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Progesterone and related progestins: potential new health benefits

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ABSTRACT

Progesterone is a steroid hormone that is essential for the regulation of reproductive function. The main physiological roles of this hormone have been widely described. Progesterone and progestins have been approved for a number of indications including the treatment of irregular and anovulatory menstrual cycles and, when combined with estrogen, for contraception, and the prevention of endometrial hyperplasia in postmenopausal hormonal replacement therapy (HRT) regimens. Lack of understanding between the differences in categories of the progestins as well as with the physiological hormone has resulted in considerable controversy surrounding the use of progestins for HRT regimens. Newer evidence suggests that there are distinct differences between the molecules and there is no progestin class effect, with regard to benefits or side-effects. In addition to its role in reproduction, progesterone regulates a number of biologically distinct processes in other tissues, particularly in the nervous system and the vessels. Recently, it has been shown in animal experiments that progesterone and the progestin Nestorone® have positive effects on neuroregeneration and repair of brain damage, as well as myelin repair. The potential benefits of natural progesterone and its related derivatives warrant further investigation. It is hoped that a better understanding of the mechanism of action of progesterone and selected progestins will help in defining better therapies for men and women.

INTRODUCTION

The name progestin can be used interchangeably with progestogen, progestagen, gestogen, and gestagen[1]. The term 'progestin' is often used to denote a synthetic progestin. To clearly differentiate the synthetic hormones from the natural progesterone, the term 'progestin' will be used in this review for all synthetic molecules as opposed to the natural hormone progesterone[1,2].

Progesterone is a steroid hormone that is secreted by the ovaries and placenta and is essential in the complex process of reproduction. It is the main hormone involved in the maintenance of pregnancy and, based on its antigonadotropic properties, also prevents further ovulation after a corpus luteum is formed. Progesterone plays a central role in the secretory transformation of an estrogen-primed endometrium, a process which prepares the endometrium to receive and nourish a fertilized ovum. In addition, depending on the level of progesterone secretion and the duration of the luteal phase, progesterone (or a progestin) can prevent the over-proliferation of endometrial tissue[1,3]. Once a pregnancy occurs, progesterone is necessary for the full differentiation of breast tissue that occurs in preparation for lactation. In the mammary gland, progesterone acts synergistically with estrogen to transform the terminal end buds into differentiated lobules necessary for milk secretion[4].

The use of progesterone and progestins has been studied extensively for the treatment of different gynecological pathologies, as contraceptive agents[5,6], and in assisted reproductive technologies[3]. In replacement cycles, the secretory transformation of an estrogen-primed endometrium depends on the antiestrogenic properties of the progestin used, as well as on the dose and duration of treatment[1,2]. In women with anovulatory cycles or in those treated with estrogen, administration of a progestin for 12–14 days per month induces secretory transformation of the endometrium. Withdrawal of the progestin induces regular withdrawal bleeding.

The main indications for which progesterone and progestins have been approved include the treatment of irregular and anovulatory menstrual cycles and, when combined with estrogen, the prevention of endometrial hyperplasia in postmenopausal hormonal replacement therapy (HRT) regimens[1,2,7]. Progestins

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can be combined with estrogen for hormonal contraception[5], but have also been used without estrogen as progestin-only contraceptive agents in premenopausal women[6]. Natural progesterone is appropriate for use during *in vitro* fertilization (IVF) programs and for the maintenance of pregnancy after embryo transfer during oocyte donation programs[3]. Progesterone, administered via vaginal ring[7], has also been approved in a number of countries for contraceptive use during lactation[8,9].

While progesterone is widely recognized for its role in reproduction, it also regulates a number of biologically distinct processes in other tissues, particularly in the nervous system. Recently, it has been shown that progesterone and Nestorone®, a novel progestin with a chemical structure close to progesterone, have positive effects on neuroregeneration[10] and myelin repair[11] in the brain as well as on decreasing ischemia in an animal model of stroke[12]. These exciting results are opening new avenues for the development of therapeutic options for progesterone and related progestins beyond the reproductive field.

PHARMACOLOGY AND MODE OF ACTION

Comparative pharmacology of synthetic progestins

Over the past decade, several new progestins have been synthesized for use in contraceptives and HRT[1,2]. The progestins used so far are structurally related either to testosterone (19-nortestosterone derivatives which include the estranes (e.g. norethisterone) and gonanes (e.g. levonorgestrel, desogestrel, gestodene) or to progesterone (17-hydroxyprogesterone derivatives or pregnanes (e.g. medroxyprogesterone acetate (MPA), cyproterone acetate (CPA)), and 19-norprogesterone derivatives or norpregnanes (e.g. Nestorone®, nomegestrol acetate (NOMAc), trimegestone)[1] (Table 1). The new progestins synthesized in the last two decades were designed with the objective of creating the 'ideal' progestin, which would produce the benefits of progesterone, with strong progestational and antiestrogenic actions on the endometrium coupled with a strong antigonadotropic effect and without any androgenic, estrogenic or glucocorticoid receptor interaction to prevent unwanted side-effects.

Table 1 Progestational and other activities of synthetic progestins related to progesterone structure

Pure progestational
Nestorone
Nomegestrol acetate
Trimegestone
Antiandrogenic
Cyproterone acetate
Drospirenone, nomegestrol acetate, chlormadinone acetate
Partly glucocorticoid
Medroxyprogesterone acetate
Antialdosterone
Drospirenone

The various progestins available for contraception and HRT not only differ in their steroid receptor interactions but also in responses to *in vivo* bioassays and in pharmacokinetic profiles (Table 2).

While both norethisterone, also named norethindrone, and levonorgestrel bind to sex hormone binding globulin (SHBG), their elimination half-lifes ($t_{1/2\beta}$) vary, the terminal half-life being around 7–8 h for norethisterone and up to 26 h for levonorgestrel. In contrast, CPA has a $t_{1/2\beta}$ of 48 h. The norpregnanes also differ in their elimination half-life, NOMAc showing the highest, close to 50 h, while Nestorone is not active orally, similarly to progesterone in its crystal form. However, it shows a long half-life of 27 h or more when applied transdermally due to the reservoir effect of the epidermis[13].

Drospirenone is an antimineralocorticoid progestin derived from spirolactone and is rapidly absorbed after oral intake, reaching a peak plasma level after 1–2 h. Its bioavailability is about 76%[1]. Dienogest has a 19-nortestosterone structure but with a 17α-cyanomethyl instead of a 17-ethinyl group[1]. Its pharmacokinetics make it suitable for oral administration, since it has a high oral bioavailability (>90%) and is rapidly absorbed; the peak level (t_{max}) is reached after 2 h and the elimination half-life is around 10 h[1].

Mechanism of action of progesterone

Progesterone acts through binding to intracellular progesterone receptors (PRs)[14]. Progesterone receptor ligands have genomic effects that are mediated by nuclear receptors which modulate transcriptional activity in reproductive tissues. Progesterone receptors have been identified in many tissues outside of the female genital tract.

Two main isoforms of the progesterone receptor (PRA and PRB) have been described[14]. Both are encoded by the same gene and have common ligand-binding and DNA-binding domains. Results from studies using PR-knockout (PRKO) mouse models suggest that PRA and PRB act in a tissue-specific manner[14]. In mice, PRA controls estradiol-induced endometrial proliferation, whereas PRB appears to control breast differentiation and proliferation[14]. Following binding of the ligand to the specific ligand-binding domain, nuclear receptors interact with the transcriptional machinery through a large molecular complex which includes co-regulators. When recruited to the transcription initiation site of a target gene, some of the co-regulators activate transcription (co-activators), whereas others decrease the level of transcriptional activation (co-repressors)[15].

In addition to binding specifically to the PR with differing binding affinities, most progestins, according to their chemical structure, could also interact with the androgen receptor (AR), estrogen receptor (ER), glucocorticoid receptor (GR) or mineralocorticoid receptor (MR), which in turn may lead to agonistic or antagonistic actions according to the co-activators or co-repressors involved in the specific receptor interaction[1,14].

Table 2 Comparison of pharmacokinetic parameters of different progestins. Estranes (norethindrone/norethisterone); non-ethinylated estrane (dienogest); gonanes (levonorgestrel, desogestrel, gestodene); norpregnane (nomegestrol acetate); spirolactone derivative (drospirenone). Source: references 1, 2 and 5

		Distribution	*Elimination*
Progestin	*Bioavailability (%)*	*Half-life (α)* (h)	*Half-life (β)* (h)
Norethindrone/norethisterone	36–60	1.5	7–8
Levonorgestrel	80–87	0.6–1.3	12.6–26
Desogestrel	80–87	0.5–1.5	11.9–23.8
Gestodene	99	1.0–1.5	11.8–22
Cyproterone acetate	100		48
Nomegestrol acetate	63	1.9–3.6	28–51
Dienogest	92	0.6–2	6.5–12
Drospirenone	76	2	25–33

Progesterone has been shown to transactivate the PR but not the AR or the GR[16]. It has been also shown to exert antiandrogenic and antialdosteronic actions. Other progestins that transactivate the PR also transactivate the AR (levonorgestrel, MPA), or the GR (MPA) and therefore induce other effects than only progestational.

MPA transactivates both AR and GR, leading to related side-effects and therefore cannot be compared with natural progesterone[16]. The 19-norpregnane group relates structurally to progesterone with a high specificity to the progesterone receptor and includes promegestone, trimegestone, NOMAc and Nestorone[1,5]. This latter group binds specifically to the PR and exerts no androgenic, estrogenic or glucocorticoid action.

ADMINISTRATION OF PROGESTERONE FOR THERAPEUTIC USE

Progesterone has a molecular formula of $C_{21}H_{30}O_2$; the structural formula is shown in Figure 1. Progesterone cannot be administered orally unless made in a micronized formulation as it is quickly metabolized and possesses a very short half-life; the bioavailability is less than 10% with oral non-micronized formulations[1]. Progesterone is insoluble in water; therefore, for therapeutic purposes, it can be administered in oil-based formulations by intramuscular injection or vaginally via gel or ring formulation, or by oral capsules in a micronized formulation. Vaginal gels and rings have been developed for the treatment of progesterone deficiency and have an advantage over intramuscular injection because they are painless and can provide continuous steady-state delivery of progesterone[7,17]. Previously, it was demonstrated that, when progesterone was delivered vaginally via gel formulation applied every other day, high concentrations were found locally in the endometrium and low levels were found in the serum, a phenomenon described as the 'first-uterine-pass effect'[18]. Therefore, vaginal delivery of progesterone may avoid the potential side-effects of systemic administration of the hormone.

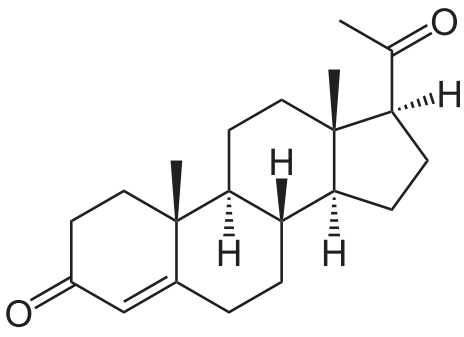

Figure 1 Structural formula of progesterone

Progesterone use for contraception during lactation

Progesterone has potential advantages for contraception during lactation[8,9]. In non-pregnant females, an elevated concentration of progesterone inhibits the cyclic release of luteinizing hormone and inhibits production of follicle stimulating hormone, thereby preventing further ovulation. The development of a non-oral, long-acting, sustained delivery system for progesterone, such as a progesterone vaginal ring (PVR), allows sustained delivery of the steroid which can reach serum levels of approximately 20 nmol/l, sufficient to inhibit ovulation[5,8]. The plasma progesterone levels required to inhibit fertility during breastfeeding with a PVR were reportedly in the range of 10–25 nmol/l[8]. In nursing mothers with lactational amenorrhea, it was shown that use of a PVR prolonged the period of lactational amenorrhea and provided effective contraception during the first postpartum year[8,19,20].

Nursing mothers who used a PVR for 1 year postpartum had a greater suckling-induced increase in prolactin and higher post-suckling prolactin levels than did those using a non-hormonal contraceptive (CuT intrauterine device), while follicle size remained smaller in women receiving progesterone[21].

Due to the rapid breakdown of progesterone after oral ingestion, infants are not affected by the small amount of the

steroid in their mother's milk. A similar effect has been described for Nestorone, which is not active orally. Therefore, progesterone and Nestorone are good candidates for use as contraceptives during lactation[22].

Progesterone indication for luteal maintenance during IVF

One of the major physiological roles of progesterone is to support and maintain pregnancy. Its use as a therapeutic agent in situations of luteal deficiency has therefore been proposed. However, given the low bioavailability of non-micronized oral progesterone, only intramuscular formulations have been developed for pregnancy maintenance. More recently, alternative routes of progesterone delivery have been developed, such as vaginal gels, rings, or suppositories[7,23].

The effects of vaginally administered progesterone on the estrogen-primed endometrium have been evaluated in premenopausal women undergoing assisted reproduction procedures[23,24]. After vaginal administration, relatively low serum or plasma concentrations of progesterone were sufficient to produce secretory transformation of the endometrium in agonadal women desiring pregnancy[23–25].

Therefore, the high tissue concentration of progesterone reached after vaginal administration would allow efficient replacement therapy despite lower serum levels than with injectables[26,27]. Besides vaginal delivery systems, new water-based injectable formulations are being developed to prevent the painfulness of injections of oily solutions.

Progesterone to prevent preterm birth

The use of exogenous progesterone during pregnancy remains controversial[28]. Progesterone and its synthetic form 17α-hydroxyprogesterone caproate offer an effective intervention when the continuation of pregnancy is at risk from immunological factors, luteal deficiencies, and myometrial hypercontractility[29]. Progesterone has been successfully used as prophylaxis in the prevention of spontaneous miscarriage, with treatment beginning from the first trimester of pregnancy. In addition, there is substantial evidence to indicate that women with idiopathic recurrent miscarriage may benefit from the immune-modulatory properties of progesterone in early pregnancy[30].

Progesterone is critical in maintaining uterine quiescence, and pharmacologic progesterone withdrawal is associated with increased uterine contractility[28,29]. The use of progesterone has therefore been proposed for preventing the recurrence of preterm birth. The proposed mechanisms supporting its use include: anti-inflammatory action and oxytocin antagonism as well as prevention of premature cervical ripening[30,31]. Trials in women at high risk for preterm delivery in Brazil using progesterone vaginal suppositories or gel demonstrated reduced risk of preterm delivery[32] and neonatal morbidity[33]. Progesterone, by its anti-inflammatory action and by modulating the mucosal immune environment, may protect against preterm birth[34].

PROGESTERONE AND RELATED MOLECULES WITH NEUROREGENERATION POTENTIAL

Neuroregeneration

Progesterone receptors have been identified in several areas of the brain[35] and have been implicated in a variety of functions, including cognition[36], neuroprotection[37,38], and dendritic remodeling[39]. In the hippocampal formation of female rats, progesterone counterbalances the effects of estrogen. Progesterone reduces the incidence of epileptic activity both directly[40] and through conversion to allopregnanolone[41]. In addition, progesterone has neuroprotective effects after traumatic or ischemic brain injury, both alone and after estrogen priming[42].

Neurogenesis

Liu and colleagues[10] in Brinton's laboratory reported on the efficacy of seven different progestins used alone or in combination with 17β-estradiol on adult rat neural progenitor cell (rNPC) proliferation and hippocampal cell viability *in vitro* and *in vivo*. *In vitro* analyses indicated that progesterone, norgestimate, Nestorone, norethynodrel, norethisterone, and levonorgestrel significantly increased rNPC proliferation, whereas norethisterone acetate was without effect, and MPA inhibited rNPC proliferation. Proliferative progestins *in vitro* were also neuroprotective. Acute *in vivo* exposure to progesterone and Nestorone significantly increased proliferating cell nuclear antigen and cell division cycle 2 expression and total number of hippocampal 5-bromo-2-deoxyuridine-positive cells, whereas levonorgestrel and MPA were without effect. In combination with 17β-estradiol, progesterone, Nestorone, levonorgestrel, and MPA significantly increased proliferating cells. However, proliferation induced by 17β-estradiol plus levonorgestrel or MPA was paralleled by a significant increase in apoptosis. A rise in Bax/Bcl-2 ratio paralleled apoptosis induced by levonorgestrel and MPA but not by progesterone or Nestorone[10]. These preclinical translational analyses indicate that clinical progestins vary dramatically in their impact on the regenerative capacity of the brain and thus are likely to have clinical implications for long-term neurological function.

Traumatic brain injury

The neuroprotective actions of progesterone have been documented by numerous experimental studies[42,43] and progesterone is now considered to be a promising therapeutic candidate for brain injuries[44]. This concept has recently

been translated into clinical practice[38,45]. Two phase-II trials have assessed the beneficial effects of progesterone after traumatic brain injury[46,47] and their encouraging outcomes have been followed by the initiation of two phase-III multicenter trials (Protect III, 2011, at http://www.clinicaltrials.gov/NCT00822900 and SyNAPSe, 2011, at http://www.synapse-trial.com).

Stroke

Stroke represents a major cause of death and neurological disability. The only approved treatment for acute stroke is thrombolysis with tissue plasminogen activator, but it can only be used in a small percentage of patients[48]. Progesterone is also a promising candidate for neuroprotective strategies after stroke[12] since experimental studies have demonstrated progesterone efficiency in reducing infarct volume and improving functional recovery after either transient or permanent occlusion of brain arteries[49–51].

Progesterone receptors are abundant not only within hypothalamic nuclei, involved in the control of reproductive functions, but also in cerebral cortex and subcortical structures[52,53]. PRs are crucial mediators of neuroprotection, as shown in a model of transient middle cerebral artery occlusion and PRKO mice[12]. Six hours after ischemia, rapid increases in progesterone and 5α-dihydroprogesterone, the endogenous PR ligands, were observed; in addition, PR deficiency, even haplo insufficiency, increases brain susceptibility to stroke damage. Within a time window of 24 h, PR-dependent signaling of endogenous brain progesterone limits the extent of tissue damage and the impairment of motor functions in mice[12]. Longer-term improvement requires additional treatment with exogenous progesterone and is also PR-dependent. These results suggest that PRs are linked to signaling pathways that influence susceptibility to stroke, and that they are key targets for both endogenous neuroprotection and therapeutic strategies after stroke. Allopregnanolone may also protect the brain against ischemic damage by other signaling mechanisms not involving PRs[54].

Liu and colleagues have also shown that the potent and selective PR agonist Nestorone was also effective in that model[12]. This 19-norprogesterone derivative shows high specific binding to the PR and is about 100 times more potent than progesterone[5,55]. $PR^{+/+}$ mice received three injections of Nestorone at doses 100 times lower than progesterone, given at 1, 6, and 24 h after middle cerebral artery occlusion, and neurological outcomes were examined at 48 h. When compared with oil treatment, Nestorone reduced the total infarct volume by 32% ($p \leq 0.01$), the ischemic lesions in cerebral cortex and in subcortical structures, respectively, by 22% ($p \leq 0.01$) and 52% ($p \leq 0.01$) and increased the time $P^{+/+}$ mice remained on the rotarod by 43% ($p \leq 0.05$)[12]. These data identified the PR as key for early endogenous neuroprotection and suggest that targeting the PR may be a realistic strategy for recovery after stroke.

Myelin repair

In two different animal models of demyelination, progesterone administered at the time of induction of experimental autoimmune encephalomyelitis (an animal model of multiple sclerosis) or cuprizone toxic demyelination partly *prevented* demyelination, either alone in intact female mice[56] or together with 17β-estradiol in male rats[57]. However, progesterone also has promyelinating actions which are now well documented by experimental studies, making it a particularly promising *therapeutic* agent for neurodegenerative diseases.

Progesterone promotes the formation of new myelin sheaths, which are necessary for axonal integrity. A role of progesterone in myelin formation was first demonstrated in peripheral nerves and dorsal root ganglia[58]. Progesterone also accelerates axonal myelination by oligodendrocytes during brain development, as shown in organotypic cultures of cerebellar slices taken from postnatal rats, at a stage when the myelination of axons is very intense[59]. The proremyelinating actions of progesterone involve the stimulation of oligodendrocyte progenitor cell (OPC) proliferation and maturation, require the presence of PR, and are not observed in PRKO rodents[59,60].

In multiple sclerosis, the endogenous capacity of myelin repair is limited and decreases progressively during relapsing/remitting episodes. Myelin repair requires recruitment, proliferation, and differentiation of OPC into mature oligodendrocytes[61]. It has been shown that steroid hormones such as progesterone and 17β-estradiol can stimulate this regenerative process. Indeed, in experimental models, progesterone and Nestorone promote the remyelination of axons by oligodendrocytes after lysolecithin-induced demyelination in organotypic cultures of cerebellar slices taken from postnatal rats or mice[11]. In contrast to Nestorone, the 17-hydroxypregnane derivative MPA had no effect. Intracellular PRs mediate the proremyelinating actions of progesterone and Nestorone, as no effect is seen in PRKO mice. Nestorone was less efficient in heterozygous mice, expressing reduced levels of PRs, suggesting PR haplo insufficiency in myelin repair. Nestorone stimulates the recruitment and maturation of OPC, two steps which are critical for efficient myelin repair. These data may thus open new perspectives for the use of progesterone and selected progestins, which target the PRs with high selectivity, to promote the endogenous regeneration of myelin[62].

Progesterone appears to be a promising therapeutic agent for the treatment of stroke, brain injury, and myelin repair. It is possible that molecules related to progesterone may also confer novel therapeutic benefits in the nervous system. Further investigation to understand the mechanism of action of progesterone and progestin is warranted.

PROGESTERONE IN HRT

There has been considerable controversy regarding the use of HRT since the Women's Health Initiative (WHI) Study was published in 2002[63]. This controversy is due, in part, to the confusion between progesterone and progestins, and the

incorrect belief that results related to MPA could be extrapolated to progesterone and all other progestins.

Stanczyk and colleagues[1] examined whether there is any reliable evidence to support the view for a general, uniform effect of progestins. The authors showed distinct differences between the progestins and progesterone, confirming the absence of a class effect[1].

Comparative activities of progestins used in HRT

17-Hydroxyprogesterone derivatives or pregnanes differ from one another and, while CPA shows the highest antiandrogenic potency, MPA – the most prescribed progestin in the USA for HRT and also used in the WHI – transactivates both the AR and the GR and exerts androgenic and glucocorticoid activities, therefore differing considerably from natural progesterone[1,16].

The 19-norpregnanes have a high specificity to bind the PRs, and not other receptors. They exert no androgenic, estrogenic or glucocorticoid action. Trimegestone seems the most potent of all progestins in the endometrial transformation test in the rabbit, and counters the uterotropic effect of estradiol in the immature mouse bioassay. In this test, trimegestone is more active than other progestins in the order trimegestone > Nestorone > levonorgestrel > MPA ≥ NOMAc.

The estrane and gonane progestins are less prescribed for HRT, although norethisterone is associated with estradiol in oral combinations, as well as transdermal patches. Levonorgestrel is the most androgenic of the nortestosterone molecules in the order levonorgestrel > desogestrel, gestodene > norgestimate, the last being slightly antiandrogenic rather than androgenic[1]. Differing from the above two categories, drospirenone and dienogest have been developed mostly for contraception. However, in postmenopausal women with mild hypertension, the combination of drospirenone with natural 17β-estradiol in hormone therapy has been shown to induce a decrease in blood pressure[1].

The antiandrogenic activity of dienogest has been found to be 40% of that of CPA, the most potent antiandrogenic progestin. Due to its high antiestrogenic effect on the endometrial tissue, dienogest has also the potential to be used in HRT and also in the treatment of endometriosis[5].

RISKS AND BENEFITS OF PROGESTINS VERSUS PROGESTERONE: VENOUS RISK

Progestins when given alone, such as progestin-only pills for contraception, carry little risk on cardiovascular outcomes and most progestins used in contraception and HRT do not modify the clotting factors[1,5]. In contrast to combinations of progestins with ethinylestradiol, when dienogest is associated with estradiol valerate[64], or NOMAc with estradiol[65], no or little changes in clotting factors were noted indicating that the hemostatic changes reported with oral contraceptivess are likely related to ethinylestradiol and not to the progestin component[5].

MPA has been shown to potentiate the vascular procoagulant effects of thrombin, an effect related to its glucocorticoid action[1,66].

The route of estrogen administration and type of progestin both seem important factors for venous risk in postmenopausal women using HRT. In a French cohort study, finding that oral but not transdermal estrogens were associated with increased thrombotic risk (hazard ratio (HR) 1.7; 95% confidence interval (CI) 1.1–2.8 and HR 1.1; 95% CI 0.8–1.8, respectively), no significant association with progesterone (HR 0.9; 95% CI 0.6–1.5), pregnane (HR 1.3; 95% CI 0.9–2.0) or nortestosterone derivatives (HR 1.4; 95% CI 0.7–2.4) was found. However, two norpregnanes when combined with estrogen showed increased risk (HR 1.8; 95% CI 1.2–2.7)[67]. The latter finding is in contrast with the absence of changes in clotting factors observed when norpregnane derivatives[65] are used alone or in combination with estradiol, and prescription biases are not excluded. However, it appears that transdermal estradiol alone or combined with natural progesterone does not increase thrombotic risk.

EFFECTS ON VESSELS

Very small structural changes in the progestin molecules may induce considerable differences in their effects on the surrogate markers of cardiovascular disease risk. The cardioprotective benefits of estrogen may be reversed by some progestins but not all molecules act similarly[1]. The cardiovascular side-effects of some progestins could be related to their androgenic or glucocorticoid effects. Some progestins may reverse the estrogen-positive effects on flow-mediated vasodilatation and peripheral vascular function.

In experiments conducted in ovariectomized female monkeys fed a highly atherogenic diet, estrogens preserved normal endothelial-dependent dilator responses of coronary arteries to acetylcholine, and progesterone did not alter this cardioprotective mechanism. While MPA diminished the endothelium-dependent vasodilation[68], NOMAc did not reverse the estrogen response[69].

Estradiol has been shown to trigger the expression of the endothelial nitric oxide synthase (eNOS) gene and increase the release of nitric oxide, causing relaxation of the vascular smooth muscle cells. Most of the arterial effects of progestins are mediated through PRs present in the arterial wall as well as through down-regulation of the ERs, but rapid effects may not be mediated by the nuclear receptor mechanism[70].

Simoncini and colleagues[71] compared the effects of drospirenone with progesterone and MPA; drospirenone led to a rapid activation of the eNOS and enhanced eNOS expression. These actions were PR-dependent[71]. Progesterone or drospirenone did not interfere with the induction or activation of eNOS by estradiol, while MPA did. NOMAc was found to preserve the beneficial impact of estradiol on nitric oxide formation in human endothelial cells[72].

Synthetic progestins with androgenic activity may decrease high density lipoprotein cholesterol, increase insulin resistance and impair glucose tolerance, important risk factors for cardiovascular disease. Drospirenone, a potent antimineralocorticoid progestin, which antagonizes the water and sodium retention effect of estrogens, may exert beneficial cardiovascular effects as it decreases blood pressure in menopausal women[1]. Natural progesterone and some of its derivatives, as well as the non-ethinylated drospirenone and dienogest, do not exert any androgenic effect and have no negative effect on the lipids or on the endothelial cells[1,70]. It is likely that progesterone and some new progestins may be neutral on cardiovascular disease risk, but this has not been confirmed by large, randomized, controlled trials.

EFFECTS ON THE BREAST

While *in vitro* studies demonstrate the dual proliferative and inhibitory effect of progestins on breast tissue growth, only long-term follow-up can determine the effect of different progestins on the risk of breast cancer.

The French E3N cohort study found that the association of estrogen–progestin combinations with breast cancer risk varied significantly according to the type of progestin: the relative risk was 1.00 (95% CI 0.83–1.22) for estrogen–progesterone, 1.16 (95% CI 0.94–1.43) for estrogen–dydrogesterone, and 1.69 (95% CI 1.50–1.91) for estrogen combined with other progestins[73]. A subsequent study has shown that the increased risk of breast cancer observed with combinations other than estrogen–progesterone and estrogen–dydrogesterone seems to apply preferentially to ER+ carcinomas, especially those ER+/PR−, and to affect both ductal and lobular carcinomas[74]. In PR− tumors, the risk may be related to the estrogen only. Observational studies may have some biases as the type of progestin selected by the prescriber according to the woman's history or breast conditions may influence the results.

Studies on breast cell proliferation have generated conflicting data according to the cell lines used and the model of experiment. When assessing effects of progesterone, MPA, drospirenone and Nestorone alone or combined with 17β-estradiol on T47-D breast cancer cell migration and invasion, it was observed that different progestins enhance the ability of breast cancer cells to move in the surrounding environment, but less for drospirenone[75].

Ten years after the WHI, the International Menopause Society has issued recent consensus statements identifying risks and benefits of the various therapies for menopausal women according to age groups[76]. Also, the United States Preventive Services Task Force recommended against the use of postmenopausal hormone therapy for the prevention of chronic conditions[77]. The recommendation included use of combined estrogen–progestin in postmenopausal women with a uterus, as well as estrogen-alone therapy in postmenopausal women who have had a hysterectomy. There was no difference made in the categories of hormones used.

The Endocrine Society has endorsed the statement but maintained a commitment to hormone therapy as an effective and relatively safe treatment for healthy women who are close in time to menopause and are seeking relief of symptoms[78]. Unfortunately, none of the recent statements addressed the difference between MPA and progesterone, even though the differences in action between the two molecules have been demonstrated in receptor interaction and transactivation[1,16] and *in vivo*, in vessels[79], brain[80], and heart[81]. It would be unfortunate if this confusion between the molecules deprives menopausal women of the well-known effects and benefits of progesterone.

Results from a recent Danish study[82] found that, when HRT, using natural 17β-estradiol instead of conjugated estrogens and norethisterone acetate instead of MPA, was started early in young postmenopausal women, there was a significant reduction in the risk of mortality, myocardial infarction, or heart failure. In addition, postmenopausal women who started the described HRT regimen early and used it for more than 10 years were not at significantly increased risk of breast cancer or stroke[82]. Here again, these results highlight the difference between the molecules used for HRT as well as the timing of therapy.

CONCLUSION

As described in this review, natural progesterone and its structurally related derivatives have a number of potential clinical benefits that warrant further investigation. The impact of progesterone and some progestins on the regenerative capacity of the brain has implications for the selection of hormonal formulations prescribed in pre- and postmenopausal women. There is considerable evidence to confirm the different effects of progestins, the distinctiveness of progesterone, and the lack of a progestin class effect[1,17]. Most of the adverse effects induced by medroxyprogesterone acetate in the WHI study should definitely not be extrapolated to the natural hormone or other molecules with high specificity for the progesterone receptor and no other steroid receptor interaction. Given the novel effects of progesterone and 19-norprogesterone identified recently in the brain, it is hoped that a better understanding of the mechanisms of action of these steroids will open up new therapeutic avenues for both women and men.

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Conflict of interest M. E. E. has no conflict of interest to declare. R. S. W. is an employee of the Population Council, a not-for-profit organization developing the progesterone vaginal ring for lactating women. She also lectured and advised Bayer Pharma and Merck on progestins used as contraception.

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