Characteristics of hormone therapy, cognitive function, and dementia: the prospective 3C Study.

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ABSTRACT

Objectives: To examine the association between hormone therapy (HT) and cognitive performance or dementia, focusing on the duration and type of treatment used, as well as the timing of initiation of HT in relation to the menopause.

Methods: Women 65 years and older were recruited in France as part of the Three City Study. At baseline and 2 and 4 year follow-up, women were administered a short cognitive test battery and a clinical diagnosis of dementia was made. Detailed information was also gathered relating to current and past HT use. Analysis was adjusted for a number of socio-demographic, behavioural, physical and mental health variables, as well as Apolipoprotein ε4 (Apoe-ε4).

Results: Among 3130 naturally postmenopausal women, current HT users performed significantly better than never users on verbal fluency, working memory and psychomotor speed. These associations varied according to the type of treatment and a longer duration of HT appeared to be more beneficial. However, initiation of HT close to the menopause was not associated with better cognition. HT did not significantly reduce dementia risk over 4 years but current treatment diminished the negative effect associated with Apoe-ε4.

Conclusions: Current HT was associated with better performance in certain cognitive domains but these associations are dependent on the duration and type of treatment used. We found no evidence that HT needs to be initiated close to the menopause to have a beneficial effect on cognitive function in later life. Current HT may decrease the risk of dementia associated with the Apoe-ε4 allele.

INTRODUCTION

Postmenopausal hormone therapy (HT) remains the most effective treatment for alleviating menopausal symptoms which affect up to 80% of women, yet its effect on cognitive ageing remains controversial[1, 2]. The discordance in findings across studies could relate to differences in study design and cognitive assessment. Compared to observational studies, randomised controlled trials (RCTs) are less susceptible to bias, in particular the healthy-user bias of women selectively taking treatment. Yet, the majority of RCTs have a shorter follow-up, and long-term changes in cognitive function over time may not be recorded. In addition, HT may have varying effects on specific cognitive functions, with evidence that it could preferentially protect verbal memory[3], although other specific domains have not been extensively studied. Most studies have focused on dementia or global function which may have ceiling effects in non-demented people. The type of HT and the duration of use might also play a role in determining its effect[4], and treatment may need to be administered around the menopause to have an optimal beneficial effect on later-life cognition[5].

While the role played by HT in cognitive dysfunction remains an important question to be addressed, it is unlikely to be answered in the near future by large-scale RCTs with long-term HT users. However, it is currently feasible to use existing data on hormone exposure and later-life cognition from longitudinal population-based studies. This study aims to determine the association between dementia or specific areas of cognition and different characteristics of HT in naturally postmenopausal women.

METHODS

The Three City Study (3C) is a multi-centre longitudinal study involving the French cities Bordeaux, Dijon and Montpellier of community-dwelling elderly[6]. Recruitment of the study

cohort took place between 1999 and 2001 with eligible participants (aged over 65 years and non-institutionalised) being randomly selected from the electoral rolls. Participants were administered standardised questionnaires by trained staff and underwent clinical examinations at baseline and each 2-year follow-up. Fasting blood samples were provided and Apoe-ε4 genotyping was also performed[7]. The data described here was gathered during the first 4 years of the study.

Standard protocol approvals, registrations and patient consents

The study protocol was approved by the Ethical Committee of the University- Hospital of Kremlin-Bicêtre (France). Written informed consent was obtained from all participants in the study.

Cognitive function

A battery of cognitive tests was administered by trained staff and assessed different areas of cognitive function (Table 1), as detailed previously[8]. All of the tests were administered at baseline, and wave 1 and 2 of follow-up, except the Trail Making test which was not given in wave 1. Consequently, analysis of these tasks involves 356 fewer participants.

Dementia diagnosis

A preliminary diagnosis and classification of dementia at each follow-up was made by 3C study clinical investigators, according to DSM-IV revised criteria[9] and was further validated by a national panel of neurologists independently of the 3C investigators. The onset of dementia was the date of the follow-up interview when dementia was diagnosed.

HT

Participants recorded current and past HT use, the duration and type of treatment, and age when treatment was first initiated. Current treatment was validated by presentation of the prescription or the medication. Menopause age and type of menopause were noted.

Statistical Analysis

Logistic regression models were deemed the most appropriate for our analysis, due to the distribution of scores on the cognitive tests. Low cognitive performance was defined as scoring in the lowest quintile for each cognitive test except the TMTA and TMTB, which were timed tasks and thus the highest quintile was considered. A substantial decline in cognitive function over follow-up was defined as the lowest quintile of the difference between baseline score and either follow-up visit, or the highest quintile for TMTA and TMTB. Multivariate models adjusted for age, education, centre, marital status, caffeine consumption, BMI, menopause age, anticholinergic medication, depressive symptoms, comorbidity, physical incapacities and Apoe-ε4. Longitudinal models also adjusted for baseline cognitive score. Cox proportional hazards models with delayed entry assessed the association between HT and dementia incidence. To avoid the non-proportionality in dementia risk with age, the time scale was age and the time origin birth[10]. Interaction terms between HT and other risk factors for cognitive function or dementia were considered. SAS (v9.1) was used for the analyses (SAS Institute,Inc.,North Carolina).

RESULTS

Of the 4429 naturally postmenopausal women initially recruited into the study, women were excluded from this analysis if they were diagnosed with possible dementia at inclusion (n=86), or if they died or were lost to follow-up (n=439). Women were also omitted if they had missing data concerning: cognitive testing at baseline or follow-up (n=400); HT use (n=118); or one of the covariates (n=256). Compared to the analysed sample (n=3130), those excluded were more likely to be older, single, depressed, never HT users and to have a lower education level, an earlier menopause age, physical incapacities, comorbidity and lower scores on all cognitive tests

($p<0.001$). Fifteen percent of the women studied reported current HT use, and a similar number were past users (Table 2). With the exception of the MMSE, the percentage of women who performed poorly on each of the cognitive tests was lowest among current HT users and highest amongst the never users.

Baseline cognitive function

In multivariate adjusted models (Table 3), current HT use was significantly associated with better performance on tasks of verbal fluency, visual memory and psychomotor speed, compared to women who had never used HT. There was no significant association between HT use and global cognitive function, verbal memory or executive function. There was no significant difference in cognitive performance between never and past HT use overall (data not shown), however recent past use (≤2yrs ago), was still associated with significantly better verbal fluency. Adjusted regression models (Table 4) further indicated that long rather than short-term HT was significantly associated with better performance on the Isaacs and TMTA tasks. However, this duration effect was not observed with visual memory. Only women initiating treatment more than 5yrs after the menopause had a reduced risk of poor verbal fluency compared to never HT users, but there was no apparent beneficial effect of later initiation with regards to visual memory and psychomotor speed. Transdermal estrogen preparations combined with a progestagen, and in particular synthetic progestin, were most significantly associated with a decreased risk of poor performance in the three areas of cognitive function examined.

Change in cognitive function

We failed to find a significant adjusted association between baseline HT use and 4-year change in global function ($p=0.32$), verbal fluency ($p=0.63$), visual memory ($p=0.65$), verbal memory ($p=0.41$) or executive function ($p=0.93$). However, HT use was associated with a decreased risk

of decline in psychomotor speed (OR=0.73, 95%CI:0.53-0.99,p=0.05). There was no difference in terms of the type or the timing of initiation of treatment.

Dementia Incidence

Over follow-up there were 79 cases of all-cause dementia and 53 were classified as Alzheimer's disease (AD). Adjusted Cox models failed to find a significant association between HT use and the incidence of dementia or AD (Table 5). However, the risk of dementia associated with Apoe-ε4 appeared to vary according to HT use. Apoe-ε4 was a strong risk factor for dementia incidence overall, and in stratified analysis increased dementia risk among never users, with a similar trend for past users. By contrast, there was no significant increase in dementia risk for women with an Apoe-ε4 allele who were current users of HT. The findings were comparable when we examined AD.

DISCUSSION

The effect of HT on cognitive function has been widely debated since publication of the results from the Women's Health Initiative Memory Study (WHIMS). In this large RCT, non-hysterectomized women administered conjugated equine estrogen (CEE) and medroxyprogesterone acetate (MPA), had a significantly increased risk of global cognitive decline[11] and of all-cause dementia[12] but not of mild cognitive impairment. Combined CEE and MPA also had a negative effect on verbal memory[13]. These negative results contrast with a number of observational studies, and our study has attempted to clarify such inconsistencies, by investigating different characteristics of HT and their association with a range of cognitive domains.

Our study supports the notion that HT has varying associations with different areas of cognitive function. We found that current HT users performed significantly better on the tasks of verbal fluency, visual memory and psychomotor speed, however there was no significant association with global function, verbal memory or executive function. Current HT was also significantly associated with a reduced risk of 4-year decline in psychomotor speed. By contrast, past HT was not significantly associated with cognitive performance except very recent past use (<2yrs) and verbal fluency. This suggests that any beneficial effect from current exposure is removed once treatment is stopped. Our findings corroborate previous meta-analyses of observational studies, showing that HT is associated with better functioning in certain specific domains[14, 15], such as working memory[16] and the WHIMS RCT also found a positive effect of HT using the same visual memory task (BVRT)[13]. However, of the previous studies which examined psychomotor speed, the majority found no association with HT use[13, 17]. Evidence indicates that HT may preferentially protect verbal performance, *i.e.* verbal fluency[18, 19] or verbal memory[2], although this has not been found consistently[13, 16]. Our study supports the beneficial effect of HT on verbal fluency, but no significant association with word recall. As we used only one measure of verbal memory, which is subject to ceiling effects in non-demented people, it is still possible that HT could be associated with other tasks assessing this same cognitive domain.

We found that the association of HT with cognitive function varied according to the duration and type of treatment, which emphasizes the importance of considering characteristics of HT. The majority of RCTs are short and only one type of treatment is administered, so important effects may not have been observed. To date, few observational studies have examined whether the type of HT used can influence the associations found. In contrast with the WHIMS where equine estrogen and MPA were used, the majority of women in our study used combined transdermal estradiol-progestagen preparations and this treatment was associated with better verbal fluency, visual memory and psychomotor speed. Interestingly however, the associations with cognitive

function remained significant after adjustment only among women using synthetic progestin, most commonly 19-norprogesterone derivatives (medrogestone or promegestone). The demographic characteristics of women using the different HT preparations were very similar, with the exception that users of estrogen alone were slightly older than women reporting other formulations. While variability in neuroprotective effect depending on the type of progestagen is not surprising, previous research suggests natural progestagens may be preferred over synthetic ones such as MPA[20]. The effect of 19-norprogesterone derivatives has however, not been evaluated in epidemiological studies, although they are among the most selective agonists of the progesterone receptor (also referred as "pure" progestagens), notably devoid of estrogenic, androgenic or glucocorticoid activities in contrast with MPA [21]. Definite conclusions cannot be drawn regarding the impact of progestagens however, as the absence of significant associations may result from the smaller sample size and thus a difference in statistical power.

Our finding that women reporting long-term rather than short-term treatment, performed significantly better on the tasks of verbal fluency and psychomotor speed, is in accordance with other studies reporting beneficial effects from increasing HT duration[17, 19]. Thus treatment may need to be administered for a sufficient length of time before any positive effects are observed. This could help explain the absence of significant effects in some RCTs, due to the shorter treatment period. Likewise, in the WHIMS, the positive and negative effects of HT on visual and verbal memory respectively, were only evident after long-term treatment.

As one of the few studies to directly examine the "critical window" hypothesis, we found no evidence to suggest that HT needs to be initiated close to the menopause to have a beneficial effect on later-life cognitive function. In fact in terms of verbal fluency, only women initiating treatment >5yrs after the menopause performed significantly better than women who had never used HT. The critical window hypothesis was thought to partially explain why positive associations between HT and cognition have been found more frequently among peri- or early

postmenopausal women, compared to older populations[2, 22]. However, only a couple of small studies have directly tested this hypothesis[23, 24]. Among 343 naturally postmenopausal women, those receiving 2-3yrs of treatment in the early postmenopausal had a decreased risk of overall cognitive impairment[23]. Similar results were also seen for women who were either current or long-term HT users, although with half the sample size, this did not reach significance. Thus this study showed that treatment in the early postmenopause can have long-lasting effects on cognitive function, but later initiation may also be beneficial[23]. In the REMEMBER pilot study [25] of 428 women aged over 60yrs, those who commenced HT before 56yrs had significantly better global function, compared with women initiating HT later, as well as better performance on the TMTA, a finding that was not supported by our data. However, with respect to the other cognitive domains examined, no significant difference was found between early and late initiators, including the positive association between HT and verbal fluency. In this study age at HT initiation was examined, rather than timing in relation to the menopause, and the authors did not differentiate current from past HT users or the duration of HT use. They also included both natural and surgically menopausal women, who may respond differently to HT in terms of cognitive outcomes[5, 26]. Hence, the "critical window" hypothesis is still awaiting definitive proof. On the other hand, additional experimental and clinical imaging observations suggest that even a delayed initiation of HT may still have beneficial effects on particular brain functions; the longer the duration of HT, the better the sparing of brain tissue and the greater the effects on brain structure with increasing age [21] .

In our study we found no association between HT use and dementia incidence over follow-up, which is in contrast with some[27-29] but not all previous longitudinal studies[22, 30]. The WHIMS RCT also failed to find a significant association between dementia incidence and unopposed estrogen treatment[31], although combined CEE+MPA was actually found to increase dementia risk[12]. The discrepancies between studies could results from differences in the age or health

status of the populations or the length of follow-up. HT may only be protective against AD when administered to younger women[22]. It is also possible that with a longer follow-up time for our study, and a higher number of incident dementia cases, we would have increased power to detect a significant association with HT.

An interesting finding from our study was that HT may modify the association between Apoe-ε4 and dementia, a possibility that has rarely been addressed previously. Women carrying the ε4 allele are known to have an increased risk for dementia, and such a significant association was found in our study. However, among current HT users, the risk of dementia was not significantly increased for Apoe-ε4 positive women. This finding is supported by a recent study[32], reporting that the 2-fold increased risk of familial AD associated with Apoe-ε4 was attenuated in women using HT. Experimental models have also shown that estradiol modulates Apoe expression. In a mouse memory model expressing human Apoe-ε4, which has reduced long-term potentiation (LTP) compared with wild-type mice, estradiol treatment selectively enhanced LTP [33]. On the other hand, another study found that oral treatment was associated with a reduced risk of global cognitive dysfunction, but only in women without an Apoe-ε4 allele[34]. It has also being reported that Apoe-ε4 negative women using unopposed estrogen had better episodic learning and memory[35], in unadjusted analysis. Surprisingly however, they found no difference in cognitive performance between Apoe-ε4 negative and positive non-HT users. Whether these different results are partly due to residual confounding remains uncertain, but these studies all highlight the importance of considering both HT and Apoe-ε4 status when examining cognition function/dementia. Importantly, the majority of previous studies on HT and the risk of incident dementia, including the WHIMS, did not control for Apoe-ε4 genotype.

There are some limitations to our study. Data concerning HT use were gathered retrospectively and are subject to recall errors. Current HT users were however encouraged to present medications or prescriptions, so that treatment could be verified. The 4-year follow-up time may

have been insufficient to examine decline in cognition, particularly as there is a component of learning with repeat cognitive tasks. Likewise, with only 79 cases of incident dementia we had reduced statistical power to detect any associations with HT. Bias due to the exclusion of participants, including women with possible dementia at inclusion may have also reduced our power to detect significant associations if they were present. The strengths of our results relate to the design of the 3C study and its community-based population. In addition to dementia, a number of specific areas of cognitive function were examined and we were able to investigate particular characteristics associated with HT. The size of the dataset and the vast information relating to each participant enabled adjustment for an extensive range of covariates, which may confound the cognition-HT relationship, reducing the risk of prescription bias in regards to women using HT.

While the association between HT and later-life cognition remains complex, our study suggests that HT is preferentially associated with certain cognitive domains and these associations vary depending on the duration) and type of treatment used which might explain some discrepancies with previous RCTs, including the WHIMS. We have no evidence that HT needs to be initiated close to the menopause to have a beneficial effect on later-life cognitive function. Our finding that HT can modify the association between Apoe-ε4 and dementia risk is potentially very interesting and thus warrants further investigation. Obviously recognised contraindications of HT use should be considered and HT prescription proceeded by a screening examination to determine probability of real benefit, rather than generalized prescription to all women.

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Table 1. Cognitive functions which were assessed and participant's baseline scores.

Cognitive Test	Domain	Range	Median Score [Interquartile Range]
Mini-Mental State Examination (MMSE) [36]	Global Function	19 – 30	28 [26 - 29]
Isaac's Set Test (Isaacs) [37]	Verbal Fluency	20 – 101	48 [42 – 55]
Benton's Visual Retention Test (BVRT) [38]	Visual Memory	0 – 15	12 [10 – 13]
Immediate and Delayed Recall (Word Recall) [39]	Verbal Memory	1 – 10	9 [8 - 10]
Trail Making test A (TMTA) [40]	Psychomotor Speed	19 – 190	50 [40 – 63]
Trail Making test B (TMTB) [40]	Executive Function	30 - 379	96 [75 - 128]

Table 2. Baseline socio-demographic, health and lifestyle characteristics of the 3130[a] participants, according to their use of hormone therapy (HT).

Characteristic	Never user (n=2169[a])	Past HT user (n=487[a])	Current HT user (n=474[a])	Test for difference Statistic	p
		Mean (S.D.)		ANOVA (*df*)	
Age	74.9 (5.3)	72.4 (4.7)	70.2 (3.3)	193.2 (2)	<0.001
Age at menopause	50.5 (4.1)	50.5 (4.3)	51.2 (4.2)	5.2 (2)	0.006
Body mass index (BMI, kg/m²)	25.5 (4.4)	24.9 (3.8)	24.4 (3.4)	16.5 (2)	<0.001
		%		χ^2 (*df*)	
≥ 12 years of education	24.1	26.1	35.0	24.0 (2)	<0.001
Married or has a partner	42.3	51.3	60.8	58.9 (2)	<0.001
High caffeine intake[b]	30.1	25.5	34.6	9.5 (2)	0.009
Depressive symptoms (CESD≥16)[c]	27.2	30.0	28.1	1.6 (2)	0.46
Carrier of the Apoe-ε4 allele	18.3	20.1	21.7	3.5 (2)	0.18
Anticholinergic medication	8.9	7.4	8.9	1.2 (2)	0.54
Comorbidity[d]	47.4	42.7	35.2	24.1 (2)	<0.001
Physical Incapacities[e]	9.8	4.3	2.5	38.6 (2)	<0.001

Centre [f]					
Bordeaux	*19.6*	*13.6*	*13.3*	23.9 (4)	<0.001
Dijon	*55.1*	*54.2*	*60.3*		
Montpellier	*25.3*	*32.2*	*26.4*		
MMSE score <26	15.3	15.8	12.9	2.1 (2)	0.35
Isaacs set test score ≤40	24.1	20.3	12.0	33.8 (2)	<0.001
Benton score ≤10	32.0	25.9	19.2	34.1 (2)	<0.001
Word recall score ≤8	30.1	26.9	21.3	15.2 (2)	<0.001
Trail Making Task A score ≥69	21.5	16.2	10.8	29.0 (2)	<0.001
Trail Making Task B score ≥139	21.9	17.3	11.9	24.0 (2)	<0.001

[a]Except for Trail Making Tasks A and B where N=2774 (1907 never users; 439 past HT users; 428 current HT user).

[b] Equivalent to three or more cups of coffee each day.

[c]The presence of depressive symptoms was assessed using the 20-item centre for epidemiology studies depression scale (CES-D), with a cut-off of ≥16.

[d]Participants were classified as having comorbidity if they had a history of vascular disease (angina pectoris, myocardial infarction, stroke, cardiovascular surgery, bradycardia or palpitations), other chronic illnesses (asthma, diabetes (fasting glucose≥7.0mmol/l or treatment), hypercholesterolemia (total cholesterol ≥6.2mmol/l), hypertension (resting blood pressure ≥160/95mm Hg or treatment) and thyroid problems) or a diagnoses of cancer within the last 2 years.

[e]Participants were classified as disabled if they were unable to complete at least two tasks from either the Instrumental Activities of Daily Living or the Activities of Daily Living (ADL) scales.

[f]Percentages given within each treatment type.

Table 3. Logistic regression models for the association between HT and cognitive function.

Use of Hormone Therapy	N	Global function, MMSE<26		Verbal fluency, Isaacs≤40		Visual memory, BVRT ≤10		Verbal memory, word recall ≤8		N	Psychomotor speed, TMTA ≥69		Executive Function, TMTB≥139	
		OR [95% CI]	*p*	*OR [95% CI]*	*p*	*OR [95% CI]*	*p*	*OR [95% CI]*	*p*		*OR [95% CI]*	*p*	*OR [95% CI]*	*p*
UNADJUSTED														
Never	2169	1		1		1		1		1907	1		1	
Current	474	0.82 [0.61-1.10]	0.18	0.43 [0.32-0.58]	0.001	0.50 [0.39-0.64]	0.001	0.63 [0.50-0.80]	0.001	428	0.44 [0.32-0.61]	0.001	0.48 [0.35-0.66]	0.001
Past: ≤2 years ago	141	1.19 [0.76-1.87]	0.44	0.52 [0.32-0.85]	0.008	0.73 [0.49-1.07]	0.11	0.89 [0.61-1.30]	0.55	129	0.84 [0.53-1.32]	0.44	0.81 [0.52-1.29]	0.38
Past: >2 & ≤12yrs ago	140	1.15 [0.73-1.80]	0.56	0.86 [0.57-1.30]	0.48	0.76 [0.52-1.12]	0.17	0.53 [0.34-0.82]	0.004	127	0.49 [0.28-0.85]	0.01	0.70 [0.44-1.14]	0.16
Past: >12 & ≤22yrs ago	116	0.82 [0.47-1.43]	0.49	0.87 [0.55-1.36]	0.54	0.68 [0.44-1.04]	0.08	0.97 [0.64-1.45]	0.87	104	0.66 [0.39-1.14]	0.14	0.60 [0.34-1.05]	0.07
Past: >22yrs ago	90	0.93 [0.51-1.70]	0.82	1.15 [0.71-1.85]	0.57	0.82 [0.51-1.31]	0.40	1.28 [0.83-1.99]	0.27	79	0.93 [0.53-1.62]	0.79	0.91 [0.52-1.58]	0.73
MULTIVARIATE[a]														
Never	2169	1		1		1		1		1907	1		1	
Current	474	1.02 [0.74-1.40]	0.90	0.64 [0.47-0.88]	0.005	0.70 [0.54-0.92]	0.009	0.89 [0.69-1.14]	0.35	428	0.66 [0.47-0.94]	0.02	0.79 [0.56-1.10]	0.16
Past: ≤2 years ago	141	1.28 [0.81-2.04]	0.30	0.59 [0.36-0.98]	0.04	0.83 [0.56-1.25]	0.38	1.01 [0.68-1.49]	0.98	129	1.01 [0.63-1.62]	0.97	0.98 [0.61-1.59]	0.95
Past: >2 & ≤12yrs ago	140	1.27 [0.78-2.04]	0.34	1.30 [0.83-2.01]	0.25	1.08 [0.72-1.62]	0.73	0.73 [0.46-1.44]	0.17	127	0.73 [0.42-1.29]	0.29	1.18 [0.71-1.95]	0.53

Past: >12 & ≤22yrs ago	116	0.85 [0.48-1.50]	0.58	1.06 [0.66-1.70]	0.80	0.79 [0.51-1.24]	0.32	1.17 [0.77-1.78]	0.46	104	0.87 [0.50-1.51]	0.61	0.78 [0.44-1.38]	0.39
Past: >22yrs ago	90	0.98 [0.53-1.84]	0.96	1.14 [0.69-1.89]	0.60	0.82 [0.51-1.33]	0.42	1.24 [0.79-1.95]	0.34	79	0.90 [0.51-1.59]	0.71	0.90 [0.51-1.61]	0.73

[a]Adjusted for centre, age, education level, age at menopause, marital status, depressive symptoms, physical incapacities, comorbidity, Apoe-ε4 allele, high caffeine consumption, anticholinergic drugs and BMI.

Table 4. Logistic regression models for the association between characteristics of current HT use and poor cognitive function at baseline.

Characteristics of current HT users	N	Unadjusted Verbal fluency, Isaacs≤40 OR [95% CI]	p	Multivariate[a] Verbal fluency, Isaacs≤40 OR [95% CI]	p	Unadjusted Visual memory, BVRT ≤10 OR [95% CI]	p	Multivariate[a] Visual memory, BVRT ≤10 OR [95% CI]	p	N	Unadjusted Psychomotor speed, TMTA ≥69 OR [95% CI]	p	Multivariate[a] Psychomotor speed, TMTA ≥69 OR [95% CI]	p
Duration of use:														
Never used	2169	1		1		1		1		1907	1		1	
<10yrs	157	0.60 [0.39-0.93]	0.02	0.85 [0.53-1.35]	0.48	0.54 [0.36-0.81]	<0.01	0.72 [0.47-1.09]	0.12	142	0.56 [0.34-0.93]	0.02	0.82 [0.49-1.38]	0.46
>10 & ≤15yrs	160	0.40 [0.24-0.66]	<0.01	0.71 [0.42-1.21]	0.21	0.45 [0.30-0.68]	<0.01	0.71 [0.46-1.10]	0.12	143	0.46 [0.27-0.78]	<0.01	0.81 [0.46-1.41]	0.45
>15yrs	127	0.33 [0.18-0.60]	<0.01	0.48 [0.26-0.90]	0.02	0.57 [0.37-0.88]	<0.01	0.78 [0.50-1.23]	0.29	116	0.35 [0.18-0.67]	<0.01	0.50 [0.26-0.98]	0.04
Initiation of use:														
Never used	2169	1		1		1		1		1907	1		1	
≤5yrs of menopause	255	0.42 [0.28-0.62]	<0.01	0.78 [0.51-1.19]	0.25	0.45 [0.33-0.64]	<0.01	0.72 [0.51-1.03]	0.08	231	0.40 [0.26-0.63]	<0.01	0.69 [0.43-1.10]	0.12
>5yrs after menopause	194	0.47 [0.30-0.72]	<0.01	0.58 [0.37-0.91]	0.02	0.60 [0.43-0.86]	<0.01	0.72 [0.50-1.04]	0.08	175	0.53 [0.33-0.83]	<0.01	0.72 [0.45-1.16]	0.18
Type of HT:														

Never used	2169	1		1		1		1		1907	1		1	
Estrogen alone	46	0.47 [0.20-1.12]	0.09	0.60 [0.25-1.46]	0.26	0.67 [0.34-1.32]	0.25	0.79 [0.39-1.61]	0.53	41	0.40 [0.14-1.11]	0.08	0.53 [0.18-1.52]	0.24
Oral estrogen & progestagen	78	0.52 [0.27-0.99]	0.05	0.85 [0.44-1.67]	0.64	0.46 [0.26-0.83]	0.01	0.69 [0.37-1.26]	0.23	73	0.39 [0.18-0.85]	0.02	0.58 [0.26-1.30]	0.19
Transdermal estrogen & progestagen	329	0.39 [0.27-0.56]	<0.01	0.60 [0.41-0.88]	0.009	0.48 [0.36-0.67]	<0.01	0.69 [0.50-0.94]	0.02	295	0.44 [0.30-0.65]	<0.01	0.69 [0.46-1.03]	0.07
Natural progesterone	*164*	*0.44 [0.27-0.71]*	*<0.01*	*0.65 [0.39-1.07]*	*0.09*	*0.58 [0.39-0.85]*	*<0.01*	*0.79 [0.53-1.19]*	*0.26*	*148*	*0.57 [0.35-0.93]*	*0.02*	*0.86 [0.52-1.42]*	*0.55*
Synthetic progestin	*165*	*0.34 [0.20-0.59]*	*<0.01*	*0.55[0.32-0.95]*	*0.03*	*0.40 [0.26-0.61]*	*<0.01*	*0.59[0.37-0.91]*	*0.02*	*147*	*0.33 [0.18-0.59]*	*<0.01*	*0.52[0.28-0.97]*	*0.04*
Other / unsure	21	Nd		Nd		Nd		Nd		19			Nd	

[a]Adjusted for centre, age, education level, age at menopause, marital status, depressive symptoms, physical incapacities, comorbidity, Apoe-ε4 allele, high caffeine consumption, anticholinergic drugs and BMI.

Table 5. Cox proportional hazards models for the incidence of dementia and Alzheimer's Disease according to HT use.

Exposure	N	Incidence of Dementia (n=79 cases) Unadjusted HR [95% CI]	p	Multivariate HR [95% CI]	p	N	Incidence of Alzheimer's Disease (n=53 cases)[b] Unadjusted HR [95% CI]	p	Multivariate HR [95% CI]	p
Never use of HT	2169	1		1		2146	1		1	
Past use of HT	487	0.76 (0.38-1.51)	0.43	0.74 (0.35-1.55)	0.42	485	0.79 (0.34-1.83)	0.57	0. 39 (0.39-2.23)	0.86
Current use of HT	474	1.04 (0.43-2.50)	0.94	0.83 (0.32-2.17)	0.70	473	1.61 (0.60-4.32)	0.34	1.36 (0.44-4.20)	0.59
Apoe-ε4 + (all women)	3130	2.35 (1.53-3.60)	<0.001	2.27 (1.48-3.49)	<0.001	3104	2.38 (1.42-4.00)	0.001	2.21 (1.31-3.74)	0.003
Apoe-ε4 - & never HT user	1773	1		1		1757	1		1	
Apoe-ε4 - & past HT user	389	0.61 (0.25-1.53)	0.30	0.54 (0.22-1.35)	0.19	387	0.56 (0.17-1.82)	0.34	0.48 (0.15-1.57)	0.23
Apoe-ε4 - & current HT user	371	1.35 (0.52-3.53)	0.54	1.05 (0.41-2.73)	0.92	370	2.01 (0.67-5.97)	0.21	1.45 (0.49-4.32)	0.51
Apoe-ε4 + & never HT user	396	2.33 (1.47-3.71)	<0.001	2.24 (1.41-3.56)	<0.001	389	2.29 (1.30-4.05)	0.004	2.13 (1.20-3.80)	0.01
Apoe-ε4 + & past HT user	98	2.58 (0.92-7.23)	0.07	2.33 (0.83-6.53)	0.11	98	3.24 (0.98-10.7)	0.05	2.54 (0.76-8.53)	0.13
Apoe-ε4 + & current HT user	103	1.13 (0.15-8.39)	0.91	0.87 (0.12-6.52)	0.89	103	2.17 (0.28-16.6)	0.46	1.47 (0.19-11.5)	0.71

[a]Adjusted for age, education level, centre, high caffeine consumption, depressive symptoms, physical incapacities, comorbidity, marital status, anticholinergic drugs, body mass index and age at menopause (adjustment was also made for Apoe-ε4 when the association with HT use was examined).