

New hormonal therapies and regimens in the postmenopause: routes of administration and timing of initiation

R. Sitruk-Ware

Population Council and Rockefeller University, New York, USA

Key words: POSTMENOPAUSE, HORMONAL THERAPY, ROUTES OF ADMINISTRATION, TIMING OF INITIATION, ESTRADIOL, PROGESTERONE, PROGESTINS, SUSTAINED-RELEASE SYSTEMS

ABSTRACT

Since the publication of the Women's Health Initiative (WHI) study followed by the results of the Million Women Study (MWS), the role of hormonal therapy in postmenopausal women has been further challenged. The risks attributed to hormone therapy have been overestimated and the data has been wrongly extrapolated to the whole class of therapies.

The trends in postmenopausal hormonal therapy seem now to favor the non-oral delivery routes for both the estrogen and the progestin for women with an intact uterus, based on the assumption that a lesser stimulation of the liver proteins and a neutral metabolic profile would be more favorable in terms of cardiovascular and venous risk.

The combination of non-oral administration of estradiol and local delivery of progesterone or a progestin such as levonorgestrel by means of gels, sprays, vaginal rings or intrauterine systems would represent new methods of replacement therapy for the menopausal woman, improving compliance and minimizing the risks of hormone replacement. Several of these systems are either available or in development.

Long-term studies on the risk/benefit of various non-oral formulations are certainly warranted.

INTRODUCTION

Since the publication of the large randomized controlled trials and observational studies of hormonal therapy (HT), the management of postmenopausal symptoms as well as long-term prevention of chronic diseases has been an area of ongoing controversies^{1–6}.

While it is well admitted that progesterone or a progestin is recommended for women with an intact uterus receiving estrogen therapy, the impact of progestins combined with estrogen on

other target organs, especially breast cells, has been challenged³. Unfortunately, the results from the Women's Health Initiative (WHI) trial, where conjugated equine estrogens (CEE) combined with medroxyprogesterone acetate (MPA) were studied, were extrapolated to all other combinations of estrogen and progestins without discrimination, while the molecules available for HT are quite different in their pharmacological and pharmacodynamic profiles. Also, non-oral delivery of the

Correspondence: Professor R. Sitruk-Ware, Center for Biomedical Research, Population Council, 1230 York Avenue, New York, New York 10021, USA

sex steroids induces different responses as compared to oral delivery.

Transdermal administration of the natural hormone estradiol in postmenopausal women prevents the increase in the liver production of estrogen-sensitive proteins such as sex hormone binding globulin (SHBG), high density lipoprotein (HDL) and angiotensinogen⁷. Recently, the risk of venous thromboembolism has been shown to differ significantly when transdermal estradiol was used as compared with oral estradiol⁸. Whether the difference is related to a differential impact of estradiol on clotting factors synthesized in the liver is not confirmed. In addition, more recent data show that the category of progestins added to estradiol modifies the risk⁹.

The risks attributed to the progestins have also been overestimated and the data from the WHI conducted with MPA have been wrongly extrapolated to the whole class of compounds, while the risk may vary according to the nature of the progestin as well as the dose and duration of use¹⁰. Recently, a trend has been observed for the use of natural progesterone derivatives, based on observational data suggesting less impact on breast cancer risk^{10,11} as well as the use of non-oral progestins and progesterone, on the assumption that very low circulating levels of progesterone will have less negative impact on breast cancer risk, if any.

Progesterone can be delivered from a gel or a ring in the vagina, leading to high local concentrations in the endometrium and low circulating serum levels, below the threshold of 10 nmol/l. This is described as a 'first uterine pass' delivering progesterone from the vagina and resulting in high local concentration in the uterus with very little systemic distribution of the hormone, avoiding the potential systemic side-effects^{12,13}. The vaginal ring delivering progesterone at a dose of 10 mg/day over 3 months would also ensure better compliance, as the system is in place for 3 consecutive months and withdrawal bleeding would occur only four times a year¹⁴.

Levonorgestrel can also be delivered at very low doses in the uterine cavity from an intrauterine system (IUS) that is active for 5 consecutive years. Here also, the local concentrations of progestin in the endometrium are far above the plasma levels measured in users of the system¹⁴. These two approaches may allow the protection of the endometrium due to the direct effect of progesterone or levonorgestrel in the tissue in postmenopausal women with an intact uterus, while very low levels reach the systemic circulation.

In addition, other approaches such as transdermal (gels and spray) or nasal delivery of estrogen and progestins are under development, although, in those cases, the endometrial protection relies on the circulating levels of the progestins that would reach, later on, the uterine target.

Combination of non-oral administration of estradiol and local delivery of progesterone or a progestin such as levonorgestrel would represent new methods of replacement therapy for the menopausal woman, improving compliance and minimizing the risks of hormone replacement. Long-term studies comparing various non-oral formulations to placebo to confirm this hypothesis are certainly warranted.

NON-ORAL ROUTES OF DELIVERY FOR ESTROGEN

Among the molecules available to the prescriber, the natural estrogens are usually preferred to synthetic steroids for substitutive estrogen therapy.

To avoid the intensive first-pass metabolism following the intake of 17 β -estradiol, other routes of administration of estradiol have been sought and delivery through injections, implants, vaginal rings, and transdermal systems (gels, transdermal sprays, patches) as well as transmucosal nasal sprays has been successfully realized. With these systems of parenteral estradiol administration, premenopausal serum levels of estradiol are achieved with lower levels of estrone, resulting in a more physiologic estradiol/estrone ratio¹⁵⁻¹⁷.

The serum levels obtained and the time to reach peak concentrations of circulating estrogen vary greatly, according to the type of estrogen used and the route of administration considered. After parenteral administration of estrogens, the first-pass gastrointestinal and liver metabolism is avoided and therefore the plasma levels measured reflect more accurately the dose delivered and absorbed.

Vaginal delivery

The vaginal epithelium rapidly absorbs estrogens, and estradiol can be delivered through silastic rings¹⁸, giving relatively stable estradiol levels over several months. Nash and colleagues¹⁹, using rings delivering 60–140 μ g/day estradiol, were effective in correcting menopausal symptoms over 6 months. Mean estradiol levels were 123 ± 48 and 307 ± 93 pmol/l with the low- and high-dosage levels, respectively. Estrone levels exceeded

estradiol levels by 1.7-fold in the high-dosage ring and 2.6-fold for the lower-dosage ring. Given the more recent recommendations for HT, these doses of estradiol are rather high and lower delivery rates should be evaluated.

Vaginal creams of estradiol can also lead to high plasma estradiol levels²⁰.

Transdermal delivery

The first method used for skin delivery of estradiol was the *percutaneous* application of estradiol dissolved into a water-alcohol solvent, in the form of a gel containing 3 mg of estradiol per 5 g of gel, leading to a plasma concentration of 110–124 pg/ml²¹ after repeated administration. The usual recommended dose with this system is 1.5 mg/day, leading to about 50–60 pg/ml in plasma, which allow the relief of symptoms. This mode of skin delivery of estradiol has been described as *percutaneous* administration and differentiated from the *transdermal* delivery using transdermal delivery systems or patches.

The first generation of rate-controlled reservoir systems was designed for a twice-weekly application. More recently, second-generation systems have been introduced using the matrix dispersion-type systems, including systems designed for a once-weekly application. The main goal of the development of second-generation transdermal delivery systems was to avoid ethanol in the reservoir system, potentially responsible for the skin irritation leading to treatment discontinuation in about 5% of the cases. The new systems have better adhesive properties and a thinner and flat appearance. Further development was then conducted in two directions: first, the addition of a progestin into the same transdermal system was made for sequential or continuous combined therapy. Second, smaller patches have been designed in order to improve their acceptability.

In order to develop smaller patches such as the 'dot' systems, different enhancers have been used²² in order to penetrate the skin on a very limited surface area, as little as 5 cm². Therefore, although the application of a steroid on the skin is absorbed proportionally to the surface of application, the technical improvement of the systems would allow the steroid delivery on a much smaller surface than with the first generation of transdermal delivery systems.

The Metered Dose Transdermal System[®] (MDTS, Acrux Pty Ltd, Melbourne, Australia) is a precisely engineered spray that is capable of transferring a number of molecules onto the skin,

from where they can be rapidly absorbed into the dermis with the assistance of enhancers, using sunscreen technology (ACROSS[®] enhancers, Acrux, Melbourne) to form a 'depot within the skin' (Patchless Patch[™] Delivery, Acrux, Melbourne). Preliminary experience with estradiol and also with testosterone suggests that this is a viable system for the low-dose delivery of steroids²³. This delivery system is under development, under the name of Evamist[®], for estradiol delivery; initial experience suggests that an 'invisible' skin delivery from a spray will present a great level of appeal to users.

Nasal delivery

The benefits of intranasal estrogen therapy have also been examined. It was shown that 300 µg/day of 17β-estradiol delivered in an aqueous spray formulation is effective at reducing symptoms, is well tolerated and well accepted by the women²⁴. The pharmacokinetics of intranasal estradiol differ from other delivery forms. Maximal plasma levels are reached within 10–30 min and then decrease to 10% of the peak value after 2 h. The plasma profile is described as a pulse-like delivery, as opposed to the sustained delivery described with transdermal or vaginal administration. Low levels of estrone and SHBG observed in women receiving estradiol nasal spray indicate that the first-pass hepatic metabolism is avoided. Although plasma levels are low between pulses, efficacy has been shown by symptom correction equivalent to that of oral estradiol and side-effects were not different between oral or nasal treatment²⁴.

PROGESTERONE AND PROGESTINS: ROUTES OF DELIVERY

Certain progestins, because of their progestational potency, are effective in low doses and thus ideally suited for sustained-release delivery via implants, vaginal rings, or transdermal systems, e.g. gel or patch²⁵. Due to the uterine first-pass effect, administration of progesterone vaginally via gels or rings allows the delivery of the steroid directly into the target organ, without high serum levels, thus limiting the systemic action of progesterone. Sustained-release systems also allow steady release of low doses of steroids for extended durations, with compliance rates generally exceeding those observed with oral therapy¹⁴.

Those women with intact uteri, taking estrogen alone, are at increased risk of developing endometrial cancer. Unopposed estrogen treatment

leads to continuous glandular cell proliferation, causing over-stimulation of the endometrial tissue. This potentially leads to endometrial hyperplasia and endometrial cancer. Shulman observed 12.8% hyperplasia with 45 $\mu\text{g}/\text{day}$ of estradiol given alone without progestin²⁶ and the Postmenopausal Estrogen/Progestin Interventions (PEPI) trial found that unopposed estrogen treatment (0.625 mg/day CEE) was associated with a significantly increased risk of adenomatous or atypical hyperplasia, compared with estrogen/progestin regimens, 34% vs. 1%²⁷. To counter estrogen's effects on the uterus, progesterone or progestins are recommended for women with intact uteri^{15,25,26}.

Although effective in preventing endometrial hyperplasia and adenocarcinoma, recent concerns have been raised regarding the apparent link between progestin use and breast cancer and coronary heart disease. The WHI trial found that women taking a combination of CEE and MPA had an increased risk of breast cancer and coronary heart disease, in contrast with findings in women given CEE alone^{1,2}.

The WHI trial studied the effects of only one dose and oral formulation of progestin, MPA, and therefore study conclusions cannot be broadly applied to all progestins. MPA, the most commonly prescribed synthetic progestin used for HT, is a derivative of 17-hydroxyprogesterone that exhibits some androgenic and glucocorticoid effects^{16,25}. This may account for some of the cardiovascular effects observed in its clinical use. It has already been shown in animals that different progestins have different cardiovascular effects and Miyagawa and colleagues²⁸, comparing the effects of progesterone and MPA on the primate model, showed that progesterone plus estradiol protected, but MPA plus estradiol failed to protect against coronary artery vasospasm. In humans, Rosano and colleagues²⁹ compared the effects of estrogen/transvaginal progesterone gel with estrogen/MPA on exercise-induced myocardial ischemia in postmenopausal women with coronary artery disease, or previous myocardial infarction, or both. They showed that the combination of estrogen and transvaginal progesterone gel increased exercise time to ischemia compared with estrogen/MPA.

There is also some indication that synthetic progestins and natural progesterone differ in their effects on target organs, particularly on breast tissue. Synthetic compounds are seen to have a detrimental effect on breast tissue compared to natural compounds. In several recent epidemiological observational studies, the use of various

synthetic progestins combined with estrogen was observed to increase the risk of breast cancer, while the use of natural progesterone did not^{3,10,11}. However, there are no randomized, controlled studies demonstrating that a natural progesterone/estradiol combination would be better than using CEE with MPA (the hormones used in the WHI study). Nevertheless, the conclusions drawn by the observational studies support the use of natural progesterone over the use of synthetic progestins in HT¹¹. But, since there is no concrete evidence on which form of progesterone is superior to another, the current trend in prescribing HT is to maintain low but therapeutic levels of progestins to ensure their beneficial effects against endometrial cancer, while avoiding high circulating levels and potential deleterious effects on the breast and heart.

New methods of sustained-release delivery recently developed include implants, vaginal rings or transdermal systems, e.g. gel, metered dose transdermal system or patch¹⁴.

Although progestins vary in progestational and antiestrogenic potency, all progestins exert a progestational effect and oppose the proliferative action of estrogen on the endometrium. The potency of individual progestins determines the dose required to achieve these effects. Molecules such as levonorgestrel and its derivatives, and Nestorone[®] are progestins that, because of their high progestational potency, pharmacokinetic properties, and activities related to their derivative molecule, have been successfully used in sustained-release delivery systems³⁰.

Nestorone is one of the most potent progestins when used parenterally and tested *in vivo*, using the McPhail index in immature rabbits and pregnancy maintenance tests in female rats. With subcutaneous administration, Nestorone is 100 times more potent than progesterone and 10 times more potent than levonorgestrel³¹. Recently synthesized trimegestone, another 19-norprogesterone derivative, is somewhat more potent than Nestorone on the McPhail index, but less potent in the inhibition of ovulation test in rats.

Progestins with a short half-life when given orally may benefit from a parenteral administration, as its delivery via a sustained-release systems makes the molecule more available to the target tissues when delivered on a continuous basis. In addition, potent progestational molecules are best suited for delivery systems, as lower doses could be used for continuous daily delivery²⁵.

A range of easy-to-use and effective methods for contraception and hormone replacement therapy

are currently available to women or are in development. The most innovative methods are sustained-release systems that generally have high compliance rates and are either user-controlled or require no attention for extended periods of time. These include vaginal rings, transdermal patches and gels for the former and intrauterine systems for the latter.

Vaginal delivery

Vaginal delivery of progesterone/progestin is being considered as a logical approach to meet the current HT prescribing trend. Cicinelli and colleagues^{12,13} have previously demonstrated that the delivery of progesterone to the vagina in a gel formulation applied twice weekly causes high local concentrations in the endometrium and low circulating serum levels. The same authors demonstrated a first-uterine-pass effect of vaginal delivery of progesterone that could be attributed to the rich network of arteries and veins linking the vagina to the uterus¹³. Therefore, delivering progesterone to the vagina would result in high local concentrations in the uterus with very little systemic distribution of the hormone, avoiding the potential side-effects of systemic administration of progestins³².

Delivery of progesterone in a vaginal gel

The recent development of a controlled and sustained-release vaginal progesterone gel allowed single daily application and made prolonged use, such as for menopause, possible. De Ziegler and colleagues³³ studied two therapeutic options for HT using natural progesterone administered vaginally. A first group of 69 menopausal women received the sustained-release vaginal progesterone gel, Crinone[®] 4% (45 mg daily) from days 1 to 10 of each calendar month with estrogens taken continuously. A second group of 67 women received Crinone 4% twice weekly in conjunction with continuous estrogen therapy. Endometrial thickness was evaluated before and after 6 months of treatment. Histological verification was obtained in all cases of abnormal bleeding. At 6 months, 63 out of 69 (91.9%) women receiving progesterone cyclically experienced predictable withdrawal bleeding. The vast majority, 54 (80.6%) of 67 women receiving Crinone in constant combined association with estrogen therapy, remained amenorrheic throughout 6 months of therapy. All cases of abnormal bleeding were biopsied and no hyperplasia was seen. These results indicate that

both regimens using the sustained-release vaginal progesterone gel controlled bleeding in HT. Combined with the lower incidence of side-effects characteristic of vaginal progesterone, it appeared that both vaginal progesterone regimens have the potential of improving HT compliance.

In another study, Cicinelli and colleagues³⁴ used capsules of oral progesterone 100 mg for vaginal application every other day and showed that this route of absorption was feasible, and, when used with estrogen for 3 years, resulted in an atrophic endometrium in all subjects.

In another study, 20 menopausal women deprived of ovarian function were given estrogen for 12 days and a combined therapy of estrogen (administered orally) and progesterone for another 12-day period³⁵. Progesterone was administered vaginally through a gel (Crinone) utilizing a bioadhesive, biocompatible polymer as a base to achieve a sustained-release effect. After the estrogen-progestogen therapy, whatever the dose of progesterone given, a secretory transformation of the endometrial mucosa occurred, mitotic activity decreased significantly, more ramified and coiled glands were observed, and a decrease in progesterone receptor content was noted in epithelial and stromal nuclei; a decrease in progesterone receptor content was also observed in epithelial nuclei but not in stromal nuclei.

Accurate new techniques of image analysis have shown that this progesterone vaginal therapy could eliminate the proliferative effects of estrogen treatment in postmenopausal women, despite doses as low as 45 mg progesterone administered vaginally every other day. The results suggest that the sustained-release effects of the vaginal gel are clinically relevant.

Delivery of progesterone from a vaginal ring

While gel application of progesterone twice weekly may be acceptable to some women, it may be less convenient to others and compliance would vary greatly according to the population studied. As an alternative, a vaginal ring delivering progesterone may meet with higher acceptability. Previous studies conducted with vaginal rings, both for contraception and hormone therapy, indicated a high acceptability of the method, as the long-acting system is under the user's control and does not require a health provider for insertion^{36,37}. In addition, the ring does not require daily attention and remains in place without being noticed by the woman or her

partner. Partner awareness has been recorded at around 20% in contraceptive studies³⁸. Therefore, a vaginal ring delivering a low dose of natural progesterone in a steady state would result in low plasma levels and protection of the endometrium in women receiving estrogen therapy³⁵. Also, it may be assumed that this low dose of systemic progesterone would not have the deleterious effect upon the breast and heart seen with MPA.

Results of a previous study with a progesterone vaginal ring in *in vitro* fertilization (IVF) programs and oocyte donation give an indication as to the progesterone doses that might be effective in HT. It was shown that 6 mg/day estradiol and 20 mg/day progesterone for 2 weeks followed by 7 weeks of 10 mg/day progesterone were sufficient to transform the endometrium into secretory tissue capable of allowing embryo implantation and pregnancy maintenance³⁵. The estrogen dosage in this case was three to six times higher than the doses recommended for HT. It was therefore concluded that lower doses of progesterone will be sufficient for endometrial protection. It would appear feasible to transform postmenopausal endometrium with half the dose of progesterone used in the IVF program. Additionally, a low dose of estrogen could be used to alleviate menopausal symptoms and, if given transdermally, would avoid the first-pass effect on the liver^{15,16}.

As the WHI study suggested, the risks of using a synthetic progestin such as MPA in HT seem to be greater than the risks observed when CEE is used alone^{1,2}. But for women who have their lives compromised by menopausal symptoms and still have their uterus, combination HT is still necessary. Other authors proposed the use of ultra-low doses of estrogen without progestins and the monitoring of endometrial changes by ultrasound, but the endometrial thickness was shown to increase, even with very low doses of unopposed estrogen³⁹. In addition, the potential benefits of progesterone and some progestins on other targets such as the brain⁴⁰ or the vascular system⁴¹ may justify its use besides the protection of the endometrium. To have available to these women an HT regimen that may offer less risk and is effective and easy to use would represent a significant therapeutic advance.

Endometrial progesterone concentrations have been shown to be higher in women receiving progesterone vaginally versus intramuscularly. Miles and colleagues compared vaginal delivery of progesterone to intramuscular administration and showed that endometrial progesterone concentrations were higher with vaginally

administered progesterone than endometrial concentrations observed in normal ovulatory women or women who consistently had the highest serum progesterone after intramuscular administration⁴². These experiments confirm that vaginal delivery would result in a higher concentration of progesterone in the uterus relative to the low levels found systemically.

Delivery of steroids by the vaginal route offers many advantages and research continues on refinements of the vaginal ring delivery method.

Progesterone/estradiol vaginal ring

One novel method currently in development is the vaginal ring releasing low doses of natural steroids for HT. Results of a clinical study testing a ring delivering both estradiol and progesterone indicate that this ring can protect the endometrium and provide sufficient steroid levels to alleviate menopausal symptoms⁴³. Fifty-five postmenopausal women used vaginal rings releasing 150 µg/day estradiol, a rather high dose of estrogen in the light of current recommendations for the use of the lowest possible dose of estrogen, and either 9 mg/day or 5 mg/day of progesterone for 6 months. Both high- and low-dose rings achieved mean serum estradiol levels of 69 pg/ml at 2 weeks and 36 pg/ml at 6 months. Mean progesterone levels with the low-dose ring were 2.8 ng/ml at 2 weeks and 1.4 ng/ml at 6 months; the high-dose ring achieved progesterone levels of 4.8 and 2.6 ng/ml at 2 weeks and 6 months, respectively. Both doses effectively inhibited endometrial proliferation, with no dose-dependent effect. Over half of the subjects on both doses were amenorrheic. A marked decline in hot flashes and night sweats was noted with use of high- and low-dose rings, and significant reductions in pain at intercourse ($p < 0.02$) and vaginal dryness ($p < 0.001$) were reported at month 1. Mood was significantly elevated in the majority of subjects during treatment. The two most frequent complaints, bleeding and spotting and breast tenderness, were generally limited to the first 2 months of ring use.

Transdermal delivery

Gels

Progestins can also be delivered through gels or transdermal delivery. The transdermal method is especially advantageous for progestins that are inactive orally yet have effective transdermal

absorption, such as Nestorone. While Nestorone given orally is rapidly metabolized and inactive, Nestorone applied transdermally is highly active, resulting in good systemic bioavailability. A Nestorone transdermal gel is currently in development. Preliminary results of a 3-month study indicate that Nestorone is readily absorbed through the skin and, because of its potent progestational and anti-ovulatory effects, has been able to suppress ovulation in the majority of subjects^{30,44}. In addition, Nestorone doses in the same range have been tested with estradiol and were shown to transform the endometrium into a secretory state in postmenopausal women³⁰. These encouraging results would open new avenues both in contraception and HT.

Transdermal patches

The transdermal patch is another sustained-release, user-controlled contraceptive method that is capable of delivering progestins through the skin to the systemic circulation. It has been used for more than a decade for HT and several doses and formulations are available for estrogen therapy. As far as the progestin is concerned, norethisterone acetate (NETA) has been essentially used for HT in combination transdermal systems also delivering estradiol. Four-day systems delivering low doses of 140 $\mu\text{g}/\text{day}$ of the progestin NETA combined with 50 $\mu\text{g}/\text{day}$ estradiol have been shown to prevent endometrial hyperplasia in postmenopausal women and also maintain bone mineral density⁴⁵. Levonorgestrel has also been used in 7-day delivery systems for HT and protects the endometrium from over-proliferation at doses as low as 10 or 20 $\mu\text{g}/\text{day}$ ⁴⁶.

Metered dose transdermal system

Fraser and colleagues aimed to test metered spray delivery of a precise dosage of Nestorone progestin as a possible transdermal progestin-only contraceptive (submitted). Six healthy postmenopausal volunteers, not recently using any hormonal therapies, volunteered for this first study. Each subject was studied on two occasions with multiple blood sampling for assay of Nestorone over a 24-h period, on the first occasion after a single dosage of $3 \times 50 \mu\text{l}$ Nestorone sprays using a specially devised, precisely metered delivery device, and on the second occasion following the fifth in a series of five daily transdermal dosages of $3 \times 50 \mu\text{l}$ of Nestorone spray. Conventional

pharmacokinetic parameters were calculated. Nestorone was assayed in serum using a specific radioimmunoassay.

Mean serum levels of Nestorone peaked at around 20 h following dosing, and levels plateaued at 285–290 pmol/l after 4–5 days of daily spray application. The apparent elimination half-life of Nestorone after the last dose on day 5 was 26.8 h. No unexpected adverse events were encountered.

This early pharmacokinetic trial of a new transdermal steroid delivery showed the feasibility of using Nestorone and reaching plasma levels that would be sufficient to block ovulation. Lower doses should be tested for HT.

Intrauterine delivery

The levonorgestrel intrauterine system (LNG-IUS or Mirena[®]), approved in the US in 2000, has been developed by the Population Council as an effective contraception for up to 5 years^{47,48}. This small T-shaped device, inserted into the uterus by a clinician or, in some countries by a nurse practitioner or a midwife, slowly releases 20 $\mu\text{g}/\text{day}$ levonorgestrel, without serum levonorgestrel fluctuations. In the first few weeks, plasma levonorgestrel levels range from 150 to 200 pg/ml. At 2 years, serum levonorgestrel levels are at 192 pg/ml and at 5 years fall to 159 pg/ml⁴⁸. The LNG-IUS prevents pregnancy primarily via a local effect on the cervix and the endometrium. In addition, the use of this system for HT has been recognized as a potential novel way of delivering progestins to the endometrium with minimal systemic effects, and the system has been approved for HT in several countries⁴⁸. New systems delivering lower levels of levonorgestrel per day are under development and should become a major route of delivering progestins for HT in the future⁴⁹.

Because of its strong antiproliferative action on the endometrium, the LNG-IUS is a logical approach for protection from endometrial hyperplasia during estrogen treatment for climacteric symptoms^{47,48}. This approach also allows the free choice of estrogen with regard to type, dose and mode of administration (oral, transdermal, etc.). The downside of this regimen is that insertion of the LNG-IUS is a specific procedure; the insertion may be more difficult in women with an atrophic uterus due to hypoestrogenism and, as such, may not be acceptable for all women. The use of the LNG-IUS is especially suitable for the perimenopausal phase (a phase beginning at the start of

menstrual irregularity and ending 1 year after the final menstrual period), when contraception is still needed but the ovaries show variable activity, with phases of both hypo- and hyperestrogenism⁵⁰. In perimenopausal women, menorrhagia associated with uterine myomas and adenomyosis is frequent, and the system has also proven effective in the management of this condition⁴⁸. The tissue concentrations of levonorgestrel in the uterus with local dosing far exceed the uterine concentrations when levonorgestrel is given systemically, which explains the marked endometrial suppression and amenorrhea.

With LNG-IUS use in women on estrogen therapy, the endometrium remains uniformly suppressed and non-proliferative, even with high systemic levels of exogenous estrogen^{51,52}. The use of oral continuous combined HT regimens is limited to the postmenopausal phase only and therefore the LNG-IUS combined with oral or transdermal estrogen is one of the few methods available to avoid the withdrawal bleedings associated with sequential HT regimens. The LNG-IUS has been shown to be highly effective in preventing estrogen-induced endometrial hyperplasia. However, spotting may occur as with other HT regimens, which should be mentioned during counseling. As the effective lifespan of the LNG-IUS is 5 years, and the size of the uterus tends to decrease (involute) after menopause, a system with smaller dimensions and with 3 years' usage time, together with a lower release rate of 10 µg/day (the menopausal levonorgestrel system, MLS) has been studied⁴⁹. With MLS, the systemic levonorgestrel concentration is approximately 40% of that of the system with the 20 µg/day release rate. The authors reported high acceptability and tolerability, although spotting was frequently observed during the first year of use. The changes in the lipid profile during use of the MLS and estrogen therapy are very close to those achieved with estrogen-only therapy.

Outcome on breast proliferation and breast cancer risk

A large prospective study assessed the relationship between breast cancer and use of the LNG-IUS⁵³. This large post-marketing study on LNG-IUS users ($n = 17\,360$) was carried out in Finland. The results present the breast cancer incidence comparison between LNG-IUS user data and the data on the average Finnish female population (derived from the Finnish Cancer Registry), between 30 and 54 years of age. Based on the

95% confidence intervals for the incidences of breast cancer, and the Fisher exact test, there was no indication of a difference between the LNG-IUS users and the average Finnish female population in any of the 5-year age groups. The incidence rates per 100 000 woman-years were, for the age groups 30–34 years, 27.2 and 25.5 ($p = 0.84$); for 35–39 years, 74.0 and 49.2 ($p = 0.056$); for 40–44 years, 120.3 and 122.4 ($p > 0.99$); for 45–49 years, 203.6 and 232.5 ($p = 0.41$); and for 50–54 years, 258.5 and 272.6 ($p = 0.85$), in the LNG-IUS group and in the Finnish female population, respectively. The results suggest that the use of the LNG-IUS is not associated with an increased risk of breast cancer.

These findings are reassuring in view of the previous publication from the Million Women Study where all progestins used in HT, including oral levonorgestrel, were found to induce a two-fold increase in breast cancer risk³. However, the occurrence of breast cancer found in the very first years of HT in that observational study seemed to point toward the promotion of latent yet undiagnosed cancers rather than to a carcinogenic effect of the treatments⁶. Also, the only large, randomized, controlled trial of the WHI showed that, in women who were non-previous users of HT (CEE + MPA) at entry in the trial, the risk of breast cancer was not significantly increased up to 6 years of use in the study (hazard ratio 1.02; 95% confidence interval (CI) 0.77–1.36)⁵⁴.

Another prospective study in a large French cohort studied the impact of various progestins used in HT^{10,11} and found no increase in risk in users of natural progesterone as compared with synthetic progestins. Although that study was not randomized, controlled but observational in design, it suggested a difference of impact according to the nature of the progestin. Based on this information, the hypothesis as to whether the action of progesterone on breast tissue differed from other progestins and also differed according to the route of administration of the steroid needs to be assessed.

Wood and colleagues⁵⁵ evaluated the potential action of progesterone on breast cell proliferation when delivered orally or from vaginal rings. This experiment was a two-way cross-over study in which 20 ovariectomized adult female cynomolgus macaques were treated (in equivalent doses for women) with oral estradiol (1 mg/day) + oral micronized progesterone (200 mg/day) or intravaginal progesterone delivered by small silastic rings (6–10 mg/day release rate). Hormone treatments lasted 2 months and were separated by a

1-month wash-out period. The primary outcome measure was breast epithelial proliferation. Serum progesterone concentrations were significantly greater following estradiol + oral progesterone (10.9 ng/ml) compared to estradiol + intravaginal progesterone (3.8 ng/ml) at 2–3 h after oral dosing ($p < 0.0001$) but not at 24–28 h after oral dosing (2.9 ng/ml for oral progesterone versus 3.2 ng/ml for intravaginal progesterone at 2 months, $p = 0.19$). Markers of breast proliferation, sex steroid receptor expression, and endometrial area did not differ significantly between estradiol + oral progesterone and estradiol + intravaginal progesterone treatments ($p > 0.1$ for all). Despite different pharmacodynamic profiles, oral and intravaginal progesterone had similar effects on biomarkers in the postmenopausal breast, although lower than previously observed with other progestins such as MPA^{55,56}.

Potential advantages of non-oral delivery of progesterone and progestins

The objective of delivering progesterone or other progestins from non-oral delivery systems has been of using the lowest possible dose of progestin necessary to oppose the estrogen effect on the endometrium with low systemic levels, and bypassing the metabolism of the steroids in the liver, as occurs after oral intake of progesterone or progestins. One of the potential advantages would be to avoid the conversion of progesterone into allopregnanolone, which may induce sleepiness and mood changes when high doses of progesterone are ingested orally. However, oral progesterone has been shown to improve sleep in postmenopausal women and this is an added benefit of the treatment⁵⁷. Another possible benefit would be to avoid the modification of liver proteins such as clotting factors. However, when progestins have been used without estrogen, little change has been observed in the hemostatic system⁵⁸. Progesterone or progestins that exert no glucocorticoid action should not modify the clotting factors, as suggested by Kuhl^{16,58}.

Many women choose not to take HT and women with contraindications to estrogen need alternative options⁵⁹. For women surviving breast cancer who need alternatives for estrogen, a tailored treatment strategy is needed. Treatments such as statins have been shown to prevent heart disease and therapies such as bisphosphonates, parathormone or calcitonin are available for both prevention and treatment of osteoporosis.

TIMING OF ADMINISTRATION

The WHI studies were designed to examine the effects of estrogen and progestin (CEE + MPA) and estrogen alone in postmenopausal women^{1,2}. About 70% of the population enrolled in the WHI was aged over 60 years, and a third of the cohort was 50–59 years old, representing those women reaching the time of menopause. The analyses of the outcomes should have been made for each age group in order to draw recommendations appropriate for each age group. The subgroup analyses were reported only a few years after the initial results were spread by the media^{4,6,60}. Grodstein and colleagues⁶⁰ reviewed the data from their observational study and prospectively examined the relation of HT to coronary heart disease (CHD), according to timing of hormone initiation relative to age and time since menopause. They found that women beginning HT near menopause had a significantly reduced risk of CHD (relative risk (RR) 0.66, 95% CI 0.54–0.80 for estrogen alone; RR 0.72, 95% CI 0.56–0.92 for estrogen with progestin), and later use after more than 10 years of menopause provided no cardiac protection.

Therefore, recent reviews and evidence from experimental studies have suggested that HT initiated at the time of menopause is associated with less development of cardiovascular disease^{4–6,60}. In contrast, if therapy is initiated over 10 years after the menopause, HT may be harmful, as the pre-existing plaques on the vessel walls may be more likely to rupture⁴.

Clark⁵ published a re-evaluation of these studies based on the graphic analysis of their tabulated data. In contrast to the conclusions reached by the WHI, he concluded that treatment of postmenopausal women with estrogen and progestin (CEE + MPA) does not increase the risks of cardiovascular disease, invasive breast cancer, stroke or venous thromboembolism. He also challenged the conclusion that an increased risk of stroke existed in women treated with estrogen alone. This re-evaluation of the WHI data, considered together with the re-appraisal of Philips and colleagues⁴ and Grodstein and colleagues⁶⁰, suggests that inappropriate consequences of the large, randomized, controlled trial occurred and a new approach should be made to the treatment recommendations for postmenopausal women⁶.

IMPACT OF HT ON COGNITIVE FUNCTION ACCORDING TO AGE

In 2003, the results of the Women's Health Initiative Memory Study (WHIMS), a randomized,

double-blind, placebo-controlled clinical trial, concluded that estrogen plus progestin therapy increased the risk for probable dementia in postmenopausal women aged 65 years or older⁶¹. In addition, estrogen plus progestin therapy did not prevent mild cognitive impairment in these women. The overall hazard ratio (HR) for probable dementia was 2.05 (95% CI 1.21–3.48; $p=0.01$). Alzheimer's disease was the most common classification of dementia in both study groups. Treatment effects on mild cognitive impairment did not differ between groups (HR 1.07; 95% CI 0.74–1.55; 63 vs. 59 cases per 10 000 person-years; $p=0.72$).

These results did not match the biological effects of estrogen and progesterone on the brain and further analysis indicates, however, that the only subgroup where the risk of dementia was increased was that of women aged 75 and over and who were first users of HT. The HR was 2.34 (95% CI 1.11–4.94) in the 803 women aged 75–79; in the 3531 women who had no prior hormone use, the HR was 1.98 (95% CI 1.13–3.47), both results being statistically significant. In contrast, in those women who had used HT in the past ($n=1001$), the HR was not significant at 2.69 (95% CI 0.52–13.85). The same wide CI and non-significant results were found in the younger age group, 65–69-year-old, who had a HR of 3.25 with a 95% CI of 0.66–16.11, including the value 1, and hence not statistically significant.

In other words, the only group where the results were significant and indicate that the treatment used in the WHI was harmful, was that of women who started HT for the first time at age 75–79. Those women who may have already developed arterial disease may have experienced a sudden worsening of their condition, triggered by the high dose of steroids they received including MPA, which has been shown to reverse the beneficial effect of estrogen on neuronal survival while progesterone and 19-norprogesterone were synergistic with estradiol⁶².

Another important point to consider relates to the cumulative effects of HT over the years of treatment. In the WHI study, more than 2000 women per treatment arm had previously used hormones prior to enrolment in the study, and some for many years. The wash-out period of 3 months before enrolment is of no value for the risks that are cumulative. From previous epidemiological studies and from data on the doubling time of breast cancer cells, it is established that the breast cancer risk observed under estrogen therapy is cumulative. Therefore conclusions on breast

cancer risk should not be made on the total cohort of women in this study but be based only on the naïve patients who did not use hormones prior to entering the WHI study. These women, 6280 in the active group and 6024 in the placebo group, should have been examined separately for all the outcomes. Indeed, the subgroup analysis of women who never used hormones before enrolment in the trial indicated no increase in risk of breast cancer (non-users: HR 1.06, 95% CI 0.81–1.38). Women who were prior users of HT before entering the study carried a much higher risk and account for the total HR of 1.26 observed for the total study population (prior use <5 years: HR 2.13, 95% CI 1.15–3.94; users for 5–10 years: HR 4.61, 95% CI 1.01–21.02)^{2,54}.

Should the excess of eight cases of breast cancer included in the global index be due essentially to the 25% of the study population who used hormones before the study, the calculation of the Global Index of risks and benefits may have been different. Should the increased risk of vascular events also be affected by the subgroup of previous hormone use, or by the age at entry as we have seen in later re-analyses, the conclusions of the study may have been different.

CONCLUSIONS

Several new options for delivery of estradiol or other estrogens, progesterone or progestins are now either under development or already available for hormonal therapy.

The hypothesis, that lower doses of the progestins that can be delivered more steadily from non-oral formulations may have less impact on the other target organs than the uterus, remain to be confirmed. However, non-oral delivery avoids the peaks and troughs in the kinetics of absorption of the steroids and therefore is likely to minimize the side-effects. Combinations of non-oral estradiol and progesterone or other progestins with a chemical structure close to the native hormone, also delivered non-orally, have been tested and proved acceptable and well tolerated.

There is a growing body of evidence that each mode of delivery, type of estrogen and progestin, and timing of therapy have distinct beneficial and adverse effects, and the results pertaining to one regimen should not be extrapolated to other types of therapy. This likely applies especially to the unwanted effects of HT, particularly on breast cancer and cardiovascular events. The ultimate effect of an HT regimen depends on the combined

effects of that particular regimen on the vessels, the brain and the breast tissue^{4,6,63}.

Use of HT for a few years at around the time of the menopause will relieve symptoms, improve quality of life and not lead to decrease or increase in osteoporosis, cardiovascular disease or cancer. With long-term use, in some women the benefits far outweigh the risks, while for others the risks outweigh the benefits. The use of HT has to be tailored to the needs and risk factors of each individual. The negative results obtained from some randomized, controlled trials in the treatment of women *with* established disease such as atherosclerosis or Alzheimer's disease, should not be extrapolated to a lack of preventive effect in women *without* such conditions. Prescribing molecules close to the natural hormones, estradiol and progesterone, with no deleterious action on the metabolic factors would be preferable. Counseling the woman on the risk/benefits of therapy and involving her in the decision process are of paramount importance.

Good medical sense should be exercised for each individual case. Recommendations would be different for women with an early onset of menopause, women who are first users of

hormones or women who are in their sixties and have previously used HT for several years.

Each woman is unique and has her own risk profile. Therefore HT should be tailored to her profile and her preferences and adjusted according to her response. Following these recommendations may lead to an increased compliance and could bring health benefits with minimal risks, enhancing the overall quality of life of the postmenopausal woman.

The contents of the manuscript reflect the personal views of the author.

Conflict of interest No conflict related to any products mentioned in paper. The author is an employee of the Population Council which initially developed the levonorgestrel IUS and the progesterone ring and which owns Nestorone[®].

Source of funding The author thanks USAID, UNFPA, NICHD, CONRAD, the Hewlett Foundation, the Noble Foundation and the Hecht Foundation who partly supported the Population Council for the studies conducted during the development of some of the delivery systems described in the manuscript.

References

- Rossouw JE, Anderson GL, Prentice RL, *et al.* 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
- The WHI Steering Committee. Effect of conjugated equine estrogen in postmenopausal women with hysterectomy: the Women's Health Initiative randomized controlled trial. *JAMA* 2004;291:1701–12
- Beral V. Million Women Study Collaborators. Breast cancer and hormone-replacement therapy in the Million Women Study. *Lancet* 2003;362:419–27
- Phillips LS, Langer RD. Postmenopausal hormone therapy: critical reappraisal and a unified hypothesis. *Fertil Steril* 2005;83:558–66
- Clark JH. A critique of Women's Health Initiative Studies (2002–2006). *Nucl Recept Signal* 2006;4:e-23
- Pines A, Sturdee DW, Birkhäuser MH, *et al.*, on behalf of the Board of the International Menopause Society. IMS Updated Recommendations on postmenopausal hormone therapy. *Climacteric* 2007;10:181–94
- De Lignieres B, Basdevant A, Thomas G, *et al.* Biological effects of estradiol-17 beta in postmenopausal women: oral versus percutaneous administration. *J Clin Endocrinol Metab* 1986; 62:536–41
- Scarabin P-Y, Oger E, Plu-Bureau G, *et al.* Differential association of oral and transdermal oestrogen-replacement therapy with venous thromboembolism risk. *Lancet* 2003;362:428–32
- Canonico M, Oger E, Plu-Bureau G, *et al.* Hormone therapy and venous thromboembolism among postmenopausal women: impact of the route of estrogen administration and progestogens: the ESTHER study. *Circulation* 2007; 115:840–5
- Fournier A, Berrino F, Riboli E, *et al.* Breast cancer risk in relation to different types of hormone replacement therapy in the E3N-EPIC cohort. *Int J Cancer* 2005;114:448–54
- Fournier A, Berrino F, Clave-Chapelon F. Unequal risks for breast cancer associated with

- different hormone replacement therapies: results from the E3N cohort study. *Breast Cancer Res Treat* 2007 Feb 27; [Epub ahead of print].
12. Cicinelli E, de Ziegler D. Transvaginal progesterone: evidence for a new functional 'portal system' flowing from the vagina to the uterus. *Hum Reprod Update* 1999;5:365-72
 13. Cicinelli E, de Ziegler D, Bulletti C, *et al.* Direct transport of progesterone from vagina to uterus. *Obstet Gynecol* 2000;95:403-6
 14. Sitruk-Ware R. Delivery options for contraceptives. *Drug Discov Today* 2005;10:977-85
 15. Sitruk-Ware R. Hormonal replacement therapy. A review. *Rev Endocrine Metab Disorders* 2002;3:243-56
 16. Kuhl H. Pharmacology of estrogen and progestogens: influence of different routes of administration. *Climacteric* 2005;8(Suppl 1):3-63
 17. Samsioe G. Transdermal hormone therapy: gels and patches. *Climacteric* 2004;7:347-56
 18. Mishell DR Jr, Moore DE, Roy S, *et al.* Clinical performance and endocrine profiles with contraceptive vaginal rings containing a combination of estradiol and d-norgestrel. *Am J Obstet Gynecol* 1978;130:55-62
 19. Nash HA, Sanchez-Alvarez F, Mishell DR Jr, *et al.* Estradiol-delivering vaginal rings for hormone replacement therapy. *Am J Obstet Gynecol* 1999;181:1400-6
 20. Rigg LA, Hermann H, Yen SSC. Absorption of estrogens from vaginal creams. *N Engl J Med* 1978;298:195-7
 21. Sitruk-Ware R, De Lignieres B, Basdevant A, *et al.* Absorption of percutaneous oestradiol in postmenopausal women. *Maturitas* 1980;2:207-11
 22. Hossain M, Quebe-Fehling E, Sergejew T, *et al.* Comparative bioequivalence studies with Estradot and Menorest transdermal systems. *Maturitas* 2003;46:187-98
 23. Humberstone AJ, Evans AM, Davis SR. A comparative study of the pharmacokinetics of testosterone (T) following administration of a Metered-Dose Transdermal Spray (MDTS) to the abdomen or forearm in healthy postmenopausal women with low serum testosterone. Presented at *The Endocrine Society's 85th Annual Meeting*; 2003, Philadelphia, USA, 2003, Abstr.
 24. Studd J, Pornel B, Marton I, *et al.* for the Aerodiol Study Group. Efficacy and acceptability of intranasal 17 β -oestradiol for menopausal symptoms: randomized dose-response study. *Lancet* 1999;353:1574-8
 25. Sitruk-Ware R. New progestogens: a review of their effects in perimenopausal and postmenopausal women. *Drugs Aging* 2004;21:865-83
 26. Shulman LP, Yankov V, Uhl K. Safety and efficacy of a continuous once-a-week 17beta-estradiol/levonorgestrel transdermal system and its effects on vasomotor symptoms and endometrial safety in postmenopausal women: the results of two multicenter, double-blind, randomized, controlled trials. *Menopause* 2002;3:195-7
 27. 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
 28. Miyagawa K, Rösch J, Stanczyk F, *et al.* Medroxyprogesterone acetate interferes with ovarian steroid protection against coronary vasospasm. *Nature Med* 1997;3:324-7
 29. Rosano GM, Webb CM, Chierchia S, *et al.* Natural progesterone, but not medroxyprogesterone acetate, enhances the beneficial effect of estrogen on exercise-induced myocardial ischemia in postmenopausal women. *J Am Coll Cardiol* 2000;36:2154-9
 30. Sitruk-Ware R, Small M, Kumar N, *et al.* Nestorone[®]: clinical applications for contraception and HRT. *Steroids* 2003;68:907-13
 31. Kumar N, Koide S, Tsong Y-Y, Sundaram K. Nestorone: a progestin with a unique pharmacological profile. *Steroids* 2000;65:629-36
 32. de Ziegler D, Fanchin R. Progesterone and progestins: applications in gynecology. *Steroids* 2000;65:671-9
 33. de Ziegler D, Ferriani R, Moraes LA, Bulletti C. Vaginal progesterone in menopause: Crinone 4% in cyclical and constant combined regimes. *Hum Reprod* 2000;15(Suppl 1):149-58
 34. Cicinelli E, de Ziegler D, Alfonso R, Nicoletti R, Bellavia M, Colafiglio G. Endometrial effects, bleeding control, and compliance with a new postmenopausal hormone therapy regimen based on transdermal estradiol gel and every-other-day vaginal progesterone in capsules: a 3-year pilot study. *Fertil Steril* 2005;83:1859-63
 35. Casanas-Roux F, Nisolle M, Marbaix E, *et al.* Morphometric, immunohistological and three-dimensional evaluation of the endometrium of menopausal women treated by oestrogen and Crinone, a new slow-release vaginal progesterone. *Hum Reprod* 1996;11:357-63
 36. Zegers-Hochschild F, Balmaceda JP, Fabres C, *et al.* Prospective randomized trial to evaluate the efficacy of a vaginal ring releasing progesterone for IVF and oocyte donation. *Hum Reprod* 2000;15:2093-7
 37. Alexander NJ, Baker E, Kaptein M, *et al.* Why consider vaginal drug administration? *Fertil Steril* 2004;82:12

38. Novak A, de la Loge C, Abetz L, *et al.* The combined contraceptive vaginal ring, NuvaRing: an international study of user acceptability. *Contraception* 2003;67:187–94
39. Carranza-Lira S, Gooch AL, Velasco-Díaz G, Solano J, Arzola-Paniagua A. Low and ultra low-dose estrogen therapy for climacteric symptom control – preliminary report. *Int J Fertil Womens Med* 2006;51:171–5
40. Sitruk-Ware R, Kumar N. Progestins and the brain. Effects on sexuality and quality of life. http://www.6thimworkshop.com/sito/inside_pages/proceedings/sitruk_ware.pdf
41. Mallareddy M, Hanes V, White WB. Drospirenone, a new progestogen, for postmenopausal women with hypertension. *Drugs Aging* 2007; 24:453–66
42. Miles RA, Paulson RJ, Lobo RA, *et al.* Pharmacokinetics and endometrial tissue levels of progesterone after administration by intramuscular and vaginal routes: a comparative study. *Fertil Steril* 1994;62:485–90
43. Maruo T, Mishell DR, Ben-Chetrit A, *et al.* Vaginal rings delivering progesterone and estradiol may be a new method of hormone replacement therapy. *Fertil Steril* 2002;78:1010–16
44. Croxatto HB, Zepeda A. Transdermal contraceptive systems: innovative technology for the twenty-first century. In Sitruk-Ware R, Bardin W, eds. *Contraception. Newer Pharmacological Agents, Devices, and Delivery Systems*. New York: Marcel Dekker, Inc, 1992:101–15
45. Rubinacci A, Peruzzi E, Modena AB, *et al.* Effect of low-dose transdermal E2/NETA on the reduction of postmenopausal bone loss in women. *Menopause* 2003;10:241–9
46. Sturdee DW, van de WP, von Holst T. Endometrial safety of a transdermal sequential estradiol–levonorgestrel combination. *Climacteric* 2002;5:170–7
47. Luukkainen T, Lahteenmaki P, Toivonen J. Levonorgestrel-releasing intrauterine device. *Ann Med* 1990;22:85–90
48. Sitruk-Ware R, Inki P. The levonorgestrel intrauterine system: long-term contraception and therapeutic effects. *Women's Health* 2005; 1:171–82
49. Sturdee DW, Rantala ML, Colau JC, *et al.* The acceptability of a small intrauterine progestogen-releasing system for continuous combined hormone therapy in early postmenopausal women. *Climacteric* 2004;7:404–11
50. Burger HG, Dudley EC, Robertson DM, Dennerstein L. Hormonal changes in the menopause transition. *Recent Prog Horm Res* 2002; 57:257–75
51. Critchley H. Endometrial effects of progestogens. *Gynecol Forum* 2003;8:6–10
52. Jones RL, Critchley HO. Morphological and functional changes in human endometrium following intrauterine levonorgestrel delivery. *Hum Reprod* 2000;15(Suppl 3):S162–72
53. Backman T, Rauramo I, Jaakkola K, *et al.* Use of the levonorgestrel-releasing intrauterine system and breast cancer. *Obstet Gynecol* 2005; 106:813–7
54. Anderson GL, Chlebowski RT, Rossouw JE, *et al.* Prior hormone therapy and breast cancer risk in the Women's Health Initiative randomized trial of estrogen plus progestin. *Maturitas* 2006 20;55(2):103–15. Epub 2006 Jul 11
55. Wood CE, Sitruk-Ware R, Tsong YY, *et al.* Effects of estradiol with oral or intravaginal progesterone on risk markers for breast cancer in a postmenopausal monkey model. *Menopause* 2007;14:1–9
56. Wood CE, Register TC, Lees CJ, Chen H, Kimrey S, Cline JM. Effects of estradiol with micronized progesterone or medroxyprogesterone acetate on risk markers for breast cancer in postmenopausal monkeys. *Breast Cancer Res Treat* 2007;101:125–34
57. Montplaisir J, Lorrain J, Denesle R, *et al.* Sleep in menopause: differential effects of two forms of hormone replacement therapy. *Menopause* 2002;8:10–16
58. Kuhl H. Effects of progestogens on haemostasis. *Maturitas* 1996;24:1–19
59. Pinkerton J, Santen R. Alternatives to the use of estrogen in postmenopausal women. *Endocrine Rev* 1999;20:308–20
60. Grodstein F, Manson JE, Stampfer MJ. Hormone therapy and coronary heart disease: the role of time since menopause and age at hormone initiation. *J Women's Health (Larchmt)* 2006;15:35–44
61. Shumaker SA, Legault C, Rapp SR, *et al.* 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
62. Nilsen J, Brinton RD. Impact of progestins on estrogen-induced neuroprotection: synergy by progesterone and 19-norprogesterone and antagonism by medroxyprogesterone acetate. *Endocrinology* 2002;143:205–12
63. Sitruk-Ware R, Plu-Bureau G. Exogenous progestagens and the human breast. *Maturitas* 2004;49:58–66

Copyright of Climacteric is the property of Taylor & Francis Ltd and its content may not be copied or emailed to multiple sites or posted to a listserv without the copyright holder's express written permission. However, users may print, download, or email articles for individual use.