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Estradiol interacts with the cholinergic system to affect verbal memory in postmenopausal women: Evidence for the critical period hypothesis

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Abstract

Estradiol has been shown to interact with the cholinergic system to affect cognition in postmenopausal women. This study further investigated the interaction of estradiol and cholinergic system functioning on verbal memory and attention in two groups of healthy younger (ages 50–62) and older (ages 70–81) postmenopausal women. Twenty-two postmenopausal women were randomly and blindly placed on 1 mg of 17-beta estradiol orally for 1 month then 2 mg for 2 months or matching placebo pills after which they participated in three anticholinergic challenge sessions when verbal memory and attention were assessed. Subjects were administered either the antimuscarinic drug scopolamine (SCOP), the antinicotinic drug mecamylamine (MECA), or placebo. After the first challenge phase, they were crossed over to the other hormone treatment for another 3 months and repeated the challenges. Results showed that estradiol pretreatment significantly attenuated the anticholinergic drug-induced impairments on a test of episodic memory (the Buschke Selective Reminding Task) for the younger group only, while estradiol treatment impaired performance of the older group. The results suggest that younger subjects may experience more cholinergic benefit from estradiol treatment than older subjects, supporting the concept of a critical period for postmenopausal estrogen use.

Keywords: Estradiol; Cholinergic system; Verbal memory; Critical period

Introduction

Cognitive changes in women after menopause have been widely reported (Halbreich et al., 1995). The ability of estrogen therapy to reverse or prevent this decline is controversial. The evidence for the beneficial effect of estrogen therapy is robust in studies of women who have undergone surgical menopause (Phillips and Sherwin, 1992; Sherwin, 1988). These studies show that women who took estrogen therapy (ET) showed less cognitive decline and even some improvement relative to those who received placebo. Additionally, a number of epidemiological studies have shown that women who take hormones during and/or after the menopause transition perform better on cognitive tests than women who do not take hormone therapy (e.g., Duka et al., 2000; Jacobs et al., 1998; Maki, 2005; Resnick et al., 1997; Smith et al., 2001).

While many studies have shown positive effects of ET on cognition after menopause, there are a number of studies that showed no benefit (Barrett-Connor and Kritz-Silverstein, 1993; Binder et al., 2001; Ditkoff et al., 1991; Grady et al., 2002; Polo-Kantola et al., 1998). Recent findings from the Women's Health Initiative (WHI) and Women's Health Initiative Memory Study (WHIMS) have shown that the risk of diagnosis of dementia in women taking conjugated equine estrogen (CEE) alone and CEE and medroxyprogesterone acetate (MPA) was twice that of women in the placebo group (Shumaker et al., 2004, 2003). However, the WHI study was not designed as a study of cognition and was primarily a study of cardiovascular prevention, thus the women were older than is typical for postmenopausal hormone therapy. Thus, the data are at best conflicting regarding the usefulness for estrogen to positively affect cognition among older women.

In the WHI/WHIMS study the women were older, an average age of 65, when the study began. One hypothesis for the lack of a beneficial effect of estrogen and even the finding of negative effects is that the subject group was too old to benefit from the

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hormone therapy. In prior studies where the subjects were younger than 65 when they started estrogen therapy, beneficial effects were seen (Joffe et al., 2006; Phillips and Sherwin, 1992; Shaywitz et al., 2003; Sherwin, 1996; Woo et al., 2003). In addition to the WHI/WHIMS study, other studies with women over 65 have shown no benefit of estrogen treatment (Almeida et al., 2006; Viscoli et al., 2005; Yaffe et al., 2006). These findings have led to the proposal of a critical period for an estrogen benefit on cognition (Resnick and Henderson, 2002; Maki, 2006; Sherwin, 2007).

The current study examined the influence of age on the ability of 17-beta estradiol (E2) to affect cholinergic system functioning. A number of studies from the animal literature show that an intact cholinergic system is necessary to see a beneficial effect of estrogen treatment (e.g., Gibbs and Aggarwal, 1998; Tinkler and Voytko, 2005). For example, the detrimental effects of SCOP on memory for passive avoidance were attenuated when ovariectomized rats were given E2 (Gibbs et al., 1998). Voytko and colleagues (Tinkler and Voytko, 2005; Voytko, 2002) have shown that E2 modulates attentional performance in primates through its interaction with the cholinergic system. After ovariectomy, monkeys were impaired during invalid cues in an attention task. This impairment was improved after E2 and not after placebo (Voytko, 2002). Thus, E2 uses the muscarinic cholinergic system to influence visuospatial attention (Tinkler and Voytko, 2005). We have also demonstrated the E2–cholinergic interaction in postmenopausal women (Dumas et al., 2006). We found that 3 months of 1 mg of E2 per day attenuated the impairment seen during anticholinergic drug challenge on tests of attention and tests with a speed component.

Further evidence examining the estrogen–cholinergic interaction in older women has been shown in a single photon emission tomography (SPET) study by Norbury and colleagues (Norbury et al., 2007). They examined the relationship between muscarinic receptor density and estrogen therapy in younger premenopausal women and two groups of older postmenopausal women who were either long-term users of estrogen therapy or never-users. Younger women had more muscarinic receptors than either group of older women. Additionally, their data showed that ET users had a higher density of muscarinic receptors than never-users (Norbury et al., 2007). Thus, these data contribute to the accumulating evidence showing an important relationship between estrogen and the cholinergic system in postmenopausal women.

In the current study, we have continued to utilize both a muscarinic antagonist, scopolamine (SCOP) and a nicotinic antagonist, mecamylamine (MECA), in our E2–cholinergic model. Our prior study (Dumas et al., 2006) found interactions of E2 with both the muscarinic and nicotinic antagonists and have extended the investigation of these interactions in the current study. While the prior animal studies (e.g., Gibbs and Aggarwal, 1998; Tinkler and Voytko, 2005) and the imaging study by Norbury et al. (2007) showed specific evidence of the estrogen–muscarinic interaction, these studies did not assess the estrogen–nicotinic interaction. In addition, there was some indication that E2 also attenuated the effects of the anticholinergic drugs on a measure of verbal memory but this effect did not reach significance in our prior study (Dumas et al., 2006). However, only one dose of E2 was tested and the possibility exists that brain systems and/or cognitive operations may be differentially sensitive to E2 dose. In the current study we examined the effect of 2 mg per day of E2 in our cholinergic challenge model to further examine the E2–cholinergic interaction in younger and older postmenopausal women.

Methods

Subjects

Subjects were 22 cognitively normal women, ages 50–81, $M=65$ (SD=10.2). Four additional subjects passed the screening but withdrew before beginning hormone treatment because of the time commitment of the study. Subjects were stratified during recruitment by age into two groups. The younger group was ages 50–62 ($M=55.8$, SD=4.3, $N=11$). The older group was ages 70–81 ($M=74.3$, SD=3.7, $N=11$). See Table 1 for demographic characteristics.

Subjects were recruited through notices and advertisements in local newspapers and direct mailings. Subjects were required to be postmenopausal, without menses for 1 year and without surgically induced menopause. Exclusion criteria included smoking, a history of breast cancer, and use of hormone therapy during the last year. Twelve subjects had previously taken hormone or estrogen therapy after menopause. The length of time of prior hormone use ranged from 1 week to 6 years (see Table 1). Medical exclusion criteria for E2 treatment included: contraindications for hormone therapy, estrogen-dependent neoplasia, untreated blood pressure greater than 160/100, history of deep vein thrombosis or other thromboembolic disease, hepatoma, severe migraines or stroke on oral contraceptives, current use of barbiturates, rifampin, insulin, carbamezepine, oral hypoglycemics, antidepressants, or lipid-lowering drugs, known intolerance to conjugated estrogens, diabetes, untreated thyroid disease, clinical osteoporosis, and a history or presence of severe menopausal symptoms. In addition, the following exclusions applied to the challenge drugs: heavy alcohol (more than an average of 1 drink per day) or coffee use (more than three cups per day), significant cardiovascular disease, asthma, active peptic ulcer, hyperthyroidism, pyloric stenosis, narrow angle glaucoma, epilepsy, or current Axis I psychiatric disorders. The alcohol criterion was used to ensure that that subjects were not alcohol dependent, and the caffeine criterion was used to ensure that subjects would not experience caffeine withdrawal on testing days.

Upon meeting these criteria, subjects were approved for further screening at the University of Vermont (UVM) General Clinical Research Center (GCRC). After signing informed consent documents, subjects gave a medical history and underwent a physical and laboratory tests assessing hematopoietic, renal, hepatic, and hormonal function. Subjects were cognitively evaluated using the Mini Mental State Exam (MMSE; Folstein et al., 1975), Brief Cognitive Rating Scale (Reisberg et al., 1988), and the Mattis Dementia Rating Scale (DRS; Jurica et al., 2001) to establish a Global Deterioration Scale score (GDS) which rated the degree of cognitive impairment (Reisberg and Ferris, 1988). Subjects were required to have an MMSE score greater than or equal to 27, a DRS score of 123 or greater, and a GDS score of 1 or 2.

Behavioral screening consisted of a partial Structured Clinical Interview for DSM-IV-TR (SCID; First et al., 2001) to establish the presence/absence of Axis I psychiatric disorders. In addition, subjects completed the Beck Depression

Table 1
Demographic characteristics for younger and older subjects (Means and standard deviations)

	Young	Old
Age **	55.8 (4.3)	74.3 (3.7)
Education	14.6 (1.4)	14.5 (2.6)
Years since menopause **	5.8 (8.6)	22.0 (5.3)
Years on estrogen	0.85 (1.9)	0.83 (1.8)
Years since estrogen *	0.80 (1.32)	12.7 (12.0)
BMI	24.8 (4.1)	27.0 (4.5)

* $p<0.01$.
** $p<0.0001$.