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Factors influencing the cognitive and neural effects of hormone treatment during aging in a rodent model

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Abstract

Whether hormone treatment alters brain structure or has beneficial effects on cognition during aging has recently become a topic of debate. Although previous research has indicated that hormone treatment benefits memory in menopausal women, several newer studies have shown no effect or detrimental effects. These inconsistencies emphasize the need to evaluate the role of hormones in protecting against age-related cognitive decline in an animal model. Importantly, many studies investigating the effects of estrogen and progesterone on cognition and related brain regions have used young adult animals, which respond differently than aged animals. However, when only the studies that have examined the effects of hormone treatment in an aging model are reviewed, there are still varied behavioral and neural outcomes. This article reviews some of the important factors that can influence the behavioral and neural outcomes of hormone treatment including the type of estrogen administered, whether or not estrogen is combined with progesterone and if so, the type of progesterone used, as well as the route, mode, and length of treatment. How these factors influence cognitive outcomes highlights the importance of study design and avoiding generalizations from a small number of studies.

Keywords

estradiol; progesterone; cognition; prefrontal cortex; memory; aging; menopause; medroxyprogesterone; estrogen; hippocampus

Aging in human females is accompanied by a cessation of the menstrual cycle, known as menopause, which usually occurs between 45–55 years of age and results from a depletion of ovarian follicles. The depletion in follicles leads to increased follicle-stimulating hormone and luteinizing hormone levels and a dramatic decrease in estrogen and progesterone levels.

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Because of the decrease in ovarian hormones, menopause is associated with several symptoms including vaginal dryness, bone loss, and hot flashes. In addition, lower levels of naturally occurring estradiol (E₂) correlated with poorer performance on a verbal task in middle-aged females (Wolf and Kirschbaum, 2002), indicating that the decline in ovarian hormones may also play a part in the cognitive decline observed during aging.

Hormone therapies, including Premarin (conjugated equine estrogens; CEE) and Prempro (CEE in combination with medroxyprogesterone acetate; MPA), have been approved to alleviate the symptoms of menopause. Moreover, studies have found beneficial effects of estradiol treatment on several cognitive tasks including measures of verbal and working memory (Joffe et al., 2006; Krug et al., 2006; LeBlanc et al., 2001). However, results from the Women's Health Initiative (WHI) indicate that CEE alone or CEE administered with MPA results in an increased risk of stroke and dementia (Anderson et al., 2004; Anderson et al., 2004; Shumaker et al., 2004; Wassertheil-Smoller et al., 2003). Because of this, whether hormone treatment has beneficial effects on cognition during aging has recently become a topic of debate.

This review aims to discuss the factors that are known to influence the cognitive outcome of hormone treatment and will focus on the rodent literature. What is known about how these factors influence neural mechanisms important for cognition will also be reviewed. As discussed in the sections below, the length of hormone deprivation and the age of the subjects tested are known to alter behavioral outcomes, and therefore this review will focus on studies that have treated middle-aged or aged females.

Length of Hormone Deprivation

There is evidence that the length of hormone deprivation influences the outcome of hormone treatment and may partially explain the negative findings of the WHI studies (Daniel and Bohacek, 2010; Gibbs, 2000; Sherwin, 2009). This research has indicated that there is a "window of opportunity", which means that waiting too long after menopause, or estropause in rats, to begin hormone treatment could lead to the treatment having no effect or negative effects on cognition. In WHIMS, the memory study of the WHI, the ages of the subjects ranged from 65–79 years which may have been too late to initiate hormone treatment in order to have beneficial effects on cognition. Animal studies have provided support for the "window of opportunity" with the length of hormone deprivation affecting both behavioral and neural outcomes. In a study by Gibbs (2000), animals treated with E₂ or E₂ in combination with progesterone immediately following ovariectomy or within three months of ovariectomy performed significantly better than ovariectomized controls on a delayed matching to sample task. This difference in performance did not occur when hormone treatment was initiated 10 months after ovariectomy (Gibbs, 2000). Furthermore, females ovariectomized at 12 months of age and immediately treated with E₂ performed better than controls on the radial arm maze when tested after 5 months of treatment (Daniel et al., 2006). In contrast, females that were ovariectomized at 12 months of age but not treated with E₂ until 17 months of age did not perform better than ovariectomized controls (Daniel et al., 2006).

Ovarian hormones affect neuroanatomy in cognitive brain regions including the hippocampus and prefrontal cortex, and not surprisingly the factors that influence behavioral outcome also influence neuroanatomy. Recent work found that E₂ treatment within 15 months of ovariectomy increased dendritic spine density in the hippocampus whereas treatment initiated after 19 months failed to alter this measure (McLaughlin et al., 2008; Smith et al., 2010). Importantly, E₂ treatment immediately following ovariectomy of 21 month old females also enhanced long term potentiation in the hippocampus indicating that

the lack of effect after 19 months of ovariectomy was a result of the length of hormone deprivation rather than aging (Smith et al., 2010). The issue of hormone deprivation has been discussed more thoroughly in several recent reviews (Daniel and Bohacek, 2010; Rocca et al., 2011). Given the evidence for the “window of opportunity”, only studies that initiated hormone treatment close to the loss of naturally circulating hormones were included in the discussion below of factors influencing outcomes of hormone treatments.

Age: Using an Appropriate Model

There is an extensive body of literature that has examined the neural and cognitive effects of hormone treatments in young ovariectomized animals (Daniel, 2006). However, studies suggest that the effects of hormone treatment in young female animals are often not the same as the effects of hormone treatment during aging. Our laboratory found that young adult females who were ovariectomized and given E₂ and progesterone were impaired in the acquisition of the Morris water maze (Chesler and Juraska, 2000), while treatment with E₂ and progesterone in ovariectomized middle-aged animals facilitated performance of the same task (Markham et al., 2002). Similarly, Foster et al. (2003) found that young ovariectomized females perform worse on the Morris water maze when treated with estradiol benzoate, whereas aged ovariectomized females receiving estradiol benzoate perform better. Other laboratories have found beneficial treatment effects in young or middle-aged animals, but no effects of hormone treatment in older animals. Although treatment with estradiol enhanced performance of ovariectomized young and middle-aged animals on the Morris water maze, estradiol treatment did not improve performance of ovariectomized aged animals (Talboom et al., 2008). Similarly, a single E₂ injection enhanced novel object recognition in 6 month old animals but not at 22 months of age (Gresack et al., 2007).

These age related differences in the effects of hormone treatments on cognition are not surprising given that the percentage of ER α immunoreactive synapses (Adams et al., 2002) and the number of ER β mRNA positive cells (Yamaguchi-Shima and Yuri, 2007) decrease in the hippocampus during aging. Studies have also found age-related differences in neural outcomes after hormone treatment. For example, E₂ increased synapse number in the hippocampus of young females, whereas treatment during aging failed to result in an increase in synapses (Adams et al., 2001). However in this study, there was an increase in NR1, a subunit of the NMDA receptor, in the hippocampus of aged females that was not observed in young animals (Adams et al., 2001). A more recent study found that E₂ in combination with progesterone increased synaptophysin in hippocampal CA1 of young rats but not middle-aged or aged animals (Williams et al., 2011). It seems that the aging hippocampus may be less responsive to ovarian hormones, which might explain some of the differences in behavioral outcomes observed between adult and aged rats after hormone treatment. Therefore, it is important to assess the effects of hormone treatment in middle-aged or aged female animals, a model that more closely mimics hormone treatment during human aging.

Effects of Estrogen on Cognition and Neuroanatomy During Aging

Like humans, middle-aged rats experience a cessation of the cycle, referred to as estropause, between 9–12 months of age; however, in contrast to human females, they continue to secrete low levels of estrogen and progesterone (Dudley, 1982; Markham and Juraska, 2002). To more closely model the hormone loss in humans, studies that use middle-aged or aged animals most often ovariectomized the animals and then initiate hormone treatment. The studies that have administered estrogen treatments have resulted in conflicting results including cognitive benefits, no effects, or impairment. Long-term chronic E₂ initiated at 13

months of age benefited acquisition of a delayed matching to position task even when animals were tested at approximately 22 months of age (Gibbs, 2000). Silastic capsules containing E₂ improved object recognition and performance on a spontaneous alternation task in aged mice (Miller et al., 1999; Vaucher et al., 2002), and infusions of E₂ into the hippocampus enhanced memory consolidation on an object recognition task in middle-aged female rats (Fan et al., 2010). In addition, chronic oral administration of E₂ improved water maze performance and novel object recognition in middle-aged rodents (Fernandez and Frick, 2004; Lowry et al., 2010). However, chronic E₂ administered both orally and via daily injections did not affect the number of working memory errors in the radial arm maze (Fernandez and Frick, 2004; Gresack and Frick, 2006), and although chronic treatment with E₂ minipellets decreased the number of reference memory errors in the radial arm maze, there was no effect on working memory errors (Heikkinen et al., 2004). Also, two studies have found that long-term chronic E₂ failed to alter performance on delayed alternation task in middle-aged or aged females (Chisholm and Juraska, 2012b; Gibbs, 2000). Studies using the operant versions of the delayed spatial alternation task consistently find that E₂ administered chronically via silastic capsules during aging impairs performance (Neese et al., 2010; Wang et al., 2009). Impaired performance was also observed on a water radial arm maze after 5 weeks of oral E₂ treatment in 18 month old mice (Fernandez and Frick, 2004).

One possible explanation for the inconsistent outcomes of estrogen treatment is the type of memory assessed. In aging females, there is somewhat more agreement among studies evaluating the effects of estrogens on reference memory rather than working memory. Several studies have found that E₂ or estradiol benzoate treatment in both middle-aged and aged female rodents improves performance on the Morris water maze, a spatial reference memory task (Foster et al., 2003; Frick et al., 2002; Frye et al., 2005; Harburger et al., 2007; Lowry et al., 2010; Markham et al., 2002; Talboom et al., 2008). Even when the type of memory tested is accounted for, the outcome of hormone treatment cannot be generalized because several other methodological factors discussed in this review can influence the behavioral outcome.

There is an extensive body of research documenting the effects of estrogen in the young adult hippocampus, and thus most studies in aging animals have investigated the effects of estrogen only treatments on the hippocampus. Decreased amounts of synaptic ER α immunoreactivity were observed in the CA1 of aged female rats (Adams et al., 2002), and estrogen treatment during aging failed to increase synapse density in this same brain region (Adams et al., 2001). In addition, E₂ did not alter the amount of synaptophysin and opioid peptides in the CA1 and dentate gyrus of aged animals (Williams et al., 2011). However, chronic E₂ treatment via silastic capsules increased brain derived neurotrophic factor (BDNF) levels in the hippocampus of middle-aged females (Kiss et al., 2012), and ten days of E₂ treatment increased choline acetyltransferase levels in the hippocampus, but not the prefrontal cortex, of middle-aged female rats (Bohacek et al., 2008). Despite the importance of the prefrontal cortex for several cognitive tasks, less is known about the effects of estrogen treatments during aging on this brain region in rodents. There have been two studies that have examined the prefrontal cortex in aged animals, and both have found that estrogen influences neuroanatomical measures involved with cognition. Aged female rats receiving long-term E₂ treatment had a greater amount of tyrosine hydroxylase and a marginally greater number of synapses in the medial prefrontal cortex (mPFC) than control animals (Chisholm et al., 2012; Chisholm and Juraska, 2012a), suggesting that the mPFC remains responsive to estrogens during aging. Although research on the prefrontal cortex is limited in rodents, the effects of estrogens on the non-human primate prefrontal cortex are well documented. Much of the non-human primate literature indicates that estrogen alters dendritic spines in the prefrontal cortex and improves performance on tasks mediated by this brain region in both young and aged females (reviewed in Bailey et al., 2011).

This section has highlighted some of the cognitive and neural outcomes of estrogen only treatments during aging. Importantly, there are many factors that may affect behavioral and neural outcomes of hormone treatment during aging including, the type of estrogen administered, whether or not estrogen is combined with progesterone and the type of progesterone, the route and mode of administration, and the length of treatment.

Factors Influencing the Outcome of Hormone Treatment

Type of Estrogen

Most animal studies that have examined the effects of estrogen on cognition have administered E_2 , which is the most potent naturally occurring estrogen. However, the conjugated equine estrogens (CEE) found in Premarin and Prempro, the treatments used in the Women's Health Initiative, consist of at least 10 different estrogens. Estrone and equilin are the primary biologically active hormones after CEE has been metabolized, and estrone can then be converted to E_2 (Mayer et al., 2008; Sitruk-Ware, 2002). These estrogens differ in their binding affinities for the estrogen receptor (Kuhl, 2005; Sitruk-Ware, 2002), and therefore may not result in similar behavioral outcomes. Recently, one study found that chronic cyclic injections of CEE prevented overnight forgetting on the Morris water maze in middle-aged female rats (Acosta et al., 2009). This result is in agreement with several other studies that have found improved performance on the Morris water maze after treatment with E_2 (Bimonte-Nelson et al., 2006; Markham et al., 2002; Talboom et al., 2008). Also, a single injection of CEE enhanced performance on an object recognition task (Walf and Frye, 2008). Factors that can influence the outcome of E_2 treatment, such as dose and route of administration, may also influence the behavioral outcome of CEE treatment. A recent study found that ovariectomized middle-aged females treated with high or medium doses of CEE made fewer errors than control animals following a six hour delay on a delayed matching to sample task (Engler-Chiurazzi et al., 2011). However in the same study, animals receiving a low dose of CEE made more working memory errors than controls on a delayed matching to sample task (Engler-Chiurazzi et al., 2011). Importantly, CEE is a mixture of estrogens and understanding how each estrogen affects cognition may provide insight into optimizing hormone treatment. The Bimonte-Nelson lab has recently examined some of the individual estrogens that make up CEE. The CEE component, delta 8,9 dehydroestrone, improved performance of middle-aged females on a delayed matching to sample task and the water maze, whereas equilin did not (Talboom et al., 2010). Administration of a high dose of estrone via osmotic pumps to middle-aged females impaired acquisition and retention of a delayed matching to sample task, but did not alter performance on the water maze (Engler-Chiurazzi et al., 2012). Testing the cognitive effects of estrone is further complicated by the fact that it is readily converted to E_2 (Kuhl, 2005). Medium and high doses of CEE increased E_2 levels, whereas the animals treated with the low dose of CEE had E_2 levels similar to controls (Acosta et al., 2009; Engler-Chiurazzi et al., 2011). Therefore, differences in behavioral outcomes between CEE components may be related to their ability to increase serum E_2 levels.

The type of estrogen administered has also been shown to affect neural outcomes in middle-aged animals. E_2 administered chronically via osmotic pumps resulted in a greater number of choline acetyltransferase immunoreactive neurons in the basal forebrain, whereas estrone did not increase this measure at any of the doses administered (Engler-Chiurazzi et al., 2012). Approximately four weeks of treatment with delta 8,9 dehydroestrone decreased cholinergic nicotinic receptors in the hippocampus of middle-aged females, while equilin did not alter receptor expression in any brain region (Talboom et al., 2010). Although not compared within the same study, chronic E_2 treatment increased the levels of BDNF protein in the entorhinal cortex of aged ovariectomized rats (Bimonte-Nelson et al., 2004), and CEE treatment increased BDNF levels in the cingulate cortex but not the entorhinal cortex.

(Engler-Chiurazzi et al., 2011). Understanding how these individual estrogens alter neuroanatomy could help explain the differences observed on cognitive tasks after treatment with the different components of CEE.

Addition of Progestagens

In women with a uterus, estrogen is administered with a progestagen to prevent endometrial hyperplasia (Whitehead et al., 1979). Animal studies have traditionally examined the behavioral effects of estrogen in combination with natural progesterone. Two studies have found that treatment with E₂ and progesterone resulted in similar improvements when compared to controls as E₂ only treatments. E₂ and E₂ in combination with progesterone prevented the overnight forgetting observed in ovariectomized control animals in the water maze (Markham et al., 2002). In addition, weekly injections of E₂ and progesterone for approximately 10 months resulted in a similar improvement as E₂ only treatment on the delayed alternation t-maze as compared to controls (Gibbs, 2000). However, other studies have found that the addition of natural progesterone can interfere with the beneficial effects of E₂. For example, progesterone administered through continuous release pellets reversed the beneficial effects of E₂ on the water maze so that middle-aged animals receiving both E₂ and progesterone did not differ from control animals (Bimonte-Nelson et al., 2006). Similarly, Lowry et al. (2010) found that long-term treatment with chronic E₂ improved performance on the fourth day of water maze testing as compared to controls, whereas animals receiving E₂ in combination with progesterone did not perform significantly different from controls (Fig. 1A).

Research comparing the neural effects of estrogen only treatments with those of estrogen in combination with natural progesterone has also produced conflicting results. Initial studies examining the neuroprotective properties of these two hormone regimens in cultured hippocampal neurons found similar outcomes with both resulting in neuroprotection (Nilsen and Brinton, 2002; Nilsen and Brinton, 2003). However, the importance of age has been previously discussed in this review, and studies that have compared these regimens in middle-aged or aged animals have resulted in divergent effects. Long-term treatment with E₂ in middle-aged females reduced neuron loss in the hippocampus after kainite lesions and the addition of progesterone prevented this neuroprotection (Carroll et al., 2008). In addition, chronic E₂ treatment via subcutaneous pellets increased BDNF and nerve growth factor levels in entorhinal cortex, whereas E₂ in combination with progesterone resulted in neurotrophin levels comparable to control animals (Bimonte-Nelson et al., 2004).

Even though studies show that in some instances natural progesterone opposes the beneficial effects of estrogen only treatments, some argue that natural progesterone may be more beneficial than MPA, the most commonly prescribed progestin. After the Women's Health Initiative found that hormone treatment in postmenopausal women failed to enhance cognition and increased the number of subjects diagnosed with either probable dementia or mild cognitive impairment (Espeland et al., 2004; Rapp et al., 2003b; Shumaker et al., 2003; Shumaker et al., 2004), researchers suggested that the presence of MPA in these hormone treatments might be responsible. MPA is a synthetic analogue of progesterone and binds to progesterone receptors, as well as androgen and glucocorticoid receptors (Bamberger et al., 1999; Bardin et al., 1983). Importantly, progesterone is readily metabolized to allopregnanolone (Majewska et al., 1986) and MPA inhibits the enzymes needed for this conversion (Jarrell, 1984; Lee et al., 1999; Penning et al., 1985). Differences in mechanisms of action between MPA and progesterone may result in divergent behavioral responses. Indeed, studies have found differences in the neuroprotective properties of these two progestagens (Jodhka et al., 2009; Nilsen and Brinton, 2003). However, the relationship between neuroprotective properties after extreme insult and changes during normal aging needs to be examined. The studies that have compared the effects of these two progestagens

on cognitive behavior during aging have resulted in varying outcomes. For example, MPA administered without estrogen impaired performance on the water radial arm maze and the spatial water maze (Braden et al., 2010). This study found that unlike MPA, natural progesterone did not impair performance on the water maze, but both MPA and progesterone impaired performance on the water radial arm maze (Braden et al., 2010). However, our laboratory has shown that long-term treatment with E₂ in combination with MPA attenuates the beneficial effect of E₂ on water maze performance in middle-aged rats, while E₂ with progesterone resulted in performance that was not different than E₂ alone (Lowry et al., 2010). Further complicating the role of progestagens, a more recent study found that long-term treatment with E₂ and MPA benefited acquisition of a delayed alternation t-maze, and E₂ in combination with progesterone resulted in a similar trend (Chisholm and Juraska, 2012b) (Fig 1B).

Similar to cognitive outcomes, the type of progestagen administered can alter the neural outcome. Most of the work comparing the neural effects of progesterone and MPA has examined the neuroprotective properties of these two progestagens. These studies typically find that progesterone results in neuroprotection whereas MPA antagonizes the neuroprotective properties of E₂ (Jodhka et al., 2009; Nilsen and Brinton, 2003).

More recently, studies have investigated the effects of these two progestagens after long-term exposure, and although they sometimes result in divergent neural outcomes, natural progesterone does not seem to be more beneficial than MPA. Braden et al. (2010) found that chronic MPA significantly and progesterone marginally decreased levels of glutamic acid decarboxylase in the hippocampus when administered via osmotic pumps. The only studies that looked at the long-term effects of these progestagens in combination with E₂ on neural structure found that E₂ in combination with MPA during aging significantly increased synaptophysin labeled boutons and tyrosine hydroxylase fibers in the mPFC whereas E₂ in combination with progesterone did not significantly increase these measures as compared to control females (Chisholm et al., 2012; Chisholm and Juraska, 2012a) (Fig 2A and B). It is important to note that studying the effects of progestagens with the addition of an estrogen may not result in the same outcomes as progestagens alone. Although some women receive estrogen only treatment, progestagens are rarely prescribed alone as treatment for menopausal symptoms. Therefore when investigating the effects of these progestagens as a model of postmenopausal aging, they should be administered with an estrogen.

Route of Administration

Although oral administration is the most common route of administration of hormone treatment in post menopausal women, animal studies more commonly use injections or silastic capsules to deliver hormone treatments. These routes of administration have different pharmacokinetics than oral administration and do not allow for first pass metabolism of the hormone which may result in different outcomes of hormone treatment. For example, oral administration of E₂ improved performance on object recognition (Fernandez and Frick, 2004); while another study from the same lab found that daily injections of E₂ did not improve performance on the same task (Gresack and Frick, 2006). Routes of administration can differ in the level and pattern of hormones achieved. Typically capsules containing E₂ produce relatively constant levels of estrogen in the physiological range; whereas injections of E₂ produce a spike in hormone levels that returns to baseline within 24–48 hours. Oral administration, through the drinking water, results in levels that increase during the dark cycle and return to near baseline during the light cycle due to circadian drinking patterns (Gordon et al., 1986). In addition, these routes of administration differ in the level of stress that an animal experiences from hormone administration. Stress has been shown to interact with hormones and alter behavioral outcomes (Rubinow et al., 2004). Therefore, although few studies directly compare the behavioral outcomes of

different routes of administration, it is an important variable that may clarify some of the inconsistencies in the literature.

Cyclic or Intermittent Versus Chronic Treatment

Studies have found that the length of hormone treatment can affect neural mechanisms that may play a role in behavioral outcomes. Levels of choline acetyltransferase increased after acute E₂ treatment but returned to normal after chronic treatment in the basal forebrain (Gibbs, 1997), and acute treatment with estrogen and progesterone resulted in decreases in GABA receptor subunit expression 12, but not 24, hours after treatment (Pazol et al., 2009). It is possible that long-term chronic hormone treatment during aging might lead to a down regulation of ovarian hormone receptors in cognitive brain regions and different behavioral outcomes than short term treatments. Indeed a down-regulation of estrogen receptors was observed after chronic long-term treatment with E₂ (Brown et al., 1996). Because hormone treatment in human females typically continues for several months or years, studies that administer hormones long-term may provide a better model for understanding how these treatments affect cognition during human aging.

Hormone treatments that more closely mimic the natural cycle of hormones might be more beneficial for cognition. However, the studies that have attempted to address this factor have produced varying results. Weekly injections of E₂ plus progesterone enhanced performance of aged female rats on a delayed match-to-position t-maze task, but this improvement was not significantly different from those treated chronically with E₂ and progesterone (Gibbs, 2000). Both continuous and intermittent E₂ improved task acquisition in middle-aged and aged animals (Bimonte-Nelson et al., 2006; Markowska and Savonenko, 2002). While no benefit of E₂ treatment was observed, Chisholm et al. (2012b) also found that chronic and cyclic E₂ treatment resulted in similar effects on a delayed alternation task (Fig 1B). However, some studies have demonstrated differences between these types of treatments. While continuous E₂ treatment had no effect on spatial working or reference memory in the radial arm maze, intermittent E₂ treatment impaired spatial reference memory (Gresack and Frick, 2006). Likewise, middle-aged females treated with long-term chronic E₂ performed better than those treated with long-term cyclic E₂ on the water maze (Lowry et al., 2010) (Fig 1A). None of the studies have found that cyclic treatment was more beneficial for task performance than chronic treatment. Importantly, most of the studies that have attempted to mimic the natural cycle of hormones have only simulated certain aspects of the cycle, and it is possible that more closely simulating the natural cycle by including fluctuations in both estrogen and progesterone levels may be more beneficial than the chronic treatment of ovarian hormones.

Other factors

In order to obtain middle-aged or aged animals, many studies use retired breeders. Reproductive experience has been shown to alter levels of ER α in the striatum with multiparous females having a higher number of ER α positive cells compared to virgin females (Byrnes et al., 2009). Given this, hormone treatments during aging may result in different neural outcomes depending on reproductive experience. In fact, injections of E₂ or estrone increased cell proliferation in the hippocampus of middle-aged multiparous females; however neither estrogen influenced cell proliferation in virgin females (Barha and Galea, 2009). Therefore, this is an important factor when comparing results across studies.

As previously discussed, to more closely model the hormone loss in humans, studies examining the behavioral effects of hormone treatment during aging most often surgically remove the ovaries in middle-aged animals. There is evidence that the type of menopause, surgical or transitional, may influence the behavioral outcome of hormone treatments. The

Bimonte-Nelson laboratory has used the 4-vinylcyclohexene diepoxide (VCD) model to induce a transitional menopause in rats that results in a depletion of ovarian follicles similar to that observed in humans. Interestingly, an increase in the number of working memory errors was observed in these animals when they were treated with CEE (Acosta et al., 2010). This is in contrast to the reduced number of errors observed after treatment with CEE in surgically menopausal females in the same study. How the type of menopause affects the cognitive outcomes of hormone treatment should be further investigated. It is important to note that although the effects of VCD treatment on peripheral tissues has been extensively studied (Van Kempen et al., 2011), it is currently unknown if VCD treatment has direct effects on the brain, and this needs to be fully investigated in order to interpret results from studies using this model.

Concluding remarks: A cautionary note for studies of postmenopausal women

Given that research in rodents indicates that many variables influence behavioral and neural outcomes from hormone treatment, it is not surprising that the studies in humans have resulted in conflicting effects on cognition. There are considerable differences in the age of subjects, length of hormone deprivation, length of hormone treatment and types of hormones administered even among the randomized controlled hormone treatment studies. Along with the rodent literature that supports the “window of opportunity”, reanalysis of the WHI has supported this idea. Whereas the initial study concluded that there was an increased risk for coronary heart disease, reanalysis of women who initiated treatment between the ages of 50–59 years did not find an increase (Rossouw et al., 2007). Importantly, all of the women that were included in the Women’s Health Initiative Memory Study (WHIMS) were 65 years of age or older, and therefore these results cannot be reanalyzed to examine how the length of hormone deprivation might affect cognitive outcomes (Espeland et al., 2004; Rapp et al., 2003a; Rapp et al., 2003b). Therefore the results of the WHIMS and the subsequent Women’s Health Initiative Study of Cognitive Aging (WHISCA) should be interpreted with caution as all of the women included in the study initiated treatment at 65 years of age or older (Rapp et al., 2003b; Resnick et al., 2006; Shumaker et al., 2004). In addition, the initial hypothesis that the negative cognitive effects observed in the WHI were due to MPA being administered with CEE has not been verified by the small number of existing studies. Although, WHISCA found that CEE in combination with MPA decreased verbal memory (Resnick et al., 2006), there have been few human studies that have looked at the effects of estrogen in combination with MPA on cognition in women younger than 65 years. In a randomized controlled trial of women between the ages of 45–55, CEE with MPA did not significantly alter short or long term verbal memory; nor did this combination of hormones impair any of the other measures tested (Maki et al., 2007). A study that examined the cognitive effects of the combination of CEE and MPA in women between the ages of 44–62 years found decreased performance on a digit span forward task; however, this hormone combination resulted in improved performance on a letter fluency task and had no effect on four of the other five measures of verbal memory tested (Maki et al., 2009). Given the small number of studies that have investigated the cognitive effects of CEE in combination with MPA and the lack of significant effects in those studies, more research should be conducted before concluding that CEE with MPA impairs memory. As is evident from the rodent literature, hormone treatments may be beneficial for some tasks in humans and detrimental for others, and our understanding is incomplete at best. We know too little to generalize about the effects of hormone treatments in human females during menopause. The research reviewed in this article highlights the complex nature of hormone treatment during aging and cognition. A better understanding of the details and specificity of the effects would aid future studies. Acknowledgements Supported by NIA AG 022499

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Figure 1.

Behavioral effects of long-term hormone treatment. (A) The spatial water maze. Mean $\pm$ SEM pathlength to find the submerged platform averaged across four days of testing for all groups. The E₂ + MPA group had significantly longer pathlengths than all other groups receiving hormone treatment (Lowry et al., 2010). (B) Spatial alternation. Mean $\pm$ SEM number of sessions to criterion during alternation trials. Animals receiving E₂ + MPA required fewer sessions to reach criterion than animals in three groups: no replacement, chronic E₂, and cyclic E₂ (Chisholm and Juraska 2012b). * $p < .05$

Figure 2.

The effects of long-term hormone treatment on the structure of the medial prefrontal cortex. (A) Synapses labeled with synaptophysin. Total number (mean + SEM) of synaptophysin labeled boutons. Animals receiving E_2 + MPA had more synaptophysin boutons than those receiving no replacement and E_2 + P (Chisholm and Juraska 2012a). (B) Dopaminergic fibers labeled with tyrosine hydroxylase. The volume of TH fibers (mean+ SEM) within the upper layers of the mPFC. Animals receiving E_2 + MPA had a greater volume of tyrosine hydroxylase fibers in the mPFC than those receiving no replacement (Chisholm et al., 2012). * $p < .05$