Hot flashes and estrogen therapy do not influence cognition in early menopausal women

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ABSTRACT

Objective: To examine how menopausal symptoms and estrogen therapy (ET)–induced symptom relief affect cognition in early menopause.

Design: There were two components. Part 1 was a cross-sectional study of 37 healthy, recently postmenopausal women with diverse menopausal symptoms. Women were categorized as having low (n = 20) or high symptoms (n = 17) based on a validated symptom questionnaire. Women completed mood and sleep questionnaires and underwent cognitive testing, which included verbal memory, visual memory, emotional memory, and verbal fluency. Thirty-two of these women went on to part 2 of the study. Fourteen were randomly assigned to receive ET and 18 to receive placebo for 8 weeks. Before treatment and at 4 and 8 weeks, women completed the same measures as in part 1 of the study.

Results: High symptom women had more negative mood (P = 0.01) and lower quality sleep (P < 0.001) than low symptom women. Despite suffering from more menopausal symptoms, worse mood, and poorer sleep, women in the high symptom group performed the same on cognitive testing as women in the low symptom group. Women receiving ET had greater improvements in menopausal symptoms and sleep compared with those receiving the placebo (P ≤ 0.05). ET did not improve mood compared with placebo. Women receiving ET did not have any improvement in cognitive performance compared with those receiving the placebo.

Conclusions: Menopausal symptoms do not impair cognition. ET does not improve cognition despite alleviating symptoms and improving sleep in recently naturally menopausal women with diverse menopausal symptoms.

Key Words: Cognition – Estrogen therapy – Hot flashes.
flashes, night sweats, mood changes, and sleep difficulties.\textsuperscript{10-17} Hot flashes are recurrent episodes of flushing, perspiration, and a sensation of heat. Sixty-five to 80\% of women experience them at some point during the menopausal transition. When hot flashes occur at night, they are referred to as night sweats and can lead to awakenings and sleep disruption.\textsuperscript{10,18-21} Sleep disruption may also occur during menopause independent of hot flashes.\textsuperscript{22} The transition into menopause is also associated with an increased risk of mood dysphoria that may be due to the fluctuations in ovarian steroids.\textsuperscript{23} Whether actual hormonal changes or the symptom and sleep problems accompanying the changes cause the mood disturbances is not clear.\textsuperscript{15,17,24-26}

Previous studies evaluating the effects of ET on cognition in menopausal women have shown conflicting results. ET does not appear to have cognitive effects in older women past the menopausal transition who are not having menopausal symptoms (hot flashes or night sweats).\textsuperscript{27-38} However, studies of younger women who had recently undergone surgical menopause\textsuperscript{6,8} have shown that ET improves performance on specific cognitive tasks, such as verbal memory,\textsuperscript{6,8,39} vigilance (sustained attention),\textsuperscript{40,41} and motor speed,\textsuperscript{40} but not cognitive functions, such as nonverbal memory or mental tracking.

There are several methodological differences among the studies that could account for the discrepant findings between younger, early postmenopausal women and older women. One difference is in the prevalence of menopausal symptoms such as hot flashes, sleep disturbances, and mood disorders among the participants. Alleviation of vasomotor symptoms or improvement in sleep and mood could underlie the improvement in cognitive performance in the studies of early menopausal women with symptoms,\textsuperscript{6,8,40-44} which older women do not have. The type of menopause could also influence how ET affects the brain and cognition. Surgically menopausal women who have very abrupt decreases in estradiol levels may have different cognitive responses to estradiol than women who have a more gradual estradiol decline during natural menopause. Thus, results from studies of the cognitive effects of ET in surgically menopausal women may differ from those of women after natural menopause and those of older menopausal women.

The timing of ET in relation to estrogen loss may also be important and might explain why early menopausal women with a short duration of estrogen deprivation experienced cognitive benefits with ET, whereas older women well past the menopausal transition did not. Data from animal models\textsuperscript{45-49} and observational human studies\textsuperscript{50-52} suggest the possibility of a critical window during which estrogen can exert positive cognitive effects. Younger brains (ie, 45-year-old brains) or continuous ET use since menopause may result in a different neural response than when estrogen is initiated or reinitiated in older brains (ie, 65-year-old brains).\textsuperscript{53} Finally, studies that used cognitive tests, such as verbal memory tasks, targeting brain areas known to be affected by estrogen, such as the hippocampus,\textsuperscript{45,54-58} may show positive effects of ET. Studies using cognitive measures that evaluate brain areas not expected to be influenced by estrogen would probably have negative findings.

This study was designed to evaluate how symptoms are related to cognitive performance both before and after ET in early menopausal women. We studied early menopause because information on the cognitive effects of ET in early, naturally menopausal women is lacking. Part 1 of the study addressed the role of vasomotor symptoms on cognitive performance, sleep, and mood in early menopause. Our hypothesis was that vasomotor symptoms would be associated with worse cognitive performance, sleep, and mood. Part 2 evaluated the effect of ET and the alleviation of symptoms on cognitive performance. We hypothesized that ET would affect cognitive performance in early, naturally menopausal women both with and without symptoms. We believed the cognitive domains of verbal memory, verbal fluency, and emotional memory would be positively affected by ET because data suggest that estrogen may affect the brain areas (hippocampus, amygdala, and prefrontal cortex) critical for these cognitive processes and/or because previous studies in humans have shown that ET improves performance on these measures in surgically menopausal women. We used a nonverbal memory measure as a control and did not expect positive effects of ET on this task.

**METHODS**

**Overall study outline**

This study had two components: a cross-sectional study, in which we examined whether menopausal symptoms are associated with cognition and mood, and a double-blind, placebo-controlled study in which we examined how ET affected symptoms, sleep, mood, and cognition (Fig. 1).

**Cross-sectional study**

**Participants**

Participants were recruited from an urban population through newspaper advertisements and fliers and
from a university medical center patient pool using recruitment letters. A screening interview included a medical history, vital sign measurements, electrocardiogram, and blood sample drawn for thyroid-stimulating hormone, hemoglobin, triglycerides, electrolytes and renal function, follicle-stimulating hormone (FSH), and estradiol. Participants were also administered the vocabulary subtest of the Wechsler Adult Intelligence Scale-Revised (WAIS-R), which provides a standardized approximation of functional intelligence. Those with a score indicating below average intelligence (scaled score $G < 8$) were excluded to ensure comparability of participant groups; three women were excluded on this basis.

Inclusion criteria required that individuals be in the late menopausal transition or early postmenopause (last menstrual period between 3 and 36 months before enrollment and an FSH level $> 20$ IU/L), have not been exposed to exogenous hormones in the last 3 months, understand English, and have adequate hearing and vision to perform computer and paper-pencil tasks (with correction if necessary). Participants with medical conditions or taking medications that could affect cognition (ie, epilepsy, stroke, or antidepressants) or that precluded estrogen use (ie, history of deep venous thrombosis, breast cancer, elevated blood pressure, or elevated triglycerides) or with medical screening abnormalities that could affect the test measures (ie, abnormal thyroid tests or anemia) were excluded. All participants were nonsmokers. Thirty-seven of the 68 women screened went on to participate in the cross-sectional study visit (Fig. 2). Women who were screened but did not participate had similar ages, education, and WAIS-R vocabulary scores ($P > 0.40$) as those who completed the study; however, they were slightly heavier (body mass index 28.96 [1.15] vs 26.82 [0.87], $P = 0.14$) and had lower FSH levels ($P = 0.01$) and higher estradiol levels ($P = 0.01$). Five of the 23 women who did not meet screening criteria were not menopausal; others were screened out because of abnormal blood pressure ($n = 5$), an abnormal thyroid test ($n = 3$), low WAIS-R vocabulary scores ($n = 3$), tobacco use ($n = 2$), abnormal electrolytes ($n = 2$), spotting ($n = 2$), or the inability to have a blood sample drawn ($n = 1$). All participants provided written informed consent and were paid for their involvement in the study. The study was approved by the Oregon Health and Science University Investigational Review Board.

Procedures

Participants completed daily menopausal symptom questionnaires, hot flash diaries, and sleep journals for 1 week before the study visit. Menopausal symptoms were monitored over 1 week because symptoms and hot flashes can vary from day to day$^{61-63}$ and because we wanted to assess the average symptomatology in the week before the visit. At the study visit, participants had a blood sample drawn for an estradiol level, completed the symptom questionnaire, hot flash diary, sleep journal, and mood evaluation, and underwent cognitive testing.

Estradiol assay. Assays were performed by the General Clinical Research Center at Oregon Health and Science University. All of the estradiol levels from a single individual (visits 1, 2, and 3 for the treatment study) were run in the same assay to avoid
interassay variability. A double-antibody radioimmunoassay (Diagnostic Systems Laboratories) was used to assay estradiol levels. The assay had a sensitivity of 2.2 pg/mL with an intra-assay coefficient of variation of 6.5% to 8.9% and interassay coefficient of variation of 7.5% to 12.2%.

Symptom questionnaires. The symptom questionnaires included 21 questions covering five symptom clusters: anxiety, depression, somatic, vasomotor, and sexual.64 Participants were divided into symptom groups according to their total score during the week before the study visit. Participants with scores greater than 10 were categorized as having high symptoms, and the remaining participants were categorized as having low symptoms based on scores in pre- and postmenopausal women in the literature.64 These categories were used for subsequent analysis.

Hot flash diary. Participants recorded the number of hot flashes they experienced in the previous 24 hours in the hot flash diary. They also recorded the severity of each hot flash (mild, moderate, severe, or very severe on a scale of 1 to 4) on the basis of example descriptions given for each level of severity. The main outcome measure was the average number of daily hot flashes. In addition, each hot flash was multiplied by its intensity and then added together to create a hot flash score, which was used in secondary analyses. This diary has been shown to be valid and reliable.65

Sleep diary. Sleep was evaluated with a sleep diary that is used clinically at the Oregon Health and Science University sleep laboratory, which included 16 questions about sleep architecture and sleep quality. For instance, it has questions about duration of sleep, number of awakenings, quality of sleep, and restfulness upon awakening. We combined seven questions about sleep quality into an overall quality index by transforming the raw scores of each of the seven questions into z scores and then averaging the seven z scores.

Mood. The Profile of Mood States (POMS) was used to measure mood over the last week. It is a short, validated measure of tension, depression, anger, vigor, fatigue, and confusion.66 Participants rated how often in the last week they experienced 65 mood adjectives using a 1 (not at all) to 5 (extremely) scale. The total score for each mood state was the outcome measure used.

Cognitive measures

The cognitive measures were chosen because (1) they were sensitive enough to detect differences in this healthy population of middle-aged women, (2) there is biological and/or animal data to suggest that estrogen may affect the brain areas (hippocampus and prefrontal cortex) critical for these cognitive processes, and (3) previous studies in humans have shown that ET improves performance on these measures. Cognitive testing took approximately 1 hour.

Verbal memory. Previous studies have shown that women with recent surgical menopause had improved performance when receiving ET compared with placebo on paragraph recall and verbal paired associates.6,8 In addition, these two verbal memory measures require use of the hippocampus,67 which is affected by estrogen.68 For both measures, the standard scoring system was used.59

For paragraph recall,59 participants were read a paragraph and asked to recall it immediately and again after a 30-minute interval. The number of phrases correctly recalled from the paragraph immediately (immediate) and after a 30-minute interval (delayed) were the measures of interest (total possible: immediate, 25; delayed, 25).

For paired associates,59 participants were read eight word pairs that were not readily associated (eg, cabbage-pen) and six pairs that were easily associated (eg, baby-cries). Participants heard each list of pairs 3 times, with a cued recall trial after each reading in which one item in a pair was given and participants recalled its pair. A single cued recall trial was given one-half hour later. The total number of difficult word pairs plus one-half the number of easy pairs recalled was the measure of interest (total possible: immediate 33; delayed 11).

Emotional memory. We studied the effects of menopausal symptoms on one category of emotional perception and memory, emotional faces, because women report that menopause affects the regulation of their emotions and because the neural basis of emotion and emotional memory overlaps with the location of estrogen receptors in the brain. Lesions of the amygdala or hippocampus and functional imaging studies suggest that this measure requires the amygdala, which modulates the hippocampus and amplifies memory for emotional information.69

Participants viewed a series of 36 faces presented individually on a computer screen.70 The faces included 11 men and 7 women. Each face was presented twice, but with unique emotional expressions. The faces expressed different emotions categorized as one of three valences: negative (sad, anger), neutral, or positive (happy). Participants rated the valence of each face on a 1 (negative) to
9 (positive) scale. After a 30-minute interval, participants completed a yes-no recognition test for those stimuli they had rated. The recognition test included 72 faces, 36 previously rated and 36 previously unseen. The latter consisted of “lures” in which half were the same people but expressing a different emotion than when previously seen and half were novel faces. Participants had to correctly recognize the same face with the same emotion.

Nonverbal memory. Three previous studies of ET and cognition have used this nonverbal memory measure, and none have found effects of estrogen. Therefore, this measure was used as a negative control.

For visual reproduction,59 women viewed four abstract designs for 10 seconds each. After each exposure, participants immediately drew what they remembered of the design. They then drew the designs again after a 30-minute interval. All of the drawings were scored by the same individual (P.E.C.) using standardized criteria (total possible: immediate, 41; delayed, 41).

Verbal fluency.71 Two large studies of older postmenopausal women without symptoms35,72 showed positive effects of ET on this measure. It was included to determine the effects of ET on fluency in younger early postmenopausal women with and without symptoms. The prefrontal cortex is critical for performance of this measure.73 Participants were given 1 minute to produce words beginning with a particular letter (ie, F, A, or S). The total number of words produced was recorded.

Treatment study
Participants
The same women participated in the treatment study as in the cross-sectional study described above. Five women did not complete the treatment study due to scheduling problems or illness (Fig. 2). Only one woman dropped out of the study because of side effects that may have been related to ET (bloating). The women who did not complete the study had demographics similar to those who completed the study ($P > 0.3$).

Procedures
After completion of the cognitive tests, participants were assigned in a double-blind manner to receive estradiol 2-mg tablets (ET) or an identical-looking placebo using a computer-generated randomization scheme. A 2-mg dose was used because it is the dose that gives maximum relief of menopausal symptoms74 and estradiol levels in the physiological range for premenopausal adult women (median 97 pg/mL, interquartile range 66-144).74 The last five participants underwent randomization stratified by the WAIS-R vocabulary subtest and estradiol level by a noninvolved third party because of a mismatch in WAIS-R vocabulary subtest and screening estradiol levels between treatment groups, which occurred by chance with randomization. The investigators conducting the testing sessions and the participants themselves were blinded to the treatment assignment.

Participants received ET or placebo for the subsequent 8 weeks and returned for test sessions at 4 and 8 weeks. This time frame was chosen because prior studies showed that there are significant improvements in hot flashes74,75 and sleep76 in women who receive ET for 4 weeks and that maximum symptom relief occurs by 8 weeks.74,75 Alternate but comparable cognitive measures for paragraph recall, verbal paired associates, visual reproduction, and verbal fluency were used for the repeated test sessions to decrease practice effects. The testing procedures at these visits were identical to those at the baseline visit except that at the third and final visit, participants were asked to complete a Likert scale about how likely it was they were receiving ET, with responses ranging from 1 (definitely not taking ET) to 5 (definitely taking ET).

At the end of the study, those who had received ET were given medroxyprogesterone acetate 5 mg for 14 days (to prevent endometrial hyperplasia), and those who had received the placebo were given an identical-looking placebo. Participants received a follow-up call 2 weeks after completing the study to check on side effects and to ensure that menstrual bleeding had occurred.

Data analysis
Repeated-measures, factorial analysis of variance (ANOVA) and $t$ tests (SPSS 12.0) were used to compare baseline characteristics, symptom and mood scores, and cognitive performance between the symptom and treatment groups. Analyses focused on two main questions. In the cross-sectional analysis, we compared cognitive performance between women with high and women with low symptoms. This was to answer the question of whether menopausal symptoms were associated with cognitive performance. For measures with only one test administration (ie, verbal fluency and emotional faces), a one-way ANOVA was used to determine whether there was a difference between symptom groups. When the task
included an immediate and delayed portion (verbal and nonverbal memory measures), a repeated-measures ANOVA was used to determine whether there were differences in memory retention from the immediate to the delayed portion of the task. Effects of ET on cognition were evaluated using a repeated-measures ANOVA with the baseline and 4- and 8-week test measurements as repeated measures. We excluded outliers (defined as a score greater than $\pm 2.5$ SD from the mean) because the values were so extreme that neither square root nor logarithmic transformations brought them within a normal range. Cross-sectional cognitive task results were covaried for WAIS-R vocabulary subtest scores because the symptom groups were marginally different on this variable. Treatment study results were covaried for body mass index. In post hoc analyses, comparisons were Bonferroni corrected for multiple comparisons.

Power analysis

We based our study size calculations on previous studies of the effects of ET on cognition in recently surgically menopausal women. These studies had 9 to 10 participants per treatment arm. Using one of these as the basis for our power analysis, we determined that we needed 17 participants per treatment arm to have 80% power to detect equivalent improvement on our primary outcome measure, immediate verbal paired associates (verbal memory).

RESULTS

Cross-sectional outcomes

Participant characteristics

Women in the high symptom group (n = 17) had an average symptom score of 16.37 (0.95) and experienced an average of 6.60 (1.02) hot flashes daily; those in the low symptom group (n = 20) had an average symptom score of 5.52 (0.72) with 2.41 (0.38) hot flashes per day (Table 1). The symptom groups had comparable background characteristics, although women who had high symptoms tended to have lower WAIS-R vocabulary scores ($P = 0.07$).

 Estradiol levels

 Estradiol levels were marginally higher in the low symptom women than in the high symptom women (31.51 [8.90] vs 14.30 [1.34], $P = 0.09$) (Table 1).

Sleep and mood

 The high symptom women had a lower quality sleep and more negative mood on the POMS than the low symptom women (Table 1). On the POMS, women in the high symptom group had more tension, depression, and fatigue, lower vigor, and increased, confusion ($P \leq 0.05$) (Fig. 3). Differences were not found for anger ($P = 0.84$).

Cognitive performance

Symptom status was not related to cognition (Table 2). Women with high and low symptoms performed similarly on paragraph recall, verbal paired associates, emotional memory, and verbal fluency ($P > 0.12$). Symptom groups were marginally different ($P = 0.08$) on visual reproduction. Low symptom women forgot visual details over 30 minutes ($P = 0.001$), whereas high symptom women did not ($P = 0.08$). When the above analyses were repeated with controls for estradiol level, cognitive performance was not different between symptom groups on any measure ($P > 0.15$).

Treatment study outcomes

Participant characteristics

Treatment groups were matched for age, education, baseline FSH and estradiol levels, symptom score, number of hot flashes, and mood (Table 3). However, by chance, the group randomly assigned to estradiol had a lower body mass index ($P = 0.02$), numerically lower WAIS-R vocabulary scores ($P = 0.07$), and poorer baseline sleep quality ($P = 0.08$).

 Estradiol levels

Women were compliant with the assigned medication (Table 4). Women receiving placebo did not

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**TABLE 1. Characteristics of symptom groups**

<table>
<thead>
<tr>
<th></th>
<th>Low symptom group (n = 20)</th>
<th>High symptom group (n = 17)</th>
<th>$P$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>52.80 (0.54)</td>
<td>51.82 (1.02)</td>
<td>0.39</td>
</tr>
<tr>
<td>Education, y</td>
<td>16.55 (0.48)</td>
<td>15.59 (0.55)</td>
<td>0.20</td>
</tr>
<tr>
<td>WAIS-R vocabulary</td>
<td>12.30 (2.49)</td>
<td>11.00 (1.58)</td>
<td>0.07</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>25.56 (1.03)</td>
<td>28.44 (1.37)</td>
<td>0.10</td>
</tr>
<tr>
<td>FSH, IU/L</td>
<td>69.85 (5.54)</td>
<td>65.88 (6.14)</td>
<td>0.63</td>
</tr>
<tr>
<td>Estradiol level, pg/mL</td>
<td>31.51 (8.90)</td>
<td>14.30 (1.34)</td>
<td>0.09</td>
</tr>
<tr>
<td>Symptom score</td>
<td>5.52 (0.72)</td>
<td>16.37 (0.95)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Hot flashes$^a$</td>
<td>2.41 (0.38)</td>
<td>6.60 (1.02)</td>
<td>&lt;0.001</td>
</tr>
<tr>
<td>Mood score$^b$</td>
<td>17.85 (5.04)</td>
<td>43.12 (8.14)</td>
<td>0.01</td>
</tr>
<tr>
<td>Sleep quality$^c$</td>
<td>–0.36 (0.14)</td>
<td>0.41 (0.15)</td>
<td>0.001</td>
</tr>
</tbody>
</table>

Values are mean (SEM). WAIS-R, Wechsler Adult Intelligence Scale-Revised$^d$; BMI, body mass index; FSH, follicle-stimulating hormone. $^a$From independent t-tests. $^b$From Greene Climacteric Scale$^{e}$; higher score indicates worse symptoms. $^c$From hot flash diary$^f$; score indicates average number of daily hot flashes. $^d$From Profile of Mood States$^{g}$; higher score indicates more negative mood. $^e$From sleep diary; lower score indicates higher quality sleep.
show a change in estradiol levels during the study, whereas women receiving ET had an increase in their estradiol levels (P < 0.001). Estradiol levels at visits 2 and 3 were higher than those at baseline in the women receiving ET (P < 0.001).

Menopausal symptoms

**Symptom score.** The placebo group had a borderline improvement in symptoms (P = 0.08), which has also been seen in other studies (Table 5). However, the ET group had significantly more symptom improvement (P < 0.001) than those given placebo (P value for difference between treatment groups = 0.03). Significant symptom relief occurred in the ET group by 4 weeks (P = 0.003).

**Hot flashes.** Treatment affected hot flash frequency (P = 0.02). Women receiving ET had significantly fewer hot flashes than those given placebo at visits 2 and 3 (P < 0.006). All participants in the ET group were averaging less than 1 hot flash per day by week 4.


![Profile of Mood States scores](image)

**FIG. 3.** Profile of Mood States scores. *P < 0.05.

We evaluated how symptom status interacted with treatment effects. This is an exploratory analysis because there were only a few participants (n = 6-11) in each comparison group. Women in the high symptom group had improvements in symptoms, hot flashes, and sleep during the study (P < 0.05). Their scores for each factor became comparable to those for the medium symptom group.

**COGNITIVE EFFECTS OF HOT FLASHES AND ET**

**TABLE 2. Cognitive performance by symptom group**

<table>
<thead>
<tr>
<th></th>
<th>High mean (n = 17)</th>
<th>Low mean (n = 20)</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Paragraph recall</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immediate</td>
<td>14.18 (0.91)</td>
<td>13.60 (0.92)</td>
<td>0.13</td>
</tr>
<tr>
<td>Delayed</td>
<td>12.76 (0.77)</td>
<td>12.70 (0.97)</td>
<td></td>
</tr>
<tr>
<td>Verbal paired associates</td>
<td>18.47 (1.20)</td>
<td>18.45 (1.22)</td>
<td>0.98</td>
</tr>
<tr>
<td>Immediate</td>
<td>8.21 (0.49)</td>
<td>8.15 (0.52)</td>
<td></td>
</tr>
<tr>
<td>Visual reproduction</td>
<td></td>
<td></td>
<td></td>
</tr>
<tr>
<td>Immediate</td>
<td>30.06 (1.61)</td>
<td>30.55 (1.22)</td>
<td>0.08</td>
</tr>
<tr>
<td>Delayed</td>
<td>28.18 (0.13)</td>
<td>25.35 (1.63)</td>
<td></td>
</tr>
<tr>
<td>Emotional facesa</td>
<td>73.50 (2.83)</td>
<td>77.17 (2.52)</td>
<td>0.93</td>
</tr>
<tr>
<td>Verbal fluencyb</td>
<td>39.25 (2.28)</td>
<td>41.53 (2.24)</td>
<td>0.48</td>
</tr>
</tbody>
</table>

Values are mean (SEM). *P value is from analysis of variance; results were covaried for Wechsler Adult Intelligence Scale-Revised; outliers were defined as a score greater than ±2 SD from the mean and were not included in the analysis.

One participant was dropped from emotional faces and two from verbal fluency because they were outliers.

**TABLE 3. Baseline characteristics of treatment groups**

<table>
<thead>
<tr>
<th></th>
<th>Estradiol (n = 14)</th>
<th>Placebo (n = 18)</th>
<th>P*</th>
</tr>
</thead>
<tbody>
<tr>
<td>Age, y</td>
<td>53.26 (0.64)</td>
<td>52.08 (0.64)</td>
<td>0.22</td>
</tr>
<tr>
<td>Education, y</td>
<td>15.95 (0.53)</td>
<td>16.54 (0.66)</td>
<td>0.49</td>
</tr>
<tr>
<td>BMI, kg/m²</td>
<td>28.59 (1.42)</td>
<td>23.95 (0.65)</td>
<td>0.02</td>
</tr>
<tr>
<td>WAI R vocabulary</td>
<td>12.37 (0.51)</td>
<td>10.95 (0.61)</td>
<td>0.07</td>
</tr>
<tr>
<td>Estradiol, pg/mL</td>
<td>19.79 (3.65)</td>
<td>32.70 (13.13)</td>
<td>0.28</td>
</tr>
<tr>
<td>FSH, IU/L</td>
<td>63.37 (5.52)</td>
<td>73.00 (5.84)</td>
<td>0.31</td>
</tr>
<tr>
<td>Symptom scoreb</td>
<td>9.4 (1.7)</td>
<td>11.1 (1.4)</td>
<td>0.48</td>
</tr>
<tr>
<td>Hot flashesc</td>
<td>5.0 (0.99)</td>
<td>3.2 (0.84)</td>
<td>0.22</td>
</tr>
<tr>
<td>Mood scored</td>
<td>26.8 (7.2)</td>
<td>36.2 (9.6)</td>
<td>0.42</td>
</tr>
<tr>
<td>Sleep qualitye</td>
<td>0.36 (0.16)</td>
<td>−0.09 (0.17)</td>
<td>0.08</td>
</tr>
</tbody>
</table>

Values are mean (SEM). WAI R, Wechsler Adult Intelligence Scale-Revised; BMI, body mass index; FSH, follicle-stimulating hormone. *P value from independent samples t test.

From Green Climacteric Scale; higher score indicates worse symptoms.

From hot flash diary; score indicates average number of daily hot flashes.

From Profile of Mood States; higher score indicates more negative mood.

From sleep diary; lower score indicates higher quality sleep. Two women in the estradiol group had missing sleep data and were not included in the analysis.

**Sleep.** There was an effect of treatment on sleep (P = 0.05). Sleep quality improved more between visits 1 and 3 in the ET group than in the placebo group (P = 0.01).

**Mood.** Mood did not improve in either group over the study visits.

**Cognitive performance**

The women taking ET did not perform better than those on placebo for any of the cognitive measures (P > 0.13) (Table 6).

**Effects of symptom status during the treatment portion of the study**

We evaluated how symptom status interacted with treatment effects. This is an exploratory analysis because there were only a few participants (n = 6-11) in each comparison group. Women in the high symptom group had improvements in symptoms, hot flashes, and sleep during the study (P < 0.05). Their scores for each factor became comparable to those for the medium symptom group.

**TABLE 4. Estradiol levels for estrogen therapy and placebo groups**

<table>
<thead>
<tr>
<th></th>
<th>Baseline, pg/mL</th>
<th>4 wk, pg/mL</th>
<th>8 wk, pg/mL</th>
</tr>
</thead>
<tbody>
<tr>
<td>Estradiol (n = 12)a</td>
<td>32.87 (14.28)</td>
<td>207.57 (31.71)</td>
<td>205.08 (30.45)</td>
</tr>
<tr>
<td>Placebo (n = 18)</td>
<td>20.07 (3.84)</td>
<td>19.31 (2.19)</td>
<td>21.86 (5.57)</td>
</tr>
</tbody>
</table>

Values are mean (SEM). *Two women were excluded from the analysis because of missing values.
low symptom women. There were no similar improvements in low symptom women, possibly because their scores left little room for improvement (floor effect). Symptom status did not interact with the effects of ET on mood or cognition.

Effectiveness of blinding

Those randomly assigned to ET were significantly more likely to think they were taking ET than those randomly assigned to placebo ($P < 0.001$). All 13 women randomly assigned to ET thought they were probably or definitely taking ET. Two women in the placebo group thought they were definitely or probably taking ET, four did not know whether they were taking ET, and 13 did not think they were taking ET. Several women guessed they were taking ET because of side effects such as breast tenderness.

DISCUSSION

The aim of the cross-sectional study was to evaluate the association between vasomotor symptoms and sleep, mood, and cognition in early menopause. Our hypothesis was that vasomotor symptoms would have negative effects on mood, sleep, and cognition. We found that symptoms were associated with a more negative mood and lower quality sleep but were not related to cognitive performance.

We had hypothesized that symptoms would be associated with worse cognitive performance because prior studies of women with menopausal symptoms after surgically induced menopause found that ET improved cognition. In addition, studies that showed cognitive effects of estradiol included women with menopausal symptoms. The studies that did not show effects of ET were those in which women were past the symptom stage of menopause. One possibility was that hot flashes affect cognition directly by decreasing cerebral blood flow or indirectly through symptom-induced discomfort, mood disturbances, or sleep disruption. Indeed, the presence of menopausal symptoms has been associated with lower reading and verbal memory scores, although these results were not adjusted for education or socioeconomic status. Lower education and socioeconomic status are

<p>| TABLE 5. Effects of estrogen therapy on menopausal symptoms |
|-------------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|</p>
<table>
<thead>
<tr>
<th></th>
<th>Visit 1 Estradiol (n = 14)</th>
<th>Visit 1 Placebo (n = 18)</th>
<th>Visit 2 Estradiol (n = 14)</th>
<th>Visit 2 Placebo (n = 18)</th>
<th>Visit 3 Estradiol (n = 14)</th>
<th>Visit 3 Placebo (n = 18)</th>
<th>$P^a$</th>
</tr>
</thead>
<tbody>
<tr>
<td>Symptom score$^b$</td>
<td>11.08 (1.40)</td>
<td>9.43 (1.66)</td>
<td>6.17 (1.42)</td>
<td>8.30 (1.34)</td>
<td>5.59 (1.00)</td>
<td>7.51 (1.36)</td>
<td>0.03</td>
</tr>
<tr>
<td>Hot flashes$^c$</td>
<td>3.20 (0.84)</td>
<td>4.41 (0.88)</td>
<td>0.25 (0.09)</td>
<td>3.46 (0.76)</td>
<td>0.23 (0.14)</td>
<td>2.71 (0.63)</td>
<td>0.02</td>
</tr>
<tr>
<td>Mood score$^d$</td>
<td>29.75 (7.61)</td>
<td>26.84 (7.15)</td>
<td>24.00 (8.95)</td>
<td>24.10 (6.74)</td>
<td>22.83 (9.71)</td>
<td>23.89 (8.74)</td>
<td>0.25</td>
</tr>
<tr>
<td>Sleep quality$^e$</td>
<td>0.36 (0.16)</td>
<td>-0.09 (0.17)</td>
<td>0.23 (0.13)</td>
<td>-0.07 (0.18)</td>
<td>0.11 (0.10)</td>
<td>0.02 (0.15)</td>
<td>0.05</td>
</tr>
</tbody>
</table>

$^a$P value from repeated-measures analysis of variance. The analysis was covaried for body mass index; outliers were defined as a score greater than $\pm 2.5$ SD from the mean and were not included in analysis.

$^b$From Greene Climacteric Scale; higher score indicates worse symptoms.

$^c$From hot flash diary; score indicates average number of daily hot flashes; one outlier was removed.

$^d$From Profile of Mood States; higher score indicates more negative mood; one outlier was removed.

$^e$From sleep diary; lower score indicates high quality of sleep. Two participants in the estrogen group had missing data for sleep and were not included in the analysis.

| TABLE 6. Effects of estrogen therapy on cognition |
|-------------------------------|-----------------|-----------------|-----------------|-----------------|-----------------|-----------------|
|                             | Visit 1 Estradiol (n = 14) | Visit 1 Placebo (n = 18) | Visit 2 Estradiol (n = 14) | Visit 2 Placebo (n = 18) | Visit 3 Estradiol (n = 14) | Visit 3 Placebo (n = 18) | $P^a$ |
| Paragraph recall             | 13.46 (1.11)    | 13.63 (0.96)    | 16.23 (0.94)    | 15.32 (0.56)    | 16.54 (0.96)    | 15.89 (0.93)    | 1.00 |
| Delayed                      | 12.54 (1.12)    | 12.32 (0.90)    | 15.00 (1.07)    | 14.53 (0.77)    | 15.15 (1.07)    | 14.63 (0.98)    | 0.89 |
| Verbal paired associates     | 18.85 (1.35)    | 17.55 (1.22)    | 22.50 (1.83)    | 22.08 (1.30)    | 24.50 (1.37)    | 24.21 (1.52)    | 0.94 |
| Delayed                      | 8.38 (0.63)     | 7.74 (0.50)     | 8.62 (0.64)     | 9.00 (0.49)     | 9.04 (0.92)     | 9.28 (0.43)     | 0.42 |
| Visual reproduction          | 28.77 (1.70)    | 30.95 (1.48)    | 33.08 (1.35)    | 32.89 (1.03)    | 32.00 (1.67)    | 32.68 (1.63)    | 0.53 |
| Emotional faces$^b$          | 80.09 (2.77)    | 71.96 (2.24)    | 87.27 (2.38)    | 78.95 (2.25)    | 80.41 (3.82)    | 79.73 (2.63)    | 0.13 |
| Verbal fluency              | 39.00 (2.04)    | 41.16 (3.48)    | 45.77 (2.49)    | 46.00 (1.75)    | 43.31 (2.78)    | 47.05 (2.46)    | 0.77 |

Values are mean (SE).

$^a$P value from repeated-measures analysis of variance; analysis was covaried for body mass index; outliers were defined as a score greater than $\pm 2.5$ SD from the mean and were not included in analysis.

$^b$One participant was dropped from emotional faces because she was an outlier.
associated with a higher likelihood of developing menopausal symptoms.\textsuperscript{78,79} With our results we would argue that when women with similar educational status are studied, menopausal symptoms are not associated with cognitive performance. In addition, the lower quality sleep and negative mood associated with hot flashes are not related to cognitive performance.

We also evaluated how ET and the resultant alleviation of symptoms affected cognitive performance. We hypothesized that ET would improve verbal memory, emotional memory, and verbal fluency in early naturally menopausal women both with and without symptoms. However, ET did not improve cognitive performance. ET did not have beneficial effects on verbal memory, emotional memory, or verbal fluency in women who had an average age of approximately 53 and were within 3 years of their last menstrual period. There was also no effect of ET on our control measure, nonverbal memory. This lack of cognitive benefit was in the setting of significant improvements in menopausal symptoms, hot flashes, and sleep. ET did not alleviate mood disturbances. Thus, it is possible that significant mood improvements might have led to improved cognition.

One of our objectives for this study was to determine whether the positive cognitive effects of ET seen in studies of recently surgically postmenopausal women also occurred in women in the early stages of natural menopause. Previous studies of younger women who had recently undergone surgical menopause\textsuperscript{6,8} have shown that ET improves performance on verbal memory,\textsuperscript{6,8} which is in contrast to our results. The gradual decline in estrogen that occurs over many years during natural menopause is quite different from the abrupt decline observed with surgical menopause. The speed and degree of estrogen loss could be critical factors in how estrogen loss affects the brain and cognitive function. Our results suggest that the more gradual decline in estrogen that occurs with natural menopause may not result in the same adverse consequences. In addition, levels of androgens, such as testosterone and androstenedione, are lower in women who have had surgical removal of their ovaries compared with women who undergo natural menopause.\textsuperscript{80} Androstenedione and testosterone can be converted into estrogen or may act directly on androgen receptors in the brain\textsuperscript{81} to affect cognitive performance. Differences in androgen levels could therefore also explain the different cognitive effects of natural versus surgical menopause.

Our study focused on women who were in early postmenopause because at least some animal work suggests that estrogen may have positive brain effects only when estrogen loss has been of short duration.\textsuperscript{45,46} We did not find in our sample of women who had undergone estrogen loss in the last 3 years that ET was beneficial for cognition during the period of active estrogen use. We cannot exclude the possibility that taking ET around the time of menopause may result in less cognitive decline later in life, as suggested by some observational studies.\textsuperscript{50-52}

Another interesting finding from our study is the effect of vasomotor symptoms on mood. Although the high symptom group had more tension, depression, and fatigue, lower vigor, and increased confusion, they did not have more anger. The reason that menopausal symptoms would not be associated with anger is not clear and requires further study. Women in the high symptom group had POMS scores that were similar to those of outpatient women aged 18 to 65 in a smoking cessation program, whereas POMS scores of women with low symptoms were more similar to those of college-aged female norms on the West Coast.\textsuperscript{82}

ET did not affect mood despite improvements in overall menopausal symptoms and hot flashes. We hypothesized that ET would improve mood because previous studies have found positive effects of ET on mood in both depressed\textsuperscript{25,83} and nondepressed women.\textsuperscript{26} However, the previous study in nondepressed women was performed with women after oophorectomy. Our study results suggest that in nondepressed women in the early stages of natural menopause, mood may not be not affected by ET.

ET did improve sleep in the ET group, but sleep was not improved in the placebo group. In an exploratory analysis in which we considered symptom status, ET improved sleep in the high symptom women. Previous studies have shown mixed results of ET on sleep; whereas some have found positive benefits of ET,\textsuperscript{19,22,84,85} others have not.\textsuperscript{86,87} Our study indicates that ET improves sleep quality, especially in women who are having menopausal symptoms. Because the analysis is exploratory, however, we are cautious in our conclusions about the relationship between sleep, symptoms, and ET. ET could improve sleep through symptom alleviation. However, the lack of effect in the low symptom women could also be the result of a lack of sleep problems in the low symptom group (floor effect).

There were several limitations of the study. This study size was relatively small; however, the number of participants was comparable to numbers in...
previous studies that showed positive effects of estrogen on cognition in surgically menopausal women. Our study suggests that the cognitive effects of ET in naturally menopausal women, early in menopause during the period of peak symptoms, may not be as robust as those in surgically menopausal women.

Eight weeks of use may not be long enough to detect effects of estrogen on cognition. In older women, hormone therapy may have cognitive effects only after long-term use. However, in recently postmenopausal women, 21 days to 3 months of use have been shown to affect brain activity and cognitive performance and cognitive performance within 72 hours after administration. We also do not believe that our negative findings can be attributed to our use of oral estradiol. Previous studies have used a range of types and doses of estrogen given both orally and transdermally, and there is no clear correlation with type of formulation and cognitive effect.

Our negative findings were also not the result of insensitive cognitive measures. Some of the tasks are the same measures that were used in studies of surgical menopause, and we did not have ceiling effects. In addition, these measures were chosen because there is biological animal data and/or human data to indicate that these measures would be influenced by estrogen.

CONCLUSIONS
We did not find that the severity of menopausal symptoms was associated with cognitive performance. Furthermore, ET did not improve cognition in women even though it relieved symptoms and improved sleep. Our results, in contrast to the positive effects of ET on cognition in surgically menopausal women, suggest that the abrupt loss of complete ovarian function may be a key factor in whether loss or replacement affects cognition. Larger studies are needed to confirm our findings and explore the effects of abrupt versus more gradual estrogen and other ovarian hormone decline on menopausal symptoms and cognition.

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