

## Frontiers proposal. National Institute on Aging “bench to bedside: estrogen as a case study”

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**Abstract** On 28–29 September 2004, the National Institute on Aging (NIA) convened scientists for a workshop on the aging female brain focused on translating into clinical practice discoveries concerning estrogens and progestogens. Workshop objectives were to examine effects of estrogen and progestogen on brain and cognitive function in relation to aging, to examine consistencies and apparent discrepancies

between Women’s Health Initiative Memory Study findings and other research on cognitive function, to determine whether additional hormone interventions could be developed in this area, and to offer advice on design of clinical trials for other interventions that might ameliorate cognitive aging. Following the workshop, participants joined by other interested scientists organized into regional work groups to

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continue the dialogue begun in Bethesda and to propose recommendations for NIA. The resulting recommendations, referred to as the “Frontiers Proposal for Estrogen and Cognitive Aging”, acknowledge the persistence of critical gaps in our understanding of how decline in ovarian steroid secretion during reproductive aging and use of ovarian steroid hormone therapy affect normal brain function and risk for late-life neurodegenerative disorders such as Alzheimer’s disease. There is a pressing need for preclinical, human, and integrated studies on the relationship between the menopausal transition and midlife exposures to estrogens, progestogens and related compounds, and risks for age-associated cognitive disorders. Research is also needed on better predictors of adverse cognitive outcomes, valid biomarkers for risks associated with hormone therapy use, enhanced tools for monitoring brain function and disease progression, and novel forms of therapy for improving long-term cognitive outcomes.

**Keywords** Aging · Alzheimer’s disease · Cognition · Dementia · Estrogen · Menopause · Progestogen

## Introduction

There is considerable interest in identifying modifiable risk and protective factors for cognitive decline associated with usual aging and with common dementing disorders such as Alzheimer’s disease (AD). Among these factors are estrogens and progestogens, ovarian hormones with profound effects on many tissues and organs, including the brain. These effects include the regulation of non-reproductive processes such as perception, cognition, mood, and motor control.

On 28–29 September 2004, the National Institute on Aging (NIA) of the United States (US) National Institutes of Health convened scientists from basic science, epidemiology, and clinical trials fields for a workshop held in Bethesda, MD. Cosponsors for the workshop, entitled “Bench to Bedside: Estrogen as a Case Study” were the National Institute of Mental Health, Office of Research on Women’s Health, and the Alzheimer’s Association. The focus of the workshop was on scientific data concerning the use of estrogens and progestogens in the aging female brain: what is already known and what will be

required to translate discoveries from the laboratory workbench, from epidemiology, and from clinical trials into everyday medical practice.

Defined workshop objectives were the following: (1) to examine apparent discrepancies between the findings of the Women’s Health Initiative Memory Study (WHIMS) on brain and cognitive function after estrogen and progestogen treatment, and basic research and epidemiological studies, which have often differed in terms of the formulations, doses and modes of administration, and in terms of treatment timing and duration of the hormone therapy (HT) employed; (2) to examine effects of estrogen and progestogen on brain and cognitive function in relation to aging; (3) to determine what is known and what information would be needed to establish whether additional hormone interventions could be developed in this area; and (4) to determine the lessons learned from studies on estrogen that might guide the design of clinical trials for other classes of drugs that might affect cognitive aging. The workshop agenda with abstracts and presentations is available on-line at [http://www.nia.nih.gov/ResearchInformation/ExtramuralPrograms/NeuroscienceOfAging/NNA\\_Conferences/WorkshopAgendaAbsPres.htm](http://www.nia.nih.gov/ResearchInformation/ExtramuralPrograms/NeuroscienceOfAging/NNA_Conferences/WorkshopAgendaAbsPres.htm); summary notes can be found at [http://www.nia.nih.gov/ResearchInformation/ExtramuralPrograms/NeuroscienceOfAging/NNA\\_Conferences/WorkshopSummaryNotes.htm](http://www.nia.nih.gov/ResearchInformation/ExtramuralPrograms/NeuroscienceOfAging/NNA_Conferences/WorkshopSummaryNotes.htm).

Following the workshop, participants joined by other interested scientists organized into regional work groups to continue the dialogue begun in Bethesda and to propose recommendations for NIA. One venue for ongoing discussion was the biennial Graylyn Conference on Women’s Cognitive Health, co-sponsored by the NIA and hosted by the Wake Forest University School of Medicine. Meetings in October 2005 and October 2007 focused on translational research and included many participants from the Bethesda Bench to Bedside workshop.

The regional work group recommendations are described below as the Frontiers Proposal for Estrogen and Cognitive Aging, implying that there remain frontiers for discoveries concerning estrogen and other sex steroids in relation to the aging brain, cognition, and cognitive disorders. The following sections provide background information, consensus recommendations by work group members, a concluding perspective, and an executive summary.

## Background

### Basic science studies

Central nervous system consequences of menopausal estrogen and progesterone loss is an area of intense study, because many actions of these sex steroids are potentially relevant to cognitive aging and dementia. Both *in vitro* and *in vivo* analyses indicate that estrogen protects against a wide spectrum of neurodegenerative insults, a substantial number of which are linked to AD. A key finding relevant to the development of AD is the ability of estrogen to reduce formation of  $\beta$ -amyloid (Petanceska et al. 2000; Yue et al. 2005), a hallmark biochemical marker of this illness. Moreover, estrogen promotes both morphological and electrophysiological correlates of learning and memory *in vitro* and *in vivo* (Brinton et al. 1997; Hao et al. 2006; Murphy and Segal 1996; Toran-Allerand et al. 1980; Woolley and McEwen 1992). Myriad neurotransmitter systems relevant to cognition (acetylcholine, serotonin, noradrenalin, dopamine, glutamate) are regulated by estrogen. Studies in adult and aged non-human primates have revealed both behavioral and neurobiological effects of estrogen, some of which counter the effects of normal aging on cognition (Hao et al. 2006; Rapp et al. 2003a, b; Tinkler et al. 2004; Voytko 2002). Overall, basic science analyses using both *in vitro* and *in vivo* model systems indicated that estrogen—typically  $17\beta$ -estradiol but also conjugated equine estrogens—protect neurons against insults associated with AD (Brinton 2005). Moreover, these same estrogens in the same model systems can activate biochemical, genomic, cellular and behavioral mechanisms of memory (Frye et al. 2007; Singh et al. 1994; Toran-Allerand 2000). An important aspect of these studies, and of virtually all of the basic science *in vitro* and *in vivo* analyses, is that neurons were healthy prior to estrogen exposure and prior to exposure to neurodegenerative insults or lesions (Brinton 2005). Typically, basic science analyses have used a preventive model, *i.e.*, neurons are pretreated with an estrogen or progestogen prior to exposure to the neurological insult. Studies on aged rodents have reinforced this concept, *i.e.*, that healthy neurons respond differently than compromised neurons, in that hippocampal synapses are less responsive to estrogen in aged rats (Adams et al. 2001), and the behavioral impact of estrogen is less impressive in aged rats than in young animals

(Daniel et al. 2006; Markowska and Savonenko 2002; Savonenko and Markowska 2003). These data have led to the notion that there may be correlates with age or with duration of time after menopause that establish a “window of opportunity” for estrogen to be effective in protecting circuits mediating mood and cognition, with decreased responsiveness once this time frame passes (Gibbs 2000; Resnick and Henderson 2002). Indeed, increasing basic science evidence indicates the importance of the hormonal state, the duration of estrogen or progesterone privation, and the impact of aging on the neural response to estrogen (Brinton 2005; Gibbs 2000; Johnson and Sohrabji 2005).

The issue of progestogen action in the brain is less well explored, but the existing data indicate that the type of progestogen is critical to functional outcome in neurons. In some model systems, progestogens such as medroxyprogesterone acetate (MPA) are associated with outcomes very different from those associated with progesterone (Nilsen and Brinton 2003). At the same time, there is increasing evidence that progesterone has neuroprotective effects that may be very useful clinically (Stein 2007).

### Women’s Health Initiative Memory Study

The Women’s Health Initiative, the parent study for WHIMS (Shumaker et al. 2003, 2004), was a multicenter, randomized, double-blind, placebo-controlled trial designed to assess effects of a commonly prescribed oral HT on cardiovascular disease, breast cancer, and other health outcomes (Anderson et al. 2003; Rossouw et al. 2002). Active treatment was with combined HT for women with a uterus [conjugated equine estrogens (CEE) 0.625 mg/day with MPA 2.5 mg/day as a continuous combined preparation; CEE+MPA trial] or with estrogen alone (CEE 0.625 mg/day; CEE-alone trial) for women without a uterus. WHIMS assessed the incidence of dementia and other cognitive outcomes among community-dwelling women who at baseline were between the ages of 65 and 79 years inclusive. After a mean follow-up of 4–5 years, 61 women in the CEE+MPA trial developed dementia, as did 47 women in the CEE-alone trial. About one-half of cases were attributed to AD, but separate analyses were not reported for specific dementia subtypes. In both trials, dementia incidence was increased for women assigned to active treatment compared to placebo [CEE+MPA hazard ratio (95% confidence interval) 2.05 (1.21–

3.48); CEE-alone, 1.49 (0.83–2.66); Shumaker et al. 2003, 2004]. Mild cognitive impairment (MCI), thought to indicate increased risk of dementia, was a secondary WHIMS outcome; an insignificant increase in MCI risk was attributed to HT in the CEE+MPA trial [hazard ratio 1.07 (0.74–1.55)] and in the CEE-alone trial [hazard ratio 1.34 (0.95–1.89); Shumaker et al. 2003, 2004]. WHIMS findings on global cognition are considered below.

#### Alzheimer's disease prevention: observational studies prior to WHIMS

WHIMS results on dementia were unanticipated based on the basic science research described above, which provided a strong rationale why estrogen might be neuroprotective, and based on observational studies that had considered the association between HT use and AD risk. Most observational studies have shown a protective association when ever-users of HT were compared to women who had never used HT, with meta-analyses suggesting overall risk reductions of about one-third (Hogervorst et al. 2000; Yaffe et al. 1998). In the Cache County prospective cohort, the protective association of HT was modified by timing, and was seen with past, but not current, HT use (Zandi et al. 2002); in the Multi-Institutional Research in Alzheimer Genetic Epidemiology case-control study, the protective association of HT was modified by age and was seen among younger, but not older, postmenopausal women (Henderson et al. 2005).

#### Cognitive aging: clinical studies prior to WHIMS

Complaints of poor memory are common around the time of the menopausal transition (Mitchell and Woods 2001), but forgetfulness is common at other ages as well (Neugarten and Kraines 1965) and memory symptoms may be attributed to a variety of cognitive and noncognitive factors (Mitchell and Woods 2001). After ovarian estrogen production is halted abruptly by oophorectomy or by use of a gonadotropin-releasing hormone analog, there is clinical trial evidence that short-term use of estrogen has a beneficial effect on cognitive performance, at least for memory for verbally encoded information (Phillips and Sherwin 1992; Sherwin and Tulandi 1996); whether other younger women may similarly benefit from HT is less apparent (Henderson and Sherwin 2007). Importantly, long-term cognitive effects of HT used in middle age are

unknown. Available evidence suggests that natural menopause per se does not have an important effect on verbal memory or most other cognitive skills (Fuh et al. 2006; Henderson et al. 2003; Kok et al. 2006), at least in the short run, and serum estrogen concentration at midlife is unrelated to cognitive performance (Henderson et al. 2003; Herlitz et al. 2007). The relationship between endogenous estradiol levels and dementia risk is also controversial (Geerlings et al. 2003; Schupf et al. 2006). However, one follow-up study of women who had participated in randomized clinical trials of HT during middle age found that those randomized to active treatment had a lower risk of cognitive impairment 5–15 years later than those randomized to placebo (Bagger et al. 2005).

Among older postmenopausal women, observational studies on HT and cognition provide conflicting evidence (Carlson et al. 2001; Kang et al. 2004), but randomized clinical trials are more consistent in showing no cognitive benefit of HT in this older age group (Grady et al. 2002; Viscoli et al. 2005; Yaffe et al. 2006; reviewed by Henderson and Sherwin 2007). These clinical trial results are similar to those reported from WHIMS, where over a mean follow-up of about 5 years participants were assessed annually with a test of global cognition (Espeland et al. 2004; Rapp et al. 2003a, b). In HT and placebo groups, global cognitive scores tended to increase over time, presumably representing practice effects. The magnitude of increase was somewhat less among CEE-MPA and CEE-alone users than placebo-users, but mean between-group differences were small and not clinically important (Espeland et al. 2004; Rapp et al. 2003a, b). In a subgroup of WHIMS participants assessed more extensively (the Women's Health Initiative Study of Cognitive Aging), combined estrogen-progestin HT had contradictory effects, impairing verbal memory but showing a trend toward improving nonverbal memory (Resnick et al. 2006). Other comparisons in this analysis were not significant, and mean magnitudes of any between-group differences were small.

#### Discordant findings between observational studies and WHIMS

Differences between results of observational studies and WHIMS are most evident with respect to dementia outcomes. Reasons for apparent discrepancies might include (1) differences in outcomes (AD vs

all-cause dementia), (2) differences in HT formulations, doses and routes of administration (although estrogen exposure in most observational studies was primarily oral CEE and progestogen exposure was primarily MPA), (3) differences in timing of HT exposures (often used at a younger age near the time of menopause in observational studies vs used only at an older age remote from the time of menopause in WHIMS), and (4) systematic bias in observational studies (Henderson 2006). Concerning the second possibility, it can be noted that women in the WHIMS trial with a uterus received HT as a continuous combined CEE+MPA preparation, putatively a less physiological approach than sequentially administered hormones and a formulation that was not yet available at the time most observational results were conducted. The third and fourth possibilities may be most crucial: are observational studies fatally flawed by biases such as recall bias or the healthy user bias (Hemminki et al. 1993; Matthews et al. 1996), or do WHIMS findings of enhanced dementia risk—using a specific HT formulation in older postmenopausal women—fail to generalize to younger women more likely to use HT for moderate to severe vasomotor symptoms?

At least two clinical trials will consider differential effects of early and late HT, the Kronos Early Estrogen Prevention Study (KEEPS) (Harman et al. 2005), and the Early versus Late Intervention Trial with Estrogen (ELITE) (<http://www.clinicaltrials.gov/ct/show/NCT00114517>). In both, cognition will be assessed as secondary outcomes, although the trials are relatively small and further studies will almost certainly be required. Results will be very important in guiding future research initiatives.

### Frontiers proposal for estrogen and cognitive aging: research agenda and work group recommendations

#### Human studies

- 1) There is evidence of disturbances in cognition and affect during the menopausal transition. Disturbances include cognitive complaints, affective symptoms (e.g., depression, anxiety), and impaired cognitive performance. How many women are affected and to what extent?

What are the predictors (including “substrate” differences such as personal historical variables and genotype) of those women who are vulnerable to such disturbances as compared to those who are resistant?

- 2) What is the nature of subjective cognitive complaints during the menopausal transition? Are they associated with objective performance deficits, and what is their course? Are they non-specific epiphenomena or subsyndromal / prodromal symptoms of a developing disorder? How do subjective complaints of cognitive impairment compare to objective performance measures? Are current tests of cognitive performance sufficiently sensitive to measure accurately impairments implied by cognitive symptoms? What instruments (including neuroimaging techniques) might be employed to evaluate disturbances in cognition or affect during the menopausal transition? What neuropsychological measures or other clinical measures can serve as valid, sensitive and reliable markers for effects of hormone therapy? Additionally, the potential efficacy of estrogen to alter the subjective complaints of cognitive impairments has not been adequately studied.
- 3) Are the changes in cognition and affect noted during the menopausal transition and early postmenopause clinically relevant? In addition to direct measures of symptom severity (e.g., checklist or a test of cognitive performance), can an operational definition of “clinically meaningful” cognitive complaints be developed that incorporates measures of disease-related morbidity, functional impairment in work or interpersonal relationships, quality of life, and burden on the health care system?
- 4) What are the best measures and experimental designs to be employed to evaluate the following:
  - (a) Long-term outcomes associated with disturbances in cognition and affect that may occur during midlife in women, and
  - (b) the potential linkage of these disturbances in affect and cognition to the menopausal transition?
- 5) What are the relevant functional groupings of women—including education level, chronologi-

cal age, reproductive status (e.g., menopausal transition vs early postmenopause vs late postmenopause), reproductive history (e.g., previous use of oral contraceptives, previous history of premenstrual dysphoric disorder), medical/psychiatric co-morbidity (e.g., vasomotor menopausal symptoms, comorbid vascular disease, metabolic indices, HPA-axis responsiveness, levels of stress, depression, or specific symptoms such as disturbed sleep), and brain health status (e.g., neurologically healthy vs diagnosed degenerative disease)—that could be employed to evaluate the effects on cognition and affect of either age-related declines in reproductive function or hormone therapy? Stated another way, how might one best characterize HT responders as compared to non-responders?

- 6) Neurodegenerative changes of AD are observed to occur years before symptoms of dementia first appear, and perhaps even years before premonitory symptoms of MCI. Both AD and MCI are uncommon at midlife, but midlife hormonal changes have the potential to influence long-term cognitive outcomes. Is there a time prior to or during the disease course when hormone intervention is beneficial? Recent findings from WHIMS indicate that initiation of oral CEE therapy in older postmenopausal women increases risk of dementia. Can hormone therapy initiated during the perimenopause, especially for a limited period (e.g., 6 months to 5 years), reduce long-term risk of MCI or AD? Can functional subgroups of women be identified of those likely to benefit from early, short-term hormone use (e.g., women with early menopause, severe hot flashes, affective symptoms, few vascular risk factors, defined genotypes)?
- 7) There is a paucity of available information related to the best clinical and surrogate markers of progression of MCI or AD, and those representing neuroprotective actions of estrogen. Studies are necessary to identify appropriate biomarkers (e.g., plasma and cerebrospinal fluid assays for  $\beta$ -amyloid or tau, measures of oxidative stress, markers of inflammatory cascade) and surrogate endpoints (e.g., structural and functional neuroimaging) of estrogen actions in patients with MCI or AD.
- 8) For younger women (i.e., women less than 65 years of age), the effect of HT in influencing the

conversion from MCI to AD is unknown. Clinical studies are needed to evaluate the potential of HT in reducing the risk of conversion. For older women, the therapeutic potential of estrogen formulations other than oral CEE (e.g., transdermal estradiol) on MCI conversion or on dementia symptoms remains to be characterized adequately.

- 9) During midlife, the potential interactions between changing levels of estradiol and progesterone, as well as changes in ovarian/adrenal androgens, and HPA-axis function should be evaluated clinically and built into animal models.
- 10) Existing datasets should be analyzed and new observational analyses should be conducted to determine the impact of oral contraceptive use and AD risk stratified by age at initiation and duration of use. Analyze existing data based on hormone formulation used. What is the impact of use on later development of AD? Are there pharmacological agents better or worse than estradiol and progesterone? Analyze WHIMS data to determine the mean age of women who converted to AD on HT vs placebo: were hormone users younger or older than placebo users?

#### Preclinical studies

- 1) What is the importance of recapitulating the physiology of the menopausal transition with respect to the molecules and the timing of hormone administration in the animal models? What aspects of the physiology of the menopausal transition in humans should be incorporated in animal models of reproductive senescence to evaluate the impact of the menopausal transition and sex hormone therapy on brain systems relevant to the regulation of cognition, the stress response, and affect? Considerations would include identifying physiologically relevant molecules, pharmacologically relevant molecules (e.g., selective estrogen receptor modulators,  $17\alpha$ -estradiol, bioidentical hormones), contextual variables that influence response (age, brain health status, developmental stage, or reproductive age), and genetic contributions to differential behavioral phenotypes. Additionally, the individual animal models of reproductive aging or hypogonadism need to be better characterized to address the effects of brain aging on cognitive test

performance, the differences in both reproductive and brain aging across species (e.g., mice vs rat vs monkey) or among animal strains, and the ability to evaluate the effects of systemic disease (e.g., cardiovascular disease) on cognitive function in these models.

- 2) How are the animal data consistent with and inconsistent with clinical observations? Are the inconsistencies due to true differences in how different species age and respond to steroids, or do they reflect flaws and limitations inherent to the animal models as designed? For example, if the species used in the model does not undergo menopause similar to humans (e.g., rat) or investigation across the menopausal transition is impractical for many experiments (e.g., monkeys) ovariectomy might be the best approach, but what is the relevance to human menopausal transition? Are there animal models that better recapitulate human menopause than surgical ovariectomy? Nonetheless, there are basic processes that seem to be similar across species even if one cannot study the menopausal transition in rats and mice in a way that informs about the human. For example, the formation/maturation of synapses in the hippocampus and cortex happens in mice, rats and monkeys. There are species differences, with mice—compared to rat and rhesus monkey—having more estrogen-stimulated maturation of spine synapses than *de novo* synapse formation in hippocampus. Yet many of the underlying mechanisms are likely to be identical, and the presence of genomic and non-genomic estrogen receptor is comparable. Are there neural actions of ovarian hormones that can be effectively studied across basic science discovery models from cell culture and *in vivo* animal models to generate sufficient understanding that will translate to the human condition?
- 3) How does endocrine status interact with the neurobiology of aging? In what ways does an aged brain react to estrogen and other gonadal steroids differently than a young brain? Does a brain with continuous exposure to sex steroids react differently than one that has been in an estrogen-depleted environment for an extended period of time? Does a brain with long-term cyclic exposure to estrogen therapy (with or without progestogen) react differently than one with long-term continuous exposure?
- 4) How do the cellular effects of estrogen, progesterone, stress hormones, and related molecules translate into alterations in the function (e.g., cholinergic system, cell signaling pathways involved in cellular resilience/neuroplasticity, vascular supply, microglia and peripheral immune cells, blood-brain barrier, neuroprotection vs cognitive enhancement) of circuits subserving cognition and affect (e.g., hippocampus, amygdala, prefrontal cortex)? Which cellular effects are most directly linked to cognitive enhancement?
- 5) Basic science analyses should be undertaken in transgenic mouse models of AD, including the use of formulation of hormone therapies used in clinical trials, with an emphasis on understanding AD pathogenesis.
- 6) In non-human primates, research should characterize potential differences in behavioral and neuroprotective effects of various estrogenic compounds, routes of delivery (oral or parenteral), and treatment regimens for both unopposed and opposed estrogenic compounds.

#### General questions for preclinical and clinical studies

- 1) How does one define and recognize the distinction between vascular and neural events, and what is the impact of estrogen and progestogen on each? What is the interaction between vascular disease and AD pathophysiology? How does HT increase vascular risk in older women or animal models? Animal models should be created for competing risks of AD and vascular dementia. Potential mechanisms underlying increased risk of dementia observed with oral conjugated estrogen therapy in WHIMS need to be characterized systematically.
- 2) How can we re-create the beneficial effects of estrogen documented in preclinical studies and minimize the risks identified in human studies? Variables to be examined include both the formulations of estrogen and progestogens (progesterone versus MPA) employed in human studies and clinical trials, dosage and levels of exposure, timing of HT exposure, duration of HT, as well as substrate differences (i.e., neuronal health status and presence of degenerative disease). The neuro-

protective potential and cognition or mood enhancing efficacy of alternatives to HT, including neuroactive selective estrogen receptor modulators and phytoestrogens, are largely unknown in animal models, healthy women, and women with cognitive and affective disorders.

## Conclusions

There is considerable evidence from both pre-clinical and human studies that ovarian steroids have widespread neuroregulatory potential throughout the nervous system. These hormones mediate effects within the brain relevant to the pathophysiology, course, and treatment response characteristics of several brain disorders. However, critical gaps exist in our knowledge of both the effects on brain function of declining ovarian steroid secretion during reproductive aging, and the role of ovarian steroid HT in the prevention or treatment of brain diseases. Ovarian steroids exert a wide range of physiologic effects within the body. Their signaling pathways are extremely complex; involve many different organ systems, and interact with a variety of other hormonal systems. Thus the manifold nature of their regulatory capacity is accompanied by a wide range of potential actions—some beneficial under certain conditions while detrimental under others. Indeed, one observation emerging from the literature is that the effects of ovarian steroids on brain function are not uniform, and several potential sources of variability have been identified that may explain these otherwise discordant results. First, both the chronological and reproductive age of the individual, as well as their health status, influence the observed response to administered ovarian steroids. Thus, the timing of administering HT is an important determinant of both the health benefits and risks to the individual. Second, the type of steroid preparation could mediate a range of diverse effects within the same organ system, resulting in benefits accrued with one hormonal preparation and either no benefits or adverse health events caused by another preparation. Finally, the schedules of administration of estrogen and progestogen (i.e., cyclic or continuous combined) are important to consider with respect to effects on brain. As a caveat, the work group membership was concerned about two trends in prior studies that have examined the role of ovarian steroids

(both endogenous and exogenous forms) in human and animal brain function: (1) the majority of clinical studies were not designed to measure brain function specifically as the primary endpoint, and (2) basic science studies were focused narrowly on healthy models and did not incorporate the spectrum of aging and disease found in the human condition.

Future efforts should focus on developing the following: (1) improved understanding of the variables that predict those women in whom the menopause transition adversely alters the course of specific brain diseases; (2) biomarkers for those women who will benefit from the introduction of ovarian HT and those for whom HT is contraindicated; (3) novel therapeutics, such as selective ovarian steroid (i.e., estrogen, progesterone, and androgen) modulators that enhance the potential therapeutic benefits of these steroids in the brain but have more acceptable long-term safety profiles; and (4) clinically relevant assessment tools of both brain function and disease progression.

## Bench to Bedside: estrogen as a case study executive summary of a national working group

The following is a Frontiers Proposal for the development of research initiatives to investigate effects of ovarian steroid hormones on cognitive aging and brain function. This proposal for the NIA of the US National Institutes of Health has four goals:

- (1) To identify unanswered questions regarding the effects of reproductive aging on brain function under both physiologic and pathologic conditions, as well as the potential role of reproductive HT in modulating these effects;
- (2) To promote the integration of both basic and clinical research in women's health;
- (3) To provide impetus for the development of new methods for the evaluation of processes related to brain aging in health and disease; and
- (4) To provide directions for *brain-specific* investigations that can serve as the basis for NIA-sponsored requests for applications.

It is recommended that the NIA develop an integrated program to coordinate NIA-sponsored basic and clinical research studies in the areas described above. Such a program would provide a firm scientific foundation for the endocrine aspects of neural aging and increase the

knowledge available to women before they enter menopausal transition, particularly with respect to confusing discrepancies between observational and clinical trial data on HT and cognition. This investigator-based initiative identified several issues that require the attention of clinicians and scientists who work in this arena:

#### (1) Human studies

- Some women report disturbances in cognition and affect before menopause, during the menopausal transition and early postmenopause. The numbers of women affected, the course of these disturbances, their clinical relevance, and the factors that predict susceptibility and resistance to these changes are not known. Additional research investigating the effects of endocrine events on measures of cognition and affect in younger women during reproductive life will provide important information for comparison purposes.
- Both subjective and performance-based measures of cognitive complaints should be studied in relation to the menopausal transition.
- Biomarkers and surrogate endpoints should be identified that are related to disease pathologies responsible for cognitive dysfunction (e.g., mild cognitive impairment or dementia), and their relationship defined with respect to the menopause and hormone therapy.
- Factors that predict either positive or negative health outcomes during hormone therapy should be identified (e.g., chronological age, reproductive status, medical/psychiatric co-morbidity, genotypes, and cognitive status). Can we recreate the beneficial effects of estrogen demonstrated in preclinical studies and minimize the risks identified in some human clinical trials?
- The possible relationship between the time at which hormone therapy is initiated and the subsequent development of cognitive complaints, mild cognitive impairment, and dementia should be clarified.

#### (2) Preclinical studies

- Animal (non-human primate, rodent) models should be developed that recapitulate the relevant physiology of the human menopausal transition with particular attention to the

dysregulation of gonadal hormone cyclicity and ovarian function.

- Transgenic mouse models of AD should be employed to investigate the relationship between HTs used in clinical trials and the development of AD.
- The impact of individual molecules and complex formulations on outcomes of relevance to cognition and neurodegenerative condition should be determined. Molecules and formulations should include physiologically relevant molecules and pharmacologically relevant molecules [e.g., selective estrogen-receptor modulators (SERMs)].
- Contextual variables that influence the response to HT (e.g., brain aging), and genetic contributions to differential behavioral phenotypes should be studied. The interactions between brain aging, reproductive history, and the form of HT that may be most beneficial to brain physiology and behavior need to be clarified.
- The specific neural systems that are both regulated by sex steroids and subserved cognition and affect should be investigated. These analyses should be conducted in collaboration or consultation with researchers investigating the impact of HT on human cognition, psychiatric function, and surrogate markers (neuroimaging).
- Non-human primate models should be refined and used to identify potential differences in behavioral and neuroprotective effects of various estrogenic compounds, routes of delivery (oral or parenteral), and treatment regimens for both unopposed and opposed estrogenic compounds.

Finally, it is recommended that NIA provide financial support for a future workshop to develop these questions into specific investigations that would warrant new requests for applications or could be integrated into the existing portfolio of ongoing NIA-sponsored studies.

## Appendix

This report was prepared by the National and Regional Chairs, with input from the membership of regional Work Groups.

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