Effects of hormone therapy on brain structure

A randomized controlled trial

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ABSTRACT

Objective: To investigate the effects of hormone therapy on brain structure in a randomized, double-blinded, placebo-controlled trial in recently postmenopausal women.

Methods: Participants (aged 42–56 years, within 5–36 months past menopause) in the Kronos Early Estrogen Prevention Study were randomized to (1) 0.45 mg/d oral conjugated equine estrogens (CEE), (2) 50 μg/d transdermal 17β-estradiol, or (3) placebo pills and patch for 48 months. Oral progesterone (200 mg/d) was given to active treatment groups for 12 days each month. MRI and cognitive testing were performed in a subset of participants at baseline, and at 18, 36, and 48 months of randomization (n = 95). Changes in whole brain, ventricular, and white matter hyperintensity volumes, and in global cognitive function, were measured.

Results: Higher rates of ventricular expansion were observed in both the CEE and the 17β-estradiol groups compared to placebo; however, the difference was significant only in the CEE group ($p = 0.01$). Rates of ventricular expansion correlated with rates of decrease in brain volume ($r = -0.58$; $p \leq 0.001$) and with rates of increase in white matter hyperintensity volume ($r = 0.27$; $p = 0.01$) after adjusting for age. The changes were not different between the CEE and 17β-estradiol groups for any of the MRI measures. The change in global cognitive function was not different across the groups.

Conclusions: Ventricular volumes increased to a greater extent in recently menopausal women who received CEE compared to placebo but without changes in cognitive performance. Because the sample size was small and the follow-up limited to 4 years, the findings should be interpreted with caution and need confirmation.

Classification of evidence: This study provides Class I evidence that brain ventricular volume increased to a greater extent in recently menopausal women who received oral CEE compared to placebo. *Neurology*® 2016;87:887–896

GLOSSARY

CEE = conjugated equine estrogens; **FLAIR** = fluid-attenuated inversion recovery; **KEEPS** = Kronos Early Estrogen Prevention Study; **WHIMS** = Women's Health Initiative Memory Study; **WMH** = white matter hyperintensity.

Hormone therapy with conjugated equine estrogens (CEE) and medroxyprogesterone acetate initiated later in menopause increased the risk of dementia in the Women's Health Initiative Memory Study (WHIMS).[1] Whether alternative formulations of hormone therapy can preserve neuronal integrity and decrease the risk of dementia when administered early in menopause remains controversial.[2–10] Determining the effects of hormone therapy during the early postmenopausal years on the risk of dementia would require decades of follow-up. However, noninvasive imaging markers related to cognitive health have been suggested as short-term surrogate outcomes to assess the effects of menopausal hormone therapy on the brain. Thus, age-associated changes in

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brain structure on MRI can be used to measure the effects of estrogens on the brains of postmenopausal women.

The Kronos Early Estrogen Prevention Study (KEEPS) tested the hypothesis that menopausal hormone therapy administered early after the onset of menopause would slow progression of atherosclerosis.[11] However, hormone therapy in KEEPS did not affect progression of atherosclerosis[12] or cognitive function[13] within the 4 years of hormone therapy. We report here the results of an ancillary study to KEEPS, conducted to determine the effects of oral CEE and transdermal 17β-estradiol therapy on changes in structural brain MRI over 4 years. The ventricular volume change was chosen as the primary outcome measure because it was the most reliable measure of the change in brain structure associated with aging and cognitive function.[14] The secondary outcome measures included changes in whole brain and white matter hyperintensity (WMH) volumes.

METHODS

Participants. KEEPS was a multicenter, randomized, double-blinded, placebo-controlled clinical trial in recently menopausal women (n = 727). Participants enrolled in KEEPS were between 42 and 59 years of age, within 5 to 36 months past their last menses, and were in good cardiovascular health.[11] An ancillary MRI study to KEEPS was conducted from February 2006 through August 2011 at the Mayo Clinic to investigate the effects of hormone treatment on brain structure. Exclusion criteria for enrollment in the ancillary KEEPS-MRI study were MRI contraindications for safety and neurologic disorders.

Baseline MRI examinations and cognitive testing were completed following randomization but with concealed allocation and before treatment initiation. The treatments were as follows: (1) oral CEE (0.45 mg/d; Premarin, Pfizer, New York, NY); (2) transdermal 17β-estradiol (17β-estradiol skin patch, 50 μg/d; Climara, Bayer HealthCare Pharmaceuticals Inc., Wayne, NJ); or (3) placebo pills and patch. Estrogens were administered through 2 different routes, oral or transdermal, because of the increased risk of venous thrombosis with oral estrogens. It was hypothesized that transdermal 17β-estradiol would have a different effect on risk factors for atherosclerosis and thromboembolic disease compared to oral CEE. Progesterone was given orally (micronized progesterone, 200 mg/d; Prometrium, AbbVie Inc., North Chicago, IL) for 12 days at the beginning of each month to both active treatment groups to protect the endometrium. Follow-up MRI examinations and cognitive testing were performed at 18 months (estrogen-only phase), 36 months (progesterone phase; to determine whether there would be an effect on the trajectory of change in outcome measures due to progesterone), and at 48 months (estrogen-only phase).[11]

Standard protocol approvals, registrations, and patient consents. The present study (NCT00154180; https://clinicaltrials.gov/ct2/show/NCT00154180) was approved by the Mayo Clinic institutional review board (no. 224104). All participants provided written informed consent.

Cognitive testing. A confirmatory factor analysis was used to assess the underlying structure of baseline cognitive data from the KEEPS cognitive and affective study, and to derive summary scores (n = 662).[15] Using standard criteria for model fit, the cognitive variables were summarized in 4 specific independent domains and a general domain representing global cognitive function.[16]

Magnetic resonance imaging. MRI studies were performed on a single 1.5-tesla system, with an 8-channel phased-array coil (GE Healthcare, Waukesha, WI). A 3-dimensional, magnetization-prepared rapid-acquisition gradient echo sequence was acquired for volumetric analysis, and fluid-attenuated inversion recovery (FLAIR) MRI was acquired for quantification of WMH volume. Changes in ventricle and whole brain volumes were calculated automatically from each registered magnetization-prepared rapid-acquisition gradient echo scan pair using the boundary shift integral and expressed in cubic centimeters of volume change from baseline for each follow-up time point as previously described.[17] The boundary shift integral is designed to monitor brain structural changes and treatment effects in clinical trials of Alzheimer disease.[18]

WMH volumes were derived from semiautomated segmentation of FLAIR images as previously described.[19] Briefly, WMH on FLAIR images were segmented using an automated slice-based seed initialization and region growing method. A trained image analyst (S.Z.), blinded to treatment group as well as to the sequence of the follow-up scans, inspected the segmented WMH mask overlaid on the FLAIR image. Every segmented image was visually compared to the unprocessed FLAIR image and false-positive WMH labels resulting from artifacts were edited to be excluded from the WMH mask. The WMH masks on follow-up scans were also visually compared to the baseline WMH mask for consistent editing of the artifacts. One of the baseline FLAIR scans failed quality control and was therefore excluded from the analysis. Longitudinal change in WMH was expressed in cubic centimeters of volume change from baseline for each follow-up MRI.

Statistical analysis. Baseline characteristics were compared among groups using analysis of variance followed by Tukey pairwise tests, 2-sample *t* tests, or Fisher exact tests, as appropriate. Changes in the outcomes from baseline were shown using plots of mean values and their associated 95% confidence intervals at each time point. Associations between the outcome variables at 48 months were shown using scatterplots with Pearson correlation coefficients adjusted for age. In a secondary analysis, the percent change in the whole brain, ventricular, and WMH volumes over 18, 36, and 48 months in CEE and 17β-estradiol groups was compared to placebo by *t* tests.

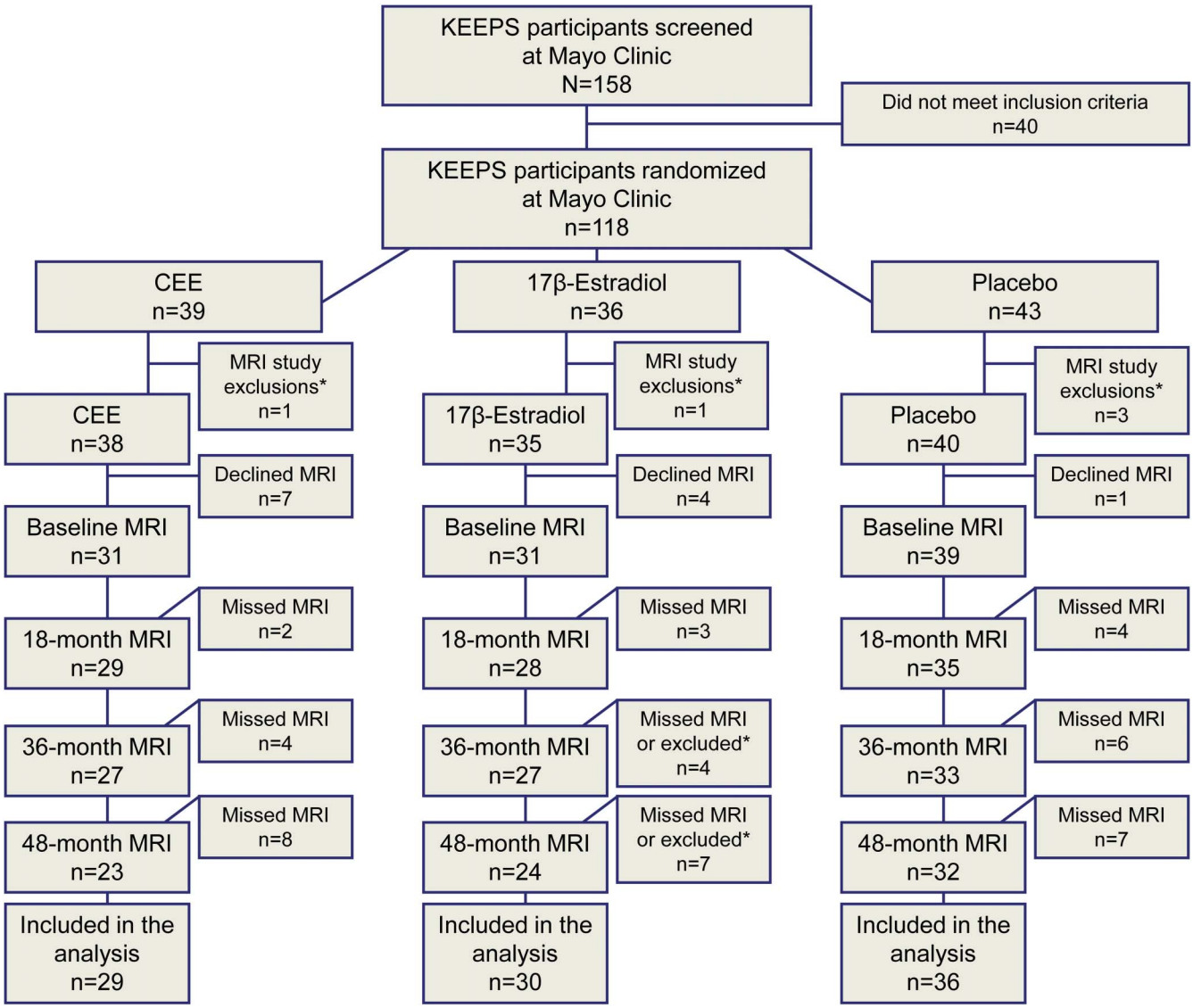
Ventricular and whole brain volumes, WMH, and global cognitive score changes over time were modeled using linear mixed models as planned a priori, with each model containing time from baseline, treatment group, and time from baseline × treatment group interactions as fixed-effects predictors. These models incorporated random effects for subject-specific intercepts and slopes. Coefficient estimates with their associated 95% confidence intervals, and categories of significance were reported for these models. Treatments were compared to each other and to placebo. Treatment by time interactions were of primary interest because any treatment effect would likely manifest as a change in the trajectories over time rather than a sudden jump in the magnitude of the outcomes. Because the primary analyses on the effects of each of the 2 hormone therapies on brain structure were determined a priori, there was no adjustment for multiple comparisons in

reporting these results. Furthermore, results from the 2 hormone therapy groups were not pooled because of the differences in formulations and route of administration.

For our primary analysis of ventricular volumes using linear mixed models, with $\alpha = 0.05$ and the smallest group size of 29 for conservative estimates, there was 80% power to detect a difference in slopes over time between groups of 0.296 cm^3 per year. For example, for a woman starting at the mean baseline ventricular volume of 19.6 cm^3, a 1.5% increase could be detected per year because of treatment (a 2.3% increase in the volume after 18 months, 4.5% after 36 months, and 6.0% after 48 months).

RESULTS All women enrolled in KEEPS at Mayo Clinic Rochester (n = 118) were invited to participate in the ancillary MRI study. Five participants were excluded because of neurologic disorders or MRI contraindications; 12 women declined participation. Eligible women underwent a baseline MRI (n = 101) before starting treatment. MRI data were analyzed for the 95 participants who repeated at least one MRI examination at 18 (n = 92), 36 (n = 87), or 48 (n = 79) months (figure 1). Baseline global cognitive performance and whole brain volumes were not different among treatment and placebo groups. However, ventricular volumes were larger in the 17β-estradiol treatment group ($p = 0.025$) and WMH volumes were larger in the CEE treatment group

Figure 1 KEEPS-MRI study participation



Participation and exclusions from the KEEPS-MRI study are shown. Participants who had a baseline MRI before onset of treatment and had at least one follow-up MRI at months 18, 36, or 48 were included in the analysis. *There were 5 exclusions at baseline MRI: MRI incompatible implant (CEE group); posterior fossa developmental abnormality and hydrocephalus (17β-estradiol group); 2 participants with multiple sclerosis and one participant with a benign brain tumor (placebo group). There was an exclusion of a participant from the 17β-estradiol group who developed breast cancer after completing the 18-month scan and underwent chemotherapy. Her structural MRI data at months 36 and 48 were excluded from analysis because of the potential adverse effects of chemotherapy on brain structure. One of the baseline fluid-attenuated inversion recovery scans failed quality control and therefore had to be excluded from the analysis. CEE = conjugated equine estrogens; KEEPS = Kronos Early Estrogen Prevention Study.

($p = 0.008$) compared to placebo at baseline after adjusting for total intracranial volume (table 1). None of the participants had silent infarcts on baseline and follow-up MRI examinations.

Whole brain volumes decreased in the CEE ($p = 0.004$) and the 17β-estradiol ($p = 0.002$) treatment groups, but not in the placebo group ($p = 0.09$) over 48 months. However, the decline in whole brain volume was not significantly different between the placebo and treatment groups (CEE and 17β-estradiol). Ventricular volumes increased in both the treatment (CEE and 17β-estradiol) and the placebo groups over 48 months ($p < 0.001$). Increases in ventricular volumes were greater only in the CEE treatment group compared to the placebo group ($p = 0.01$). WMH volume increased in the CEE ($p = 0.004$) and 17β-estradiol ($p = 0.002$) groups, but not in the placebo group ($p = 0.42$) over 48 months. However, the increase in WMH volumes was not different when comparing the CEE ($p = 0.10$) and 17β-estradiol ($p = 0.06$) groups to the placebo group (figure 2). The results of mixed-effects models are summarized in table 2. Changes in any of the MRI measures did not differ between the CEE and 17β-estradiol groups (table e-1 at Neurology.org).

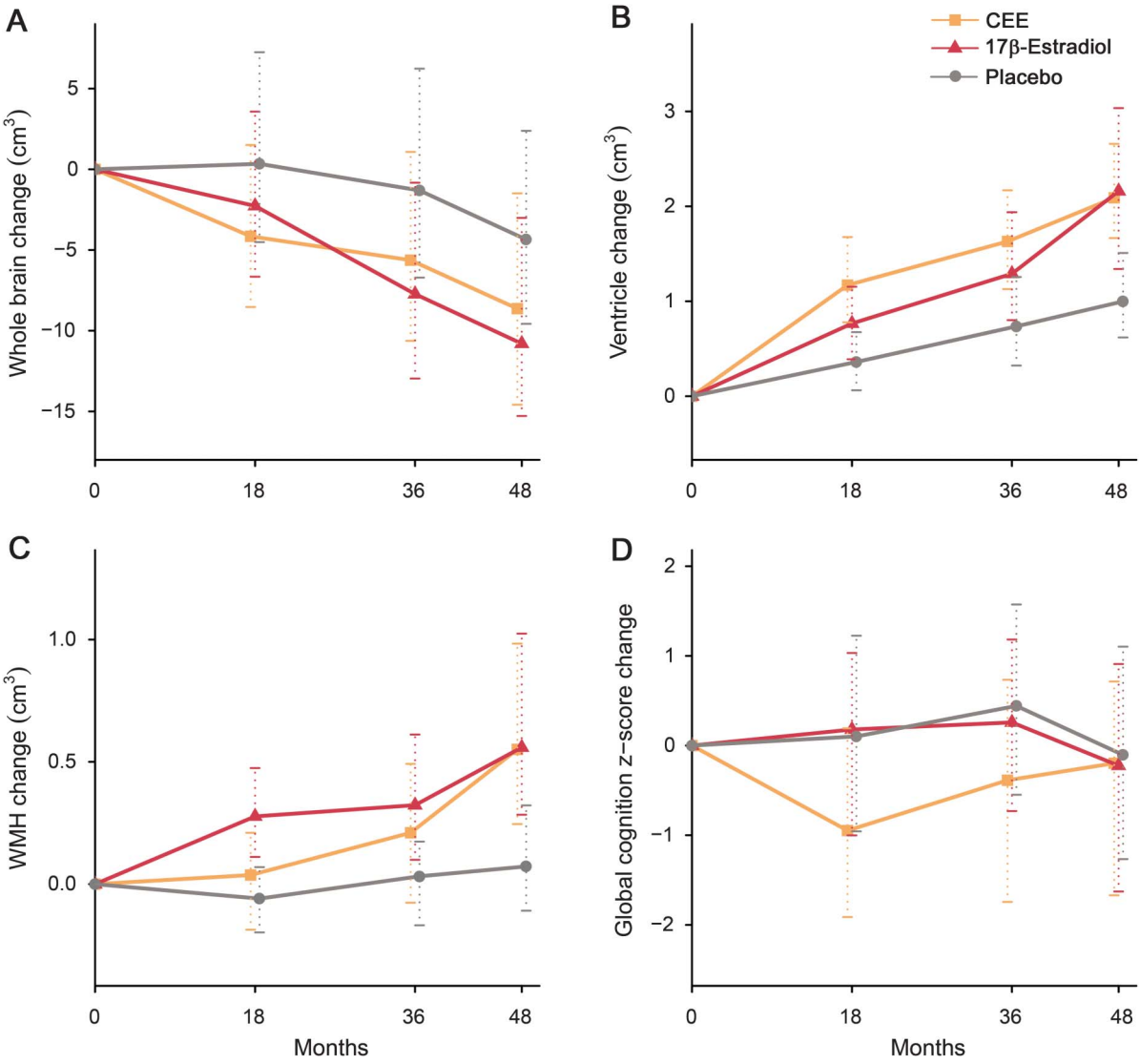
Because baseline WMH volumes were higher in the CEE group than in the placebo group, we tested whether baseline WMH modified the treatment × time interactions by adding baseline WMH and baseline WMH × time to the mixed models. The parameter estimates and p values for treatment × time were then compared with and without the baseline adjustment to see if they differed. The primary interest was in possible effects of baseline WMH on the significant CEE vs placebo difference in ventricular volume change. After adjusting for baseline WMH, increases in ventricular volumes in the CEE treatment group remained greater than in the placebo group ($p = 0.011$), but did not reach statistical significance in

Table 1 Baseline characteristics of the participants

	CEE (n = 29)	17β-Estradiol (n = 30)	Placebo (n = 36)
Age, y	53 (52, 54)	53 (52, 54)	53 (52, 54)
Education			
High school or less	2 (7)	1 (4)	3 (8)
Some college/college graduate	19 (70)	18 (64)	22 (61)
Some graduate/graduate	6 (22)	9 (32)	11 (31)
Smoking status			
Nonsmoker	15 (68)	12 (55)	24 (73)
Smoker (past or current)	7 (32)	10 (45)	9 (27)
Time past menopause, mo	21 (17, 25)	20 (17, 23)	17 (14, 20)
Treatment onset past baseline MRI, d	13 (3, 23)	20 (10, 30)	28 (7, 49)
APOE ε4 carrier	4 (15)	13 (45)	7 (21)
Migraines	3 (10)	0 (0)	4 (11)
Global cognitive function scores	−0.19 (−0.48, 0.10)	0.16 (−0.12, 0.44)	0.14 (−0.08, 0.36)
Mean systolic blood pressure, mm Hg	122 (117, 127)	120 (114, 126)	122 (118, 126)
Mean diastolic blood pressure, mm Hg	77 (74, 80)	73 (70, 76)	76 (74, 78)
Waist circumference, cm	87 (82, 92)	83 (79, 87)	84 (80, 88)
Body mass index, kg/m^2	28 (26, 30)	26 (25, 27)	27 (26, 28)
Coronary arterial calcification present	4 (14)	3 (10)	4 (11)
Carotid intima-media thickness	0.71 (0.67, 0.75)	0.72 (0.68, 0.76)	0.71 (0.68, 0.74)
Low-density lipoprotein, mg/dL	134 (124, 144)	137 (125, 149)	130 (119, 141)
High-density lipoprotein, mg/dL	59 (54, 64)	63 (58, 68)	60 (55, 65)
Triglycerides, mg/dL	93 (78, 108)	101 (81, 121)	91 (76, 106)
Whole brain volume, cm^3	1,334 (1,300, 1,368)	1,316 (1,285, 1,347)	1,302 (1,269, 1,335)
Ventricular volume, cm^3	19.6 (16.8, 22.4)	22.1 (18.7, 25.5)[a]	17.6 (15.5, 19.7)
White matter hyperintensity volume, cm^3	2.7 (2.1, 3.3)[b]	2.3 (1.8, 2.8)	1.8 (1.6, 2.0)

Abbreviation: CEE = conjugated equine estrogens.
Data are n (%) or mean (95% confidence interval).
[a] Pairwise comparison to placebo $p < 0.05$.
[b] Pairwise comparison to placebo $p < 0.01$.

Figure 2 Changes in the MRI raw value measurements and global cognitive function in treatment groups



Mean change (cm³) and 95% confidence intervals at 18, 36, and 48 months for whole brain volume (A), ventricular volume (B), and WMH volume (C). Mean change in global cognition *z* scores and 95% confidence intervals at 18, 36, and 48 months (D) is displayed. Increases in ventricular volumes were greater in the CEE treatment group than in the placebo group ($p = 0.01$), but not in the 17β-estradiol group compared to the placebo group ($p = 0.09$). CEE = conjugated equine estrogens; WMH = white matter hyperintensity.

the 17β-estradiol group compared with the placebo group ($p = 0.06$) (table e-2).

Percent increases in ventricular volume from baseline to 18, 36, and 48 months were greater in the CEE group but not in the 17β-estradiol group compared to placebo at all time points. On the contrary, percent increases in WMH volume from baseline were greater in the CEE group only at 48 months and in the 17β-estradiol group only at 18 months compared to placebo (table 3).

Global cognitive function did not change in the treatment or the placebo group, and there were no differences in global cognitive change between CEE, 17β-estradiol, and placebo groups over 48 months (figure 2).

Over the 48 months of follow-up, the increase in ventricular volume correlated with the decline in whole brain volume ($r = -0.58$; $p \leq 0.001$) and with the increase in WMH volume ($r = 0.27$; $p = 0.01$) after adjusting for age.

In the CEE group, increases in ventricular volume at 18 months correlated with timing of initiation of treatment in relation to the onset of menopause, after adjusting for age. Women who initiated hormone therapy later in menopause had greater increases in ventricular volume at 18 months ($r = 0.40$; $p = 0.03$). This association was weaker at 36 months ($r = 0.35$; $p = 0.07$) and at 48 months ($r = 0.26$; $p = 0.24$). By contrast, there were no correlations between increases in ventricular volume and timing

Table 2 Annual change in imaging markers on mixed-effects model

	Whole brain, cm^3, estimates (95% CI)	Ventricle, cm^3, estimates (95% CI)	WMH, cm^3, estimates (95% CI)
Intercept	1,303 (1,273, 1,333)[a]	17.5 (15.1, 20)[a]	1.78 (1.39, 2.17)[a]
Time from baseline	−1.12 (−2.43, 0.19)	0.27 (0.13, 0.41)[a]	0.03 (−0.04, 0.09)
CEE	30.9 (−14.3, 76.1)	2.41 (−1.27, 6.09)	0.85 (0.26, 1.43)[b]
17β-estradiol	12.8 (−31.9, 57.6)	4.4 (0.78, 8.1)[c]	0.58 (−0.007, 1.16)
CEE × time from baseline	−1.08 (−3.06, 0.9)	0.28 (0.07, 0.48)[b]	0.08 (−0.015, 0.18)
17β-estradiol × time from baseline	−1.14 (−3.1, 0.81)	0.18 (−0.03, 0.39)	0.09 (−0.002, 0.19)

Abbreviations: CEE = conjugated equine estrogens; CI = confidence interval; WMH = white matter hyperintensity.
Linear mixed-effects models with time from baseline, treatment group, and their interaction as predictors, whole brain, ventricle, and WMH volume change as outcomes, and random subject-specific intercepts and slopes are reported for the CEE, 17β-estradiol, and placebo groups. Coefficient estimates with their associated 95% CIs and categories of significance are reported for these models. Treatment × time interactions were of interest because any treatment effects would likely manifest as a change in the trajectories over time.
[a] $p < 0.001$.
[b] $p \leq 0.01$.
[c] $p < 0.05$.

of initiation of 17β-estradiol treatment, after adjusting for age.

DISCUSSION The major finding of this study is that the rates of increase in ventricular volumes were greater in recently menopausal women who received oral CEE therapy compared to placebo over 4 years without significant differences in changes in global cognition. Although the rates of changes in MRI measures did not differ between the CEE and 17β-estradiol groups, the rates of increase in ventricular volume did not reach statistical significance in the 17β-estradiol group, compared to placebo, probably because of our limited sample size and short follow-up. Furthermore, increases in ventricular volume over 18 months of treatment were greater in the CEE group if hormone therapy was initiated later in menopause.

It is difficult to compare the findings of the present study with the findings of other studies in the literature, because the earlier studies were conducted in older women, and the structural MRI assessments were cross-sectional and not longitudinal, and were performed many years after initiation of hormone therapy. However, given these limitations, greater rates of structural brain changes with CEE therapy compared to placebo in the present study are consistent with the finding of lower regional brain volumes in women who initiate hormone therapy at age 65 years or older in the WHIMS-MRI, when the structural MRI changes were observed after the study medications were stopped.[20] Our findings also agree with observational studies that found larger ventricular volumes in women using hormone therapies[21,22] but contradict other studies that showed larger brain volumes in women who used postmenopausal hormone therapy compared to women who did not.[23–26]

Although measurements of both whole brain and ventricular volumes are surrogates for the change in global brain structure and correlate with each other, differences between CEE treatment and placebo groups were statistically significant when comparing the rates of ventricular volume change but not the rates of whole brain volume change. This may be attributable to the differences in noise and measurement variability. Indeed, smaller sample sizes are needed to detect treatment effects using ventricular volume change compared to whole brain volume change.[14]

Brain volumes quantified on MRI decrease and ventricular volumes increase during physiologic aging.[27] The decrease in brain volume accelerates in cognitively normal older adults several years before they develop mild cognitive impairment[14] or dementia.[28] Although structural MRI changes are adequate surrogate markers for tracking the progression of preclinical neurodegenerative brain pathology many years before the onset of cognitive decline, they may not be ideal markers for detecting the effects of specific treatments that might modify brain function or pathology.[29] Evidence of hormonal effects on brain structure in younger women is limited to studies in premenopausal women. Premenstrual decreases in estrogen and progesterone levels are associated with increased ventricular volumes,[30] and postmenstrual increases in estrogen levels are associated with preservation of ventricular structure and even increases in hippocampal volume.[31] Furthermore, larger gray matter volumes were found in oral contraceptive users compared to nonusers.[32] Both cyclic increases in

Table 3 Percentage of change in imaging markers

	Whole brain volume				Ventricular volume				White matter hyperintensities			
Months	CEE mean change, %	Placebo mean change, %	Mean % difference (95% CI)	*p* Value	CEE mean change, %	Placebo mean change, %	Mean % difference (95% CI)	*p* Value	CEE mean change, %	Placebo mean change, %	Mean % difference (95% CI)	*p* Value
18	−0.3	0.034	−0.34 (−0.91, 0.24)	0.25	6.42	2.11	4.31 (1.57, 7)	0.003	3.88	−1.56	5.45 (−5.2, 16.1)	0.31
36	−0.42	−0.09	−0.33 (−0.97, 0.32)	0.31	9.35	3.96	5.39 (1.58, 9.2)	0.006	10.7	5.61	5.07 (−11.3, 21.4)	0.54
48	−0.64	−0.33	−0.31 (−1, 0.39)	0.38	12.3	5.84	6.5 (2.27, 11)	0.003	26.7	2.6	24.1 (1.88, 46.4)	0.03
Months	17β-Estradiol mean change, %	Placebo mean change, %	Mean % difference (95% CI)	*p* Value	17β-Estradiol mean change, %	Placebo mean change, %	Mean % difference (95% CI)	*p* Value	17β-Estradiol mean change, %	Placebo mean change, %	Mean % difference (95% CI)	*p* Value
18	−0.18	0.034	−0.21 (−0.8, 0.38)	0.48	3.94	2.11	1.83 (−0.81, 4.5)	0.17	12.6	−1.56	14.1 (2.11, 26.1)	0.02
36	−0.6	−0.09	−0.51 (−1.2, 0.17)	0.14	6.43	3.96	2.47 (−1.05, 6)	0.17	11.4	5.61	5.8 (−7.98, 19.6)	0.40
48	−0.82	−0.33	−0.49 (−1.1, 0.15)	0.13	10.1	5.84	4.28 (−0.119, 8.7)	0.06	28.5	2.6	25.9 (−5.68, 57.4)	0.10
Months	CEE mean change, %	17β-Estradiol mean change, %	Mean % difference (95% CI)	*p* Value	CEE mean change, %	17β-Estradiol mean change, %	Mean % difference (95% CI)	*p* Value	CEE mean change, %	17β-Estradiol mean change, %	Mean % difference (95% CI)	*p* Value
18	−0.3	−0.18	−0.13 (−0.7, 0.45)	0.66	6.42	3.94	2.48 (−0.267, 5.2)	0.08	3.88	12.6	−8.67 (−20.6, 3.24)	0.15
36	−0.42	−0.6	0.18 (−0.48, 0.84)	0.59	9.35	6.43	2.92 (−0.825, 6.7)	0.12	10.7	11.4	−0.728 (−17.2, 15.8)	0.93
48	−0.64	−0.82	0.18 (−0.5, 0.87)	0.59	12.3	10.1	2.22 (−2.54, 7)	0.35	26.7	28.5	−1.77 (−36.4, 32.9)	0.92

Abbreviations: CEE = conjugated equine estrogens; CI = confidence interval.
Percentage of change in whole brain volumes, ventricular volumes, and white matter hyperintensities compared in the CEE and 17β-estradiol and placebo groups at 18, 36, and 48 months.

endogenous hormones and exogenous administration of hormones appear to preserve ventricular volumes and increase brain volumes in premenopausal women, in contrast to the findings of the present study with exogenous administration of hormones in postmenopausal women. Therefore, transient retention of sodium and water, which has been implicated in the premenstrual increases in ventricular volume when endogenous estrogen is low, would not explain the finding of increased ventricular volume in postmenopausal women who received estrogens.

WMH volume gradually increased only in the CEE and 17β-estradiol treatment groups but not the placebo group. These increases in WMH volumes over 48 months correlated with the increases in ventricular volumes, suggesting that similar mechanisms may be driving both of these structural changes. Accelerated volumetric changes in brain structure and WMH are associated with cognitive decline in older adults, including ischemic small vessel disease and Alzheimer disease.[33] A relationship between longitudinal increases in WMH and ventricular volumes has been reported in a cohort with atherosclerosis, slightly older than the participants of the KEEPS-MRI study (mean age 53 vs 57 years) and with a high percentage of men, suggesting that cerebral small vessel disease may underlie the parallel increases in WMH and ventricular volumes.[34] Although the pathologic underpinnings of WMH and ventricular volume increases in women with good cardiovascular health is unclear, there is evidence that the WMH increases in participants of the KEEPS-MRI study were associated with thrombogenic microvesicles in the blood at baseline.[19] Thrombogenic microvesicles are related to the progression of atherosclerosis.[35]

In the CEE group, ventricular volumes increased less in women who initiated hormone therapy earlier than later in menopause, albeit only during the first 18 months. This association gradually weakened at 36 and 48 months of treatment. Since the mean time to starting CEE was 20 months past menopause (range: 5–36), the window in which hormone therapy causes less ventricular expansion appears to be limited to approximately 3 years into menopause. This time window may be associated with a gradual loss of estrogen receptor α in the postmenopausal brain.[36]

Women participating in the KEEPS-MRI study were in good cardiovascular and neurologic health, and well educated, which may limit generalization of findings to a broader population. However, in this homogeneous population of relatively healthy women, hormone therapy effects on brain structure were independent of concurrent cardiovascular or neurologic diseases. Furthermore, the sample size and the MRI changes in brain volumes were small,

limiting statistical power. Therefore, the changes observed in the 17β-estradiol group compared to placebo may reach statistical significance with a longer duration of the study. Further investigation including larger numbers of participants and longer follow-up is warranted. Even though the treatment and placebo groups did not differ at baseline regarding cardiovascular risk factors, the CEE group had greater WMH volume than the placebo group. Some of the differences observed across groups at baseline are not surprising given the relatively small number of women included in the MRI ancillary study. Randomization does not guarantee a balanced allocation across treatment groups when the numbers are small. We found no evidence that greater WMH volume at baseline influenced the findings on ventricular volume change. We acknowledge, however, that post hoc statistical comparisons are subject to biases.

Among older women who received estrogen treatment in the WHIMS-MRI, lower regional brain volume was associated with greater cognitive decline or dementia.[37] By contrast, we observed a change in brain structure over 4 years of hormone therapies that did not correlate with changes in cognitive function of recently menopausal women. The interpretation of this absence of an association between imaging findings and cognitive findings remains uncertain. On one hand, it is possible that the women who experienced structural brain changes with hormone therapies early in menopause will develop cognitive decline during an extended follow-up, and if proven, the imaging findings would be useful early surrogates of future clinical events. On the other hand, it is possible that these brain changes will subside after cessation of hormone therapy, and women who had structural brain changes during the 4 years of postmenopausal hormone therapy will not have an increased risk of cognitive decline over a longer follow-up.[38] In the WHIMS-MRI studies, even though the brain volumes were smaller in women who received CEE therapies compared to placebo years after the end of the treatment phase, the rates of decline in brain volumes remained similar to the placebo group in the years following the cessation of CEE therapies.[39] Thus, the short-term effects of estrogen treatment on the vascular system and on the brain may be different from long-term effects, and even opposed. Participants of the KEEPS-MRI study are being followed to determine whether these changes in brain structure are persistent after 4 years of hormone treatment and whether women with structural brain changes will develop clinically detectable cognitive decline.

Comment: Hormone therapy in the early postmenopausal stage—Safe for the brain?

Recent clinical trials, including the Women's Health Initiative Memory Study of Younger Women,[1] provide reassurance that hormone therapy (HT) confers no cognitive risk when taken early in the postmenopausal period. In contrast, results from this ancillary neuroimaging study from the Kronos Early Estrogen Prevention Study (KEEPS)[2] suggest a potential adverse effect of HT—increased ventricular volume—in women randomized to conjugated equine estrogen (CEE) early in the postmenopausal stage. These findings deserve consideration because brain changes related to Alzheimer disease are detectable earlier than neuropsychological changes.

This neuroimaging study is unique because it was embedded in a blinded randomized controlled trial, included 2 HT formulations, and repeatedly assessed multiple neuroimaging outcomes over a relatively long 4-year follow-up. Despite these strengths, the findings should be interpreted with caution because sample sizes were quite small and there were some differences in baseline neuroimaging measures across treatment groups. Critically, despite an emphasis on the adverse CEE outcomes, the ventricular volume findings in the transdermal 17β-estradiol group were similar to the CEE group but the effect just missed statistical significance ($p = 0.06$). Indeed, there were no differences in ventricular volume between CEE and 17β-estradiol groups, and their ventricular volumes were nearly identical at the end of the study.

Continued follow-up in KEEPS is warranted to determine the reproducibility of the cognitive and neuroimaging findings as women age and to address an issue of critical clinical importance that KEEPS is uniquely positioned to address. HT remains the standard treatment for moderate to severe vasomotor symptoms in women with no contraindications to HT. Approximately 28.5% of postmenopausal women younger than 55 years experience moderate to severe vasomotor symptoms, and these symptoms interfere with health- and work-related quality of life.[3] Increased awareness of sex difference in Alzheimer disease underscores the need to identify sex-related risk factors for cognitive decline and dementia.

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AUTHOR CONTRIBUTIONS

Dr. Kantarci: design or conceptualization of the study, data collection, analysis and interpretation of the data, drafting the manuscript, study funding. Ms. Tosakulwong: analysis or interpretation of the data, revising the manuscript. Mr. Lesnick: analysis or interpretation of the data, revising the manuscript. Ms. Zuk: analysis or interpretation of the data, revising the manuscript. Dr. Gunter: analysis or interpretation of the data, revising the manuscript. Dr. Gleason: data collection, analysis or interpretation of the data, revising the manuscript. Dr. Wharton: data collection, analysis or interpretation of the data, revising the manuscript. Dr. Dowling: analysis or interpretation of the data, revising the manuscript. Dr. Vemuri: analysis or interpretation of the data, revising the manuscript. Mr. Senjem: analysis or interpretation of the data, revising the manuscript. Dr. Shuster: analysis or interpretation of the data, revising the manuscript. Dr. Bailey: analysis or interpretation of the data, revising the manuscript. Dr. Rocca: analysis or interpretation of the data, revising the manuscript. Dr. Jack: analysis or interpretation of the data, revising the manuscript. Dr. Asthana: data collection, analysis or interpretation of the data, revising the manuscript. Dr. Miller: design or conceptualization of the study, data collection, analysis or interpretation of the data, revising the manuscript, study funding.

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