) of HRT with two different types of continuous combined hormone replacement therapy in women starting treatment, and women switching from mainly sequential treatment.
3. To describe changes in wellbeing during long-term treatment (one year) with two different types of continuous combined hormone replacement therapy in women starting treatment, and women switching from mainly sequential treatment.
4. To investigate the effects on lipids and lipoproteins in postmenopausal women given two different types of continuous combined hormone replacement therapy.

## SUBJECTS AND METHOD

---

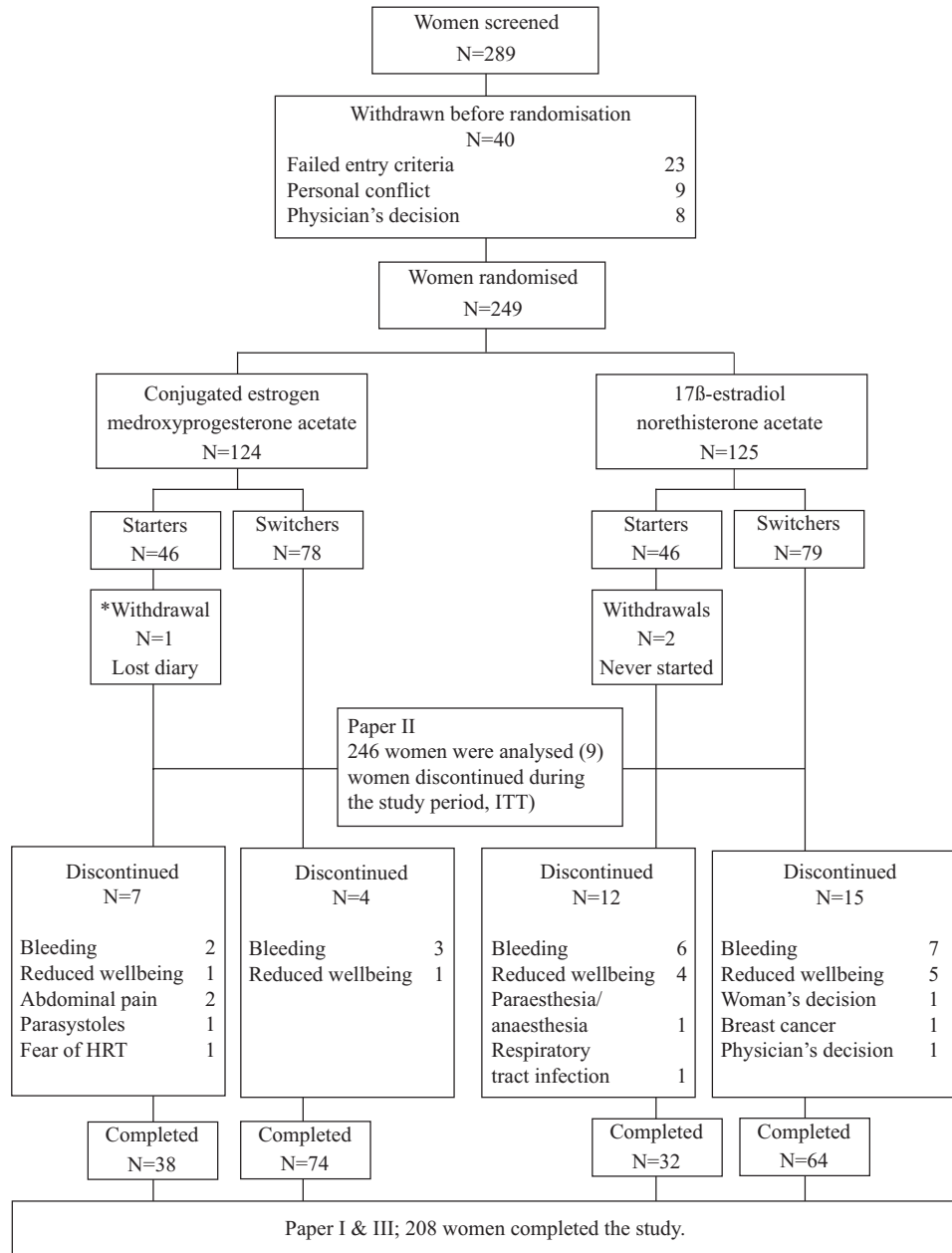
### SUBJECTS

In this clinical trial a total of 249 women were included and randomised into two different continuous combined estrogen-progestogen treatment groups, figure 1. At enrolment, the women were stratified into "starters" who had not received HRT during the previous two months or "switchers" who were currently using HRT at the time of inclusion. All women were asked to keep a daily record of vaginal bleedings in a diary. They also registered daily symptoms of wellbeing on a cyclicity diagnoser scale. Sixty-two of the 249 women who entered the main study were included in the lipid profile study, figure 2.

To be eligible for inclusion, all participants had to be healthy women aged 52 years or more with an intact uterus, who had not menstruated spontaneously for at least 2 years and who either had climacteric symptoms or were using HRT. To be eligible for the lipid sub-study, women had to have abstained from use of all forms of HRT for at least 2 months prior to the study.

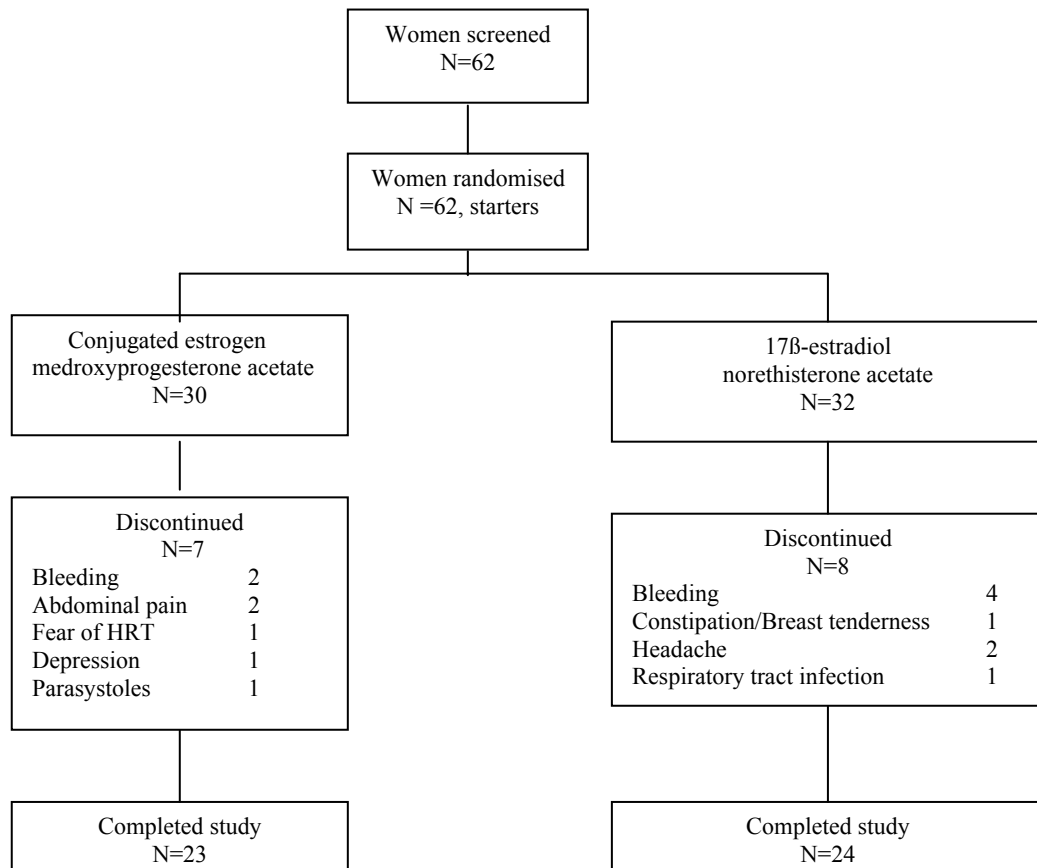
Exclusion criteria included adenomatous hyperplasia with or without atypia, undiagnosed vaginal bleeding, history of cancer of any kind, cardiovascular or thromboembolic disease, poorly regulated hypertension, diabetes, and chronic medication with barbiturates, benzodiazepines, antidepressants, or anti-epileptic medication. Use of steroid hormones (other than study medication) was not permitted during the study period. In addition, women in the lipid study were excluded if they were smokers or had a BMI above  $\geq 31 \text{ kg/m}^2$ . However, lipid and lipoprotein values outside the normal range at screening did not constitute an exclusion criterion. The women included in the lipid profile study were not permitted to use lipid lowering agents during the study. The participants were recruited by newspaper advertisements or opportunistically during clinic visits. Women with and women without a history of PMS were included.

**Figure 1. Flow chart of screened, randomised, withdrawn, discontinued and analysed women during the study.**



\*The woman who lost her diary cards was classified as a withdrawal patient in the studies of well-being but was included in the bleeding study, in which she was classified as a dropout due to bleeding.

**Figure 2. Flow chart of screened, randomised, withdrawn, discontinued and analysed women in the lipid profile sub-study.**



## Ethical considerations

The research protocol was approved by the ethics committee of each centre and by the Swedish Medical Products Agency following the rules of the Revised Declaration of Helsinki (Hong Kong 1989). Oral and written informed consent was obtained from all participants.

## Multicentre study sites

The study involved eight private clinics, two university hospitals, three health centres and one local hospital. For practical reasons only five of these centres took part in the lipid profile study, (one private clinic, two

university hospitals and two health centres) and each included between seven and 26 participants.

## Demographic data

Table 3 summarises the demographic data of the study population. No significant differences in baseline characteristics were found between the two treatment groups, starters and switchers, except for lower diastolic blood pressure, higher BMI and thinner endometrium in the starter group.

**Table 3. Demographic data of women who enrolled in the study (mean±SEM)**

	CE/MPA 0.625mg/5mg	E2/NETA 2mg/1mg	Starters	Switchers
	N=123	N=123	N=89	N=157
Age (years)	55.7 ± 0.27	56.0 ± 0.29	55.9 ± 0.34	55.8 ± 0.25
Time to menopause (years)	5.6 ± 0.35	5.4 ± 0.27	5.3 ± 0.35	5.6 ± 0.28
Body mass index (kg/m <sup>2</sup> )	24.9 ± 0.31	25.1 ± 0.32	25.5 ± 0.38*	24.7 ± 0.27
Systolic blood pressure (mm Hg)	135 ± 1.2	135 ± 1.4	133 ± 1.4	136 ± 1.2
Diastolic blood pressure (mm Hg)	81 ± 0.79	81 ± 0.82	79 ± 0.89**	82 ± 0.73
Parity (n)	2.0 ± 0.10	2.0 ± 0.10	2.0 ± 0.12	2.0 ± 0.08
History of premenstrual syndrome, (n)	37 ± 4.2	30 ± 3.9	21 ± 4.5	46 ± 3.6
Endometrial thickness (mm)	4.1±0.20	4.1±0.19	2.9±0.16***	4.7±0.17

\* Higher than switchers, p<0.05. \*\* Lower than switchers, p<0.01. \*\*\* Thinner than switchers, p<0.001.

Lipid profiles at baseline did not differ significantly between the two treatment groups. The results of serum lipid and lipoprotein concentrations at baseline and after a year of treatment are shown in table 6, together with reference values.

## METHOD

This was a one year, prospective, double blind, randomised, parallel-group, multicentre study conducted between March 1997 and January 1999. The study was conducted and monitored according to the Nordic Guidelines for Good Clinical Trial Practice (GCP). In addition, Standard Operation Procedures (SOP) defined by the sponsor were used.

Screening was carried out one month prior to enrolment and included patient history, physical examination including breast and gynaecological examinations, ultrasound estimation of endometrial thickness and endometrial biopsy. Blood pressure, weight and height were measured. Cervical smear and mammography were done in those women who had not had these tests done during the year prior to the start of the study. Plasma lipids and lipoproteins were measured in the women who participated in the lipid profile study. Kidney, liver and thyroid function were measured for safety screening at baseline and after one year of treatment. Enrolment and randomisation were performed at the baseline visit. A randomisation list in blocks of four was computer-generated by a statistician, and kit numbers were assigned in ascending order at each investigation site. In the lipid study, a limited number of starters were enrolled. The women were given continuous daily oral treatment with either a combination of 0.625mg conjugated estrogen and 5mg medroxyprogesterone acetate or a combination of 2mg 17 $\beta$ -estradiol and 1mg norethisterone acetate. In order to keep the medication blinded, a double-dummy technique with dark coated tablet blisters was used. Apoteksbolaget AB (Stockholm, Sweden) performed the randomisation and blinding. The CE/MPA 0.625mg/5mg-combination and the corresponding placebo tablets were provided by Wyeth-Ayerst (Philadelphia, USA). Novo Nordisk (Copenhagen, Denmark) provided the E2/NETA 2mg/1mg-combination, and the corresponding placebo tablets were provided by Pharma-Vinci Medical Production (Frederiksvaerk, Denmark). Compliance was checked by counting unused tablets.

The reasons for any premature discontinuation of the study medication were documented by the investigator in the clinical report form by selecting one of the following predefined categories: adverse reaction, other medical event, failed to return, unsatisfactory response/efficacy, protocol violation, other non-medical event and patient request unrelated to the study. Clinical examination was carried out at 2, 6, and 12 months. The 12 month visit also included pelvic and breast examination, ultrasound estimation of endometrial thickness, endometrial biopsy and, if

necessary, a mammogram was performed (mammogram for starters after one year and for switchers after two years). At the end of the study women were offered HRT of their own choice.

### **Bleeding diary and medication**

Daily records of vaginal bleeding were made prospectively on a bleeding diary card for 13 consecutive 28-day cycles. At enrolment, women were asked to mark each day if they had no bleeding (0), bleeding (B) or spotting (S), appendix 1. Bleeding was defined as bloody vaginal discharge that required the use of pads or tampons and spotting was defined as bloody vaginal discharge that did not require such protection. Daily intake of study medication was also registered on the diary card. Diary cards and treatment compliance were checked at every follow-up visit.

### **Daily symptom ratings**

A modified form of the Cyclicity Diagnoser (CD) scale was used. The CD scale has been validated against a visual analogue scale as an instrument for diagnosing cyclical symptoms in PMS and has also been used during sequential HRT in postmenopausal women (Sundström et al, 1999, Björn et al, 2000, Björn et al, 2002, Björn et al, 2003). It is well suited for repeated measurement analysis. Each symptom is scored every night according to the intensity of the symptoms experienced during the day. The subject is instructed to estimate symptom severity rapidly and not to think for too long before filling in the scale. The symptom rating scale includes 12 items, which are aggregated into seven domains as follows: physical symptoms (breast tenderness and swelling), positive symptoms (cheerful and energetic), negative symptoms (tension, irritability, fatigue and depression), sweats, insomnia, headache, and negative effects on daily life. The CD scale is a Likert scale (an ordinal scale), graded from one to nine, where one indicates complete absence of a particular symptom and nine represents maximum symptom intensity, appendix 2. One step on the scale is enough to detect a difference in mood experience as indicated by a study on symptom severity in women with PMS (Seippel & Bäckström 1998). Changes in domains and in separate items were used as outcome variables. Symptoms were rated by the starter group for four weeks before initiation of treatment. Both starters and switchers rated symptoms each



day during the first four weeks (cycle 1), weeks 5-8 (cycle 2), weeks 23-26 (cycle 6) and weeks 49-52 (cycle 13) of the treatment.

## The diagnosis of premenstrual syndrome

Participants were interviewed about PMS criteria at the screening visit, appendix 3. They were asked whether they had experienced, during their fertile life, negative mood symptoms or physical symptoms that had affected their daily life during the premenstrual period and whether the symptoms decreased or disappeared at the onset of menstruation. In addition, information about previous psychiatric treatment and sleep disturbances was obtained.

## Lipid and lipoprotein assays

Fasting (minimum 12 hours overnight) blood samples from the antecubital vein were used to determine serum lipid and lipoprotein levels. Kidney, liver and thyroid function tests were assessed from the same samples. Blood was centrifuged within 30-60 minutes at 3000 rev/min for 10 minutes. The fresh serum samples were analysed consecutively within 24 hours at a central laboratory - Nova Medical CALAB Clinical Trials Centre, St Görans Hospital, Stockholm, Sweden. Laboratory procedures met all criteria for Good Laboratory Practice. Laboratory personnel were blinded to treatment arms.

Levels of total cholesterol were measured by an enzymatic photometric test using cholesterol oxidase and peroxidase (Artiss & Zak 1997, Deeg & Ziegenhorn 1983), and the interassay coefficient of variation was 1.9%. Triglyceride levels were assayed by a colorimetric enzymatic test using glycerol-3-phosphateoxidase (Cole et al 1997), and the coefficient of variation was 3.5%. Concentrations of HDL-cholesterol were determined by a direct enzymatic method (Nauck et al 1998) and concentrations of LDL-cholesterol were calculated according to the method of Friedewald et al (1972) with an interassay coefficient of variation of 4.7%. Lp(a) concentrations were measured by an immunoturbidimetric analysis (Levine et al 1991) with an interassay coefficient of variation of 8.8%.

## STATISTICS

Statistical analyses were performed using the standard statistical package SPSS, versions 9.0, 10.0 and 11.0. In all statistical tests, a p-value of  $<0.05$  was considered an indication of statistical significance. Analyses were made by conventional statistical methods. When normality of data could not be rejected by the Kolmogorov-Smirnov test, one-sample paired and two independent sample t-tests were used to analyse within-group changes and between-group comparisons. When the Kolmogorov-Smirnov test yielded a statistically significant result (indicating non-normality of data) Wilcoxon non-parametric tests were used. Analyses of four-field tables were done using Fisher's exact test, the McNemar or the binomial test.

In multivariate analyses multiple step-wise forward and backward regression and logistic regression analyses were performed. Kaplan-Meier analyses and Cox regression were applied in life-table analyses. When analysing changes over time, repeated measures analysis of variance and Friedman's test were used.

The mass-significance problem was assessed using Eklund's rule (Eklund & Seeger 1965). Most analyses were performed on a per-protocol basis. However, in papers 2 and 3 in some analyses, variables were analysed on an intention-to-treat basis (ITT). When ITT was used, the principle of last observation carried forward was applied.

## RESULTS

---

### Baseline characteristics

Of the 249 women who met the inclusion and failed the exclusion criteria, two never started their treatment, leaving a total of 247 women who participated in the study, figure 1. Sixty- seven women (37 in the CE/MPA group and 30 in the E2/NETA group) were retrospectively adjudged to have a history of premenstrual syndrome during their fertile life. A previous history of psychiatric treatment was reported by 11% of the women – an equal percentage in both treatment groups. Ninety-two women had not used HRT for at least two months before study start (starters) and 157 women were currently using HRT (switchers). Of 157 switchers, 120 (76%) were on sequential therapy.

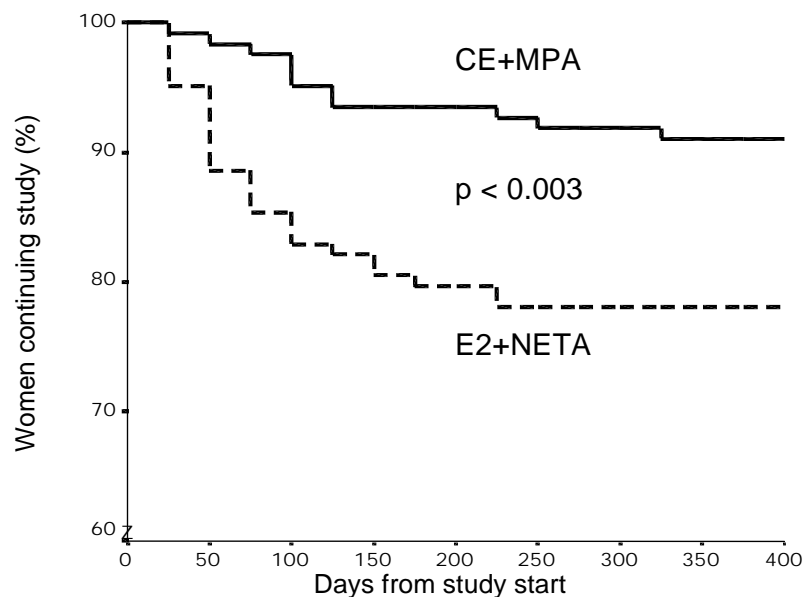
### Serious adverse events

Eleven serious adverse events were reported during the whole study period. Four were reported as possibly related to the study medication (one case of breast cancer diagnosed by mammography at the final visit, acute salpingitis/endometriosis, dizziness and headache, suspected cerebral ischemia/thrombosis producing weakness of the left arm), and seven were not related (pain in the chest and left arm, herpes zoster, nasopharyngeal surgery, diverticulitis leading to colon resection, planned hip surgery, gastritis and breast cancer).

### Discontinuation

Twenty-six out of the 246 participants discontinued the study during the first three months of treatment, significantly more women in the E2/NETA group than in the CE/MPA group, 21/123 and 5/123 respectively ( $p<0.001$ ).

**Figure 3. Time to discontinuation and proportion of women remaining in the study.**



The main reasons given for dropping out were vaginal bleedings and reduced wellbeing (negative mood symptoms, breast tenderness and headache). Twice as many women dropped out of the E2/NETA group because of bleedings than out of the CE/MPA group, and four times as many because of reduced wellbeing, figure 1.

Other reasons given for dropping out were: abdominal pain, breast cancer, respiratory tract infection, doctor opposed to HRT, unwillingness to continue, fear of HRT, feeling unwell, paraesthesia/anaesthesia, constipation/breast tenderness, and palpitations, figure 1.

During the first two months of the study, more bleeding days were noted in dropouts compared to completers ( $p < 0.05$ ). Dropouts also had more symptoms than completers as regards aggregated negative symptoms (including depression and tension) ( $p < 0.05$ ), headache ( $p < 0.05$ ) and negative effects on daily life ( $p < 0.005$ ).

Time to dropout during the whole study period differed significantly between the two treatment groups ( $p < 0.003$ ), figure 3.

In a multivariate logistic regression analysis, a significantly higher risk of premature discontinuation of the study was noted among women treated with E2/NETA (odds ratio (OR) = 4.48; 95% CI: 1.58 – 12.72,  $p < 0.005$ ), and among women reporting a deterioration in aggregated negative symptoms, headache and negative effects on daily life (OR = 4.77; 95% CI: 1.79 – 12.71,  $p < 0.005$ ). The predictor *troublesome bleedings* (= bleeding more than three days) did not reach statistical significance (OR = 1.57; 95% CI: 0.632 – 3.89, NS).

During treatment cycles 4-13, twelve more patients dropped out from the study - six from each treatment group. The reasons given for discontinuing during this part of the study were vaginal bleeding (7), negative mood symptoms (1), both bleeding and negative mood symptoms (1), and other reasons (3).

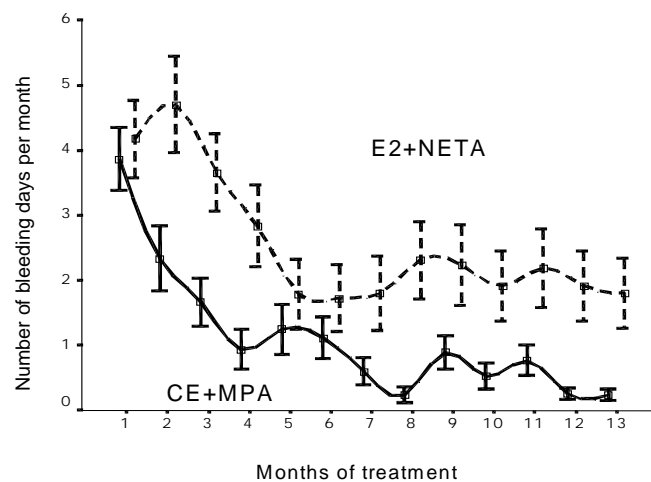
In the study of wellbeing at onset of HRT, nine women out of 246 had dropped out by the end of the first month due to headache (3), negative mood changes (1), bleeding (1), not feeling well (1), constipation/breast tenderness (1), paraesthesia/anaesthesia (1) and palpitations (1) but were included in the ITT analysis.

In the lipid profile study, 47 women completed the study and 15 discontinued, figure 2, mainly due to bleeding problems (6); other reasons given were headache (2), abdominal pain (2), fear of HRT (1), constipation/breast tenderness (1), depression (1), respiratory tract infection (1) and palpitations (1).

## Bleeding patterns

Data from 208 women who completed the study showed a decrease in the number of bleeding days during the first four months ( $p < 0.002$ ), thereafter the number of bleeding days remained unchanged throughout the year. A comparison between the two treatment groups showed significantly more bleeding days during the whole study period in the E2/NETA treatment group compared to the CE/MPA group ( $p < 0.005$ ), figure 4.

**Figure 4. Number of bleeding days each month, according to treatment (mean±SEM). The difference between the treatment groups was significant during months 2-4 ( $p<0.002$ ) and months 8-13 ( $p<0.003$ ).**

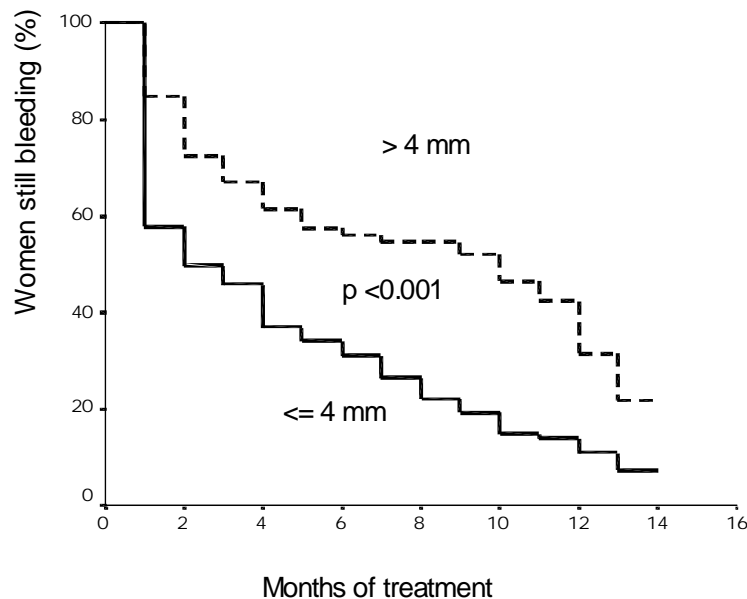


The percentage of women with amenorrhoea from the start and during the remainder of the treatment period was similar between the CE/MPA and E2/NETA treatment groups (33% and 33%, respectively). Regardless of the type of treatment, 92% of the women in the starter group reported amenorrhoea at the end of the study. The corresponding figure for the switcher group was 94% after one year of treatment with CE/MPA, but only 77% after treatment with E2/NETA ( $p<0.05$ ).

Seven women had forgotten to take the study medication for 15 days or more during the whole study. Five came from the CE/MPA group and two from the E2/NETA group. The number of missed tablets and treatment days did not significantly affect the pattern of bleedings.

Endometrial thickness measured by ultrasound at baseline correlated negatively with time to amenorrhoea. Women with an endometrial thickness  $\leq 4$  mm had a shorter time to amenorrhoea than women with an endometrium  $>4$  mm, ( $p<0.001$ ), figure 5.

**Figure 5. Time to amenorrhoea depending on endometrial thickness (more or less than 4 mm).**



### Wellbeing at onset of HRT

In order to be able to detect early changes in wellbeing and climacteric symptoms, the women in the starter group were asked to rate their symptoms four weeks prior to and during the first month of treatment.

Table 4 shows the significant changes, in individual items and domains after four weeks of treatment in the two treatment groups, in starters and switchers, and a comparison between the changes in the two treatment groups. The arrows show deterioration ( $\uparrow$ ) and improvement ( $\downarrow$ ) in wellbeing. This means that, for instance, increased scores in positive mood symptoms and decreased scores in negative mood symptoms are both illustrated by a downward arrow ( $\downarrow$ ). Multivariate analyses of the symptoms all show an advantage for CE/MPA treatment, (last column in the table).

**Table 4. Changes in wellbeing week one versus week four. Only statistically significant changes are indicated by an arrow (p<0.05).**

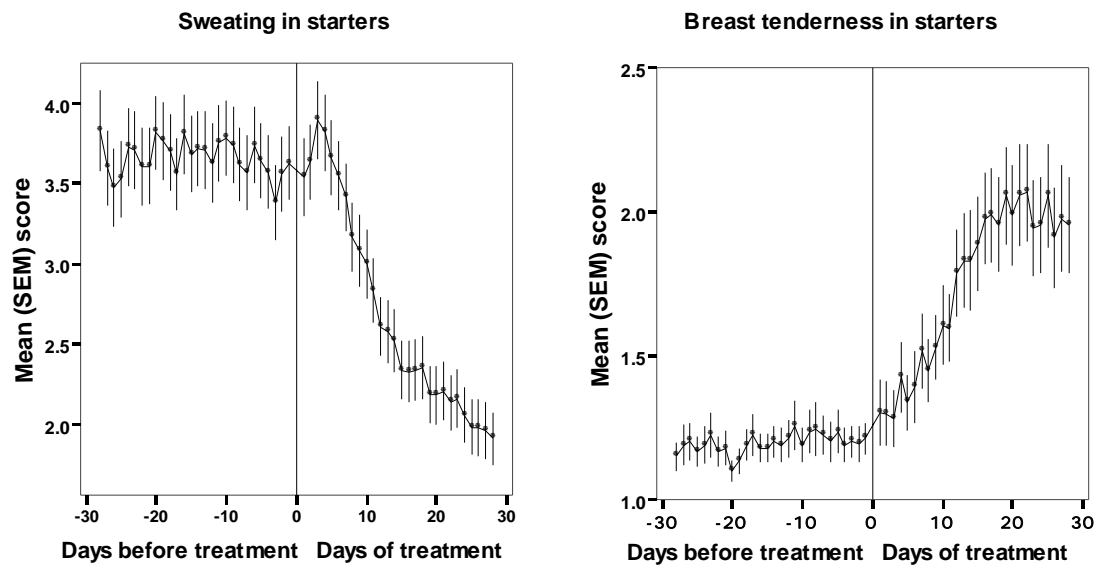
	CE/MPA starters	E2/NETA starters	CE/MPA switchers	E2/NETA switchers	Starters	Switchers	CE/MPA vs E2/NETA
Sweats	↓	↓			↓	↓	
Agg. physical	↑	↑	↑	↑	↑	↑	↑
Breast tenderness	↑	↑	↑	↑	↑	↑	↑
Swelling		↑	↑	↑	↑	↑	
Agg. positive				↑		↑	↑
Cheerfulness				↑		↑	↑
Energy							
Agg. negative							↑
Tension		↑					↑
Irritability							↑
Fatigue							
Depression				↑		↑	↑
Insomnia		↓			↓		
Headache							
Negative effects on daily life		↓		↓	↓	↑	↑

Agg. = aggregated

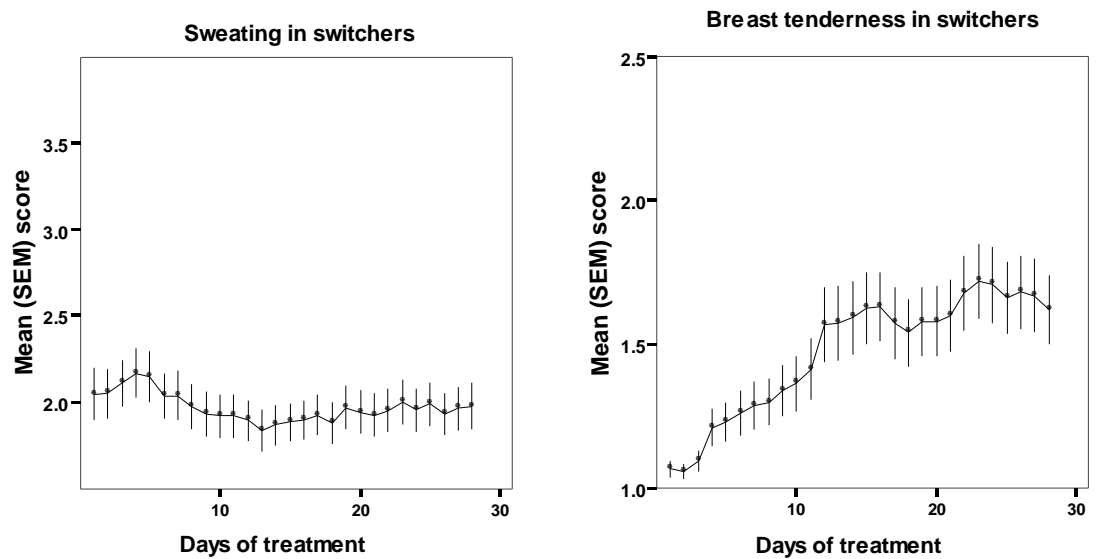
At the onset of treatment, (first day of medication), starters and switchers differed significantly in their symptoms, figures 6 and 7. Sweats (p<0.001), physical symptoms (breast tenderness and swelling) (p<0.001), negative symptoms (tension, irritability, fatigue and depression) (p<0.005), insomnia (p<0.001) and negative effects on daily life (p<0.001) were scored worse by women in the starter group compared to women in the switcher group, figures 6 and 7.



**Figure 6. Daily rating scores in the starter group.**



**Figure 7. Daily rating scores in the switcher group.**



Starters reported improvement within a week in aggregated negative symptoms ( $p<0.001$ ), tension ( $p<0.001$ ) and fatigue ( $p<0.005$ ) compared to pre-treatment ratings.

Both starters and switchers found that side effects appeared more quickly than beneficial symptom relief. Already by day four starters were reporting a deterioration in aggregated physical symptoms ( $p<0.05$ ) and by day seven increased breast tenderness and swelling ( $p<0.05$ ). Women in the switcher group reported a deterioration in aggregated physical symptoms by day three ( $p<0.05$ ), and by days five to seven they were reporting deterioration in negative effect on daily life, breast tenderness, depression and cheerfulness ( $p<0.05$ ).

In the multivariate analysis, women who had reported PMS during fertile life experienced a deterioration in wellbeing compared to women without a history of PMS during the first month. These differences were significant during study treatment as regards aggregated physical symptoms ( $p<0.05$ ), swelling ( $p<0.05$ ), depression ( $p<0.05$ ), irritability ( $p<0.05$ ), tension ( $p<0.05$ ), headache ( $p<0.001$ ) and negative effects on daily life ( $p<0.001$ ).

## Wellbeing during long-term treatment with HRT

In order to investigate changes in wellbeing during prolonged HRT both starters and switchers were also asked to rate their symptoms during cycles 2, 6 and 13.

Significant changes compared to baseline ( $\uparrow$ = deterioration and  $\downarrow$ = improvement) during the different rating periods in individual symptoms are presented separately for starters, switchers and treatment groups as shown in table 5.

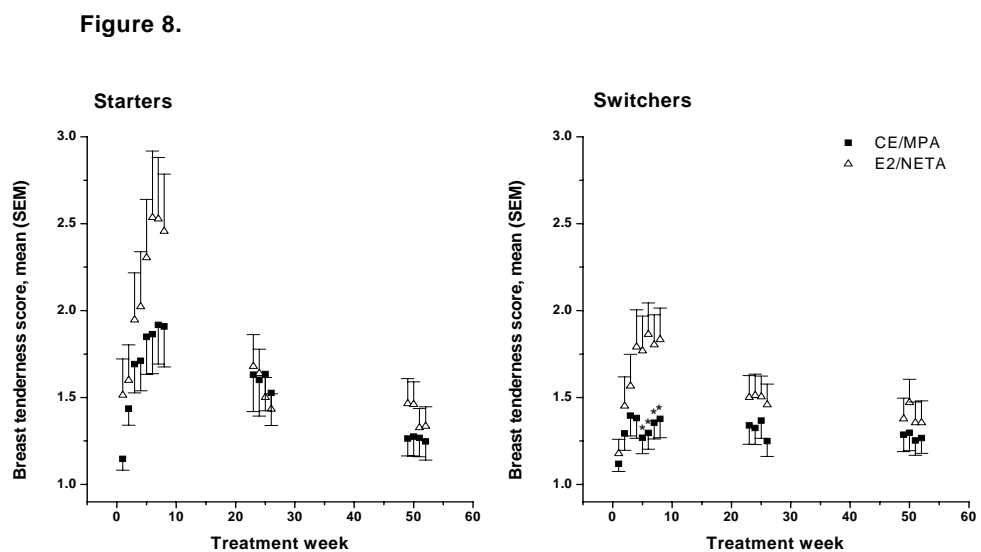
**Table 5. Changes in wellbeing during one year (cycles 1, 2, 6 and 13 compared). Only statistically significant changes are indicated by an arrow (p<0.05).**

Cycles	CE/MPA Starters	E2/NETA Starters	CE/MPA Switchers	E2/NETA Switchers	Starters	Switchers	CE/MPA Group	E2/NETA Group
	1 2 6 13	1 2 6 13	1 2 6 13	1 2 6 13	1 2 6 13	1 2 6 13	1 2 6 13	1 2 6 13
Sweats				----↑	-----↓-----	-----↓-----	-----↓-----	-----↓-----↑
Agg. Physical					--↑-----↓	--↑-----↓		
Breast tenderness	--↑	--↑		--↑	--↑-↓-----	--↑-0-----		
Swelling								
Agg. Positive			-----↓					
Cheerful			-----↓					
Energetic			-----↓					
Agg. Negative	-----↓							
Tension						--↓0-----		
Irritable					--↓-----	--↓0-----		--↓-----
Fatigue					--↓-----	--↓0-----		
Depression								
Insomnia					--↓-----	--↓-0----		
Headache								
Negative effects on daily life					-----↓			

- ↑ = deterioration up to cycle 2
- ↓ = improvement whole study period
- ↑ = deterioration from cycle 6 up to cycle 13
- ↓----- = improvement up to cycle 6 thereafter stable
- ↓-----↑ = improvement up to cycle 6 thereafter deterioration up to cycle 13
- ↓----- = improvement up to cycle 2 thereafter stable
- ↓----- = improvement first cycle thereafter stable
- ↑-----↓ = deterioration up to cycle 2 thereafter improvement rest of the study
- ↓0----- = improvement up to cycle 2 thereafter back to baseline and remained
- ↓-0----- = improvement up to cycle 2 back to baseline up to cycle 6 and remained
- ↑--↓----- = deterioration up to cycle 2 thereafter improvement up to cycle 6 and remained
- ↑-0----- = deterioration up to cycle 2 thereafter back to baseline up to cycle 6 and remained

The women treated with E2/NETA scored worse in breast tenderness during the whole study period compared to the CE/MPA group ( $p<0.001$ ), figure 8.

**Figure 8. Daily rating scores for breast tenderness, (mean $\pm$ SEM), during treatment cycles 1, 2, 6 and 13. A difference between treatment groups was evident during cycle 2 in the switcher group ( $p<0.05$ ).**



Symptom changes over time were different in starters and switchers. Starters experienced more sweats than switchers throughout the whole study period ( $F=24.42$ ,  $p<0.001$ ). Starters also experienced more breast tenderness during the first two treatment cycles ( $F=6.75$ ,  $p<0.01$ ), but after 6 cycles of treatment there was no discernible difference between starters and switchers. Switchers felt more cheerful than starters throughout the study period ( $F=5.19$ ,  $p<0.05$ ) but also suffered more tension than starters during the first two cycles ( $F=6.80$ ,  $p<0.01$ ).

A multivariate analysis was used to investigate possible predictors for changes in wellbeing. The analysis showed that, compared to other women, women with a high BMI were more prone to sweats ( $p<0.05$ ), irritability ( $p<0.001$ ), aggregated physical symptoms ( $p<0.05$ ) and swelling ( $p<0.05$ ). Multiparous women reported more breast tenderness ( $p<0.05$ ) and aggregated physical symptoms ( $p<0.01$ ) compared to women

with one or no children. Furthermore, women with a history of PMS scored higher in insomnia ( $p<0.05$ ), negative effects on daily life ( $p<0.005$ ), depression ( $p<0.01$ ) and aggregated negative symptoms ( $p<0.05$ ).

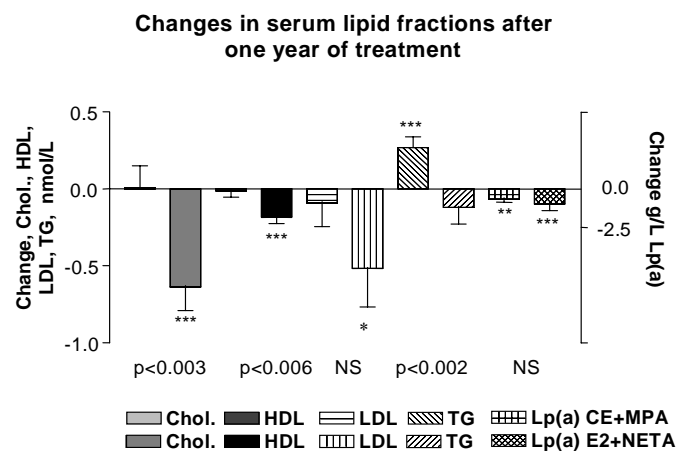
## Lipids and lipoproteins

**Table 6. Serum lipid and lipoprotein levels at baseline (B) and changes after a year of treatment (F), together with reference values.**

	Visit	Normal range	CE/MPA n=23 Mean $\pm$ SEM	E2/NETA n=24 Mean $\pm$ SEM
S-Cholesterol, mmol/L	B	<6.5	6.1 $\pm$ 0.18	6.5 $\pm$ 0.23
S-Cholesterol, mmol/L	F		6.1 $\pm$ 0.21	5.9 $\pm$ 0.17
S-Triglycerides, mmol/L	B	0.6-2.2	1.1 $\pm$ 0.10	1.2 $\pm$ 0.13
S-Triglycerides, mmol/L	F		1.3 $\pm$ 0.13	1.1 $\pm$ 0.07
S-Lp(a), g/L	B	<0.30	0.20 $\pm$ 0.04	0.30 $\pm$ 0.06
S-Lp(a), g/L	F		0.13 $\pm$ 0.03	0.20 $\pm$ 0.04
S-HDL, mmol/L	B	$\geq$ 1.15	1.7 $\pm$ 0.09	1.8 $\pm$ 0.07
S-HDL, mmol/L	F		1.7 $\pm$ 0.07	1.6 $\pm$ 0.06
S-LDL, mmol/L	B	<5.0	3.9 $\pm$ 0.18	4.1 $\pm$ 0.25
S-LDL, mmol/L	F		3.8 $\pm$ 0.20	3.6 $\pm$ 0.19

The results of the study show that treatment with CE/MPA did not change levels of TC or HDL whereas treatment with E2/NETA significantly lowered TC and HDL levels ( $p<0.001$ ). The differences in changes in both TC and HDL were significant between the two treatment groups ( $p<0.01$ ). In both treatment groups LDL levels decreased, but not significantly in the CE/MPA group. However, when the results from both treatment groups were merged, a significant decrease in LDL concentrations was observed ( $p<0.05$ ). TG levels increased significantly in the CE/MPA group, but they remained unchanged in the E2/NETA group. A significant decrease in Lp(a) levels was seen in both treatment groups, and no significant difference between the treatment groups was noted, figure 9.

**Figure 9. Serum lipid and lipoprotein levels after one year of treatment, (mean±SEM). In the pairs of bars the two treatments are shown (CE/MPA on the left and E2/NETA on the right). The differences between treatment groups are shown with p-values under/over the bars to a level of significance as indicated by the stars.**



In a multivariate regression analysis of changes in TG levels, including as predictors baseline TG, treatment and age, only age was positively correlated with change ( $p<0.01$ ) indicating that older women increased their TG levels more than younger women during HRT.

## DISCUSSION

---

This study found that the main reasons for discontinuing hormone replacement treatment were irregular bleedings and reduced wellbeing, and that three times more women given E2/NETA discontinued HRT, compared to those given CE/MPA. Indeed, women given the combination of E2/NETA had more trouble with irregular bleedings throughout the whole study, compared to those given CE/MPA. More women switching from sequential HRT to CE/MPA achieved amenorrhoea compared to those switching to E2/NETA. The thickness of the endometrium, as measured by ultrasound, correlated with the frequency of bleeding problems at the beginning of treatment or when changing from sequential to continuous HRT. Both starters and switchers reported unpleasant side effects such as breast tenderness and swelling after only a few days of treatment. Unexpectedly, unwanted effects appeared more quickly than the expected benefits of treatment, e.g. a reduction in sweats. Both treatment groups improved significantly as regards sweats during the first six months of the study. However, sweats in women in the switcher sub-group treated with E2/NETA became more frequent during the latter half of the study period. All the women treated with CE/MPA scored better on wellbeing during the first two treatment months compared to women treated with E2/NETA. Thereafter, no significant difference in wellbeing was noticed between women treated with the two regimens. Women with a history of PMS during their fertile lives reported a significant deterioration in wellbeing compared to women without a history of PMS. As regards lipid metabolism, comparison between the two regimens showed no substantial differences after a year of treatment. Both regimens significantly reduced Lp(a), the most important risk factor for CVD.

### Methodological strengths and weaknesses

This study compares two of the most commonly used forms of combined HRT. These two treatments contain different estrogens and progestogens in different doses and are therefore not easily compared. In the doses used in the study, conjugated estrogen and 17 $\beta$ -estradiol have been shown to be equivalent in their biological activity in bone tissue and in the endometrium (Lindsay 1984, Whitehead et al 1981). It is unclear if they

are equipotent as regards brain and breast tissue. In addition, the two regimens use different progestogens. MPA is derived from 17 $\alpha$ -hydroxyprogesterone which has more progesterone-like effects, and NETA is derived from 19-nortestosterone which has more androgenic effects. The potency of a progestogen is measured by its effect on the endometrium, but this does not necessarily translate into an equivalent potency in other tissues. Furthermore, the women who participated in this study showed individual variations in response to the same treatment.

The number of women in the study is easily sufficient for studying wellbeing, especially when consecutive daily ratings are used. Furthermore, a proper dropout analysis was performed as well as an intention to treat analysis - all factors adding to the solidity of the study. On the other hand, no placebo group was included. Placebo-controlled studies show a substantial placebo-effect on all, not just vasomotor, symptoms. (Campbell & Whitehead 1977, Notelovitz et al 2000). A placebo arm would have enabled interesting comparisons of sweating episodes at the onset of treatment.

Studying wellbeing is, to say the least, a challenge. There are many difficulties and potential confounders associated with subjective symptom ratings. HRT affects wellbeing in several ways. Firstly, it relieves climacteric symptoms, secondly, it provides an estrogen-mediated improvement of mood and it could produce progestogen related side effects. The women in our study rated a large number of symptoms, both positive and negative, each day for several months, making it possible to detect changes immediately. In earlier studies of wellbeing during HRT, screening instruments have been used once at baseline and once at the end of the study (Hays et al 2003). Such studies are poorly equipped to study the temporal relationship between treatment and changes in parameters of wellbeing.

The method used to diagnose PMS represents a further weakness in the present study. As women were already postmenopausal at inclusion, the PMS diagnosis was bound to be retrospective. Usually, prospective symptom ratings during two consecutive menstrual cycles are required for a diagnosis of PMS/PMDD (Hammarbäck, et al 1989<sup>1</sup>, American Psychiatric Association 1994). To meet the criteria for PMDD, the more severe form of PMS, women are required to report at least five distressing symptoms during the luteal phase, and the symptoms should interfere with social or occupational functioning. In addition, they should not have any ongoing mood or anxiety disorder. Women participating in the present study were given a diagnosis of previous PMS based on reported cyclical negative mood symptoms which decreased or disappeared at the onset of



menstruation. At inclusion, the women were asked whether they had been treated for any psychiatric condition, and psychotropic medication was not allowed during the study period. In the present trial, 27% of the women had experienced premenstrual symptoms, which is similar to the expected number of cases found in a population-based study. Only 2-6% of an average female fertile population are said to suffer from severe PMDD (Sveindóttir & Bäckström 2000). Naturally, any diagnosis of previous PMS made when counselling women with climacteric problems must be retrospective and based on symptom history.

A post-hoc power calculation showed that the small size of the lipid study gave it inadequate power (<80%) for comparisons of changes in LDL and Lp(a) concentrations. A larger study could have produced a different result. However, Lp(a) decreased significantly in both treatment groups and LDL in the E2/NETA group. Nevertheless, when the analysis was performed on both treatment groups combined, LDL levels fell significantly. In previous similar studies the number of women included in each investigated group has varied between 15 and 20 (Sporrong et al 1989, Stadberg et al 1996<sup>2</sup>).

The strength of this kind of study is that comparisons are made between two equally well-regarded preparations with different compositions and used for the same indications, and this can provide new information about effects and side effects. The new knowledge may help to refine treatment, assist counselling and enable the possibility of individualising the choice of HRT both at the onset of treatment and as a response to unwanted side effects such as irregular bleedings.

## Reasons for dropout

Trice as many women dropped out from the group treated with E2/NETA than from the group given CE/MPA during the study. The main reasons for discontinuing treatment prematurely were irregular bleedings and reduced wellbeing. This is consistent with a previous study of compliance with HRT in which the authors concluded that progestogen side effects and/or bleedings were the commonest reasons for dropping out (Björn & Bäckström 1999). The scored negative effects on wellbeing were worse during the first months of treatment. Thereafter, symptoms improved continuously in both starters and switchers. Women who chose to discontinue their treatment prematurely because of side effects such as bleeding problems or reduced wellbeing did so early in the study. We do not know how these women would have felt if they had continued with

their treatment. In theory, some of those who suffered from irregular bleedings would have gone on to achieve amenorrhoea. The cumulative frequency of amenorrhoea increased during the whole study period. Breast tenderness and negative mood might well have decreased with the passage of time. Others, in the normal run of events, would have had their HRT doses adjusted or changed to a different type. This study therefore illustrates the importance of providing information and support with regard to the risk of developing side effects and bleeding problems at the onset of HRT. This ought to promote better compliance and greater treatment benefits. However, it is important to continue the search for new regimens that can more reliably avoid irregular bleedings and other undesirable side effects.

## Effects on bleeding patterns

The study shows that women in the CE/MPA group had fewer bleeding days during the year of treatment compared to women treated with E2/NETA. Furthermore, in switchers, more women treated with CE/MPA had amenorrhoea by the end of the study. Generally, women who developed amenorrhoea had, initially, a thinner endometrium than those who went on to develop bleeding problems.

The cumulative frequency of amenorrhoea was greater in women with an endometrial thickness of  $\leq 4$  mm versus  $>4$  mm, measured ultrasonically. Ultrasonography can therefore represent a valuable tool for estimating the risk of vaginal bleeding and spotting at the onset of HRT, or on switching from sequential to combined HRT. Not all women who experienced bleeding problems chose to discontinue the study. Presumably, these women persevered with the treatment in the hope of spontaneous improvement in episodes of sweats and the positive effects on mood. It is conceivable that with the onset of amenorrhoea, women feel an immediate and tangible benefit and find it easier to accept the treatment. The explanation for the difference in bleeding patterns between the groups may be found in the differing pharmacology of the treatments. The combination CE/MPA - a more estrogen-dominant regimen - may build up a more resilient endometrium, whereas E2/NETA - a more androgenic combination - produces a thinner endometrium. Use of non-steroidal anti-inflammatory agents (NSAIDs) and aspirin during the study did not show any relation to bleeding pattern. Nor did missing study medication seem to have influenced bleeding patterns.

In the present study, the cumulative frequencies of amenorrhoea were 92% in the CE/MPA group and 77% in the E2/NETA group after one year of treatment. Earlier studies have reported 87% amenorrhoea for CE/MPA users (Archer et al 1994) and 100% amenorrhoea for E2/NETA users (Stadberg et al 1996<sup>1</sup>) - in both cases after one year of treatment. The women who took part in the latter study were all HRT-naïve at inclusion and their results can be compared to those found in the starter group in the present study – the group that achieved the greater degree of amenorrhoea. However, a recently published placebo-controlled study that compared 1mg NETA/5µg ethinylestradiol and 0.625mg CE/2.5mg MPA showed that the former gave better control of bleeding over a year of treatment and this is not in agreement with our results. The authors stated that the cumulative amenorrhoea frequencies were 88% and 68% in the two treatment groups respectively (Simon et al 2003). One explanation for the conflicting results might be that NETA was combined with another estrogen compound whereas concentrations of MPA were lower than in our study.

Perimenopausal women produce variable amounts of endogenous ovarian estradiol that causes endometrial proliferation and may cause irregular bleedings during HRT. Treatment with sequential regimens of estrogen and progestogen are therefore commonly used to regulate bleeding patterns in perimenopausal women. Sequential HRT produces recurrent periods of proliferation and this might lend a certain instability to the endometrium when switching to continuous combined treatment. As expected, the switcher group in the present study reported significantly more bleeding days during the first month of treatment compared to starters. Most of the switchers started the study with a scheduled bleeding on sequential estrogen-progestogen treatment. The reason for developing continuous combined regimens with estrogen and progestogen was to induce endometrial inactivity and produce a thinner, near-atrophic, endometrium and thereby avoid periodic withdrawal bleeding. Continuous progestogens down regulate both estrogen and progesterone receptors in the endometrium. Bleeding problems may also occur due to an imbalance between estrogen and progestogen, with estrogen inducing proliferation of the endometrium resulting in irregular bleeding. Van de Weijer et al (1999) studied breakthrough bleedings during continuous combined HRT and found a relation between serum estradiol levels and the incidence of bleedings in postmenopausal women. At the start of continuous combined HRT, estrogen may induce proliferation of the endometrium in spite of the opposing effect of progestogens resulting in breakthrough bleeding, and this might have been the main reason for the early bleeding problems in

our study. Over time, bleeding due to extreme atrophy of the endometrium probably became more common. Studies of endometrial atrophy and bleeding problems during HRT and hormonal contraceptives have shown that an extremely thin/atrophic endometrium has a greater tendency to bleed because of vascular fragility and a thin stroma (Hickey et al 1996). A shrinking stroma gives less support to the fragile vessels and that may contribute to bleedings (Casper 1996). One study has shown that the endometrium of younger women treated with sub-dermal progestogen implants for contraception, when studied with hysteroscopy, showed areas with dilated superficial vessels that bled easily when the intrauterine pressure was increased (Hickey et al 1996). The exact mechanisms leading to vascular fragility and breakthrough bleedings remain unknown, but obviously there are great inter-individual variations in response to continuous combined treatment.

## Measuring wellbeing

Quality of life is a broad term that represents the summation of wellbeing and satisfaction which in this study is estimated by positive effects on wellbeing items, improvement in episodes of sweats and the absence of progestogen side effects. Measuring QoL provides information about how the menopause is experienced by women and which symptoms impinge on their daily lives, as well as providing information about the individual woman's treatment response. Today we have good methods for measuring QoL in the form of properly validated rating scales.

Psychiatric rating scales have been used in several climacteric studies. The problem with this type of instrument is that they have been developed for screening and diagnostic purposes in psychiatric disorders and not specifically for the postmenopausal period. The Kupperman index has frequently been used in Sweden to quantify episodes of climacteric symptoms. The 11 most common symptoms are graded 0-3 and are multiplied to a total score which is used to monitor the severity of the climacteric complaints (Place et al 1985). The Women's Health Questionnaire (WHQ) was one of the first questionnaires developed for measuring specific symptoms during and after the menopause (Hunter et al 1986). This questionnaire has been used in several clinical trials to assess treatment-induced changes. Other commonly used instruments include the Menopause-Specific Quality of Life Instrument (MENQOL) (Hilditch et al 1996) and the Greene Climacteric Scale (Greene 1998). In the recently published WHI study a battery of scales was used. QoL was

assessed using the RAND-36-Item Health Survey (RAND-36) (Hays et al 1993) and a modified checklist of menopausal symptoms (Moos & Leiderman 1978). Other scales that have been developed to measure typical menopausal symptoms include the Menopause Rating Scale (MRS) (Schneider et al 2000) and the Menopausal Symptom List (Perz 1997). These scales are meant for use at fixed time points during a study, using scores ranging from 0-100, a four to seven-point response scale or a 7-point Likert scale. The number of symptoms assessed by these different scales varies, and many of the symptoms are aggregated into domains that measure different aspects of the climacteric. Common domains constitute symptoms related to physical, psychosocial, sleep or sexual dysfunctions, vasomotor symptoms and bleedings.

In the present study a modified form of an earlier developed self rating scale, the cyclicity diagnoser (CD) scale was used. This is a Likert scale designed for discovering cyclical symptoms and symptom changes. The Likert scale is similar to the visual analogue scale (VAS) and is used to rate subjective symptoms. VAS was first used for evaluating pain intensity and comes in the form of a continuous 10 cm line, in contrast to the CD/Likert scale which has 9 discrete visually defined steps labelled 1-9. Continuous daily ratings enable the analysis of symptoms and are superior to intermittent ratings in that they can pick up subtle changes over time. This type of rating scale is increasingly being used to study symptom differences between groups and symptom variations within individuals over time. Difficulties arise, however, when trying to make direct comparisons between the symptom ratings of different individuals. Today, the VAS is used to compare the nature and intensity of patient symptoms in a broad range of applications (Aitken 1969, Maxwell 1978).

Seippel & Bäckström (1998) have used the VAS scale to study a group of women with PMS during two consecutive menstrual cycles and to compare the menstrual symptoms of two different groups of women. The differences between the two groups were identical when raw scores from the scales and absence/presence of a particular symptom were used. By demonstrating that the results of these two investigations were compatible, the authors wished to show that it is possible to measure the intensity of subjectively felt physical and psychological symptoms. The VAS/Likert scales can easily be used to assess how the symptoms of a particular individual or group change over time and, to some extent, to make comparisons between groups. Consideration should however be given to possible individual differences in baseline scores and, as with all instruments of self-assessment, in the interpretation of inter-individual scores. The Likert scale has been validated with high internal consistency

and time reproducibility in occupational studies on perceived stress (Consoli et al 1997).

The CD scale is easy to use and makes it possible to make many consecutive measurements. Although it would have been possible to carry out daily symptom ratings throughout the whole study period, we felt that compliance would have suffered. The scale that we chose for this study is made up of 12 items which are further aggregated into seven different domains. Aggregating several symptoms into domains in this way gives a general picture that better corresponds with the concept of wellbeing. The design of the CD scale makes optical data scanning possible - increasing the reliability of data collection. Subjective ratings could mean that an equally intense symptom rated 4 by one woman may be rated 5 by another. For this reason, data were corrected for baseline scores when comparisons between groups were made.

## Effects on wellbeing

As expected, women in the starter group had more sweats at onset of treatment, but these symptoms improved during the first six months. Other studies have shown that sweats improves successively during longer periods of treatment (Holst et al 1989, Notelovitz et al 2000). Even the switcher group, where most women had switched from sequential HRT, showed an improvement in episodes of sweats – implying that earlier sequential therapy had not fully suppressed this symptom. One possible explanation for this is that added late-cycle progestogen reduces the effect of estrogen and thereby increases the likelihood of sweats attacks. Women treated with E2/NETA reported a deterioration in sweats during the later part of the study. NETA, which has the more androgenic progestogen, has possibly a greater effect than MPA on estrogen and its ability to relieve sweats. It would have been interesting to see what would have happened if women had continued to record daily ratings up to 6 months beyond the point at which they reported optimised amelioration of sweats. Unwillingness to rate symptoms over a long period of time has been seen by many as a hindrance, but experience shows that the majority of women welcome the opportunity to report how symptoms and treatment affect their daily lives (Fallowfield et al 1987). Therefore, the value of future studies would be enhanced if women were to rate their climacteric and wellbeing symptoms daily over a longer time-span. One would have expected that, after the initial differences in wellbeing between starters and switchers on the same treatment, the two groups would rate more or

less equally in scores at the end of the one year study. However, this was not the case. At no time during the study did starters and switchers show equivalent symptom patterns.

Women treated with CE/MPA rated significantly better on wellbeing than women given E2/NETA treatment. The difference was discernible during the first 2 months of treatment, decreasing thereafter and disappearing by the end of the study. Noticeably, switchers treated with E2/NETA had more frequent sweats after 6 months of treatment than switchers treated with CE/MPA. The explanation may lie in the difference in composition of the two regimens, see above. In a randomised double-blind cross-over study of 51 postmenopausal women treated with continuous estrogen, the effect of 10mg MPA was compared to 1mg NETA, added sequentially. The study showed that MPA produced fewer negative symptoms and more positive symptoms than NETA (Björn et al 2000). The study also reported more physical symptoms on MPA than on NETA, in contrast to the findings of the present study. The reason might be that continuous progestogens were used in the latter.

In the WHI study, no significant effects on wellbeing in women without climacteric symptoms were reported. At enrolment women with moderate or severe menopausal symptoms were discouraged, but not excluded, from participating in the trial. Of all the included women only 12% reported vasomotor symptoms at baseline. A battery of quality of life scales were used, at baseline, at one year and in a subgroup after three years of treatment (Hays et al 2003). In the present study the symptoms were rated daily, during the first 2 cycles, cycle 6 and cycle 13 using the sensitive instrument of CD scales. Starters showed, as expected, more symptoms than switchers at the onset of HRT. In spite of fewer and less intensive symptoms, even the switchers scored improvement in symptoms during the study. Furthermore, the study showed that side effects of HRT appeared during the first week of treatment – much sooner than any positive effect, which appeared later during the first two months of treatment. This could not be demonstrated in WHI in which QoL was measured at a follow up after one year.

Studies show that ERT is effective in relieving vasomotor symptoms, enhancing wellbeing and improving mood in women with climacteric symptoms (Sherwin 1999, Wiklund et al 1993). Women with climacteric symptoms who experience improvement on HRT also appear to report improved wellbeing. The present study did not aim to investigate the effect of unopposed estrogens on mood, but as the estrogen arm of the WHI study is continuing we can look forward to gaining valuable information about the effect of estrogen on wellbeing.

The scope of the present study does not permit any deductions to be made as to whether it is the estrogen, progestogen or the combination of the two that affects the changes in mood symptoms during the treatment year.

## Side effects

In the present study women in both switcher and starter groups developed side effects sooner than they experienced any benefits from treatment. Already by day 4 and 5 women were reporting physical symptoms. This information was made possible by the use of daily ratings of the symptoms already at the onset of treatment, and the results are in line with clinical experience. As far as we know, this has not been reported previously. The physical side effects included breast tenderness which increased during the first treatment months in both starters and switchers and in both treatment groups. After 6 months of treatment women reported that breast tenderness had returned to pre-treatment levels. It is impossible to say whether a concomitant fall in negative mood changes related to progestogen intolerance took place, since a positive estrogen-mediated effect on mood was assumed. However, irregular bleeding subsided and an increasing number of women achieved amenorrhoea during the study period. The present study shows that women who discontinue HRT are likely to do so at the beginning of treatment. This demonstrates the importance of providing information about early side effects and suggests that good support at the onset of treatment can improve compliance.

Without doubt, women with previous PMS were at greater risk of developing side effects than women without a history of PMS. In our study retrospective analysis showed that 67 of 247 women had a history of PMS during their fertile life. These women showed a significant deterioration during the study in physical symptoms, including swelling, depression, irritability, tension, headache and effects on daily life. These results are in agreement with earlier studies in postmenopausal women with a history of PMS during treatment with sequential HRT (Björn et al 2000, Björn et al 2002). In contrast, Kirkham et al (1991) could not demonstrate a difference between PMS-like mood changes in oophorectomised women treated with sequential HRT and those given placebo. In that study PMS was diagnosed prospectively. Hammarbäck<sup>2</sup> et al (1989) have shown that high levels of estradiol and progestogen produce more pronounced PMS compared to low levels of these hormones. In addition, women with PMS are more sensitive to mood changes when using OCs



than women without PMS (Cullberg 1972). Wang et al (1996) have studied the relationship between steroids and symptoms in women with PMS. They found that in women with PMS, higher estrogen levels in the luteal phase were related to more severe symptoms compared to cycles with lower estrogen levels in the luteal phase. Another study used a sequential regimen with higher dose of estrogen, 3mg E2, in combination with 10mg MPA and found that a higher dose of estrogen during the progestogen phase was associated with deterioration in mood and physical symptoms (Björn et al 2003). Björn et al (2000) compared the effects of MPA 10mg and NETA 1mg on changes in negative mood and physical symptoms. The authors concluded that women without PMS scored less on negative and more on positive mood when treated with MPA. Therefore the choice of estrogen dose, the choice of progestogen, and their combined effect on wellbeing play a vital role in patient acceptance and treatment outcome. Previous PMS should be taken into account. This information may help clinicians counsel women more effectively about the benefits of HRT and assist in managing treatment-related side effects.

In the present study the proportion of women in the starter group reporting breast tenderness during the first two cycles was the same for both treatments. Switchers receiving E2/NETA also reported breast tenderness. Breast discomfort and pain may be associated with increased mammographic density (McNicholas et al 1994). Unopposed estrogen regimens show less breast density compared to estrogen plus progestogen (Greendale et al 1998). One study has found significantly increased mammographic density in women receiving E2/NETA compared to those receiving tibolone and placebo RR 8.3 (CI 95% 2.7-25.0) (Lundström et al 2002). They also reported significant differences in breast tenderness, reported as adverse events, between tibolone (2.4%) and E2/NETA (18%). Studies have found increased mammographic breast density to be a strong and independent risk factor for breast cancer (Boyd et al 1998). In future, it may be possible to identify women on HRT who are at risk of developing breast cancer by monitoring changes in breast density.

## HRT and cardiovascular risk

Estrogen has a positive effect on lipid metabolism and promotes vasodilatation, which is thought to reduce the risk of developing arteriosclerosis (Mendelsohn & Karas 1999). But treatment with different HRT regimens also produces a variety of changes in haemostasis, and levels of several coagulation and anticoagulation factors are adversely affected (Lowe et al 2001, Kjellberg et al 1999). As a result, the risk of thrombosis rises 2-3 fold, primarily during the first treatment year (Hulley et al 1998).

Lipoproteins are products of a long, complicated and only partially understood metabolic chain that includes a series of metabolic steps. Many studies have investigated the effects of ERT and HRT on lipid metabolism. The knowledge that HDL levels fall whilst levels of LDL and TG rise in women over 50 years has made these lipoprotein fractions interesting as predictors of CVD. Low levels of HDL and high levels of LDL are theoretically associated with a surplus of cholesterol in peripheral tissues and vessel walls. This indicates a defect in the cholesterol transport system and would imply a greater risk of developing atherosclerosis leading to cardiovascular disease. This relation between atherosclerosis and CVD has been confirmed in epidemiological studies (Asplund et al 2003). It has been postulated that this is the result of decreasing levels of endogenous estrogen. Studies of ERT show that estrogen influences HDL, LDL and TG levels in a favourable direction (The Writing Group for the PEPI Trial 1995). The addition of progestogen to ERT reduces these favourable changes in levels of HDL, LDL and TG, depending on the doses and types of progestogens used. TC levels are also affected favourably by ERT and HRT (Hodis et al 2003). The final results of the WHI study reported a relation between high levels of LDL at baseline and greater excess risk of CVD when using HRT (Manson et al 2003). The lipoprotein fraction Lp(a) is another factor that is strongly predictive of CVD (Stein & Rosenson 1997). In the present study both treatments significantly reduced Lp(a). In the CE/MPA treatment group, TG levels were significantly raised compared to baseline, compared to E2/NETA group where a slight fall in TG levels failed to reach statistical significance. One reason for this might be that CE affects the synthesis of TG in the liver more than E2 (Barrett-Connor & Bush 1991). In addition, the effect of MPA on TG removal may be less than that of NETA. Studies have looked at doses, treatment forms and different hormone preparations. Geographical differences have been shown in lipid patterns and this

further complicates matters. A study involving five European countries looked at lipid metabolism in postmenopausal women who were treated mainly with transdermal E2/NETA. It showed differences in TG, HDL and Lp(a) levels between different nationalities (Ranta et al 2002).

Many observational studies have been made over the past decades, and these point to a reduction in CVD risk among users of ERT and HRT (Stampfer & Colditz 1991). But at the end of 1990's HERS was published. This was a large, randomised, prospective, placebo controlled secondary prevention study which included women with clinically apparent CVD or who were at high risk of CVD. It showed that women treated with HRT had more cardiovascular events than the placebo group during the first treatment year (Hulley et al 1998). The validity of the findings has been brought into question as the study was discontinued early, despite the fact that protective effects could be demonstrated by the fourth year of treatment. More women in the placebo group used statins and significantly more of the cardiovascular events in the treatment group took the form of venous thrombosis as opposed to arterial events. However, a huge randomised, placebo-controlled primary prevention study – WHI - with the same treatments as in HERS, also suggested an overall increase in risk of CVD with a RR of 1.29 (CI 95% 1.02-1.63) over the whole study period (Rossouw et al 2002). The study was ended early, after an average of 5.2 years of follow-up, because it was found that the health risks associated with estrogen plus progestogen exceeded the benefits. The women in WHI were treated with 0.625mg CE/MPA 2.5mg, (the same combination as in the present study), a regimen that is commoner in the USA than in Europe where E2 is the most commonly prescribed estrogen. In the WHI trial, 34% of women had a BMI  $\geq 30\text{kg/m}^2$  and were aged between 50-79 years and could have developed sub-clinical atherosclerosis years before the trial began. CVD prevention in early menopause was not a primary endpoint in the WHI study. Atherosclerosis takes years to develop, and the cardiovascular events associated with HRT in these two randomised, controlled trials seem to occur soon after the onset of the treatment. It seems likely, therefore, that thromboembolism plays a greater role than atherogenicity. The HERS study was conducted in elderly women approximately 20 years past menopause, which means that these results cannot be generally applied to the majority of women who start HRT for relief of their climacteric symptoms. Manson et al (2003) published the final results from the WHI study and reported an increased risk of CVD (RR 1.81 CI 95% 1.09-3.01) in the first year, and during years 2-5 a smaller and non-significant excess risk on treatment with estrogen combined with progestogen. The treatment group showed a

clear trend towards lower risk over time. In fact, a sub-group analysis revealed that women who were more than 20 years postmenopausal when starting treatment contributed most of the excess risk. The result of the WHI study adds to our knowledge of HRT and cardiovascular effects, but much remains to be discovered. Whether estrogen therapy in other forms has similar effects on the cardiovascular system is uncertain. New, prospective, randomised, controlled studies on women starting therapy at menopause with different doses and regimens are required to clarify these issues.

## Conclusions and some thoughts for the future

The number of women aged 50 years or older is constantly increasing due to increasing life expectancy, and so an ever-increasing proportion of the world's female population is postmenopausal. Studies show that 75% of postmenopausal women suffer from sweats and hot flushes. Estrogens are known to give prompt and effective relief from these symptoms, and yet negative reports such as WHI may frighten women into refraining from or discontinuing HRT – medication that could help them considerably. This type of reaction has been seen before – when alarm reports regarding oral contraceptives led to much more restrictive prescribing. Such public reaction may lead to women being withheld the complete and balanced information they need to make informed decisions about starting or discontinuing treatment. This is a worrying development. Women's need for help with menopausal problems may become forgotten as the debate shifts to negative reports, and effective treatment may be denied them. Another visible consequence of these reports is the increasing sale of herbal and other alternative treatments. This would suggest that women still need help and are actively seeking other treatment options. The efficacy of these 'alternatives' on menopausal symptoms is not scientifically established and many may be worthless, or even harmful, as efficacy and adverse reactions are not monitored. Women seeking advice about HRT need, now more than ever, expert medical advice that can take a broad and dispassionate view of all the evidence and provide adequate and detailed information. Only this quality of information can enable the woman to come to a balanced decision about which, if any, form of HRT is the best for her at this particular time. It is all too easy to misrepresent scientific research and dismiss HRT as dangerous. These negative reports still leave many questions unanswered. Large randomised placebo-controlled studies are still needed to improve knowledge about these

complex issues. Proper scientific studies are still the best available methods upon which to base wise decisions that can stand the test of time, but the recent premature termination of two large studies makes it unlikely that such large and expensive studies will be undertaken in the near future. Research into effective treatments for menopausal symptoms such as changes in mood and wellbeing, free from risk of adverse effects such as breast disease, is vital for the health and quality of life of all women. The safety of menopausal treatments must stand up to the most stringent scrutiny, and treatment must be tailored to the individual needs of modern woman, in all her diversity.

**‘There is nothing either good or bad, but thinking makes it so’.** *Hamlet II*

## SUMMARY AND GENERAL CONCLUSIONS

---

- Women discontinue treatment with E2/NETA three times more often during the first three months than women treated with CE/MPA, and the most common reasons are bleedings and reduced wellbeing.
- Treatment with CE/MPA causes fewer bleeding problems than treatment with E2/NETA.
- A thin endometrium as measured by ultrasound at the start of continuous combined HRT predicts amenorrhoea.
- Women starting HRT experience an improvement in sweats, as do women who switch from sequential therapy.
- Women on continuous combined HRT with E2/NETA could experience worsening of sweats even after six months of treatment.
- Both regimens produce progestogen side effects within 3-4 days, much sooner than any perceived benefits.
- After one year of treatment, women on continuous combined CE/MPA and E2/NETA experience comparable effects on mood.
- Effects on wellbeing are different in women starting HRT compared to women switching from mainly sequential estrogen/progestogen combinations, even after one year of treatment.
- A history of PMS increases the risk of severe progestogen side effects during treatment with continuous combined HRT.
- Continuous treatment with either CE/MPA or E2/NETA lowers LDL and Lp(a) - an effect that is considered beneficial for cardiovascular protection in women.

## ACKNOWLEDGEMENTS

---

I should like to express my gratitude to all the generous and helpful people who have given me their support. It is only with their help that I have been able to carry out this study and develop it further into this thesis.

This study was carried out with the support of *Wyeth Lederle Nordiska AB*, to whom I should like to offer my heartfelt thanks for the support I received in my work, right in the middle of my own menopause!

Thanks to all the investigators and research nurses, for all their hard work, all the support, the stimulating discussions, the shared coffee and laughter. Torbjörn Bäckström, Umeå, Marie Bixo, Umeå, Inger Björn, Piteå, Arne Eliasson, Karlstad, Göran Skarin, Uppsala, Margareta Åkesson, Borås, Mahmod Moinian, Västra Frölunda, Magnus Haeger, Göteborg, Paula Atterfeldt, Göteborg, Göran Frick, Helsingborg, Eva Ingemarsson, Eslöv, Jörgen Nielsen, Malmö, Gunilla Björk, Strängnäs, Karin von Hamos, Stockholm and Claes Jüllig, Stockholm.

To *Sven-Eric Olsson*, medical director, for introducing me to this work, and for his enthusiasm, generosity, encouragement and confidence in me.

To *Siw Eriksson*, medical director, for her support and belief in me.

To *Ann-Katrin Lenkel* for excellent guidance at the onset and diligent care with the collected data.

All the *women* who participated.

**In particular I wish to express my warm and sincere gratitude to the following friends and colleagues:**

First of all I like to express my deeply felt gratitude to my supervisor *Marie Bixo*, my boss and mentor, for her support and trust, for having confidence in me, for always being there, for helping to create a place for me at work and in science, for stimulating discussions and fruitful advice in the field of scientific thinking and writing, and for her never ending support, generosity and enthusiasm.

Professor *Torbjörn Bäckström*, co-mentor, who introduced me to this fascinating subject. For his invaluable guidance and advice, constructive criticism, never-failing encouragement, patience and support, open mind, and for leading me in the ways of science.

Co-mentor *Björn Jonsson*, my deepest gratitude for stimulating discussions, generous advice, perfect data handling and surefooted guidance through the minefield of statistics. For his patience and stamina when faced with my incomprehension, his ready availability with prompt responses, and for putting up with long and often coffee-depleted working days.

A special thank you to:

*Ann Lalos, Marianne Wulff, Annika Idahl, Anna-Carin Wihlbäck, Anneli Kero, Birgit Ericson, Birgitta Renström, Carina Jonsson, Mats Löfgren and Peter Lindgren* for always extending a helping hand and for creating a wonderful, relaxing haven in the staff room.

Everybody at *Umeå Neurosteroid Research Centre* - always helpful and encouraging- for making me feel at home during my study days in Umeå.

*Sigrid Nyberg, Magdalena Taube, Inger Björn and Inger Sundström Poromaa* for support and pleasant moments at the research mill.

*Viveca Odling* for reading parts of this thesis and for wise advice.

*Stefan and Aino Fianu Jonasson* for their friendship and support.

*Umeå University and specially Department of Clinical Sciences Obstetrics and Gynecology, Umeå* for warmth, cheerfulness and a positive attitude to a postgrad from the sticks.

*Medical Library, Umeå University hospital* for all the support and positive reception.

*Lisa and Gruffydd Jones* for polishing up my English text.

*Berit Thorsell*, dear friend, and wine-specialist for support in many different ways.



*Elisabeth Hagelin Pettersson and Margareta Bondestam* for years of friendship, support and lighter moments on the straight and narrow path of research.

*The dancing team* for still being there, even though I haven't always been able to.

*Bibban Jonsson* my new found friend, for all the hours I have spent in the world of science with her husband Björn, through whom we got to know each other.

*Ann-Christine Skyttberg*, my friend, for her constant encouragement and for being a fantastic shipmate.

My friend *Marju Dahmoun* for encouragement, stimulating discussions, scientific and philosophical, on land and on the high seas.

*Ingrid Andersson*, my dear friend, for her infectious curiosity about life, and for her equally infectious laughter.

*Solfrid Sveen-Praesthus*, dear friend since youth, for her clear-sighted ability to spot life's opportunities and for sharing life's darker moments.

*Eva Levin*, dear friend, colleague and conversation partner on many a late evening, for always being there for me, even if it's on the end of a phone, and for encouraging me on my path through life with her perceptive conversation and positive attitude to life.

*Hazze* for his inexhaustible patience and endurance

My wonderful daughters *Hanna* with husband *Peter*, *Karolina* with boyfriend *Johan*, who provide the unlimited joy that give life its true significance, for always supporting and believing in me, come what may. And for still being able to believe that *mamma* is the best.

To all of you, and you are many, not mentioned but nevertheless not forgotten, who have supported and encouraged me during this work - my deepest thanks!

## REFERENCES

---

Aitken RCB. Measurement of feelings using visual analogue scales. Section of Measurement in Medicine 1969 October;62:989-93

Alder EM. How to assess quality of life: problems and methodology. In Schneider HPG (ed) Hormone replacement therapy and quality of life. Carnforth: Parthenon Publishing, 2002, pp 11-21

American Psychiatric Association. Diagnostic and statistical manual of mental disorders, 4<sup>th</sup> edition. Department of Health and Human Services, Washington, DC, 1994, pp 714-8

Anderson A, Sklovsky E, Sayers L, Steele P, Turnbull AC. Comparison of serum oestrogen concentrations in post-menopausal women taking oestrogen sulphate and oestradiol. British Medical Journal 1978;1:140-2

Andréen L, Bixo M, Nyberg S, Sundström-Poromaa I, Bäckström T. Progesterone effects during sequential hormone replacement therapy. Eur J Endocrinol 2003;148:571-7

Archer DF, Pickar JH, Bottiglioni F. Bleeding patterns in postmenopausal women taking continuous combined or sequential regimens of conjugated estrogens with medroxyprogesterone acetate. Obstet Gynecol 1994;83:686-92

Artiss JD, Zak B. Measurement of cholesterol concentration. In: Rifai N, Warnick GR, Dominiczak MH, eds. Handbook of lipoprotein testing. Washington: AACC Press 1997, pp 99-114

Asplund K, Berglund G, Lindgren S, Lindholm N. Internmedicin. Stockholm: Liber AB 2003, pp 122-33

Avis NE, Stellato R, Crawford S, Bromberger J, Cain V, Kagawa-Singer M. Is there a menopausal syndrome? Menopausal status and symptoms across racial/ethnic groups. Soc Sci Med 2001;52:345-56

Banks E, Beral V, Bull D, Reeves G. Breast cancer and hormone-replacement therapy in the million women study. *Lancet* 2003;362:41-27

Barrett-Connor E, Bush TL. Estrogen and coronary heart disease in women. *JAMA* 1991;265:1861-7

Bell SE. Sociological perspectives on the medicalization of menopause. *Ann NY Acad Sci* 1990;592:173-8

Björn I, Bäckström T. Drug related negative side-effects is a common reason for poor compliance in hormone replacement therapy. *Maturitas* 1999;32:77-86

Björn I, Bixo M, Strandberg-Nöjd K, Nyberg S and Bäckström T. Negative mood changes during hormone replacement therapy: A comparison between two progestogens. *Am J Obstet Gynecol* 2000; 183: 419-26

Björn I, Bixo M, Strandberg-Nöjd K, Collberg P, Nyberg S, Sundström-Poromaa I, Bäckström T. The impact of different doses of medroxyprogesterone acetate on mood symptoms in sequential hormonal therapy. *Gynecol Endocrinol* 2002;16:1-8.

Björn I, Sundström-Poromaa I, Bixo M, Nyberg S, Bäckström G, Bäckström T. Increase of estrogen dose deteriorates mood during progestin phase in sequential hormonal therapy. *J Clin Endocrinol Metab* 2003;88(5):2026-30

Bourne T, Hillard T, Whitehead M, Crook D, Campbell S. Oestrogens, arterial status, and postmenopausal women. *Lancet* 1990;335:1470-1

Boyd NF, Lockwood GA, Byng JW, Trichler DL, Yaffe MJ. Mammographic densities and breast cancer risk. *Cancer Epidemiol Biomark Prev* 1998;7:1133-44

Bäckström T, Sanders D, Leask R, Davidson D, Warner P, Bancroft J. Mood, sexuality, hormones, and the menstrual cycle. II. Hormone levels and their relationship to the premenstrual syndrome. *Psychosom Med* 1983;45:503-7

Bäckström T. Symptoms related to the menopause and sex steroid treatments. In Eds Baulieu EE, Bock G, Goode J. Non-reproductive actions of sex steroids. CIBA-Foundation Symposium 191. John Wiley & Sons Ltd. Chichester 1995, pp 171-86

Campbell S, Whitehead M. Oestrogen therapy and the menopausal syndrome. *Clinics in Obstetrics and Gynaecology* 1977;4:31-46

Cardozo L, Bachmann G, McClish D, Fonda D, Birgersson L. Meta-analysis of estrogen therapy in the management of urogenital atrophy in postmenopausal women: second report of the hormones and urogenital therapy committee. *Obstet Gynecol* 1998;92:722-7

Casper RF. Regulation of estrogen/progestogen receptors in the endometrium. *Int J Fertil* 1996;41:16-21

Chlebowski R, Hendrix S, Langer R, Stefanick M, Gass M, Lane D, Rodabough R, Gilligan MA, Gyr M, Thomson C, Khandekar J, Petrovitch H, McTiernan A, for the WHI investigators. Influence of estrogen plus progestin on breast cancer and mammography in healthy postmenopausal women. The women's health initiative randomized trial. *JAMA* 2003;289:3243-53

Cole TG, Klotzsch SG, McNamara JR. Measurement of triglyceride concentration. In: Rifai N, Warnick GR, Dominiczak MH, eds. *Handbook of lipoprotein testing*. Washington: AACC Press 1997, pp 115-26

Collins P, Shay J, Jiang C, Moss J. Nitric oxide accounts for dose-dependent estrogen-mediated coronary relaxation after acute estrogen withdrawal. *Circulation* 1994;90:1964-8

Collins A, Landgren BM. Psychosocial factors associated with use of hormonal replacement therapy in a longitudinal follow-up of Swedish women. *Maturitas* 1997;28:1-9

Consoli SM, Taine P, Szabason F, Lacour C, Metra PCV. Development and validation of a perceived stress questionnaire recommended as a follow-up indicator in occupational medicine. *Encephale* 1997;23:184-93

Corson SL. A practical guide to prescribing estrogen replacement therapy. *Int J Fertil* 1995;40:229-47

Cullberg J. Mood changes and premenstrual symptoms with different gestagen/estrogen combinations. A double blind comparison with a placebo. *Acta Psychiatrica Scandinavica* 1972 Suppl. 236:1-86

Deeg R, Ziegenhorn J. Kinetic enzymatic method for automated determination of total cholesterol in serum. *Clin Chem* 1983;29:1798-802

Delgado RC, Lopez DML. Prognosis of breast cancer detected in women receiving hormone replacement therapy. *Maturitas* 2001;38:147-56

Deligdisch L, Holinka C. Endometrial carcinoma: two diseases? *Cancer Detection and Prevention* 1987;10:237-46

Dennerstein L, Burrows GD, Hyman G, Wood C. Menopausal hot flushes: a double blind comparison of placebo, ethinyl oestradiol and norgestrel. *Br J Obstet Gynaecol* 1978;85:852-6

Dennerstein L, Burrows G, Hyman G, Sharpe K. Hormone therapy and affect. *Maturitas* 1979;1:247-59

Dennerstein L. Well-being, symptoms and the menopausal transition. *Maturitas* 1996;23:147-57

Derby C, Lamont Hume A, McPhillips J, McFarland Barbour M, Carleton R. Prior and current health characteristics of postmenopausal estrogen replacement therapy users compared with nonusers. *Am J Obstet Gynecol* 1995;173:544-50

Derman R, Dawood MY, Stone S. Quality of life during sequential hormone replacement therapy- a placebo-controlled study. *Int J Fertil* 1995;40:73-8

Diaz M, Frei B, Vita J, Keaney J. Antioxidants and atherosclerotic heart disease. *N Engl J Med* 1997;337(6):408-16

Diczfalussy E. Menopause, developing countries and the 21st century. *Acta Obstet Gynecol Scand Suppl* 1986;134:45-57

Dören M, Schneider HPG. The impact of different HRT regimens on compliance. *Int J Fertil* 1996;41:29-39

Eklund G, Seeger P. Masssignifikansanalys, Statistical Review Central Bureau of Statistics, Stockholm. 1965;5:355-65

Erlík Y, Meldrum DR, Judd HL. Estrogen levels in postmenopausal women with hot flushes. *Obstet Gynecol* 1982;59:403-7

Ettinger B, Black D, Mitlak B, Knickerbocker R, Nickelsen T, Genant H, Christiansen C, Delmas P, Zanchetta J, Stakkestad J, Glüer C, Krueger K, Cohen F, Eckert S, Ensrud K, Avioli L, Lips P, Cummings S. Reduction of vertebral fracture risk in postmenopausal women with osteoporosis treated with raloxifene. Results from a 3-year randomized clinical trial. *JAMA* 1999;282:637-45

Fallowfield L, Baum M, Maguire GP. Do psychological studies upset patients? *J Roy Soc Med* 1987;80:59

Freeman RR. Biochemical, metabolic, and vascular mechanisms in menopausal hot flashes. *Fertil Steril* 1998;70:332-7

Friedewald WT, Levy RI, Fredrickson DS. Estimation of the concentration of low-density lipoprotein cholesterol in plasma without use of the preparative ultracentrifuge. *Clin Chem* 1972;18:499-502

Fuh J-L, Wang S-J, Lu S-R, Juang K-D, Chiu LM. The Kinmen women-health investigation (KIWI): a menopausal study of a population aged 40-54. *Maturitas* 2001;39:117-24

Ganger K, Vyas S, Whitehead M, Crook D, Meire H, Campbell S. Pulsatility index in internal carotid artery in relation to transdermal oestradiol and time since menopause. *Lancet* 1991;338:839-42

George T. Menopause: some interpretations of the results of a study among a non-western group. *Maturitas* 1988;10:109-16

Godsland IF. Effects of postmenopausal hormone replacement therapy on lipid, lipoprotein, and apolipoprotein (a) concentrations: analysis of studies published from 1974-2000. *Fertil Steril* 2001;75:898-915

Gold EB, Sternfeld B, Kelsey JL, Brown C, Mouton C, Reame N, Salamone L, Stellato R. Relation of demographic and lifestyle factors to symptoms in a multi-racial/ethnic population of women 40-55 years of age. *Am J Epidemiol* 2000;152:463-73

Grady D, Wenger N, Herrington D, Khan S, Furberg C, Hunninghake D, Vittinghoff E, Hulley S, for the Heart and Estrogen/progestin Replacement Study Research Group. Postmenopausal hormone therapy increases risk for venous thromboembolic disease. *Ann Internal Med* 2000;132:689-96

Grady D, Herrington D, Bittner V, Blumenthal R, Davidson M, Hlatky M, Hsia J, Hulley S, Herd A, Khan S, Newby L, Waters D, Vittinghoff E, Wenger N for the HERS research group. Cardiovascular disease outcomes during 6.8 years of hormone therapy. Heart and Estrogen/progestin Replacement Study. Follow-up (HERS II). *JAMA* 2002;288:49-57

Greendale GA, Reboussin BA, Hogan P, Barnabei VA, Shumaker S, Johnson S, Barrett-Connor E. Symptom relief and side effects of postmenopausal hormones: results from the postmenopausal estrogen/progestin interventions trial. *Obstet Gynecol* 1998;92:982-8

Greene JG. Constructing a standard climacteric scale. *Maturitas* 1998;29:25-31

Grodstein F, Manson J, Colditz G, Willett W, Speizer F, Stampfer M. A prospective, observational study of postmenopausal hormone therapy and primary prevention of cardiovascular disease. *Ann Intern Med* 2000;133:933-941

Grodstein F, Manson JA, Stampfer M. Postmenopausal hormone use and secondary prevention of coronary events in the nurses' health study. *Ann Intern Med* 2001;135:1-8

Guyatt GH, Bombardier C, Tugwell PX. Measuring disease-specific quality of life in clinical trials. *Can Med Assoc J* 1986;134:889-95

Hammar M, Lindgren R, Berg G, Möller C, Niklasson M. Effects of hormonal replacement therapy on the postural balance among postmenopausal women. *Obstet Gynecol* 1996;88:955-60

Hammar M, Christau S, Nathorst-Böös J, Rud T, Garre K. A double-blind, randomized trial comparing the effects of tibolone and continuous combined hormone replacement therapy in postmenopausal women with menopausal symptoms. *Br J Obstet Gynaecol* 1998;105:904-11

Hammar M, Ekblad S, Lönnberg B, Berg G, Lindgren R, Wyon Y. Postmenopausal women without previous or current vasomotor symptoms do not flush after abruptly abandoning estrogen replacement therapy. *Maturitas* 1999;31:117-22

Hammarbäck S, Bäckström T, Holst J, von Schoultz B, Lyrenäs S. Cyclical mood changes as in the premenstrual tension syndrome during sequential estrogen-progestagen postmenopausal replacement therapy. *Acta Obstet Gynecol Scand* 1985;64:393-7

Hammarbäck S, Bäckström T, MacGibbon-Taylor B<sup>1</sup>. Diagnosis of premenstrual syndrome: description and evaluation of a procedure for diagnosis and differential diagnosis. *J Psychosom Obstet Gynecol* 1989;10:25-42

Hammarbäck S, Damber JE, Bäckström T<sup>2</sup>. Relationship between symptom severity and hormone changes in women with premenstrual syndrome. *J Clin Endocrinol Metab* 1989;68:125-30

Hays RD, Sherbourne CD, Mazel RM. The rand 36-item health survey 1.0. *Health Econ* 1993;2:217-27

Hays J, Ockene JK, Brunner RL, Kotchen JM, Manson JAE, Patterson RE, Aragaki AK, Shumaker SA, Brzyski RG, LaCroix AZ, Granek IA, Valanis BG, for the Women's Health Initiative Investigators. Effects of Estrogen plus progestin on health-related quality of life. *N Engl J Med* 2003;348: 1839-54

Hickey M, Fraser I, Dwarte D, Graham S. Endometrial vasculature in Norplant® users: preliminary results from a hysteroscopic study. *Hum Reprod* 1996;11(2):35-44



Hilditch JR, Lewis J, Peter A, van Maris B, Ross A, Franssen E, Guyatt GH, Norton PG, Dunn E. A menopause-specific quality of life questionnaire: development and psychometric properties. *Maturitas* 1996;24:161-75

Hill DA, Weiss NS, Beresford SAA, Voigt LF, Daling JR, Stanford JL, Self S. Continuous combined hormone replacement therapy and risk of endometrial cancer. *Am J Obstet Gynecol* 2000;183:1456-61

Hlatky MA, Boothroyd D, Vittinghoff E, Sharp P, Whooley MA, for the HERS Research Group. Quality-of-life and depressive symptoms in postmenopausal women after receiving hormone therapy. Results from the heart and estrogen/progestin replacement study (HERS) trial. *JAMA* 2002;287:591-7

Ho SC, Chan SG, Yip YB, Cheng A, Yi Q, Chan C. Menopausal symptoms and symptom clustering in Chinese women. *Maturitas* 1999;33:219-27

Hodis HN, Mack WJ, Azen SP, Lobo RA, Shoupe D, Mahrer PR, Faxon DP, Cashin-Hemphill L, Sanmarco ME, French WJ, Shook TL, Gaarder TD, Mehra AO, Rabbani R, Sevanian A, Shil AB, Torres M, Vogelbach KH, Selzer R for the Women's Estrogen-Progestin Lipid-Lowering Hormone Atherosclerosis Regression Trial Research Group. Hormone therapy and the progression of coronary-artery atherosclerosis in postmenopausal women. *N Engl J Med* 2003;349:535-45

Holst J, Bäckström T, Hammarbäck S, von Schoultz B. Progestogen addition during oestrogen replacement therapy – effects on vasomotor symptoms and mood. *Maturitas* 1989;11:13-20

Holte A. Influences of natural menopause on health complaints: a perspective study of healthy Norwegian women. *Maturitas* 1992;14:127-41

Hulley S, Grady D, Bush T, Furberg C, Herrington D, Riggs B, Vittinghoff E for the heart and estrogen/progestin replacement study (HERS) research group. Randomized trial of estrogen plus progestin for secondary prevention of coronary heart disease in postmenopausal women. *JAMA* 1998;280:605-13

Hunter M, Battersby R, Whitehead M. Relationships between psychological symptoms, somatic complaints and menopausal status. *Maturitas* 1986;8:217-28

Hunter M. Emotional well-being, sexual behaviour and hormone replacement therapy. *Maturitas* 1990;12:299-314

Karlberg J, Mattsson L-Å, Wiklund I. A quality of life perspective on who benefits from estradiol replacement therapy. *Acta Obstet Gynecol Scand* 1995;74:367-72

Kirkham C, Hahn PM, Van Vugt DA, Carmichael JA, Reid RL. A randomized, double-blind, placebo-controlled, cross-over trial to assess the side effects of medroxyprogesterone acetate in hormone replacement therapy. *Obstet Gynecol* 1991;78:93-7

Kjellberg U, Andersson N-E, Rosén S, Tengborn L, Hellgren M. APC resistance and other haemostatic variables during pregnancy and puerperium. *Thromb Haemost* 1999;81:527-31

Klaiber EL, Broverman DM, Vogel W, Peterson LG, Snyder MB. Relationships of serum estradiol levels, menopausal duration, and mood during hormonal replacement therapy. *Psychoneuroendocrinology* 1997;22:549-58

Knight DC, Eden JA. A review of the clinical effects of phytoestrogens. *Obstet Gynecol* 1996;87:897-904

Kronenberg F. Hot flashes: epidemiology and physiology. *Ann N Y Acad Sci.* 1990;592:52-86

Kronenberg F. Hot flashes: phenomenology, quality of life, and search for treatment options. *Exp Gerontol* 1994;29:319-36

Leibenluft E. Do gonadal steroids regulate circadian rhythms in humans? *J Affect Disord* 1993;29:175-81

Levine DM, Parker TS, Sloan BJ, Albers JJ, Marcovina SM, Donner JE, Lorenz JD, Heinzerling RH. Automated measurement of Lp(a) by immunoturbidimetric analysis. *Clin Chem* 1991;37:919

Lindgren R, Berg G, Hammar M, Zuccon E. Hormonal replacement therapy and sexuality in a population of Swedish postmenopausal women. *Acta Obstet Gynecol Scand* 1993;72:292-7

Lindsay R, Hart DM, Clark DM. The minimum effective dose of estrogen for prevention of postmenopausal bone loss. *Obstet Gynecol* 1984;63:759-63

Lip GYH, Beevers M, Churchill D, Beevers DG. Hormone replacement therapy and blood pressure in hypertensive women. *J Hum Hypertens* 1994;8:491-4

Lobo RA, Pickar JH, Wild RA, Walsh B, Hirvonen E, for the menopause study group. Metabolic impact of adding medroxyprogesterone acetate to conjugated estrogen therapy in postmenopausal women. *Obstet Gynecol* 1994;84:987-95

Lock M. Contested meanings of the menopause. *Lancet* 1991;337:1270-2

Lowe G, Upton M, Rumley A, McConnachie A, O'Reilly D, Watt G. Different effects of oral and transdermal hormone replacement therapies on factor IX, APC resistance, t-PA, PAI and C-reactive protein. *Thromb Haemost* 2001;86:550-6

Lundström E, Christow A, Kersemaekers W, Svane G, Azavedo E, Söderqvist G, Mol-Arts M, Barkfeldt J, von Schoultz B. Effects of tibolone and continuous combined hormone replacement therapy on mammographic breast density. *Am J Obstet Gynecol* 2002;186:717-22

MacLennan A, Lester S, Moore V. Oral oestrogen replacement therapy versus placebo for hot flushes (Cochrane Review). In *The Cochrane Library*, Issue 2. Oxford: Update Software, 2001

Magos A, Brewster E, Singh R, O'Dowd T, Brincat M, Studd J. The effects of norethisterone in postmenopausal women on oestrogen replacement therapy: a model for the premenstrual syndrome. *Br J Obstet Gynecol* 1986;93:1290-6

Manson JA, Hsia J, Johnson KC, Rossouw JE, Assaf AR, Lasser NL, Trevisan M, Black HR, Heckbert SR, Detrano R, Strickland OL,

Wong ND, Crouse JR, Stein E, Cushman M. Estrogen plus progestin and the risk of coronary heart disease. *N Engl J Med* 2003;349:523-34

Matthews KA, Kuller LH, Wing RR, Meilahn EN, Plantinga P. Prior to use of estrogen replacement therapy, are users healthier than nonusers? *Am J Epidemiol* 1996;143:971-8

Matthews K, Cauley J, Yaffe K, Zmuda JM. Estrogen replacement therapy and cognitive decline in older community women. *J Am Geriatr Soc* 1999;47:518-23

Mattsson L-Å. What do women want? *Br J Obstet Gynaecol* 1996;103(Suppl3):104-7

Maxwell C. Sensitivity and accuracy of the visual analogue scale: a psycho-physical classroom experiment. *Br J Clin Pharmacol* 1978;6:15-24

McKinley SM, Brambilla DJ, Posner JG. The normal menopause transition. *Maturitas* 1992;14:103-15

McNagny SE. Prescribing hormone replacement therapy for menopausal symptoms. *Ann Intern Med* 1999;131:605-16

McNicholas MMJ, Heneghan JP, Milner MH, Tunney T, Hourihane JB, MacErlaine DP. Pain and increased mammographic density in women receiving hormone replacement therapy: a prospective study. *Am J Roentgenol* 1994;163:311-5

Mendelsohn M, Karas R. The protective effects of estrogen on the cardiovascular system. *N Engl J Med* 1999;304:1801-11

Mohyi D, Tabassi K, Simon J. Differential diagnosis of hot flashes. *Maturitas* 1997;27:203-14

Montgomery JC, Studd JW. Psychological and sexual aspects of the menopause. *Br J Hosp Med* 1991;45:300-2

Moos RH, Leiderman DB. Toward a menstrual cycle symptom typology. *J Psychosom Res* 1978;22:31-40

Morse C, Smith A, Dennerstein L, Green A, Hopper J, Burger H. The treatment-seeking women at menopause. *Maturitas* 1994;18:161-73

Munk-Jensen N, Ulrich L, Obel E, Nielsen S, Edwards D, Meinertz H. Continuous combined and sequential estradiol and norethindrone acetate treatment of postmenopausal women: Effect on plasma lipoproteins in a two-year placebo-controlled trial. *Am J Obstet Gynecol* 1994;171:132-8

Nachtigall LE. Enhancing patient compliance with hormone replacement therapy at menopause. *Obstet Gynecol* 1990;75(Suppl 4): 77-80

Naessen T, Lindmark B, Larsen H-C. Better postural balance in elderly women receiving estrogens. *Am J Obstet Gynecol* 1997;177: 412-6

Nauck M, März W, Wieland H. New immunoseparation-based homogenous assay for HDL-cholesterol compared with three homogenous and two heterogeneous methods for HDL-cholesterol. *Clin Chem* 1998;44:1443-51

Nikander E, Kilkkinen A, Metsä-Heikkilä M, Adlercreutz H, Pietinen P, Tiitinen A, Ylikorkala O. A randomized placebo-controlled crossover trial with phytoestrogens in treatment of menopause in breast cancer patients. *Obstet Gynecol* 2003;101:1213-20

Notelovitz M, Lenihan J, McDermott M, Kerber I, Nanavati N, Arce J-C. Initial 17 $\beta$ -estradiol dose treating vasomotor symptoms. *Obstet Gynecol* 2000;95:726-31

Oldenhave A, Jaszmann LJB, Haspels AA, Everaerd W. Impact of climacteric on well-being. A survey based on 5213 women 39 to 60 years old. *Am J Obstet Gynecol* 1993;168:772-80

Padwick ML, Siddle NC, Lane G, Endacott JA, Cooper H, Pryse-Davies J, Whitehead MI. Oestriol with oestradiol versus oestradiol alone: a comparison of endometrial, symptomatic and psychological effects. *Br J Obstet Gynaecol* 1986;93:606-12

Pearce J, Hawton K, Blake F. Psychological and sexual symptoms associated with the menopause and the effects of hormone replacement therapy. *Br J Psychiatry* 1995;167:163-73

Persson I, Adami HO, Bergkvist L, Lindgren A, Pettersson B, Hoover R, Schairer C. Risk of endometrial cancer after treatment with oestrogens alone or in conjunction with progestogens: results of a prospective study. *Br Med J* 1989;298:147-51

Perz JM. Development of the menopause symptom list: a factor analytic study of menopause associated symptoms. *Women and Health* 1997;25:53-69

Place VA, Powers M, Darley PE, Schenkel L, Good WR. A double-blind comparative study of Estraderm and Premarin in the amelioration of postmenopausal symptoms. *Am J Obstet Gynecol* 1985;152:1092-9.

Prior JC, Alojado N, McKay DW, Vigna YM. No adverse effects of medroxyprogesterone treatment without estrogen in postmenopausal women: double-blind, placebo-controlled, crossover trial. *Obstet Gynecol* 1994;83:24-8

Ranta V, Oksanen H, Arrenbrecht S and Ylikorkala O. National differences in lipid response to postmenopausal hormone replacement therapy. *Maturitas* 2002;42:259-65

Rapp SR, Espeland MA, Shumaker SA, Henderson VW, Brunner RL, Manson JAE, Gass MLS, Stefanick ML, Lane DS, Hays J, Johnson KC, Coker LH, Daiely M, Bowen D, for the WHIMS investigators. Effect of estrogen plus progestin on global cognitive function in postmenopausal women. The women's health initiative memory study: a randomized controlled trial. *JAMA* 2003;289:2663-72

Rebar RW, Trabala J, Mortola J. Low-dose esterified estrogens (0.3 mg/day): long-term and short-term effects on menopausal symptoms and quality of life in postmenopausal women. *Climacteric* 2000;3:176-82

Rossouw JE. Writing Group for the Women's Health Initiative Investigators Risks and benefits of estrogen plus progestin in healthy

postmenopausal women. Principal results from the women's health initiative randomized controlled trial. *JAMA* 2002;288:321-33

Ryan PJ, Harrison R, Blake GM, Fogelman I. Compliance with hormone replacement therapy (HRT) after screening for postmenopausal osteoporosis. *Br J Obstet Gynaecol* 1992;99:325-8

Sacks FM, McPherson R, Walsh BW. Effect of postmenopausal estrogen replacement on plasma Lp(a) lipoprotein concentrations. *Arch Intern Med* 1994;154:1106-10

Schmidt PJ, Nieman L, Danaceau MA, Tobin MB, Roca CA, Murphy JH, Rubinow DR. Estrogen replacement in perimenopause-related depression: a preliminary report. *Am J Obstet Gynecol* 2000;183:414-20

Schneider HPG, Heinemann LAJ, Roemeier H-P, Potthoff P, Behre HM. The menopause rating scale (MRS): reliability of scores of menopausal complaints. *Climacteric* 2000;3:59-64

Seippel L, Bäckström T. Luteal-phase estradiol relates to symptom severity in patients with premenstrual syndrome. *J Clin Endocrinol Metab* 1998;83:1988-92

Shaver J, Giblin E, Lentz M, Lee K. Sleep patterns and stability in perimenopausal women. 1988;11:556-61

Shaver J, Zenk S. Sleep disturbance in menopause. *J Women's Health Gend Based Med* 2000;9:109-18

Shelley J, Smith A, Dudley E, Dennerstein L. Use of hormone replacement therapy by Melbourne women. *Aust J Health* 1995;19:387-92

Sherwin BB, Gelfand MM. Differential symptom response to parenteral estrogen and/or androgen administration in the surgical menopause. *Am J Obstet Gynecol* 1985;151:153-60

Sherwin BB. The impact of different doses of estrogen and progestin on mood and sexual behaviour in postmenopausal women. *J Clin Endocrinol Metab* 1991;72:336-43

Sherwin BB, Tulandi T. "Add-back" estrogen reverses cognitive deficits induced by a gonadotropin-releasing hormone agonist in women with leiomyomata uteri. *J Clin Endocrinol Metab* 1996;81:2545-9

Sherwin BB. Progestogens used in menopause. Side effects, mood and quality of life. *J Reprod Med* 1999;44:227-32

Shumaker S, Legault C, Rapp S, Thal L, Wallace R, Ockene J, Hendrix S, Jones B, Assaf A, Jackson R, Kotchen J, Wasserheil-Smoller S, Wactawski-Wende J, for the WHIMS Investigators. Estrogen plus progestin and the incidence of dementia and mild cognitive impairment in postmenopausal women. The women's health initiative memory study: a randomized controlled trial. *JAMA* 2003;289:2651-62

Simon J, Liu J, Speroff L, Shumel B, Symons J. Reduced vaginal bleeding in postmenopausal women who receive combined norethindrone acetate and low-dose ethinylestradiol therapy versus combined conjugated equine estrogens and medroxyprogesterone acetate therapy. *Am J Obstet Gynecol* 2003;188:92-9

Soares C, Almeida O, Joffe H, Cohen L. Efficacy of estradiol for the treatment of depressive disorders in perimenopausal women. *Arch Gen Psychiatry* 2001;58:529-34

Soma M, Fumagalli R, Paoletti R, Meschia M, Maini MC, Crosignani P, Ghanem K, Gaubatz J, Morrisett JD. Plasma Lp(a) concentration after oestrogen and progestagen in postmenopausal women. *Lancet* 1991;337:612

Sporrøng T, Hellgren M, Samsioe G, Mattsson LÅ. Metabolic effects of continuous estradiol-progestin therapy in postmenopausal women. *Obstet Gynecol* 1989;73:754-8

Stadberg E, Mattsson LÅ, Uvebrant M<sup>1</sup>. 17β-estradiol and norethisterone acetate in low doses as continuous combined hormone replacement therapy. *Maturitas* 1996;23:31-9

Stadberg E, Mattsson LÅ, Uvebrant M<sup>2</sup>. Low doses of 17β-estradiol and norethisterone acetate as continuous combined replacement



therapy in postmenopausal women: lipid metabolic effects. *Menopause* 1996;3:99-6

Stahl SM. Effects of estrogen on the central nervous system. *J Clin Psychiatry* 2001;62:317-8

Staland B. Continuous treatment with a combination of estrogen and gestagen-a way of avoiding endometrial stimulation. *Acta Obstet Gynecol Scand Suppl.* 1985;130:29-35

Stampfer M, Colditz G. Estrogen replacement therapy and coronary heart disease: a quantitative assessment of the epidemiologic evidence. *Prev Med* 1991;20:47-63

Stein J, Rosenson R. Lipoprotein Lp(a) excess and coronary heart disease. *Arch Intern Med* 1997;157:1170-6

Steiner M. Premenstrual dysphoric disorder. An update. *General Hospital Psychiatry* 1996;18:244-50

Stevenson DW, Delprato DJ. Multiple component self-control program for menopausal hot flashes. *J Behav Ther Exp Psychiatry* 1983;14:137-40

Stevenson J, Crook D, Godsland I. Influence of age and menopause on serum lipids and lipoproteins in healthy women. *Atherosclerosis* 1993;82:638-43

Studd J, Arnala I, Kicovic P, Zamblera D, Kröger H, Holland N. A randomized study of tibolone on bone mineral density in osteoporotic postmenopausal women with previous fractures. *Obstet Gynecol* 1998;92:574-9

Sturdee D, Brincat M. The hot flush. In Studd J, Whitehead M, editors. *The Menopause*. Oxford: Blackwell Scientific Publications, 1988, pp 24-42

Sundström I, Ashbrook D, Bäckström T. Reduced benzodiazepine sensitivity in patients with premenstrual syndrome: A Pilot Study. *Psychoneuroendocrinology* 1997;22 (1): 25-38

Sundström I, Nyberg S, Bixo M, Hammarbäck S, Bäckström T. Treatment of premenstrual syndrome with gonadotropin-releasing hormone agonist in a low dose regimen. *Acta Obstet Gynecol Scand* 1999;78:891-9

Sveindóttir H, Bäckström T. Prevalence of menstrual cycle symptom cyclicity and premenstrual dysphoric disorder in a random sample of women using and not using oral contraceptives. *Acta Obstet Gynecol Scand* 2000;79:405-13

The writing group for PEPI trial. Effects of estrogen or estrogen/progestin regimens on heart disease risk factors in postmenopausal women. The postmenopausal estrogen/progestin interventions (PEPI) trial. *JAMA* 1995;273:199-208

The writing group for the PEPI trial. Effects of hormone replacement therapy on endometrial histology in postmenopausal women. The Postmenopausal Estrogen/Progestin Interventions (PEPI) Trial. *JAMA* 1996;275:370-5

Topo P, Køster A, Holte A, Collins A, Landgren BM, Hemminki E, Uutela A. Trends in the use of climacteric and postclimacteric hormones in Nordic countries. *Maturitas* 1995;22:89-95

Torgerson DJ, Bell-Syer SEM. Hormone replacement therapy and prevention of nonvertebral fractures. A meta-analysis of randomised trials. *JAMA* 2001;285:2891-7

Ulrich LG, Barlow DH, Sturdee DW, Wells M, Campbell MJ, Nielsen B, Bragg AJ, Vessey MP for UK continuous combined HRT study investigators. Quality of life and patient preference for sequential versus continuous combined HRT: the UK Kliofem® multicenter study experience. *Int J Gynaecol Obstet* 1997;59 suppl.1:11-7

Utian WH. Overview on menopause. *Am J Obstet Gynecol* 1987;156:1280-3

Vickers M, Meade T, Darbyshire J. WISDOM: history and early demise-was it inevitable? *Climacteric* 2002;5:317-25

Voigt LF, Weiss NS, McKnight B, Chu J, Daling JR, van Belle G. Progestagen supplementation of exogenous oestrogens and risk of endometrial cancer. *The Lancet* 1991;338:274-7

Walker SR and Rosser RM. *Quality of life: assessment and application*. Lancaster, England: MTP Press Limited, 1988

Wang M, Seippel L, Purdy R, Bäckström T. Relationship between symptom severity and steroid variation in women with premenstrual syndrome: study on serum pregnenolone, pregnenolone sulfate, 5 $\alpha$ -pregnane-3,20-dione and 3 $\alpha$ -hydroxy-5 $\alpha$ -pregnan-20-one. *J Clin Endocrin Metab* 1996;81:1076-82

Weiderpass E, Adami HO, Baron JA, Magnusson C, Bergström R, Lindgren A, Correia N, Persson I. Risk of endometrial cancer following estrogen replacement with and without progestins. *J Natl Cancer Inst* 1999;91:1131-7

van de Weijer P, Barentsen R, de Vries M, Kenemans P. Relationship of estradiol levels to breakthrough bleeding during continuous combined hormone replacement therapy. *Obstet Gynecol* 1999;93:551-7

Whitehead MI, Townsend PT, Pryse-Davies J, Path FRC, Ryder TA, King RJB. Effects of estrogens and progestins on the biochemistry and morphology of the postmenopausal endometrium. *N Engl J Med* 1981;305:1599-605

Whitehead M, Townsend P, Pryse-Davies J, Ryder T, Lane G, Siddle N, King R. Effects of various types and dosages of progestogens on the postmenopausal endometrium. *J Reprod Med* 1982;27:539-48

Wiklund I, Karlberg J, Mattsson LÅ. Quality of life of postmenopausal women on a regimen of transdermal estradiol therapy: A double-blind placebo-controlled study. *Am J Obstet Gynecol* 1993; 168:824-30

Woodruff JD, Pickar JH. Incidence of endometrial hyperplasia in postmenopausal women taking conjugated estrogens (Premarin) with medroxyprogesterone acetate or conjugated estrogens alone. *Am J Obstet Gynecol* 1994;170:1213-23

Wyon Y, Lindgren R, Lundeberg T, Hammar M. Effects of acupuncture on climacteric vasomotor symptoms, quality of life, and urinary excretion of neuropeptides among postmenopausal women. *Menopause* 1995;2:3-12

Wyon Y, Frisk J, Lundeberg T, Theodorsson E, Hammar M. Postmenopausal women with vasomotor symptoms have increased urinary excretion of calcitonin gene-related peptide. *Maturitas* 1998; 30:289-94

Yaffe K, Sawaya G, Lieberburg I, Grady D. Estrogen therapy in postmenopausal women: effects on cognitive function and dementia. *JAMA* 1998;279(9):688-95

Ziel HK. Estrogen's role in endometrial cancer. *Obstet Gynecol* 1982; 60:509-15

APPENDIX 1

Tag med detta kort vid varje besök

Start tabl

Dag

Man

Ar

PAT. ID

Rand. nr

Initialer

BLÖDNINGAR

B = blödning    S = spotting    0 = ingen blödning, ingen spotting

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
Jan																														
Feb																														
Mars																														
April																														
Maj																														
Juni																														
Juli																														
Aug																														
Sept																														
Okt																														
Nov																														
Dec																														
Reservmån.																														
Reservmån.																														

TABLETTER

2 = 2 tabletter    1 = 1 tablett    0 = 0 tablett

1	2	3	4	5	6	7	8	9	10	11	12	13	14	15	16	17	18	19	20	21	22	23	24	25	26	27	28	29	30	31
Jan																														
Feb																														
Mars																														
April																														
Maj																														
Juni																														
Juli																														
Aug																														
Sept																														
Okt																														
Nov																														
Dec																														
Reservmån.																														
Reservmån.																														

Anteckning: (specifera dag/månad/år för din notering)

Stop tabl

Dag

Man

Ar

Instruktion: Uttrycket blödning används för all blödning från slidan som kräver användning av någon form av binda eller tampong. Termen spotting används för all blödning från slidan som inte kräver skydd. Markera varje dag när blödning inträffar med stora B och varje dag som spotting inträffar med stora S. Om ingen blödning eller spotting, markera med 0. Markera tabletttaget med en eita (1) för en tablett och tvåa (2) för två tabletter. Glömmer du ta tabletterna markera med 0.

Ansvarig prövare, signatur:

## APPENDIX 2

										HUVUDVÄRK										SVULLNAD										GLAD										SPÄND OCH OROLIG									
										9	6	3	1	9	6	3	1	9	6	3	1	9	6	3	1	9	6	2	1																				
										Huvud- värk				Svullnad				Glad				Spänd och orolig																											
										Namn:.....																																							
										Födelsedatum:.....																																							
										Månad 1...../ 2.....																																							
										Årtal.....																																							
										"Cyklitets-diagnoser " av Professor Torbjörn Bäckström Skattningsskala nummer 1.																																							
										299																																							

Dag 1

Start datum: 

1

Dag 1

Start datum: 

2

9 6 3 1 9 6 3 1 9 6 3 1 9 6 2 1

Sömnstörningar Energisk Irritabel Trött och orkeslös

SÖMNSTÖRNINGAR

Namn:.....

ENERGISK

Födelsedatum:.....

Månad 1..... / 2.....

Årtal.....

IRRITABEL

"Cykllicitets-diagnoser" av Professor Torbjörn Bäckström  
Skattningsskala nummer 2.

299(

TRÖTT OCH  
ORKESLÖS

Dag 1

Start datum: 

3

9631963196319621

Nedstämd  
Bröst-  
spänningar

Svettningar

Påverkan

- 1=Ingen Påverkan
- 2=Jag märker
- 3=Familjen märker
- 4=Stör relationer i familjen
- 5=Undviker socialt umgänge
- 6=Avstått från socialt umgänge
- 7=Svårigheter att arbeta
- 8=Ej klarat arbetet
- 9=Frånvaro från arbetet

Namn:.....

Födelsedatum:.....

2..... Månad 1...../.....

Årtal.....

Torbjörn Bäckström  
"Cyklicitets-diagnoser" av Professor Skattningskala nummer 3.

2990

NEDSTÄMD

BRÖSTSPÄNNINGAR

SVETTNINGAR

PÅVERKAN





## APPENDIX 3

### HORMONRELATERADE EFFEKTER PÅ HUMÖRET

PREMELLE 9601 SW

Center	Patient no	Patient Initial	Screening	Date of interview
□ □ □	□ □ □	— — —		____/____/____ Day Month Year

**Markera med X i lämplig ruta för frågorna 1-5 samt 8-9**  
**Frågorna 6 och 7 är öppna frågor.**

ja      nej

1. Har du tidigare haft humörsvängningar före menstruation (PMS) tex irriterabel..... ☐      ☐  
nedstämd..... ☐      ☐

Om nej gå vidare till fråga 4.

2. Vilken påverkan har det haft på Dig?  
Ingen påverkan..... ☐  
Jag märker..... ☐  
Familjen märker..... ☐  
Störd relation i familjen..... ☐  
Undviker socialt umgänge..... ☐  
Avstått från socialt umgänge..... ☐  
Svårigheter att arbeta..... ☐  
Ej klarat arbete..... ☐  
Frånvaro från arbete..... ☐

3. Dessa besvär försvann de när menstruationen startade?..... ☐      ☐

4. Om Du tidigare ätit p-piller, har Du då upplevt negativa humörförändringar eller humörbiverkningar?..... ☐      ☐

Om ej använt p-piller gå vidare till fråga 6.

5. Om ja, Var det anledning till att Du slutade med Dina p-piller?..... ☐      ☐

Om ej varit gravid, gå vidare till fråga 8.

6. Hur har Du psykiskt mått under Din graviditet/graviditeter? \_\_\_\_\_  
\_\_\_\_\_

7. Hur mårde Du efter att barnet/barnen fötts? \_\_\_\_\_  
\_\_\_\_\_

8. Har Du tidigare behandlats vid något tillfälle för psykisk åkomma, såsom depression..... ☐      ☐  
ångest..... ☐      ☐

9. Sömnstörningar, brukar Du använda eller har Du använt sömnmedel under perioder i Ditt liv?  
Aldrig..... ☐  
Någon gång, vid speciell händelse..... ☐  
Regelbundet periodvis..... ☐  
Alltid..... ☐