Alzheimer's Disease: Efficacy of Transdermal 17-ß Estradiol

Sanjay Asthana, M.D., F.R.C.P. (C)

Alice B. Thistlethwaite Chair in Geriatrics
Head, Section of Geriatrics & Gerontology, Department of Medicine
University of Wisconsin Medical School

Director, Geriatric Research, Education and Clinical Center (GRECC)
Wm S. Middleton Memorial Veteran's Hospital

Associate Director, Wisconsin Alzheimer's Institute

Womens' Health Initiative

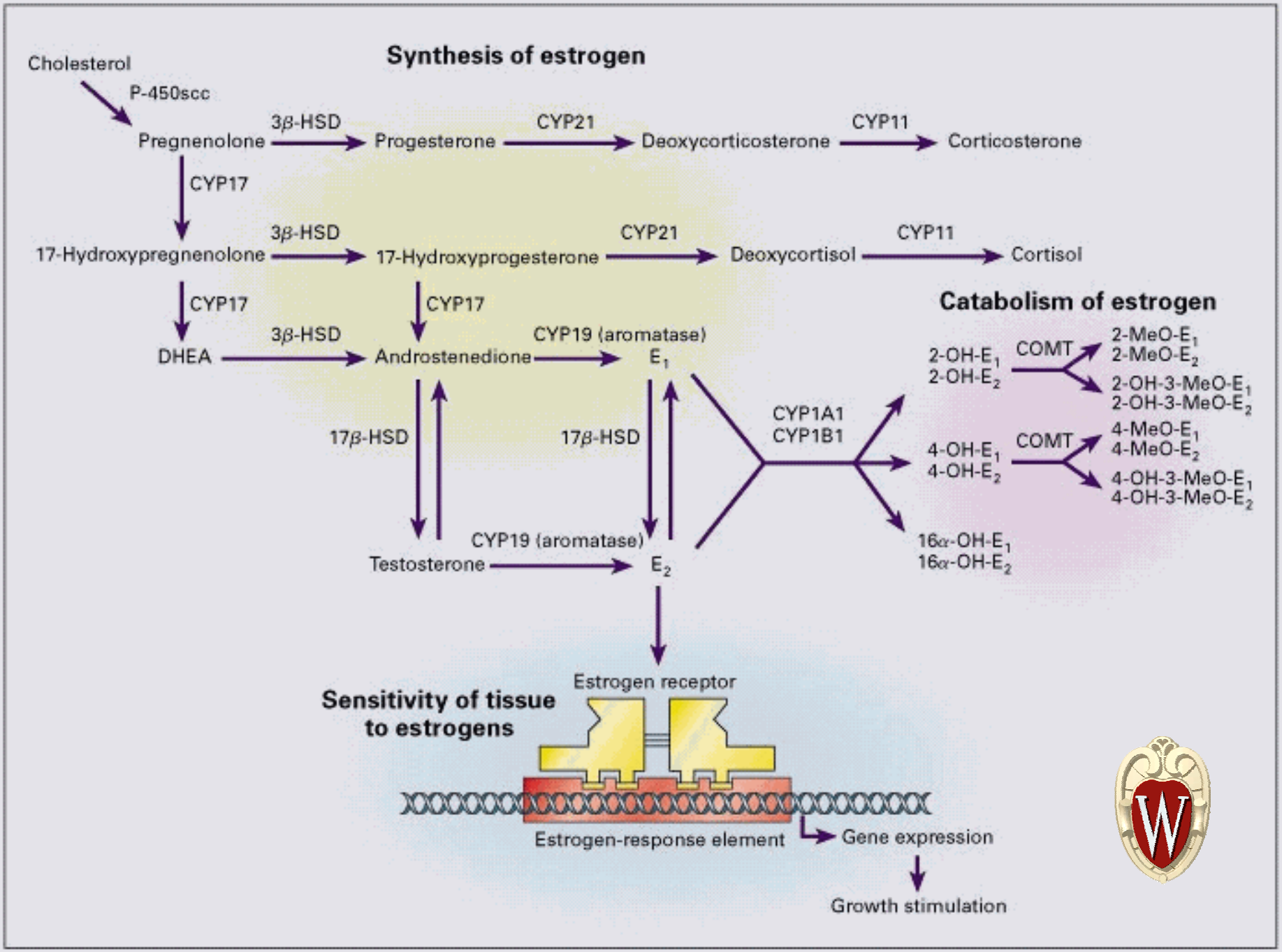
1. What is the best **form of estrogen** for evaluation in clinical studies?
2. What is the best **route of administration**?
3. What are the most sensitive **cognitive measures** for inclusion in clinical studies?
4. What is the best **form of progestin**?
5. Administration: **Cyclic** vs. **Continuous**
6. **Perimenopausal vs. Postmenopausal** women

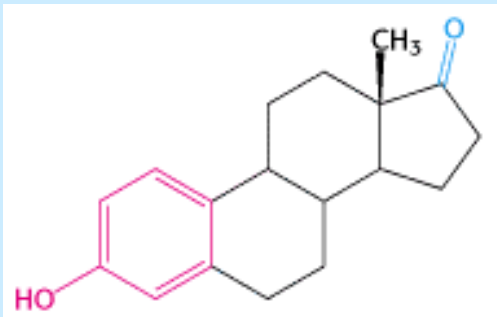
Pharmacology & Biochemistry of Hormones

Estrogen Forms:

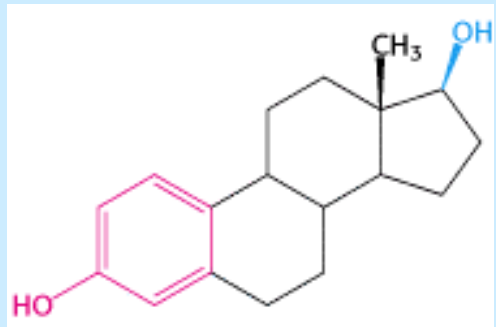
- 17-β-Estradiol - most potent and natural form of estrogen
- Estrone
- Estriol

> metabolites of Estradiol


Synthesis of estrogen
Cholesterol
P-450scc
Pregnenolone
3β-HSD
Progesterone
CYP21
Deoxycorticosterone
CYP11
Corticosterone
CYP17
17-Hydroxypregnenolone
3β-HSD
17-Hydroxyprogesterone
CYP21
Deoxycortisol
CYP11
Cortisol
CYP17
CYP17
DHEA
3β-HSD
Androstenedione
CYP19 (aromatase)
E1
Catabolism of estrogen
2-OH-E1
2-OH-E2
COMT
2-MeO-E1
2-MeO-E2
2-OH-3-MeO-E1
2-OH-3-MeO-E2
17β-HSD
17β-HSD
CYP1A1
CYP1B1
4-OH-E1
4-OH-E2
COMT
4-MeO-E1
4-MeO-E2
4-OH-3-MeO-E1
4-OH-3-MeO-E2
CYP19 (aromatase)
Testosterone
E2
16α-OH-E1
16α-OH-E2
Estrogen receptor
Sensitivity of tissue to estrogens
Estrogen-response element
Gene expression
Growth stimulation



Estrone



Estradiol

Endocrinology of Menopause

	Premenopausal	Postmenopausal
Predominant Source	17-ß-Estradiol	Estrone & Estrone Sulfate
Source of Production	Ovaries	a) Peripheral conversion of androstenedione b) The major source of Estradiol is conversion of Estrone
Plasma Hormone Concentrations	**17-ß-Estradiol Levels** a) Early follicular – 40 pg/mL b) Pre-ovulatory – 250 pg/mL c) Mid-ovulatory – 100 pg/mL	**Estradiol Levels** 15 pg/mL
	Estrone Levels a) Early follicular – 50 μg b) Pre-ovulatory – 350 μg c) Mid-ovulatory – 250 μg	**Estrone Levels** 0-10 μg/day
Gonadotropins	Normal	Elevated

Commercially Available Hormone Preparations

Preparation		Dosage (mg)	Estradiol (pg/mL)	Estrone (pg/mL)
Oral	Conjugated Equine Estrogen	0.625	30-50	153
		1.25	40-60	120-200
	Piperazine Estrone Sulfate	0.625	34	125
		1.25	30-50	150-300
		2.5	126	356
	Micronized estradiol	1	30-50	150-300
		2	114±65	575±280
	Estradiol valerate	1	50	160
		2	60-70	185-300
Vaginal	Conjugated equine estrogen	1.25	33±7	73±9
	Micronized estradiol	0.5	250	130
	Estradiol vaginal ring	100	40-50	
		200	70-80	
		400	1470	55
Parenteral	Transdermal estradiol	0.05	33-62	38-45
		0.1	48-89	32-64
	Percutaneous estradiol	1.5	68±27	90
		3	103±40	45-155
	Estradiol pellets	25	50-70	30
		50	100-120	72

SG Levrant, R.B., *Pharmacology of Estrogen*, in *Treatment of the Postmenopausal Woman*, R. Lobo, Ed. 1996, Lippincott-Raven: Philadelphia. p. 57-68.

Conjugated Equine Estrogen

- Extracted from the urine of pregnant mares
- Composition
  - Estrone Sulfate – 45%
  - Equlin Sulfate – 25%
  - 17 α-dihydroequilin – 15%
  - Trace amounts of:
    - Equilin, 17 b-estradiol
    - Equilenin
    - 17 β-dihyrdroequilin
    - 17 β-dyhydroequilenin
- Peak levels of estrone are accomplished in 6 – 10 hours and return to baseline in 48 hours – importance of compliance.

Salutory Effects of Estrogen on the Neurobiology of Alzheimer's Disease

Basic Research

- Facilitates Neurotransmission
  - Cholinergic
  - Catecholaminergic
  - Serotonergic
- Neurotrophic Effects
  - Synaptogenesis
  - Axonal arborization
  - Neurotrophins
  - Increased survivability of neurons in culture
  - Potential Effects on ß-amyloid processing

Clinical Research

- Enhances cognitive function – memory & attention
- Reduced risk of getting AD
- Antidepressant activities

Healthy Aging: Estrogen Treatment Studies

YEAR	INVESTIGATOR(S)	ESTROGEN PREPARATION	OUTCOME	YEAR	INVESTIGATOR(S)	ESTROGEN PREPARATION	OUTCOME
1952	Caldwell & Watson	Estradiol benzoate	Benefit	1999	Wolf et al.	17 ß-estradiol	Benefit
1975	Rauramo et al.	Estradiol valerate	No Benefit	2000	Duka et al.	17 ß-estradiol	Benefit
1976	Vanhulle & Demol	Estriol	No Benefit	2000	Janowsky et al.	CEEs	No Benefit
1976	Hackman & Galbraith	Piperazone sulphate	Benefit	2000	Rudolph et al.	Estradiol valerate	Benefit
1977	Fedor-Freyburgh	17 ß-estradiol	Benefit	2001	Linzmayer et al.	Estradiol valerate w/ or w/o dienogest	Benefit
1977	Campbell & Whitehead	CEEs	Benefit	2001	Binder et al.	CEEs	No Benefit
1988	Sherwin	Estradiol valerate	Benefit	2002	Grady et al.	CEEs	No Benefit
1989	Honjo et al.	CEEs	Benefit	2003	Saletu	Estradiol valerate	Benefit
1990	Sherwin & Phillips	Estradiol valerate	Benefit	2003	Woo et al.	CEEs	Benefit
1991	Ditkoff et al.	CEEs	No Benefit	2003	Shaywitz et al.	CEEs	Benefit
1992	Phillips & Sherwin	Estradiol valerate	Benefit	2003	Pan et al.	CEEs	Benefit
1998	Polo-Kantola et al.	17 ß-estradiol	No Benefit	2003	Kugaya et al.	17 ß-estradiol	Benefit
1999	Hogervorst et al.	17 ß-estradiol	Benefit	2003	WHIMS: Rapp et al. & Shumaker, et al.	CEEs	Harmful
1999	Shaywitz et al.	CEEs	Benefit & No Benefit				

Healthy Aging: Estrogen Treatment Studies

Healthy Aging Studies

Form of Estrogen	Total # of Studies	Positive Findings	Negative Findings	% Positive
Estradiol (both oral and transdermal)	14	12	2	86%
CEE	12	6	6	50%

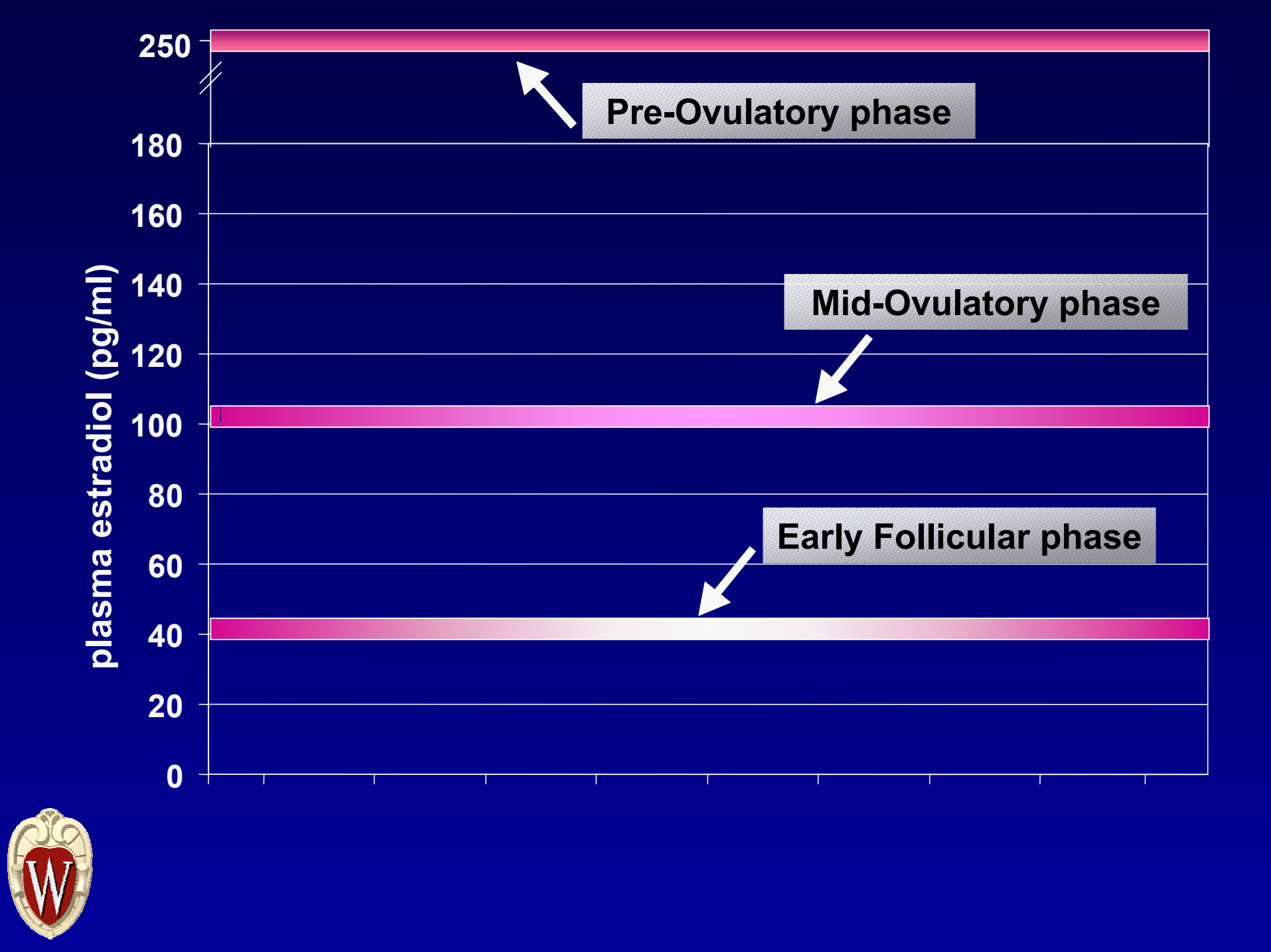
Alzheimer's disease: Estrogen Treatment Studies

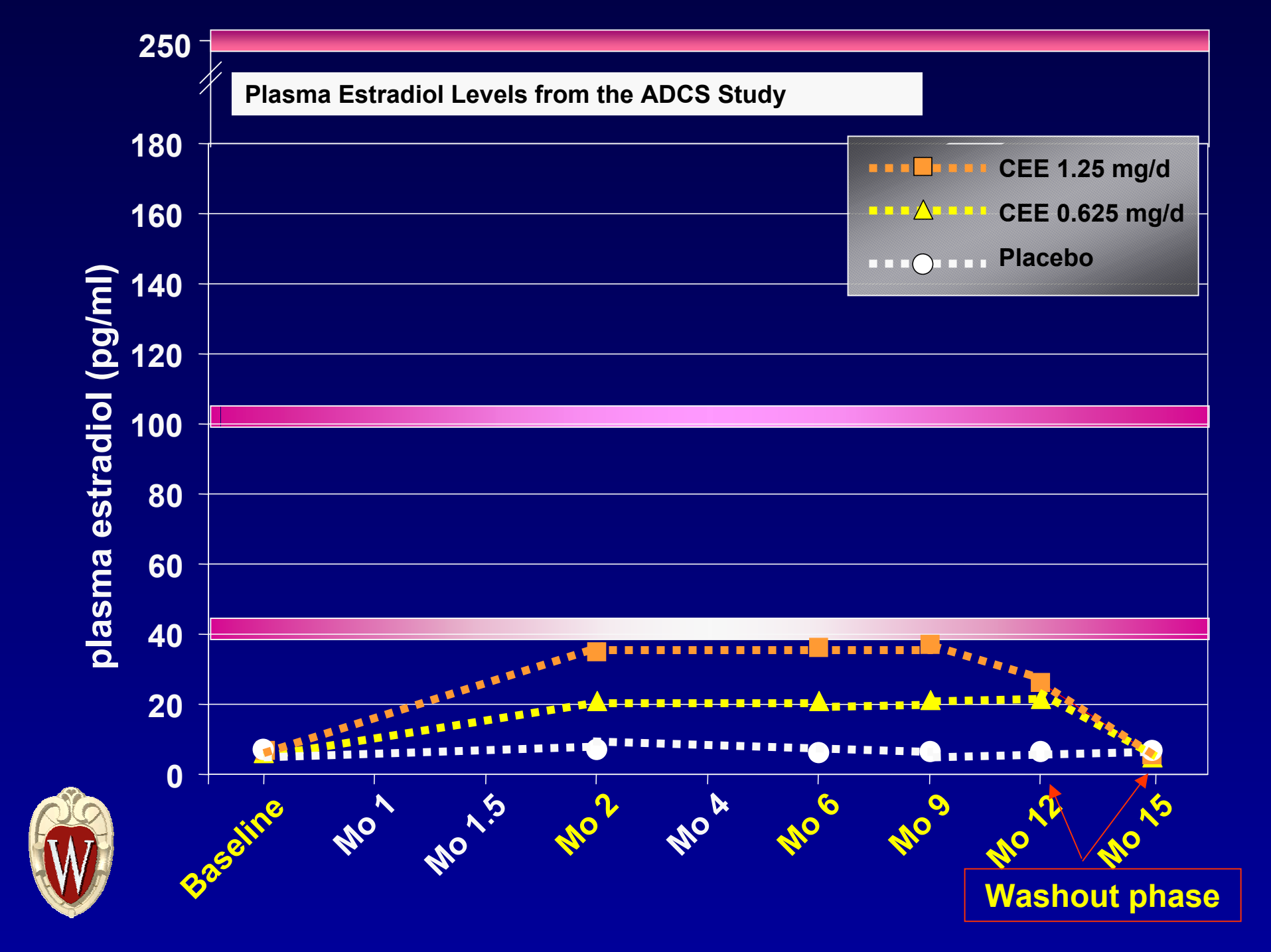
YEAR	INVESTIGATOR(S)	ESTROGEN PREPARATION	OUTCOME
1986	Fillit et al.	Estradiol	Benefit
1994	Okura et al.	CEEs	Benefit
1994	Okura et al.	CEEs	Benefit
1995	Okura et al.	CEEs	Benefit
1996	Schneider et al.	Several forms of ERT	Benefit
1999	Asthana et al.	17 ß-estradiol	Benefit
2000	Wang et al.	CEEs	No Benefit
2000	Henderson et al.	CEEs	No Benefit
2000	Mulnard et al.	CEEs	No Benefit
2001	Asthana et al.	17 ß-estradiol	Benefit
2003	Yoon et al.	CEEs	Benefit

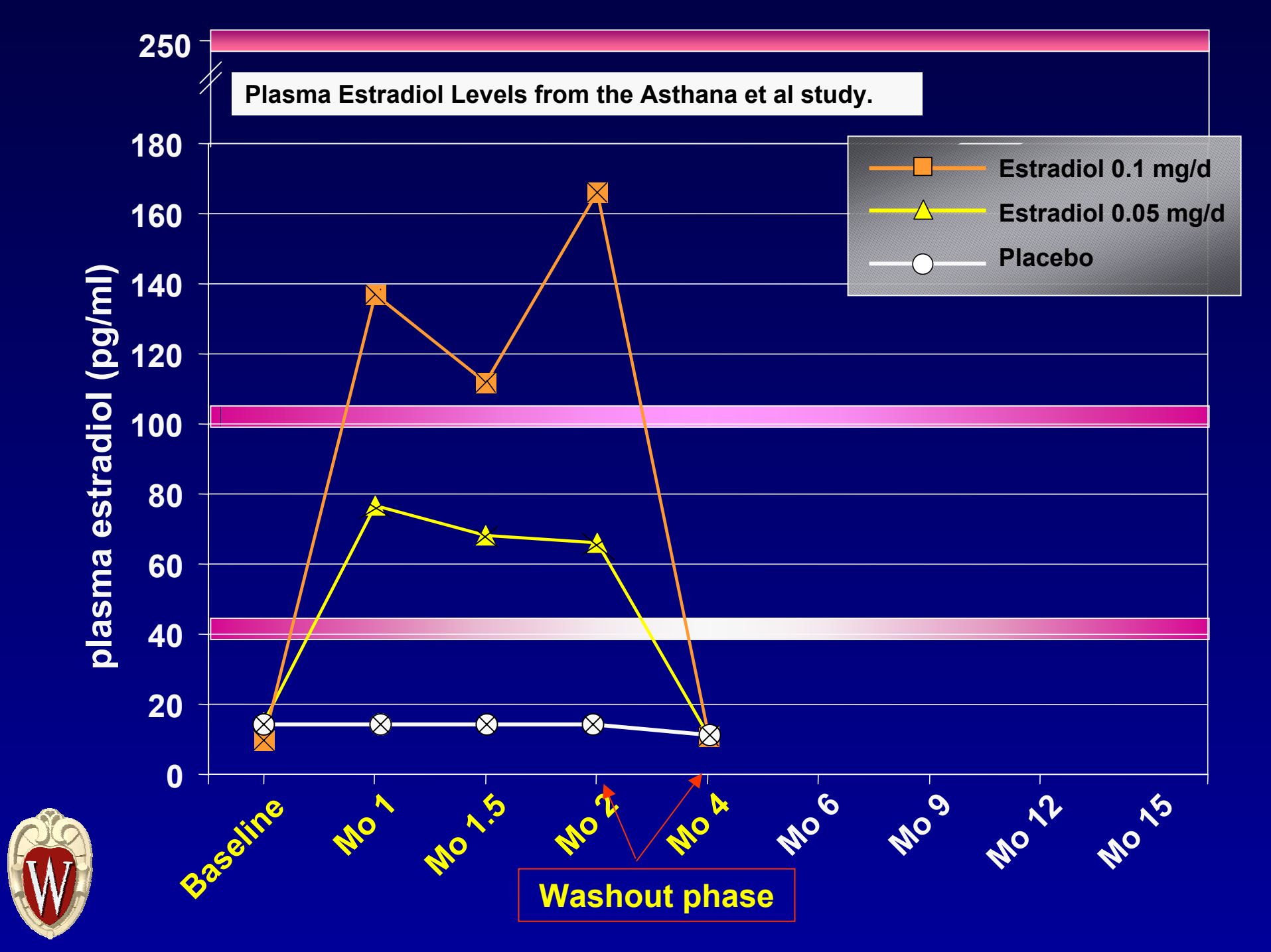
Alzheimer's disease: Estrogen Treatment Studies

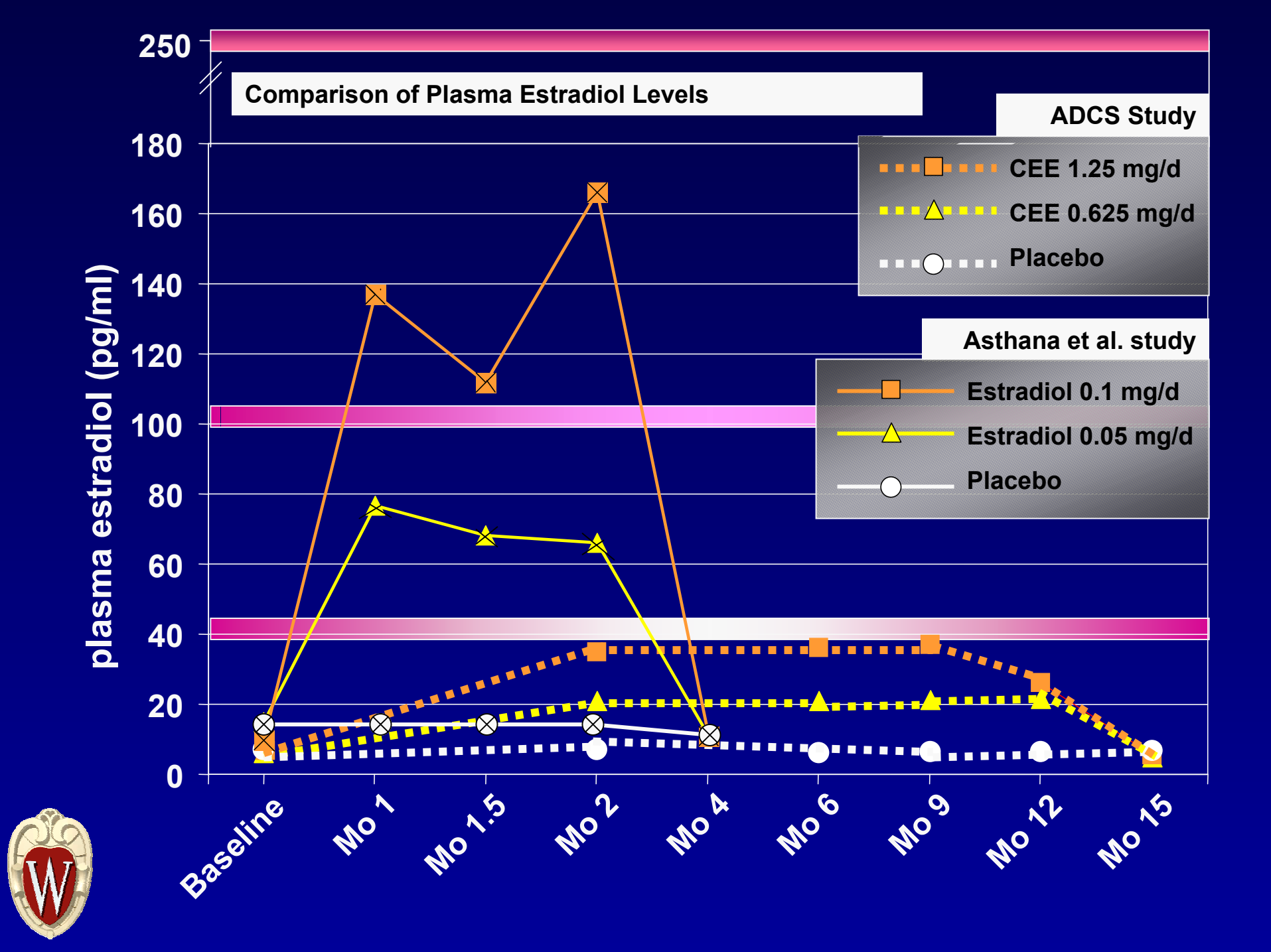
YEAR	INVESTIGATOR(S)	ESTROGEN PREPARATION	OUTCOME
2000	Mulnard et al.	CEEs	No Benefit
2001	Asthana et al.	17 ß-estradiol	Benefit
2003	Yoon et al.	CEEs	Benefit

Alzheimer's Disease Studies				
Form of Estrogen	Total # of Studies	Positive Findings	Negative Findings	% Positive
Estradiol (both oral and transdermal)	3	3	0	100%
CEE	7	4	3	57%


250
180
160
140
120
100
80
60
40
20
0
plasma estradiol (pg/ml)
Pre-Ovulatory phase
Mid-Ovulatory phase
Early Follicular phase


Plasma Estradiol Levels from the ADCS Study
plasma estradiol (pg/ml)
250
180
160
140
120
100
80
60
40
20
0
CEE 1.25 mg/d
CEE 0.625 mg/d
Placebo
Baseline
Mo 1
Mo 1.5
Mo 2
Mo 4
Mo 6
Mo 9
Mo 12
Mo 15
Washout phase


Plasma Estradiol Levels from the Asthana et al study.
plasma estradiol (pg/ml)
250
180
160
140
120
100
80
60
40
20
0
Estradiol 0.1 mg/d
Estradiol 0.05 mg/d
Placebo
Baseline
Mo 1
Mo 1.5
Mo 2
Mo 4
Mo 6
Mo 9
Mo 12
Mo 15
Washout phase


Comparison of Plasma Estradiol Levels
plasma estradiol (pg/ml)
250
180
160
140
120
100
80
60
40
20
0
Baseline
Mo 1
Mo 1.5
Mo 2
Mo 4
Mo 6
Mo 9
Mo 12
Mo 15
ADCS Study
CEE 1.25 mg/d
CEE 0.625 mg/d
Placebo
Asthana et al. study
Estradiol 0.1 mg/d
Estradiol 0.05 mg/d
Placebo

The most important finding of WHI is the increased incidence of thromboembolic complications of CEE

WHI Opposed (Prempro®)

	Hazard Ratio	Nominal 95% CI	Adjusted 95% CI
Venous thromboembolic complications	2.11	1.58-2.82	1.26-3.55
Deep Vein Thrombosis	2.07	1.49-2.87	1.14-3.74
Pulmonary Embolism	2.13	1.39-3.25	0.99-4.56
Stroke	1.41	1.07-1.85	0.86-2.31
Fatal	1.20	0.58-2.50	0.32-4.49
Non-Fatal	1.50	1.08-2.08	0.83-2.70

Rossouw, J.E., et al., *Risks and benefits of estrogen plus progestin in healthy postmenopausal women: principal results From the Women's Health Initiative randomized controlled trial.* Jama, 2002. **288**(3): p. 321-33.

The most important finding of WHI is the increased incidence of thromboembolic complications of CEE

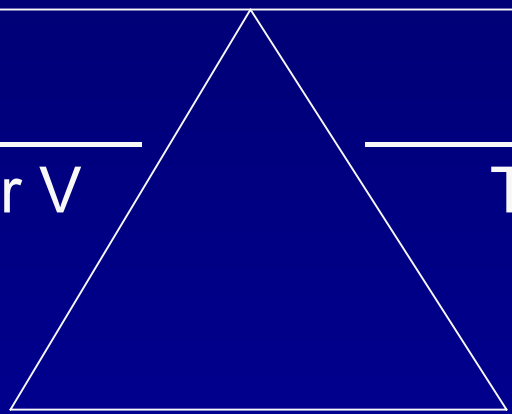
WHI Unopposed (Premarin®)

	Hazard Ratio	Nominal 95% CI	Adjusted 95% CI
Venous thromboembolic complications	1.33	0.99-1.79	0.86-2.08
Deep Vein Thrombosis	1.47	1.04-2.08	0.87-2.47
Pulmonary Embolism	1.34	0.87-2.06	0.70-2.55
Stroke	1.39	1.10-1.77	0.97-1.99
Fatal	1.13	0.54-2.34	0.38-3.36
Non-Fatal	1.33	0.99-1.79	0.86-2.47

Anderson, G.L., et al., *Effects of conjugated equine estrogen in postmenopausal women with hysterectomy: the Women's Health Initiative randomized controlled trial.* Jama, 2004. **291**(14): p. 1701-12.

Estrogen & Thrombosis

Coagulation	Anti-Coagulation
Pro-Enzymes	**Regulatory Proteins**
Factor VII	Factor IX	Protein C
Factor XI	Factor X	Protein S
Prothrombin	Anti-Thrombin III
Pro-Cofactors	**Pro-Cofactors**
Factor VII	Factor V	Thrombomodulin
Tissue Factor	Heparin



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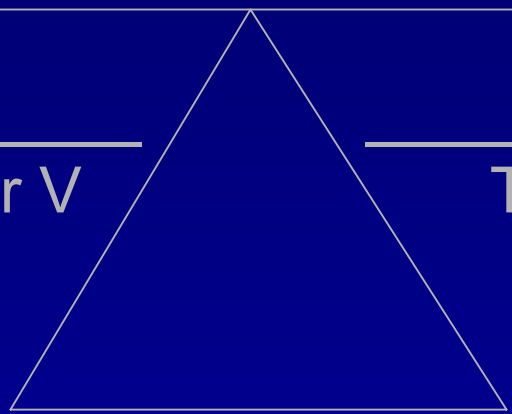
Estrogen & Thrombosis

Coagulation	Anti-Coagulation
Pro-Enzymes	**Regulatory Proteins**
Factor VII	Factor IX	Protein C
Factor XI	Factor X	Protein S
Prothrombin	Anti-Thrombin III
Pro-Cofactors	**Pro-Cofactors**
Factor VII	Factor V	Thrombomodulin
Tissue Factor	Heparin



SG Levrant, R.B., *Pharmacology of Estrogen*, in *Treatment of the Postmenopausal Woman*, R. Lobo, Editor. 1996, Lippincott-Raven Publishers: Philadelphia. p. 57-68.

Differential Effects of Estrogen Therapy on Thromboembolic Complications: Oral vs. Transdermal

	Oral Estrogen	Transdermal Estrogen
Odds Ratio for Venous Thromboembolic Events	3.5 (1.8-6.8)	0.9 (0.5-1.6)

95% Confidence Interval

Scarabin, P.Y., E. Oger, and G. Plu-Bureau, *Differential association of oral and transdermal oestrogen-replacement therapy with venous thromboembolism risk.* Lancet, 2003. **362**(9382): p. 428-32.

Advantages of Transdermal 17 β-Estradiol over oral CEE

- Contains the most potent and natural form of estrogen
- Achieves higher levels of estradiol in plasma than those achieved with comparable doses of CEE
- More studies employing estradiol reported positive findings than those using CEE
- May have a low incidence of venous thromboembolic complications
- Less induction of SHBG leading to increased levels of free estradiol in plasma
- Better compliance, especiallly in those with impaired cognitive function

Estrogen & Measures of Cognition in Healthy Aging Studies

COGNITIVE FUNCTION		POSITIVE/TOTAL TESTS ₸	EXPLANATION OF RESULTS AND MAGNITUDE OF EFFECT
MEMORY			
	Memory Battery	1/1	Increase of 8.0 with use
	Immediate Verbal Recall	4/9	Paragraph recall: increase of 2.2, 5.9, and 11.5 with use; Selective reminding: increase of 2.4 and 2.8 with use; Associate learning: increase of 1.7 and 14.0 with use
	Delayed Verbal Recall	3/8	Paragraph recall: change of -5.4 and 1.52 with use; Selective reminding: increase of 16.6 and 21.6 with use; Associate learning: increase of 2.6 and 19.3 with use
	Visual Memory	1/9	Fewer errors made by users in 1 study; 8 measures in 5 other studies were negative
ATTENTION			
	Working Memory	0/5	Increase of 0.2, 0.7 and 3.2 with use
	Complex Attention	2/9	Positive findings on 2 tests not repeated by other studies; 1 was of borderline significance; 4 studies found no effect on digit symbol; 2 studies found no effect on trail making
	Mental Tracking	2/14	1 of 5 studies had improvement on digit span: change of -1.67, 2.25, & 11.25 with use
	Vigilance		5 different tests used: in 1 study, visual search improved by 0.4 to 4 min. and sorting improved by 3 to 4 min. with use; other positive result was of borderline significance
CONCEPT FORMATION			
		2/3	Abstract reasoning: increase of 3.4 and 11.0 with use
MOTOR SPEED			
		2/3	Clerical speed & accuracy increase of 9.5; & reaction time improved 160-msec with use
VERBAL FUNCTIONS/LANGUAGE			
		1/4	Category fluency and retrieval: increase of 3.4 and 6.0 with use
MENTAL STATUS			
		2/5	Dementia screening examinations: increase of 0.89 and 0.90 with use

Table adapted from: LeBlanc, E.S. et al. *Hormone replacement therapy and cognition: A systematic review and meta-analysis*. **JAMA**; March 2001; **285**(11), p 1489-1499.

₸ Positive test indicates that women on estrogen scored significantly better than nonusers. Total tests refers to the number of test sessions on that cognitive measure. The same test may have been used by more than one study, and some studies may have used more than one type of test to measure that cognitive function

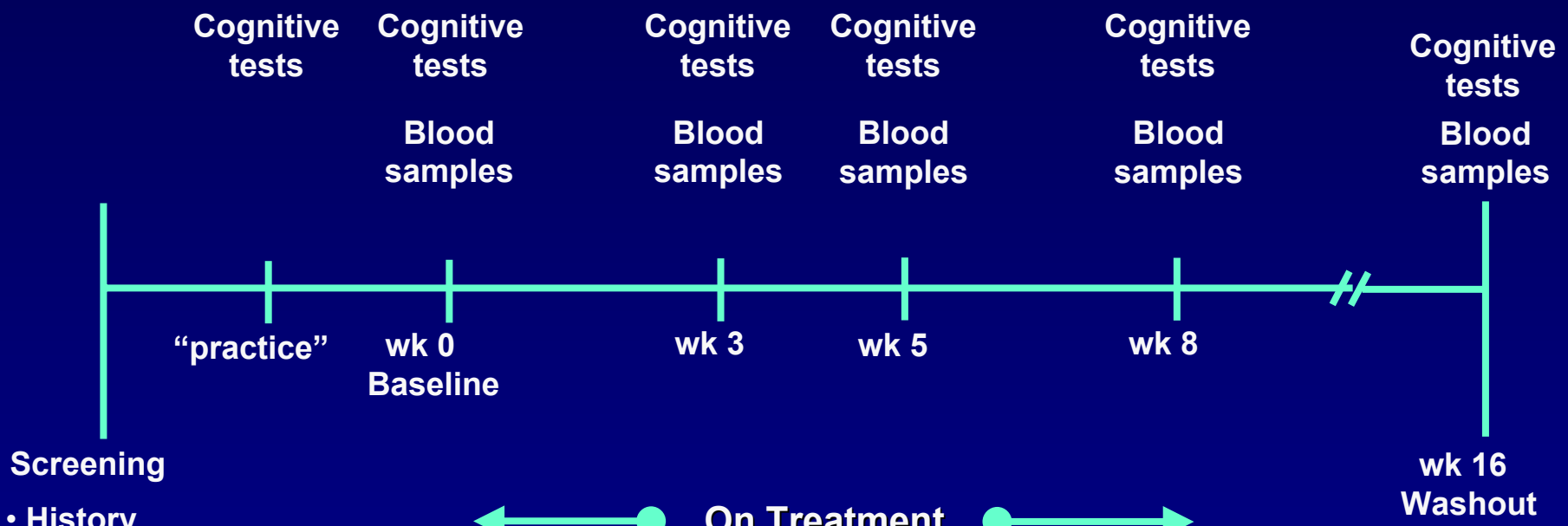
Estrogen & Measures of Cognition in Healthy Aging Studies

DOMAIN	TEST USED	PSYCHOMETRIC PROPERTIES ₸	# OF STUDIES EMPLOYING THE TEST	# OF STUDIES REPORTING POSITIVE FINDINGS
VERBAL LEARNING				
	California Verbal Learning	0.77-0.86	1	1
	Buschke Selective Reminding	0.41-0.62	5	1
	Paired Associates Learning	0.46-0.65 *	5	1
	Logical Memory/Paragraph Recall	0.55-0.67	3	2
ATTENTION/EXECUTIVE FUNCTION				
	Digit Span	0.80-0.92	7	1
	Digit Symbol	0.82	8	0
	Trail Making Test	0.60-0.90	7	2
VISUAL MEMORY				
	Figural Memory	0.19 *	1	0
	Visual Paired Associates	0.52-0.66 *	1	0
	Visual Reproduction	0.46-0.59	4	1
	Benton Visual Retention Test	0.76	3	0
VERBAL EXPRESSION				
	Boston Naming Test	0.71-0.82	4	2
	Letter Fluency	0.82	2	0
	Category Fluency	0.88	4	1
ABSTRACT REASONING				
	Similarities	0.86	2	1

Table adapted from: Rice, K. & Morse, C. *Measuring cognition in menopause research: A review of test use.***Climacteric**; March 2003; **6**(1), p 2-22.
₸ Split-half or test-retest reliability
* Estimated in populations ranging in age from 54 to 74 years old

Low Dose Study : AD Women

Study Design



- History
- Medical exam
- Labs
- Neuropsychological Assessment

- Estrogen-treated groups
  0.05 mg/day of 17β-estradiol via skin patch
  (Estraderm™, Ciba-Geigy Corporation)
- Placebo group
  inactive skin patch

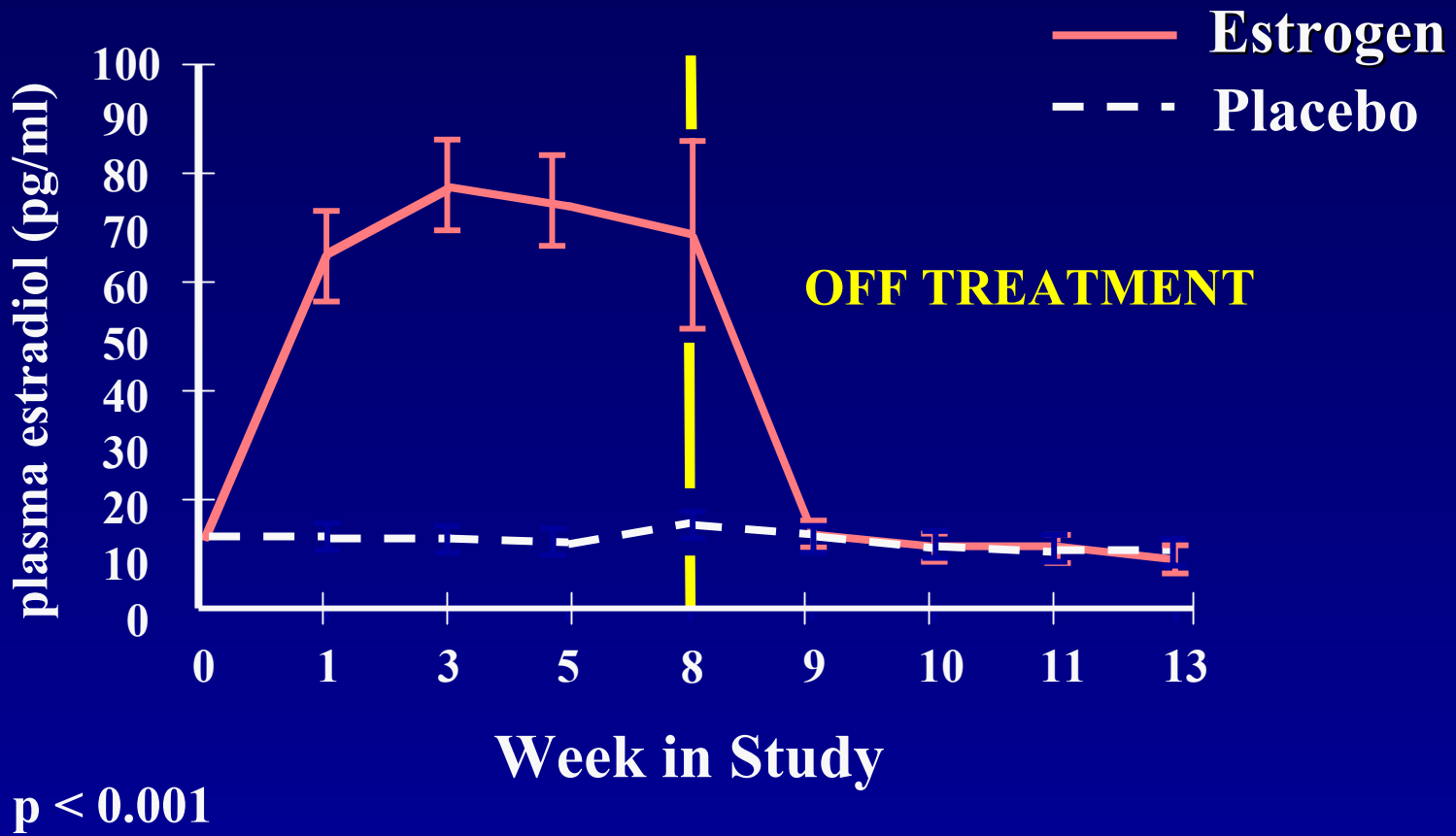
Low Dose Study : AD Women

Subject Characteristics At Study Entry

	Estrogen (n=6)			Placebo (n=6)		
	Mean	(sd)	Range	Mean	(sd)	Range
Age (yrs)	79.5	(7.9)	66-89	77.6	(6.6)	70-86
Mini-Mental State Test Score	21.4	(2.5)	19-25	20.0	(2.7)	17-23
Blessed Memory Information Concentration Test Score	21.2	(3.9)	18-30	23.6	(5.3)	18-30
Blessed Dementia Score	9.0	(3.9)	5-16	6.8	(2.6)	4-10
Duration of Symptoms (yrs)	3.3	(1.7)	2-5	3.8	(1.4)	2-5.5

Low Dose Study : AD Women

Plasma Estradiol Levels



$p < 0.001$

Cognitive Battery

- Assessed domains of cognition commonly impaired in AD:
  - Verbal and Visual memory
  - Attention
  - Language production and comprehension
  - Visuospatial skills

Stroop Color Word Interference Test

attention

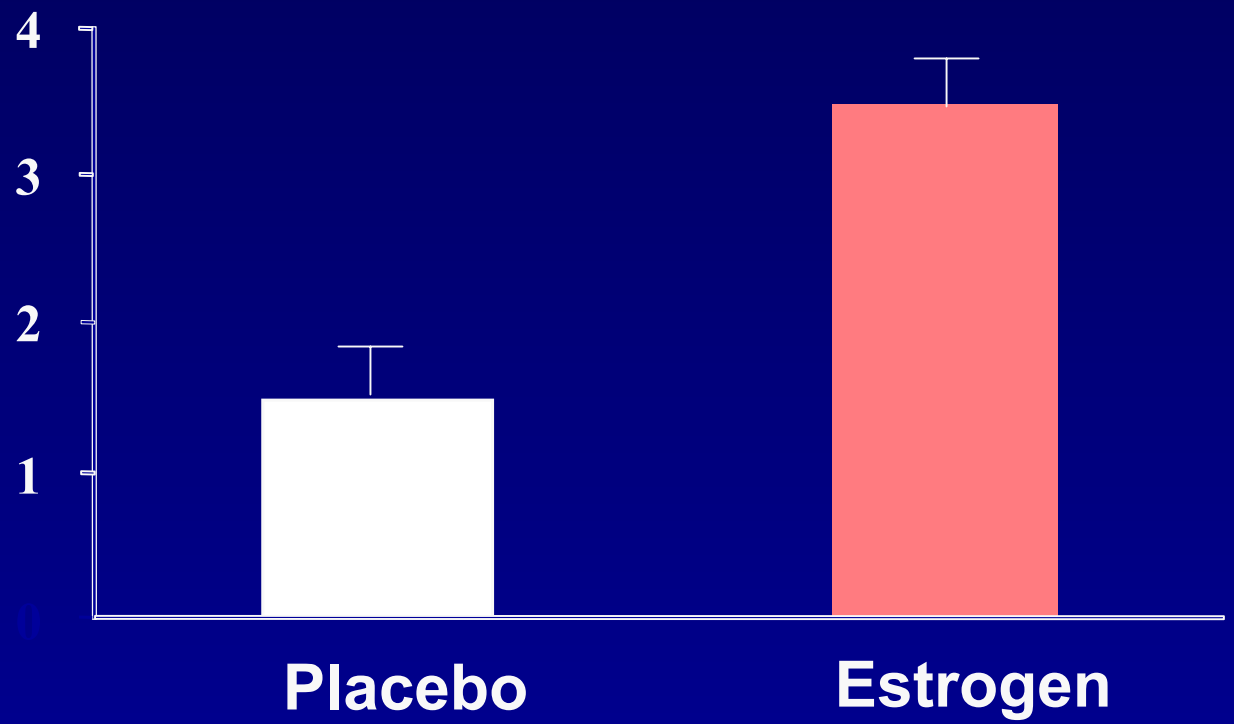
GREEN	BLUE
RED	RED
GREEN	BLUE
BLUE	GREEN
RED	GREEN
BLUE	RED

“Name the ink color”

Low Dose Study : AD Women

Stroop Color Word Interference Test

number of self-corrections



$p < 0.03$

ON Treatment

Buschke Selective Reminding Test

verbal memory

List learning task (8 trials)

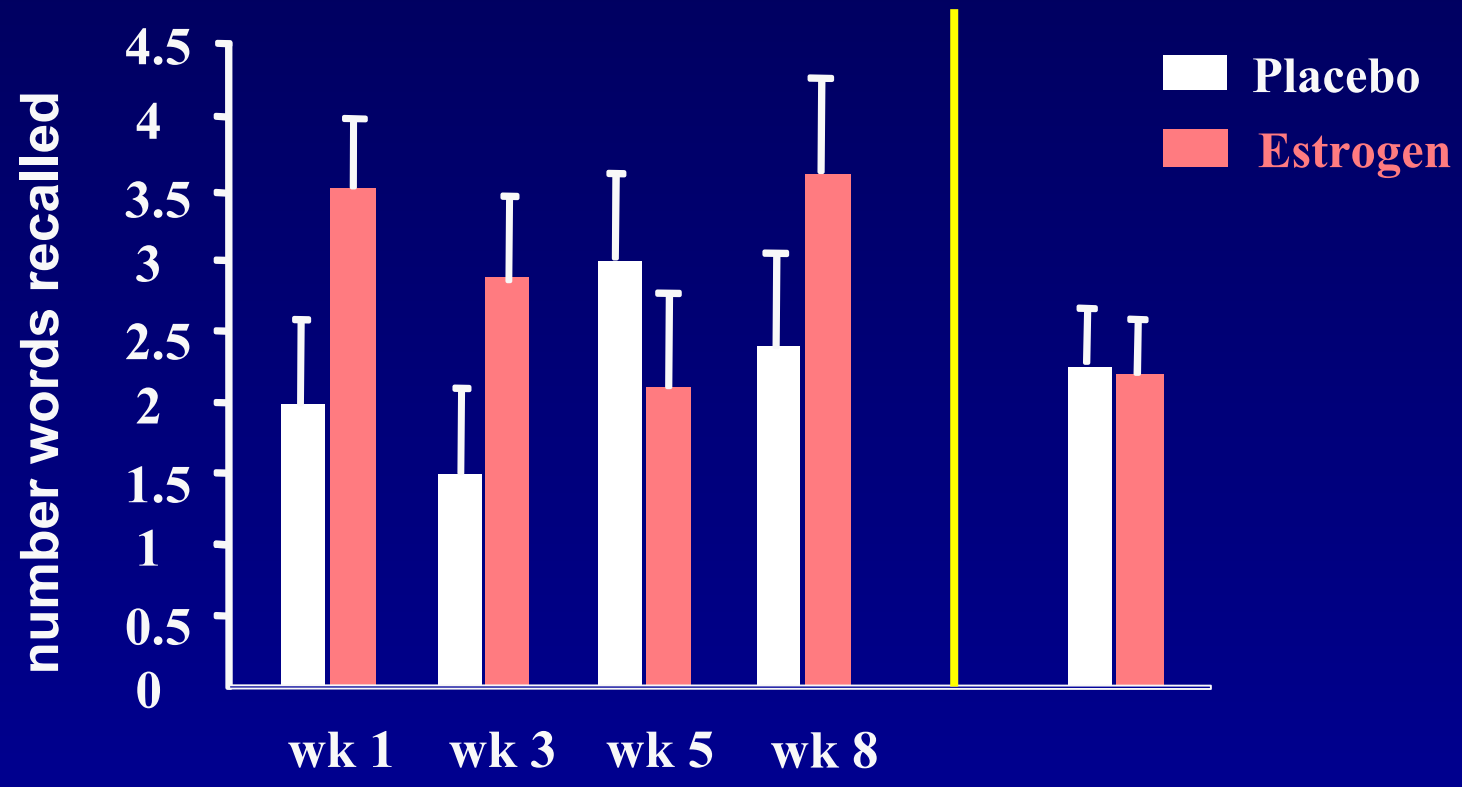
(20 minutes)

Delayed recall

Low Dose Study : AD Women

Buschke Selective Reminding Test

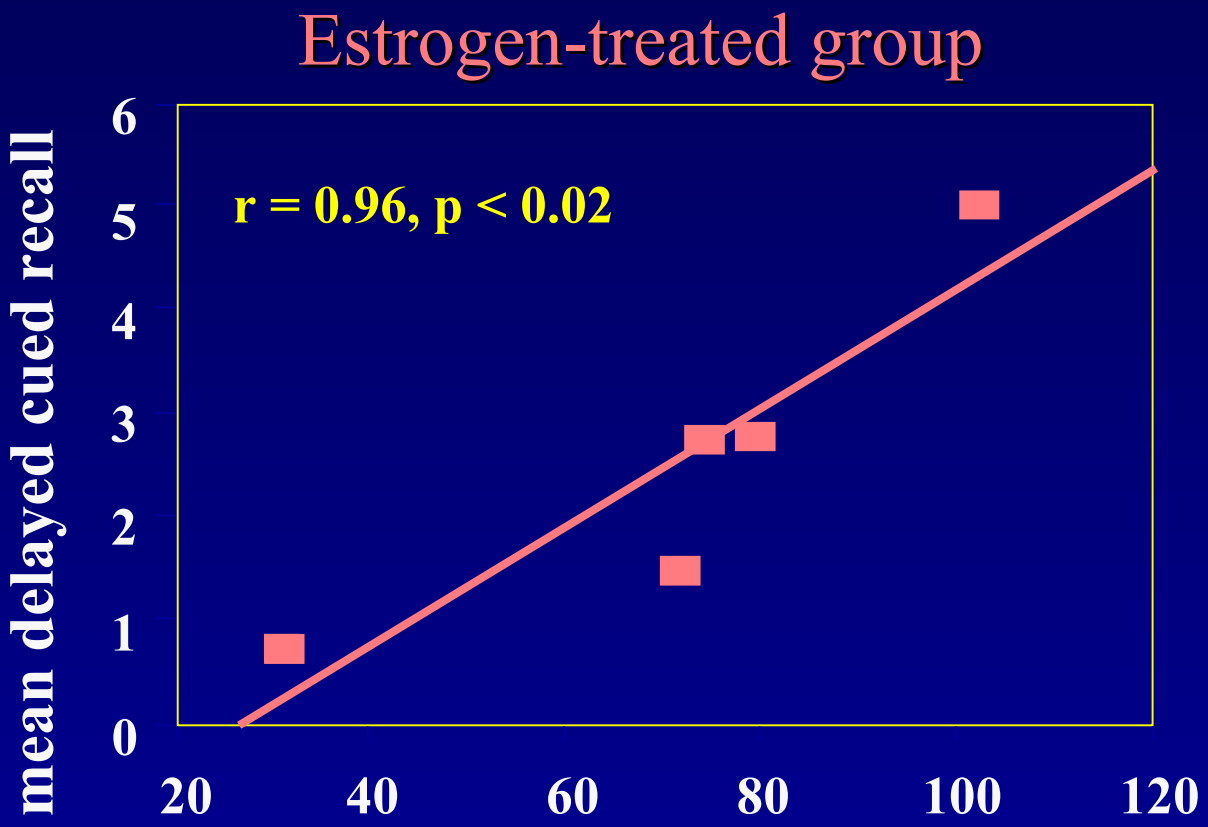
delayed cued recall



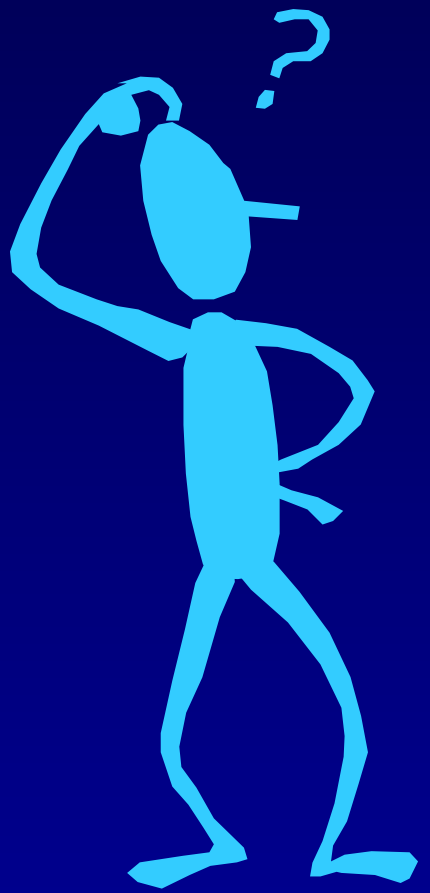
$p < 0.02$

Low Dose Study : AD Women

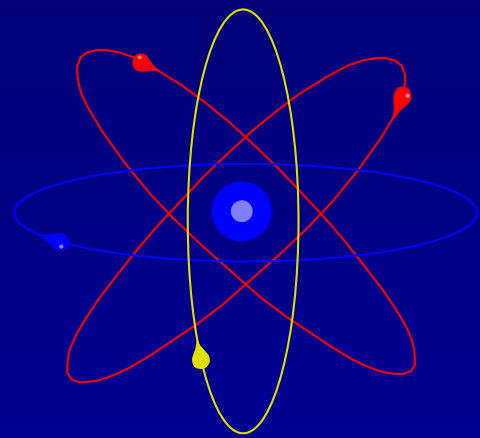
Plasma Estradiol and Delayed Cued Recall (Buschke)



Unanswered Question



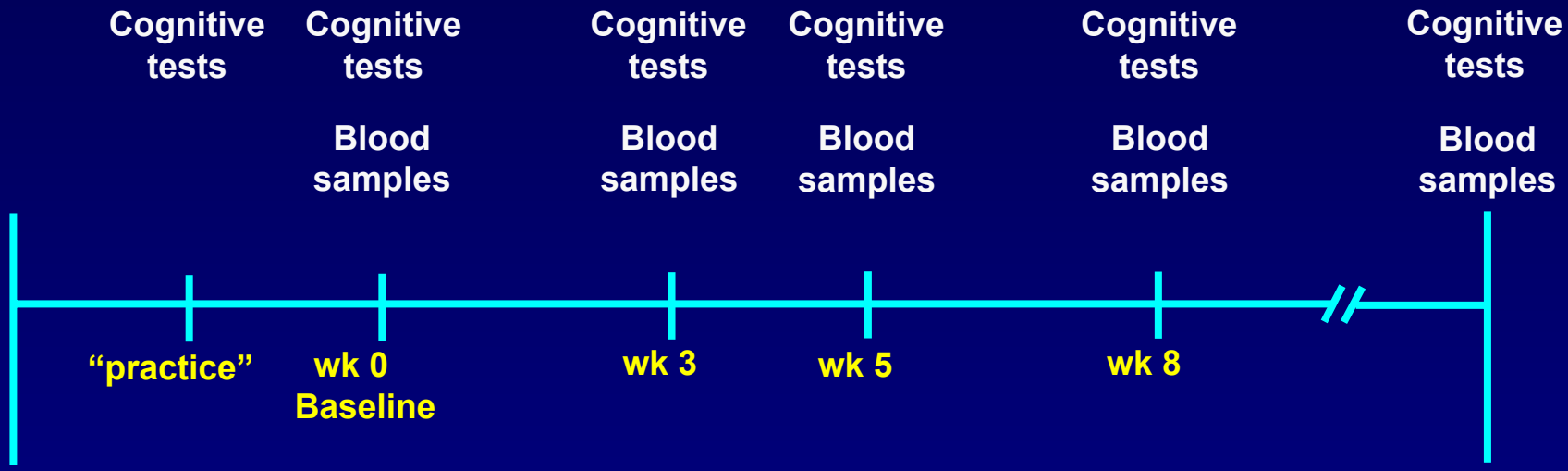
Is there a dose-dependent relationship between administration of estrogen and enhancement in cognitions?



High Dose Study : AD Women

High Dose Study : AD Women

Study Design



- History
- Medical exam
- Labs
- Neuropsychological Assessment

On Treatment

- **Estrogen-treated groups**
  0.10 mg/day of 17β-estradiol via skin patch
  (Estraderm™, Ciba-Geigy Corporation)
- **Placebo group**
  inactive skin patch

High Dose Study : AD Women

Subject Characteristics At Study Entry

	Estrogen (n=10)		Placebo (n=10)	
	Mean (sd)	Range	Mean (sd)	Range
Age (yrs)	79.0 (9.7)	61-90	80.2 (6.7)	68-90
Mini-Mental State Test Score	20.0 (6.3)	10-29	21.2 (4.2)	14-29
Blessed Memory Information Concentration Test Score	22.1 (8.7)	10-37	24.6 (5.1)	12-27
Duration of Symptoms (yrs)	3.3 (1.7)	2-5	3.8 (1.4)	2-5.5

Stroop Color Word Interference Test

attention

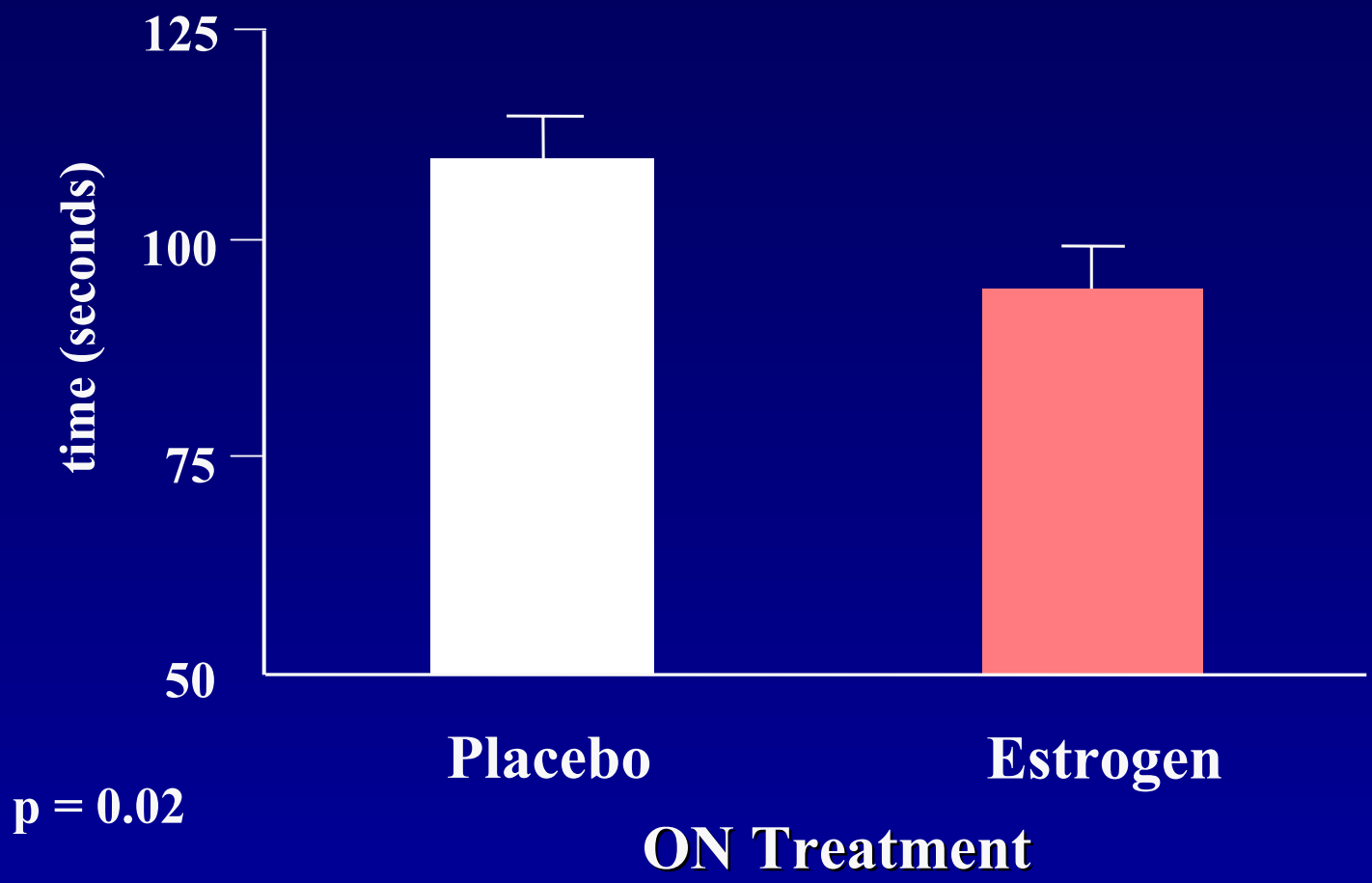
GREEN	BLUE
RED	RED
GREEN	BLUE
BLUE	GREEN
RED	GREEN
BLUE	RED

“Name the ink color”

High Dose Study : AD Women

Stroop Color Word Interference Test

time to complete the Interference condition



p = 0.02

Buschke Selective Reminding Test

verbal memory

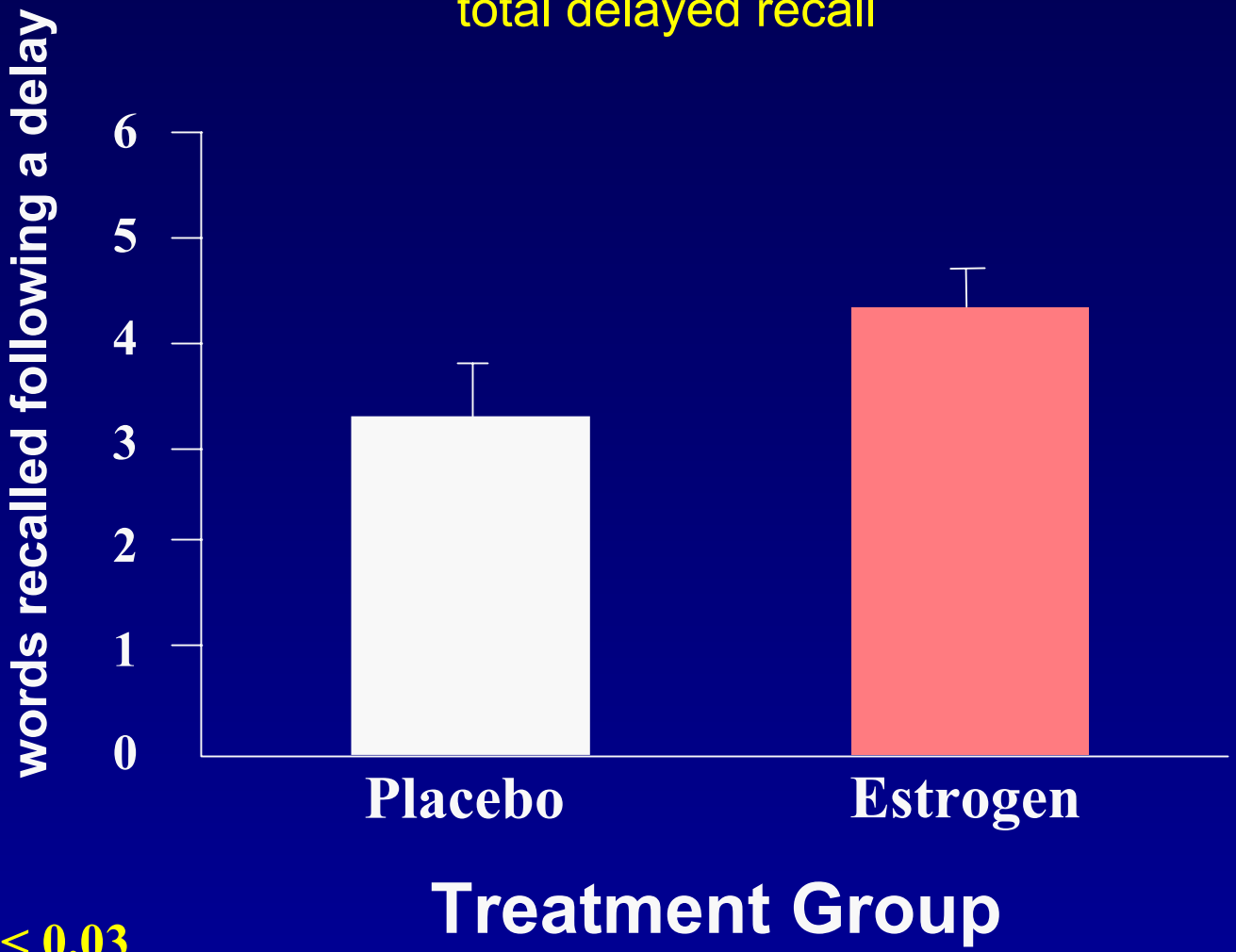
List learning task (8 trials)

(20 minutes)

Delayed recall

High Dose Study : AD Women

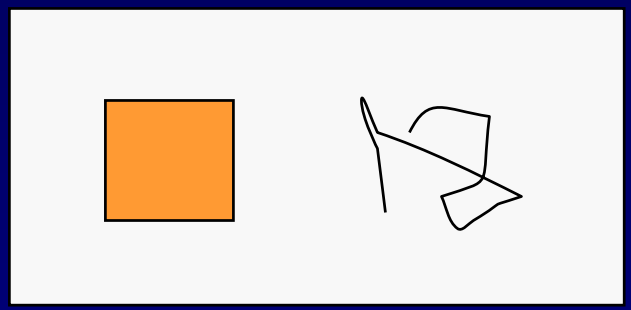
Buschke Selective Reminding Test
total delayed recall

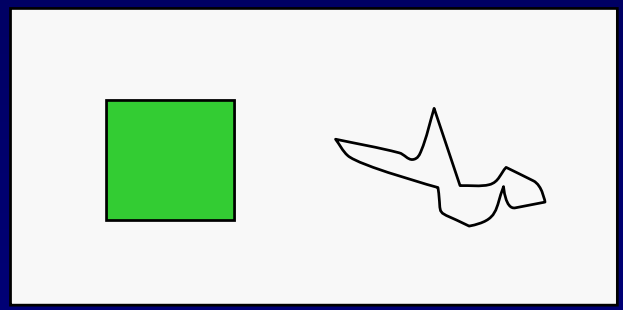


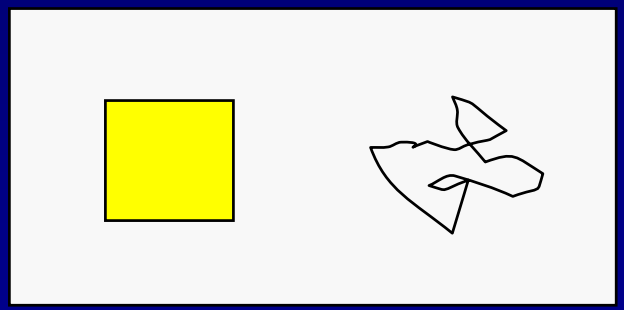
$p < 0.03$

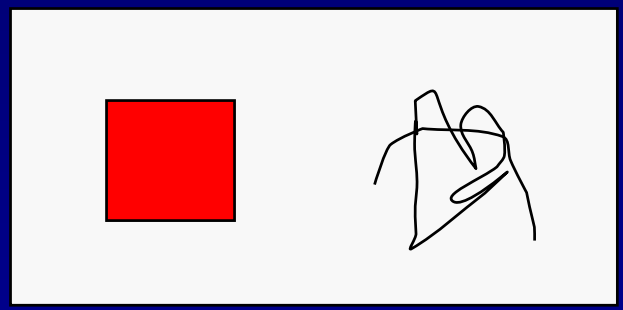
Visual Paired Associates

visual memory



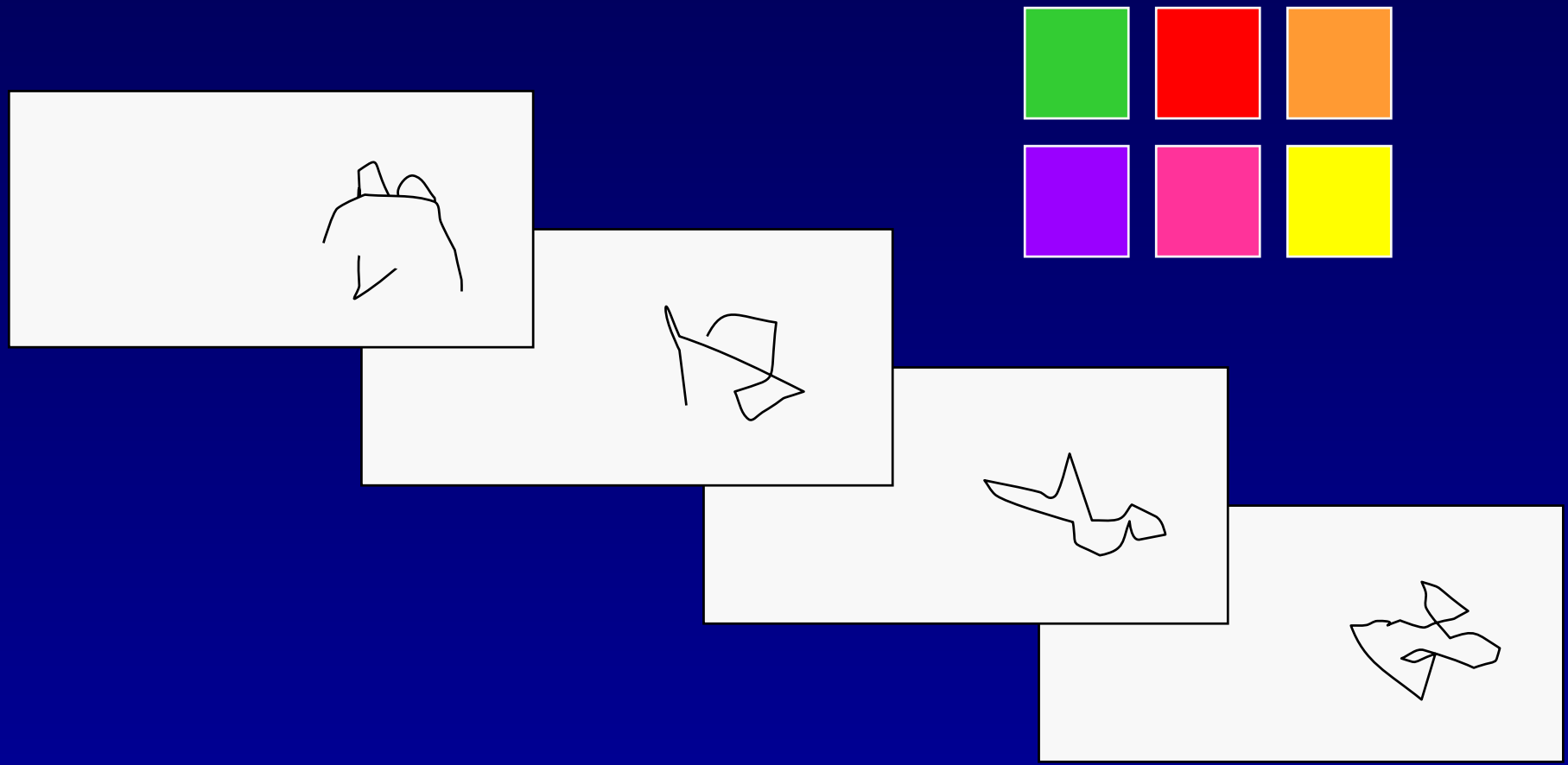






Visual Paired Associates

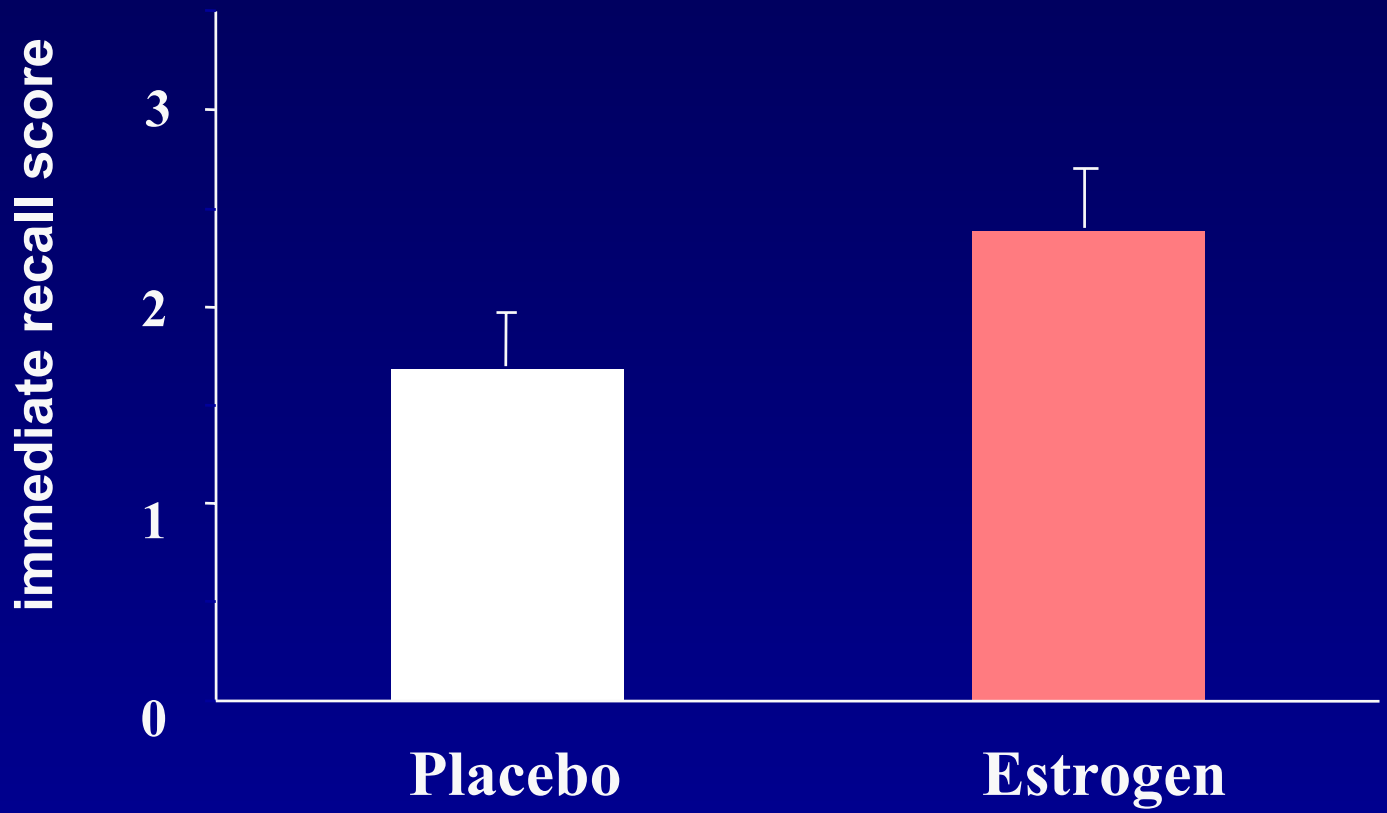
visual memory



High Dose Study : AD Women

Visual Paired Associates

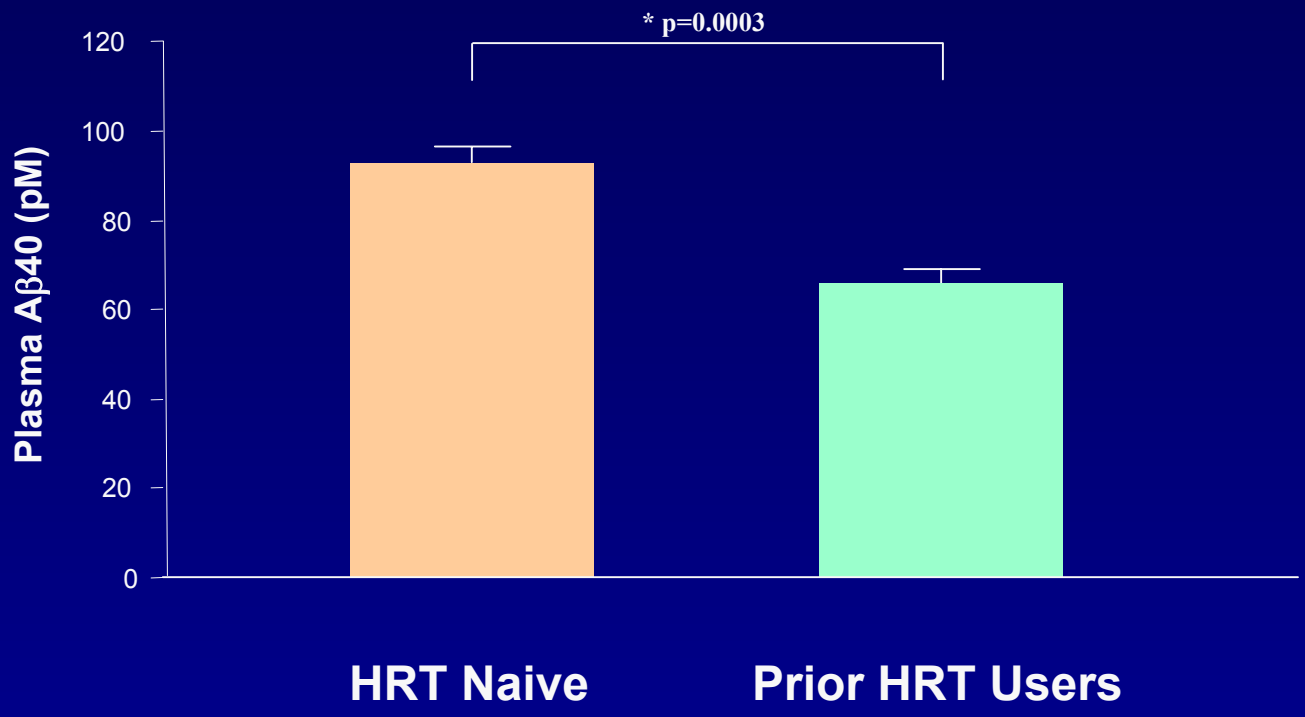
immediate recall



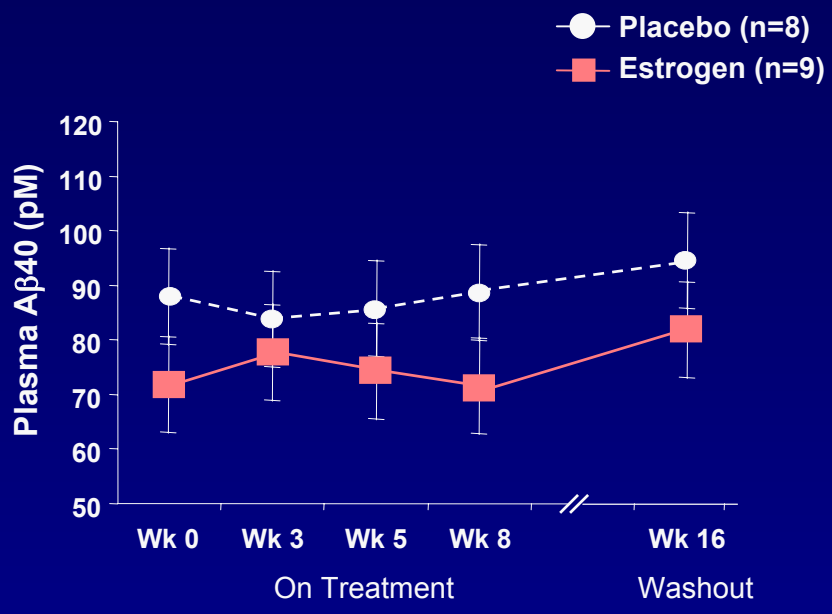
$p < 0.03$

ON Treatment

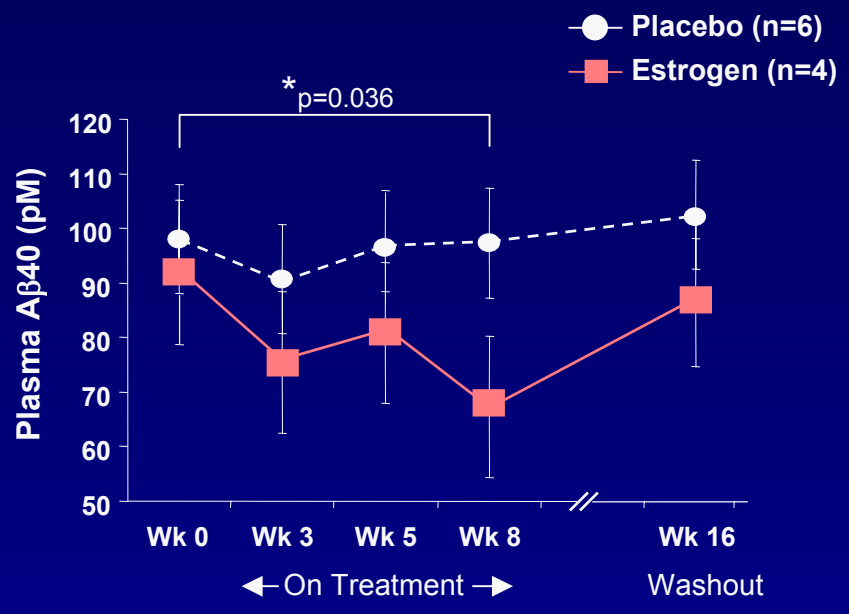
Plasma Aβ40 Levels at Baseline



Plasma Aβ40 Levels During Study Participation



All Subjects



HRT Naïve Subjects Only

Wisconsin Memory Program: Ongoing Studies in Hormone Therapy and Related Compounds

- NIA-funded study evaluating efficacy of extended therapy with transdermal estradiol in postmenopausal women with AD.
- UW GCRC-funded study of raloxifene therapy in women with AD.
- NIA-funded study of isoflavones administration in patients with AD.
- KEEPS (Kronos Early Estrogen Prevention Study) Cognitive Study evaluating differential cognition-enhancing efficacy of CEE and transdermal estradiol in healthy perimenopausal women.
- ALLADIN Study assessing the efficacy of leuprolide in older men with AD.
- Non-feminizing estrogen analogue (I.e., 17 α-estradiol) treatment studies in postmenopausal women with AD.

Conclusion

"The last chapter in the story of estrogen, cognition and dementia has not yet been written"