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# Could androgens maintain specific domains of mental health in aging men by preserving hippocampal neurogenesis?☆

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## Abstract
Interest surrounds the role of sex-hormones in regulating brain function outside of reproductive behaviour. Declining androgen production in aging males has been associated with cognitive impairment, depression and increased risk of developing Alzheimer's disease. Indication for testosterone replacement therapy is based on biochemically determined low circulating testosterone combined with manifest symptoms. However, which aspects of age-related cognitive decline are attributable to low circulating testosterone remain ambiguous. Studies examining cognition in aging men receiving testosterone replacement therapy have yielded equivocal results. The exact role of testosterone in maintaining cognitive function and the underlying neural mechanisms are largely unknown, though it would appear to be domain specific. Clarity in this area will provide clinical direction toward addressing an increasing healthcare burden of mental health decline coincident with increasing longevity. The premise that androgens contribute to maintaining aspects of mental health in aging men by preserving hippocampal neurogenesis will be used as a forum in this review to discuss current knowledge and the need for further studies to better define testosterone replacement strategies for aging male health.

## Key Words
androgen; hippocampus; neurogenesis; aging; cognition; male; testosterone

## Research Highlights
(1) The population of aged men is growing, together with their health care burden.
(2) Testosterone production progressively falls in aging men and some studies associate low testosterone with the development of depression and impaired spatial ability.
(3) Animal models link depression and spatial ability with hippocampal function and neurogenesis.
(4) The degree to which low testosterone participates in the functional decline of these specific cognitive domains remains uncertain.
(5) The merit of testosterone therapy for aged men is a contentious issue.
(6) Further research is required to evaluate a causative relationship between age-related declines in hippocampal function, testosterone production and hippocampal neurogenesis.
(7) New research may determine whether testosterone therapy could maintain hippocampal neurogenesis levels and thereby preserve hippocampal function in aging men.

## Abbreviations
LH, luteinising hormone; FSH, follicle stimulating hormone; SHBG, sex-hormone binding globulin; BrdU, 5-bromo-2'deoxyuridine; FGF, fibroblast growth factor; BDNF, brain derived neurotrophic factor

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## INTRODUCTION

Considerable improvements in health care during the course of the last century have led to unprecedented increases in current life expectancies. However, longevity continues to favor women and as a consequence Chinese and Australian national health policies recognize the need to address male health in aging[1-3]. While increased life expectancy is a broad consequence of quality health care, it also produces an aging population whose health care burden suggests wellbeing has not equally paralleled longevity. Several areas of male health decline with age with increasing incidence of impaired spatial ability and depression being significant barriers to mental health.

A sex difference in longevity intuitively points to a role of sex-hormones in maintaining health during aging. The sex-hormone testosterone is the primary androgen produced in men and circulating levels progressively decrease from 30 years onward so that by 60 years of age, up to 1 in 4 men have developed androgen deficiency[4-5]. With increasing longevity the proportion of androgen deficient or hypogonadal men is likely to grow, along with predicted adverse consequences for male health[6]. Although clinically diagnosed hypogonadism is remediable with testosterone replacement therapy, understanding a causative relationship between age-related physiological decline and reduced androgen levels will enable better defined interventional strategies.

The hippocampus is a key component of the temporal lobe and is notable for retaining ongoing adult neurogenesis within the dentate gyrus subregion[7]. The dentate gyrus is implicated in declining hippocampal function in aging humans[8-9]. Impaired hippocampal function resulting from neurogenesis deficits is hypothesised to contribute to the etiology of both depression and spatial memory impairment[7, 10-12]. In considering the collective clinical and rodent model data, a coincident age-related decline in circulating testosterone, hippocampal neurogenesis and hippocampal function becomes evident. However, a causal relationship between the age-related decline of these factors has yet to be established. This review raises a hypothesis that testosterone contributes to preserving hippocampal function in aging men by maintaining neurogenesis levels. Herein, the intention is to briefly discuss the available evidence and highlight the necessity of further studies to establish, or refute, clinically relevant causal relationships. The key elements of this review are schematically represented in Figure 1.

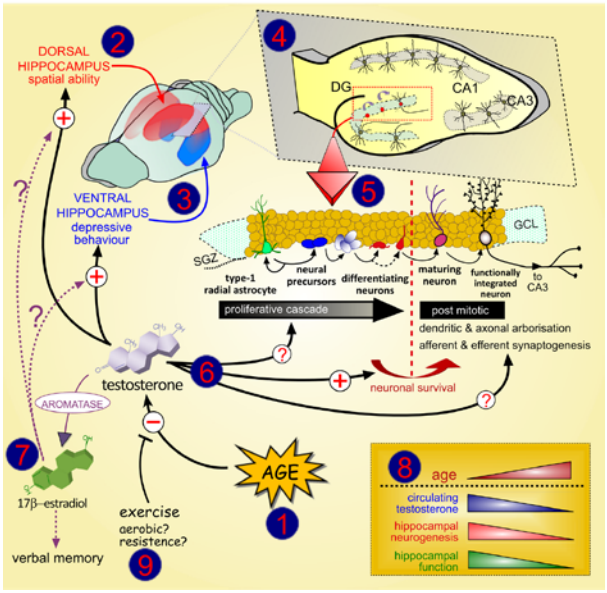


Figure 1 Production of the primary male androgen testosterone is reduced as men age (1).

Circulating testosterone targets the hippocampus, which has been functionally demarcated into dorsal (2) and ventral segments (3).

Testosterone can support spatial memory (2) and provide anti-depressant efficacy (3).

Intrinsic hippocampal circuitry comprises a well described tri-synaptic connection between CA1, CA3 and dentate gyrus (DG) subregions (4).

Ongoing adult neurogenesis proceeds in the sub-granular zone (SGZ) of the DG through sequential proliferative and post-mitotic steps to produce functionally integrated neurons in the granule cell layer (GCL) of the DG (5).

Androgen receptors are expressed in the DG but whether testosterone can directly or indirectly affect neurogenesis remains unresolved (6).

Dosage and duration of testosterone and aromatase-mediated metabolism into 17β-estradiol may be important determinants of efficacy (7).

Declines in circulating testosterone levels, hippocampal neurogenesis and hippocampal function are coincident in aging (8).

Exercise therapy may mediate mental health benefits through androgen maintenance (9).

## ANDROGEN PRODUCTION DURING MALE AGING

Testosterone is the primary male androgen and is produced through the co-ordinated actions of the hypothalamic-pituitary-testes (HPT) axis[13]. While there is normal variation among men, serum total testosterone in men aged under 35 years generally exceeds 20 nM[14-15]. According to several endocrinology health

bodies serum concentrations of total testosterone below 12 nM constitute a clinical diagnosis of hypogonadism, which can be further classified according to a taxonomy that directs remedial strategies[16]. For example, testicular dysfunction, or primary hypogonadism, manifests in the presence of high luteinizing hormone/follicle stimulating hormone (LH/FSH) and is potentially indicated for testosterone replacement therapy. Secondary hypogonadism occurs when hypothalamic/pituitary pathology fails to produce LH/FSH in sufficient levels to stimulate testicular androgen production. In this case, exogenous LH and/or FSH might be indicated to restore testicular function including testosterone production. Age-related low circulating testosterone can be the result of either primary or secondary hypogonadism[4-5]. Otherwise normal aging is associated with reduced total circulating testosterone levels (below 20 nM) concomitant with increased sex-hormone binding globulin (SHBG)[17-18]. Testosterone bound to SHBG is not bioactive and can affect the clearance rate of testosterone by the liver[17]. However, the age-related increase in SHBG levels has been shown to be independent of the age-related decline in bioactive testosterone levels[18].

Testosterone replacement therapy is advised based on symptomatic low circulating testosterone levels[16, 19]. However, applying this criterion to aging men presents potential limitations. Firstly the age at which testosterone levels become low is an important determinant of resulting symptoms[16]. Age-related, or late-onset hypogonadism, may therefore elicit a unique spectrum of symptoms that are yet to be fully established. Secondly, controversy surrounds the reliability of widely used measures of total and free serum testosterone to detect low circulating levels[20]. As symptoms other than sexual dysfunction and frailty are not readily attributed to low circulating testosterone, symptomatic testosterone deficiency could be more common than generally recognised[21]. Testosterone therapy in aging men has hitherto been directed toward treating sexual and cardiovascular dysfunction, muscle mass loss and immobility[16]. Emerging evidence now points to an added role for androgens in maintaining cognitive health[22]. Hence, the symptom profile of late-onset hypogonadism may well include declining mental health.

## COGNITIVE IMPAIRMENT WITH ADVANCING AGE

Although there are cross-sectional variations in measures of cognitive ability, a more robust decline is observed in longitudinal analyses[23-24]. Notwithstanding the view held by some that age-related cognitive decline is normal, maintaining cognition in longevity at young levels would have an undeniable impact on wellbeing. Memory is a chief cognitive function that declines with age[25]. Importantly, not all types of memory decline with normal age. Procedural memory remains largely unaffected, whereas declarative memory, particularly episodic memory shows demonstrable decline with increasing age[26-27]. Declarative memory as a whole is closely associated with temporal lobe function and abilities related to spatial memory are intimately coupled with the hippocampus[28-29]. The decline in spatial ability of aging men is a domain of cognition particularly studied in relation to testosterone levels[30]. This interest may be attributed to the fact that aspects of spatial ability are superior in men compared to women[31-34]. A sex bias in spatial ability is also reflected in rodent studies that have been largely demonstrated using maze navigation tasks[35]. Men and women employ the same brain regions, including the hippocampus, when undertaking spatial navigation tasks[36-37]. These observations suggest that testosterone may target the hippocampus in a manner that enhances spatial ability in men.

Notions of sex differences in brain anatomy and function have centred largely on the effects of sex hormones in development[38]. Evidence supports a possible organisational role of androgen signalling during brain development that imparts superior spatial cognitive ability in adulthood[39-43]. Longitudinal studies showing a correlation between age-related cognitive decline and reduced testosterone[14, 23], raise the possibility that testosterone may perform a similar, but activational role in the mature brain. This idea led to studies demonstrating that testosterone therapy promotes spatial ability in aged men[44]. Additionally, the substantial evidence demonstrating pro-cognitive effects of oestrogen replacement therapy in postmenopausal women, including enhanced hippocampal plasticity and function[45-47], underscores the possibility of a similar mechanistic role of testosterone in aging men. Oestrogen is a major metabolite of testosterone in the male brain[48] further suggesting that low circulating testosterone may be a readily modifiable factor predisposing aging men to cognitive decline.

## ANDROGENS AND HIPPOCAMPAL PLASTICITY AND FUNCTION

Androgens protect the adult brain from degenerative insults such as stroke and traumatic brain injury[49-50].

Although neurodegenerative diseases increase with advancing age, their consideration needs to be distinguished from normal age-related cognitive decline[51]. Along with the dentate gyrus the CA1 and CA3 subregions form anatomical demarcations of the hippocampus. Androgen receptors are expressed in the hippocampus of humans, non-human primates, mice and rats with higher levels in CA1 compared to the CA3 or dentate gyrus subregions[52-55]. Evidence derived from young rodents shows that androgen withdrawal in development will reversibly reduce CA1 and CA3 synaptogenesis and subsequent adult spatial ability[56-58]. Synaptic loss during aging may underlie functional decline of the hippocampus[59-60], (however also see [61]). Studies from rodents and non-human primates have provided evidence that serum androgens support CA1 synaptic density in young adulthood[62-63]. These data provide a mechanism by which androgens could participate in the long-term preservation of hippocampal function[64]. Considered in reverse, the data suggests that age-related androgen deficiency may lead to CA1 synaptic loss and hippocampal dysfunction. As acknowledged above however, longitudinal studies indicate that the dentate gyrus subregion is more closely associated with age-related hippocampal impairment[8]. Evidence that sex-hormones increase dentate gyrus synaptogenesis has been extensively derived from examining the effects of oestrogen[65]. In contrast, there is a relative paucity in studies reporting the effects of testosterone on dendritic structure and synaptic density of dentate gyrus granule neurons. It remains plausible that circulating androgens also target the aging dentate gyrus to preserve mechanisms of neuronal plasticity and hippocampal function. Modifiability of brain structure through neuronal plasticity underpinned developing theories of brain function[66]. However, these theories were immersed in the basic tenet that the mature brain lacks ongoing neuronal production. This dogma was overturned with the widespread acceptance that adult neurogenesis occurs in mammalian brains[67]. Although spatially restricted, neurons generated in the adult dentate gyrus provide a functional form of neuronal plasticity[7, 68-70]. Despite ongoing debate regarding the function of adult hippocampal neurogenesis, the weight of associative evidence points to a role of adult-generated granule neurons in spatial ability and perhaps emotional behaviour related to depression[71-74]. In contrast to the CA1 subregion, androgens could preserve functional plasticity in the dentate gyrus through the addition of new granule neurons. Levels of adult hippocampal neurogenesis decline with age, which has been hypothesised as a contributing factor to age-related decline in spatial ability[75-77]. Importantly, age-related declining adult hippocampal neurogenesis and spatial ability are reversible with factors such as physical exercise[78]. If androgens play a critical role in maintaining adult hippocampal neurogenesis levels then late-onset hypogonadism may be a remediable cause of hippocampal dysfunction in aging men.

## ANDROGENS AND THE REGULATION OF ADULT HIPPOCAMPAL NEUROGENESIS

Adult hippocampal neurogenesis is tightly regulated by physiological, environmental and pharmacological factors[79]. Adult hippocampal neurogenesis proceeds through a series of sequential steps: asymmetric division of neural stem cells, proliferative expansion of neural progenitors, neuronal differentiation and finally survival of mature neurons including both efferent and afferent synaptogenesis[7, 80-81]. Intrinsic proliferation and differentiation markers together with exogenous markers such as 5-bromo-2'deoxyuridine (BrdU; which is incorporated into dividing cells and can assess proliferation and survival at short and longer time-points, respectively) are widely used to measure adult hippocampal neurogenesis[80, 82-84]. Mechanisms of age-induced reductions in adult hippocampal neurogenesis include decreased progenitor proliferation and neuronal survival[76, 78, 85-87], but factors mediating these mechanisms remain unknown. Notions of sex-specific regulation of adult hippocampal neurogenesis were a significant impetus for exploring the effects of sex hormones on proliferation, differentiation and survival of adult-generated hippocampal neurons (reviewed in [88]). There is more hippocampal neurogenesis in neonatal male rats than females[89] and several studies have demonstrated sex-specific rates of hippocampal neurogenesis in adult rodents[90]. Whether there is a sex-specific decline in adult hippocampal neurogenesis with advancing age remains hitherto unestablished.

Early studies withdrawing androgen through gonadectomy in young male rats showed no effect on proliferation but reduced neuronal survival as demonstrated by a reduction in the number of 30-day-old BrdU-labelled cells in the dentate gyrus[91]. Repletion of testosterone at high but not low levels in these animals restored adult hippocampal neurogenesis in an androgen receptor-dependent manner[91]. In contrast, another study showed that exogenous injections of nandrolone, a synthetic derivative of testosterone and androgen receptor agonist, reduced adult hippocampal

neurogenesis by decreasing neuronal precursor proliferation in young adult male rats[92]. Our own recent work demonstrated that chronic testosterone therapy in young male adult mice had no effect on precursor proliferation as measured by the intrinsic marker Ki67, but did promote adult hippocampal neurogenesis through increased neuronal survival and differentiation 28-days after BrdU administration[55]. We measured hippocampal neurogenesis during the last 4 weeks of 3 months of supraphysiological doses of testosterone through sustained-release pellets[55], whereas a similar dosage limited to 3 days *via* repeated injections had no effect on neurogenesis[93]. Another study of young male mice demonstrated that precursor proliferation also measured by Ki67 expression was unchanged, while the generation of maturing neurons measured by another intrinsic marker doublecortin, was decreased in the absence of androgen through gonadectomy[94].

Single administration of BrdU to measure precursor proliferation in gonadectomised young adult male rats untreated or made replete with physiological levels of testosterone showed no effect of androgen withdrawal[95]. However, the same study showed that testosterone exacerbated the up-regulated proliferation in response to the anti-depressant imipramine. In a contrasting finding, Ki67 was used to demonstrate a decrease in precursor proliferation in gonadectomised young male rats compared to sham-surgery controls[96]. This study also employed BrdU to further examine survival and neuronal differentiation, showing that androgen withdrawal through gonadectomy decreased surviving 24-day-old BrdU-labelled cells in the dentate gyrus without change in neuronal differentiation[96]. Furthermore, polysialic acid-neural cell adhesion molecule (PSA-NCAM) was used to demonstrate androgen withdrawal decreases maturing/migrating neurons[96]. Using a different approach, intact male rats given placebo or testosterone and BrdU to measure proliferation, survival and differentiation showed no effect of exogenous androgen on any of these hippocampal neurogenesis parameters[97].

Manipulating testosterone levels in young male rats by gonadectomy or exogenous administration to intact rats showed that testosterone therapy had a capacity to decrease hippocampal neurogenesis[98]. The authors of this study make a pertinent argument that testosterone's effects on hippocampal neurogenesis may be dose and duration-dependent and specific to the developmental stage of newly-generated neurons. In addition, the mode of hormone delivery, repeated injections versus constant release implants likely impacts on serum testosterone levels[98]. Hence differences in delivery mode could confound direct comparisons between studies in which serum testosterone levels are reported[55, 98]. This notion is reflected clinically, where testosterone therapy through long-term skin patches could provide better efficacy compared to repeated surges that accompany periodic injections[16]. Further potential confounds reside in the methodology used to measure hippocampal neurogenesis, for example intrinsic markers such as Ki67, PSA-NCAM and doublecortin versus the timing and dose of exogenous markers like BrdU. More importantly, the manipulations of testosterone levels in young male rodents in these studies do not recapitulate the late-adult onset and gradual decrease in circulating androgen manifest in normal aging. Hence, extrapolating these data to explain a possible causal relationship between low testosterone and declining adult hippocampal neurogenesis in age is limited and specific longitudinal studies examining models of normal aging are required[99].

## ANDROGEN RECEPTOR-MEDIATED EFFECTS ON ADULT HIPPOCAMPAL NEUROGENESIS

Androgen receptor signal transduction incorporates both genomic mechanisms and non-genomic pathways including; cAMP response element-binding protein (CREB), extracellular signal-regulated kinase (ERK) and Akt signalling[100]. These non-genomic signalling pathways in particular could represent intrinsic cellular mechanisms by which testosterone directly effects hippocampal neurogenesis[101]. However, despite androgen receptor expression in the dentate gyrus region[52-55], whether neural stem cells, transient precursor intermediates or maturing neurons exhibit differential expression is unclear. Cultured neural stem cells have been found to express androgen receptor, though these in vitro studies used cells derived from the sub-ventricular zone of adult female rats[92]. Hence it remains to be determined if hippocampal neurogenesis can be directly affected by testosterone or whether effects are transmitted by cellular and/or molecular intermediates.

The local microenvironment, or neurogenic niche, is considered to play a significant role in regulating neurogenesis levels in the adult hippocampus[102]. Astrocytes are an integral cellular component of the neurogenic niche that support hippocampal neurogenesis through mechanisms not fully understood but includes promoting neuronal differentiation and maturation[103]. One mechanism by which astrocytes

stimulate hippocampal neurogenesis is through the secretion of soluble neurotrophic factors such as fibroblast growth factor (FGF)-2. Expression of FGF-2 by astrocytes residing in the hippocampal neurogenic microenvironment decreases during aging and may contribute to the age-related decline in hippocampal neurogenesis[104-105]. Hippocampal astrocytes in male rats express androgen receptor[52] and hence testosterone could influence hippocampal neurogenesis indirectly. Testosterone triggers FGF-2 production in germ-line stem cells of the adult male testis that may play a role in spermatogenesis[106]. Whether such signalling also occurs in neural stem cells to promote hippocampal neurogenesis and whether reductions in this signalling precipitate age-related decline in hippocampal neurogenesis remains untested.

Brain derived neurotrophic factor (BDNF) is another compound that both maintains and mediates up-regulation of hippocampal neurogenesis in rodents and is decreased in the aged hippocampus[107-108]. The ability of testosterone to promote survival of adult-generated neurons in adult canaries is mediated by BDNF[109-110]. Testosterone plays a role in regulating BDNF levels in the neuromuscular system of rodents[111]. However, fluctuations in circulating testosterone during the early development of male mice are independent of hippocampal BDNF levels[112]. Studies using aging rats have shown no sex-specific changes in declining hippocampal BDNF levels, nor any correlations between levels of serum oestrogen or testosterone with hippocampal BDNF in aging male rats[113-114]. The collective evidence would suggest that age-related declining serum testosterone and hippocampal BDNF levels are independent and do not interact to cause falling hippocampal neurogenesis. However, BDNF could still mediate stimulatory effects of testosterone therapy on hippocampal neurogenesis. Evidence supports a direct association between circulating and hippocampal BDNF levels in rats[115]. This association could be important in aging men given the recent findings that circulating BDNF levels are; higher in aging women compared to men, negatively correlated with age in men and positively correlated with bioavailable circulating testosterone levels in men[116]. These observations raise a possibility that BDNF could be a systemic factor mediating the cognitive efficacy of testosterone therapy. Moreover, age-related decline in cellular and molecular elements including signal transduction pathways necessary to mediate testosterone's effects could significantly impact on the effectiveness of androgen therapy and underscores the importance of their elucidation.

The physiological metabolism of testosterone is another important consideration in elucidating the mechanisms of cognitive efficacy. Testosterone is metabolised by 5α-reductase into the high-affinity androgen receptor ligand dihydrotestosterone and by aromatase into the oestrogen receptors and ligand 17β-estradiol. Thus, whether it be intrinsic production or the result of exogenous therapy, effects of circulating testosterone can be mediated through either androgen or oestrogen receptors. However, while both androgen and oestrogen mediated signalling contribute to preserving CA1 synaptic density in females, the same effect in males is androgen receptor dependent[62, 117]. Although one study using rats showed that testosterone therapy tended to increase circulating levels of 17β-estradiol (albeit a non-statistically significant increase), oestrogen activity was not responsible for the increased adult hippocampal neurogenesis[91].

## EFFECTS OF TESTOSTERONE THERAPY ON COGNITION IN ELDERLY MEN

The ability of testosterone therapy to restore declining spatial ability in aging rats was tested in advance of the general acceptance of adult hippocampal neurogenesis[118]. Intriguingly, this study showed no efficacy of testosterone therapy in preserving spatial ability. More recent studies administering testosterone to aged rats however, have shown restorative effects on declining spatial ability[113]. Moreover, studies examining the effects of androgens on spatial abilities in rodents across life-span have yielded complex results[119-120] that more generally reflect the collective clinical data examining aging men.

The relationship between testosterone levels and different parameters of cognition in aging men has been recently reviewed in detail[121]. Decreased circulating testosterone in aging men is linked to poorer performances on tasks requiring spatial ability[122]. Several clinical studies show that spatial ability is improved in aging men receiving testosterone therapy[123-124]. This effect of testosterone is also seen in men suffering mild cognitive impairment and even Alzheimer's disease[125]. However, others have shown no effect of testosterone therapy on cognitive parameters in healthy and mildly cognitive impaired aged men[126-127]. The magnitude of demonstrated spatial ability improvements have varied and may dependent on the dose of testosterone and the length of therapy[128-130]. Spatial ability varies with endogenous circulating testosterone levels in aging men and appears to respond better to intermediate doses of exogenous testosterone

compared low and supraphysiological doses that show no efficacy[122, 129]. Circulating 17β-estradiol levels could change in elderly men receiving testosterone therapy as a consequence of peripheral aromatisation. However, the efficacy of testosterone therapy on spatial ability in aging men appears to be independent of oestrogen activity, though 17β-estradiol may mediate concomitant improvements in verbal memory[128, 131]. Prospective clinical studies of prostate health support the idea that both testosterone and 17β-estradiol levels impact on male aging[132]. Both rodent and clinical studies suggest spatial ability in aged males is enhanced when high testosterone is coincident with low 17β-estradiol levels[44, 113, 122, 133]. Thus, in parallel with the notion of optimal testosterone levels, is the view that the balance of testosterone and 17β-estradiol plays a significant role in spatial ability[134].

Debate over the function of adult hippocampal neurogenesis has evolved to include a specific role in pattern separation, which is to encode and retrieve similar events and is linked to dentate gyrus function[135-136]. The strongest evidence from rodent studies also suggest an association between age-related decline in adult hippocampal neurogenesis and impaired pattern separation ability[137-138]. Emerging evidence now supports a reduction in pattern separation ability in aging humans[139-140]. Given the relative infancy of interest in age-related pattern separation impairment, there is hitherto no definitive evidence for sex differences in the decline of this domain of spatial cognition. It therefore remains possible that testosterone therapy in aging men may preserve pattern separation ability, which could be mediated through maintaining neurogenesis levels in the aging dentate gyrus.

The notion that androgens subserve specific domains of mental health includes effects on emotion-based behaviours. Furthermore, along with spatial ability, adult hippocampal neurogenesis has also been linked with emotion-based behaviour. A hypothesis based on several associative links posits that impaired adult hippocampal neurogenesis elicits depression and that efficacy of antidepressant interventions is mediated through adult hippocampal neurogenesis[12, 73-74, 141]. How does a link between depression and adult hippocampal neurogenesis relate to testosterone levels and advancing age? As with memory loss, age is a risk factor for depression[142]. Gender difference in the incidence of depression disappears with advancing age[143] and the occurrence of depression in elderly men is associated with low testosterone[144-146]. To explore these observations in the context of declining adult hippocampal neurogenesis requires a consideration of a functional distinction in hippocampal structure. The discussion has hitherto observed the classic tri-synaptic circuitry intrinsic to the hippocampus mediated through the interconnectiveness of the dentate gyrus, CA1 and CA3 subregions. However, on a more global level the rodent hippocampus has been regionalised into dorsal (septal pole or posterior in humans) and ventral (temporal pole or anterior in humans) functional subregions[147]. This regionalisation has partly been based on functional assessments, differential gene expression and connectivity, with spatial ability and emotion-based behaviour preferential to the dorsal and ventral hippocampal regions, respectively[148]. Neurogenesis rates may be higher in the dorsal compared to the ventral region of young adult hippocampus[77, 149], but this regional difference attenuates with advancing age, indicating a relatively higher impact of age on dorsal hippocampal neurogenesis. Clinical efficacy of antidepressant treatment is associated with specific increases in adult neurogenesis in the anterior hippocampus in depressed patients[150]. Whether androgen deficiency with advancing age differentially affects neurogenesis rates along the septal-temporal axis in the adult hippocampus in parallel with functional deficits such as depression and impaired spatial ability is not known.

The question of whether antidepressant and spatial ability efficacy of testosterone therapy for aging men is mediated by region-specific hippocampal neurogenesis remains intriguing. Intriguing in part, because addressing this cause-effect relationship will help refine the neurogenesis-depression and neurogenesis-cognition hypotheses[151] and also elucidate links between impaired neural substrates, hypogonadism and symptoms of cognitive dysfunction. This latter point will in turn help define position statements that advocate testosterone therapy based on low testosterone in conjunction with manifest symptoms[19, 152]. Clinical studies using testosterone therapy in elderly men showing either benefit or no effect rather than deterioration in cognition have employed inconsistent and even inadequate measures of mental health[126, 153-154]. A significant point to consider in evaluating this literature is the notion that androgens likely affect mental health in a domain-specific manner[155]. Moreover, conclusions drawn from clinical studies will depend significantly on the cognitive measures used.

## CONCLUDING REMARKS

Uncertainty as to what biochemically measured level of circulating free testosterone constitutes late-onset

hypogonadism and associated symptoms in aging men is a significant impediment to standardised testosterone replacement therapy[19, 145, 156-157]. The prevailing view that age-related mental health decline is not an indicator for testosterone therapy is because of inconclusive rather than negative evidence[16, 152]. Ethical and technical limitations in clinical studies, including varied cognitive assessment methodologies, study inclusion criteria and population size and age range of participants have played a large part in generating this equivocal evidence[121]. This review premises that reduced testosterone is a mediating factor of impaired spatial ability and depression incidence in aging males because of reduced dorsal and ventral hippocampal neurogenesis, respectively. In discussing the known effects of both androgen withdrawal and therapy on hippocampal function and plasticity, it is clear new understanding will be needed to establish a causal relationship between these factors in the context of normal male aging.

There remain several key questions concerning the role androgens play in regulating adult hippocampal neurogenesis, including age-related decline and mediation of stimulatory factors such as exercise[158]. An active lifestyle predicts higher circulating testosterone levels in aging men; could testosterone mediate the ability of physical activity to promote neurogenesis and function in the aged hippocampus[78, 159-160]? If so, given that intensity could determine the effects of exercise on testosterone and neurogenesis levels[161-163], is aerobic or anaerobic exercise best? Emerging studies showing the cognitive benefit of resistance training in the elderly could provide a paradigm shift in the way physical activity is prescribed to preserve mental health[164-166]. Testosterone promotes synaptogenesis in the CA1 subregion; does testosterone support synaptogenesis in neurons generated in the aging dentate gyrus?[81, 167]. Menopause-mediated oestrogen withdrawal is posited play a role in cognitive decline and depressive symptoms in women that manifest early than in males[168]. Because menopause is a far more precipitous event compared to the gradual decline in male androgen production, aging men retain higher levels of 17β-estradiol through peripheral aromatisation[168-169]. What role in maintaining hippocampal neurogenesis and function in aging men does testosterone aromatisation into 17β-estradiol play? The hippocampus expresses receptors for LH and gonadotropin-releasing hormone (GnRH) and their role, directly or indirectly through promoting local steriodogenesis, in age-related decline is uncertain[134, 170-171].

Studies examining these questions will address the paucity in knowledge regarding the areas of impaired mental health to which low testosterone predisposes and also elucidate ongoing debate surrounding the clinical relevance of adult hippocampal neurogenesis[172]. New clinical and basic research endeavours that encompass measures of hippocampal-dependent spatial memory, pattern separation and depression in conjunction with serum measures of HPT axis hormones will shed further light on physiological roles of testosterone in maintaining mental health in aging men.

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## REFERENCES

[1] Australian Institute of Health and Wellbeing. Older Australians at a Glance. 2007, AIHW: Canberra.
[2] Meryn S. Growing global focus on men's health. JMH. 2007;4:382-385.
[3] Sun Y, Liu Z. Men's health in China. JMH. 2007;4:13-17.
[4] Baker HW, Burger HG, de Kretser DM, et al. Changes in the pituitary-testicular system with age. Clin Endocrinol (Oxf). 1976;5:349-372.
[5] Feldman HA, Longcope C, Derby CA, et al. Age trends in the level of serum testosterone and other hormones in middle-aged men: longitudinal results from the Massachusetts male aging study. J Clin Endocrinol Metab. 2002;87:589-598.
[6] de Kretser DM. Determinants of male health: the interaction of biological and social factors. Asian J Androl. 2010;12:291-297.
[7] Song J, Christian K, Ming GL, et al. Modification of hippocampal circuitry by adult neurogenesis. Dev Neurobiol. 2012.
[8] Song J, Christian K, Ming GL, et al. A pathophysiological framework of hippocampal dysfunction in ageing and disease. Nat Rev Neurosci. 2011;12:585-601.
[9] Small SA, Tsai WY, DeLaPaz R, et al. Imaging hippocampal function across the human life span: is memory decline normal or not? Ann Neurol. 2002;51:290-295.
[10] Aimone JB, Deng W, Gage FH. Adult neurogenesis: integrating theories and separating functions. Trends Cogn Sci. 2010;14:325-337.

[11] Kempermann G, Krebs J, Fabel K. The contribution of failing adult hippocampal neurogenesis to psychiatric disorders. Curr Opin Psychiatry. 2008;21:290-295.
[12] Kempermann G, Kronenberg G. Depressed new neurons--adult hippocampal neurogenesis and a cellular plasticity hypothesis of major depression. Biol Psychiatry. 2003;54:499-503.
[13] Mooradian AD, Morley JE, Korenman SG. Biological actions of androgens. Endocr Rev. 1987;8:1-28.
[14] Vermeulen A. Declining androgens with age: An overview. In: Oddens BJ, Vermeulen A, eds. Androgens and the Aging Male. New York: Parthenon. 1996
[15] Simon D, Nahoul K, Charles MA. Sex hormones, aging, ethnicity and insulin sensitivity: An overview of the TELECOM study. In: Oddens BJ, Vermeulen A, eds. Androgens and the Aging Male. New York: Parthenon. 1996.
[16] Corona G, Rastrelli G, Forti G, et al. Update in testosterone therapy for men. J Sex Med. 2011;8: 639-654.
[17] Vermeulen A, Andó S. Metabolic clearance rate and interconversion of androgens and the influence of the free androgen fraction. J Clin Endocrinol Metab. 1979;48: 320-326.
[18] de Ronde W, van der Schouw YT, Pierik FH, et al. Serum levels of sex hormone-binding globulin (SHBG) are not associated with lower levels of non-SHBG-bound testosterone in male newborns and healthy adult men. Clin Endocrinol (Oxf). 2005;62:498-503.
[19] Andrology AS. Testosterone replacement therapy for male aging: ASA position statement. J Androl. 2006;27: 133-134.
[20] Sikaris K, McLachlan RI, Kazlauskas R, et al. Reproductive hormone reference intervals for healthy fertile young men: evaluation of automated platform assays. J Clin Endocrinol Metab. 2005;90: 5928-5936.
[21] Trinick TR, Feneley MR, Welford H, et al. International web survey shows high prevalence of symptomatic testosterone deficiency in men. Aging Male. 2011;14: 10-15.
[22] Galea LA, Uban KA, Epp JR, et al. Endocrine regulation of cognition and neuroplasticity: our pursuit to unveil the complex interaction between hormones, the brain, and behaviour. Can J Exp Psychol. 2008;62:247-260.
[23] Albert M, Duffy FH, Naeser M. Nonlinear changes in cognition with age and their neuropsychologic correlates. Can J Psychol. 1987;41:141-157.
[24] Drachman DA. Memory and cognitive function in normal aging. Dev Neuropsychol. 1986;2:277-285.
[25] Crook T, Bartus RT, Ferris SH, et al. Age-associated memory impairment: proposed diagnostic criteria and measures of clinical changes-report of a national institute of mental health work group. Dev Neuropsychol. 1986;2:261-276.
[26] Balota DA, Dolan PO, Duchek JM. Memory changes in healthy young and older adults. In: Tulving E, Craik FIM, eds. Handbook of Memory. Oxford University Press. 2000.
[27] Barnes CA. Memory changes in normal aging: Neurobiological correlates. In: Martinez Jr JA, Kesner RP, eds. Neurobiology of Learning and Memory. San Diego: Academic Press. 1998.
[28] Eichenbaum H. A cortical-hippocampal system for declarative memory. Nat Rev Neurosci. 2000;1:41-50.
[29] Eichenbaum H. Hippocampus: mapping or memory? Curr Biol. 2000;10:R785-787.
[30] Beauchet O. Testosterone and cognitive function: current clinical evidence of a relationship. Eur J Endocrinol. 2006;155:773-781.
[31] Astur RS, Ortiz ML, Sutherland RJ. A characterization of performance by men and women in a virtual Morris water task: a large and reliable sex difference. Behav Brain Res. 1998;93:185-190.
[32] Kaufman SB. Sex differences in mental rotation and spatial visualization ability: Can they be accounted for by differences in working memory capacity? Intelligence. 2007;35:211-223.
[33] Parsons TD, Larson P, Kratz K, et al. Sex differences in mental rotation and spatial rotation in a virtual environment. Neuropsychologia. 2004;42:555-562.
[34] Postma A, Jager G, Kessels RP, et al. Sex differences for selective forms of spatial memory. Brain Cogn. 2004;54: 24-34.
[35] Jonasson Z. Meta-analysis of sex differences in rodent models of learning and memory: a review of behavioral and biological data. Neurosci Biobehav Rev. 2005;28: 811-825.
[36] Blanch RJ, Brennan D, Condon B, et al. Are there gender-specific neural substrates of route learning from different perspectives? Cereb Cortex. 2004;14: 1207-1213.
[37] Maguire EA, Frackowiak RS, Frith CD. Learning to find your way: a role for the human hippocampal formation. Proc Biol Sci. 1996;263:1745-1750.
[38] Hines M. Sex-related variation in human behavior and the brain. Trends Cogn Sci. 2010;14:448-456.
[39] Hines M, Fane BA, Pasterski VL, et al. Spatial abilities following prenatal androgen abnormality: targeting and mental rotations performance in individuals with congenital adrenal hyperplasia. Psychoneuroendocrinology. 2003;28:1010-1026.
[40] Isgor C, Sengelaub DR. Prenatal gonadal steroids affect adult spatial behavior, CA1 and CA3 pyramidal cell morphology in rats. Horm Behav. 1998;34:183-198.
[41] Mueller SC, Temple V, Oh E, et al. Early androgen exposure modulates spatial cognition in congenital adrenal hyperplasia (CAH). Psychoneuroendocrinology. 2008;33:973-980.
[42] Roof RL, Havens MD. Testosterone improves maze performance and induces development of a male hippocampus in females. Brain Res. 1992;572:310-313.

[43] Williams CL, Barnett AM, Meck WH. Organizational effects of early gonadal secretions on sexual differentiation in spatial memory. Behav Neurosci. 1990;104:84-97.
[44] Janowsky JS, Oviatt SK, Orwoll ES. Testosterone influences spatial cognition in older men. Behav Neurosci. 1994;108:325-332.
[45] Frick KM. Estrogens and age-related memory decline in rodents: what have we learned and where do we go from here? Horm Behav. 2009;55:2-23.
[46] Gibbs R. Preclinical data relating to estrogen's effects on cognitive performance. In: Rasgon NL, ed. The Effects of Estrogen in Brain Function. Baltimore: The John Hopkins University Press. 2006.
[47] Spencer JL, Waters EM, Romeo RD, et al. Uncovering the mechanisms of estrogen effects on hippocampal function. Front Neuroendocrinol. 2008;29:219-237.
[48] Garcia-Segura LM. Aromatase in the brain: not just for reproduction anymore. J Neuroendocrinol. 2008;20: 705-712.
[49] Pike CJ, Nguyen TV, Ramsden M, et al. Androgen cell signaling pathways involved in neuroprotective actions. Horm Behav. 2008;53:693-705.
[50] Pike CJ, Nguyen TV, Ramsden M, et al. Androgens exert direct neuroprotective effects on the brain: a review of pre-clinical evidences. JAS. 2010;17:49-55.
[51] Albert MS. Memory decline: the boundary between aging and age-related disease. Ann Neurol. 2002;51:282-284.
[52] Tabori NE, Stewart LS, Znamensky V, et al. Ultrastructural evidence that androgen receptors are located at extranuclear sites in the rat hippocampal formation. Neuroscience. 2005;130:151-163.
[53] Kerr JE, Beck SG, Handa RJ. Androgens modulate glucocorticoid receptor mRNA, but not mineralocorticoid receptor mRNA levels, in the rat hippocampus. J Neuroendocrinol. 1996;8:439-447.
[54] Beyenburg S, Watzka M, Clusmann H, et al. Androgen receptor mRNA expression in the human hippocampus. Neurosci Lett. 2000;294:25-28.
[55] Hannan AJ, Ransome MI. Deficits in Spermatogenesis but not Neurogenesis are Alleviated by Chronic Testosterone Therapy in R6/1 Huntington's Disease Mice. J Neuroendocrinol. 2012;24:341-356.
[56] Isgor C, Sengelaub DR. Effects of neonatal gonadal steroids on adult CA3 pyramidal neuron dendritic morphology and spatial memory in rats. J Neurobiol. 2003;55:179-190.
[57] Zhang JM, Konkle AT, Zup SL, et al. Impact of sex and hormones on new cells in the developing rat hippocampus: a novel source of sex dimorphism? Eur J Neurosci. 2008;27:791-800.
[58] Zhang JM, Tonelli L, Regenold WT, et al. Effects of neonatal flutamide treatment on hippocampal neurogenesis and synaptogenesis correlate with depression-like behaviors in preadolescent male rats. Neuroscience. 2010;169:544-554.
[59] Bliss TV, Collingridge GL. A synaptic model of memory: long-term potentiation in the hippocampus. Nature. 1993;361:31-39.
[60] Desai AK. Revitalizing the aged brain. Med Clin North Am. 2011;95:463-475, ix.
[61] Burke SN, Barnes CA. Neural plasticity in the ageing brain. Nat Rev Neurosci. 2006;7:30-40.
[62] Leranth C, Petnehazy O, MacLusky NJ. Gonadal hormones affect spine synaptic density in the CA1 hippocampal subfield of male rats. J Neurosci. 2003;23:1588-1592.
[63] Leranth C, Prange-Kiel J, Frick KM, et al. Low CA1 spine synapse density is further reduced by castration in male non-human primates. Cereb Cortex. 2004;14:503-510.
[64] MacLusky NJ, Hajszan T, Prange-Kiel J, et al. Androgen modulation of hippocampal synaptic plasticity. Neuroscience. 2006;138:957-965.
[65] Hajszan T, Milner TA, Leranth C. Sex steroids and the dentate gyrus. Prog Brain Res. 2007;163:399-415.
[66] Berlucchi G, Buchtel HA. Neuronal plasticity: historical roots and evolution of meaning. Exp Brain Res. 2009;192: 307-319.
[67] Gross CG. Neurogenesis in the adult brain: death of a dogma. Nat Rev Neurosci. 2000;1:67-73.
[68] Lledo PM, Alonso M, Grubb MS. Adult neurogenesis and functional plasticity in neuronal circuits. Nat Rev Neurosci. 2006;7:179-193.
[69] ALTMAN J. Are new neurons formed in the brains of adult mammals? Science. 1962;135:1127-1128.
[70] ALTMAN J. Autoradiographic investigation of cell proliferation in the brains of rats and cats. Anat Rec. 1963;145:573-591.
[71] Deng W, Aimone JB, Gage FH. New neurons and new memories: how does adult hippocampal neurogenesis affect learning and memory? Nat Rev Neurosci. 2010;11:339-350.
[72] Dupret D, Revest JM, Koehl M, et al. Spatial relational memory requires hippocampal adult neurogenesis. PLoS One. 2008;3:e1959.
[73] Drew MR, Hen R. Adult hippocampal neurogenesis as target for the treatment of depression. CNS Neurol Disord Drug Targets. 2007;6:205-218.
[74] Jacobs BL, van Praag H, Gage FH. Adult brain neurogenesis and psychiatry: a novel theory of depression. Mol Psychiatry. 2000;5:262-269.
[75] Heine VM, Maslam S, Joëls M, et al. Prominent decline of newborn cell proliferation, differentiation, and apoptosis in the aging dentate gyrus, in absence of an age-related hypothalamus-pituitary-adrenal axis activation. Neurobiol Aging. 2004;25:361-735.
[76] Kuhn HG, Dickinson-Anson H, Gage FH. Neurogenesis in the dentate gyrus of the adult rat: age-related decrease of neuronal progenitor proliferation. J Neurosci. 1996;16: 2027-2033.
[77] Snyder JS, Ramchand P, Rabbett S, et al. Septo-temporal gradients of neurogenesis and activity in 13-month-old rats. Neurobiol Aging. 2011;32:1149-1156.

[78] van Praag H, Shubert T, Zhao C, et al. Exercise enhances learning and hippocampal neurogenesis in aged mice. J Neurosci. 2005;25:8680-8685.
[79] Gage FH. Molecular and cellular mechanisms contributing to the regulation, proliferation and differentiation of neural stem cells in the adult dentate gyrus. Keio J Med. 2010;59:79-83.
[80] Kempermann G, Jessberger S, Steiner B, et al. Milestones of neuronal development in the adult hippocampus. Trends Neurosci. 2004;27:447-452.
[81] Toni N, Sultan S. Synapse formation on adult-born hippocampal neurons. Eur J Neurosci. 2011;33: 1062-1068.
[82] Manning EE, Ransome MI, Burrows EL, et al. Increased adult hippocampal neurogenesis and abnormal migration of adult-born granule neurons is associated with hippocampal-specific cognitive deficits in phospholipase C-beta1 knockout mice. Hippocampus. 2012;22:309-319.
[83] Ransome MI, Turnley AM. Systemically delivered Erythropoietin transiently enhances adult hippocampal neurogenesis. J Neurochem. 2007;102:1953-1965.
[84] Ransome MI, Turnley AM. Growth hormone signaling and hippocampal neurogenesis: insights from genetic models. Hippocampus. 2008;18:1034-1050.
[85] Kempermann G, Kuhn HG, Gage FH. Experience-induced neurogenesis in the senescent dentate gyrus. J Neurosci. 1998;18:3206-3212.
[86] Olariu A, Cleaver KM, Cameron HA. Decreased neurogenesis in aged rats results from loss of granule cell precursors without lengthening of the cell cycle. J Comp Neurol. 2007;501:659-667.
[87] Rao MS, Hattiangady B, Abdel-Rahman A, et al. Newly born cells in the ageing dentate gyrus display normal migration, survival and neuronal fate choice but endure retarded early maturation. Eur J Neurosci. 2005;21: 464-476.
[88] Galea LA. Gonadal hormone modulation of neurogenesis in the dentate gyrus of adult male and female rodents. Brain Res Rev. 2008;57:332-341.
[89] Bowers JM, Waddell J, McCarthy MM. A developmental sex difference in hippocampal neurogenesis is mediated by endogenous oestradiol. Biol Sex Differ. 2010;1:8.
[90] Galea LA, Spritzer MD, Barker JM, et al. Gonadal hormone modulation of hippocampal neurogenesis in the adult. Hippocampus. 2006;16:225-232.
[91] Spritzer MD, Galea LA. Testosterone and dihydrotestosterone, but not estradiol, enhance survival of new hippocampal neurons in adult male rats. Dev Neurobiol. 2007;67:1321-1333.
[92] Brännvall K, Bogdanovic N, Korhonen L, et al. 19-Nortestosterone influences neural stem cell proliferation and neurogenesis in the rat brain. Eur J Neurosci. 2005;21:871-878.
[93] Zhang Z, Yang R, Zhou R, et al. Progesterone promotes the survival of newborn neurons in the dentate gyrus of adult male mice. Hippocampus. 2010;20:402-412.
[94] Benice TS, Raber J. Castration and training in a spatial task alter the number of immature neurons in the hippocampus of male mice. Brain Res. 2010;1329:21-29.
[95] Carrier N, Kabbaj M. Testosterone and imipramine have antidepressant effects in socially isolated male but not female rats. Horm Behav. 2012;61:678-685.
[96] Wainwright SR, Lieblich SE, Galea LA. Hypogonadism predisposes males to the development of behavioural and neuroplastic depressive phenotypes. Psychoneuroendocrinology. 2011;36:1327-1341.
[97] Buwalda B, van der Borght K, Koolhaas JM, et al. Testosterone decrease does not play a major role in the suppression of hippocampal cell proliferation following social defeat stress in rats. Physiol Behav. 2010;101: 719-725.
[98] Spritzer MD, Ibler E, Inglis W, et al. Testosterone and social isolation influence adult neurogenesis in the dentate gyrus of male rats. Neuroscience. 2011;195: 180-190.
[99] Gallagher M, Bizon JL, Hoyt EC, et al. Effects of aging on the hippocampal formation in a naturally occurring animal model of mild cognitive impairment. Exp Gerontol. 2003; 38:71-77.
[100] Bennett NC, Gardiner RA, Hooper JD, et al. Molecular cell biology of androgen receptor signalling. Int J Biochem Cell Biol. 2010;42:813-827.
[101] Suh H, Deng W, Gage FH. Signaling in adult neurogenesis. Annu Rev Cell Dev Biol. 2009;25:253-275.
[102] Seki T. Microenvironmental elements supporting adult hippocampal neurogenesis. Anat Sci Int. 2003;78 69-78.
[103] Lie DC, Song H, Colamarino SA, et al. Neurogenesis in the adult brain: new strategies for central nervous system diseases. Annu Rev Pharmacol Toxicol. 2004;44:399-421.
[104] Jin K, Sun Y, Xie L, et al. Neurogenesis and aging: FGF-2 and HB-EGF restore neurogenesis in hippocampus and subventricular zone of aged mice. Aging Cell. 2003;2: 175-183.
[105] Shetty AK, Hattiangady B, Shetty GA. Stem/progenitor cell proliferation factors FGF-2, IGF-1, and VEGF exhibit early decline during the course of aging in the hippocampus: role of astrocytes. Glia. 2005;51:173-186.
[106] Gonzalez-Herrera IG, Prado-Lourenco L, Pileur F, et al. Testosterone regulates FGF-2 expression during testis maturation by an IRES-dependent translational mechanism. FASEB J. 2006;20:476-478.
[107] Hattiangady B, Rao MS, Shetty GA, et al. Brain-derived neurotrophic factor, phosphorylated cyclic AMP response element binding protein and neuropeptide Y decline as early as middle age in the dentate gyrus and CA1 and CA3 subfields of the hippocampus. Exp Neurol. 2005;195: 353-371.
[108] Bekinschtein P, Oomen CA, Saksida LM, et al. Effects of environmental enrichment and voluntary exercise on neurogenesis, learning and memory, and pattern separation: BDNF as a critical variable? Semin Cell Dev Biol. 2011;22:536-542.

[109] Nottebohm F. From bird song to neurogenesis. Sci Am. 1989;260:74-79.
[110] Rasika S, Alvarez-Buylla A, Nottebohm F. BDNF mediates the effects of testosterone on the survival of new neurons in an adult brain. Neuron. 1999;22:53-62.
[111] Verhovshek T, Cai Y, Osborne MC, et al. Androgen regulates brain-derived neurotrophic factor in spinal motoneurons and their target musculature. Endocrinology. 2010;151:253-261.
[112] Hill RA, Wu YW, Kwek P, et al. Modulatory effects of sex steroid hormones on brain-derived neurotrophic factor-tyrosine kinase B expression during adolescent development in C57Bl/6 mice. J Neuroendocrinol. 2012;24:774-788.
[113] Bimonte-Nelson HA, Singleton RS, Nelson ME, et al. Testosterone, but not nonaromatizable dihydrotestosterone, improves working memory and alters nerve growth factor levels in aged male rats. Exp Neurol. 2003;181:301-312.
[114] Bimonte-Nelson HA, Granholm AC, Nelson ME, et al. Patterns of neurotrophin protein levels in male and female Fischer 344 rats from adulthood to senescence: how young is "young" and how old is "old"? Exp Aging Res. 2008;34:13-26.
[115] Klein AB, Williamson R, Santini MA, et al. Blood BDNF concentrations reflect brain-tissue BDNF levels across species. Int J Neuropsychopharmacol. 2011;14:347-353.
[116] Golden E, Emiliano A, Maudsley S, et al. Circulating brain-derived neurotrophic factor and indices of metabolic and cardiovascular health: data from the Baltimore Longitudinal Study of Aging. PLoS One. 2010;5:e10099.
[117] Leranth C, Hajszan T, MacLusky NJ. Androgens increase spine synapse density in the CA1 hippocampal subfield of ovariectomized female rats. J Neurosci. 2004;24:495-499.
[118] Goudsmit E, Van de Poll NE, Swaab DF. Testosterone fails to reverse spatial memory decline in aged rats and impairs retention in young and middle-aged animals. Behav Neural Biol. 1990;53:6-20.
[119] Gibbs RB, Johnson DA. Sex-specific effects of gonadectomy and hormone treatment on acquisition of a 12-arm radial maze task by Sprague Dawley rats. Endocrinology. 2008;149:3176-383.
[120] Spritzer MD, Daviau ED, Coneeny MK, et al. Effects of testosterone on spatial learning and memory in adult male rats. Horm Behav. 2011;59:484-496.
[121] Holland J, Bandelow S, Hogervorst E.Testosterone levels and cognition in elderly men: a review. Maturitas. 2011;69: 322-337.
[122] Muller M, Aleman A, Grobbee DE, et al. Endogenous sex hormone levels and cognitive function in aging men: is there an optimal level? Neurology. 2005;64:866-871.
[123] Tan RS, Pu SJ. A pilot study on the effects of testosterone in hypogonadal aging male patients with Alzheimer's disease. Aging Male. 2003;6:13-17.
[124] Janowsky JS. The role of androgens in cognition and brain aging in men. Neuroscience. 2006;138:1015-1020.
[125] Cherrier MM, Matsumoto AM, Amory JK, et al. Testosterone improves spatial memory in men with Alzheimer disease and mild cognitive impairment. Neurology. 2005;64:2063-2068.
[126] Kenny AM, Fabregas G, Song C, et al. Effects of testosterone on behavior, depression, and cognitive function in older men with mild cognitive loss. J Gerontol A Biol Sci Med Sci. 2004;59:75-78.
[127] Haren MT, Wittert GA, Chapman IM, et al. Effect of oral testosterone undecanoate on visuospatial cognition, mood and quality of life in elderly men with low-normal gonadal status. Maturitas. 2005;50:124-133.
[128] Cherrier MM, Asthana S, Plymate S, et al. Testosterone supplementation improves spatial and verbal memory in healthy older men. Neurology. 2001;57:80-88.
[129] Cherrier MM, Matsumoto AM, Amory JK, et al. Characterization of verbal and spatial memory changes from moderate to supraphysiological increases in serum testosterone in healthy older men. Psychoneuroendocrinology. 2007;32:72-79.
[130] Gray PB, Singh AB, Woodhouse LJ, et al. Dose-dependent effects of testosterone on sexual function, mood, and visuospatial cognition in older men. J Clin Endocrinol Metab. 2005;90:3838-3846.
[131] Cherrier MM, Matsumoto AM, Amory JK, et al. The role of aromatization in testosterone supplementation: effects on cognition in older men. Neurology. 2005;64:290-296.
[132] Gann PH, Hennekens CH, Longcope C, et al. A prospective study of plasma hormone levels, nonhormonal factors, and development of benign prostatic hyperplasia. Prostate. 1995;26:40-49.
[133] Barrett-Connor E, Goodman-Gruen D, Patay B. Endogenous sex hormones and cognitive function in older men. J Clin Endocrinol Metab. 1999;84:3681-3685.
[134] Conrad CD, Bimonte-Nelson HA. Impact of the hypothalamic-pituitary-adrenal/gonadal axes on trajectory of age-related cognitive decline. Prog Brain Res. 2010; 182:31-76.
[135] Leutgeb JK, Leutgeb S, Moser MB, et al. Pattern separation in the dentate gyrus and CA3 of the hippocampus. Science. 2007;315:961-966.
[136] Marr D. Simple memory: a theory for archicortex. Philos Trans R Soc Lond B Biol Sci. 1971;262:23-81.
[137] Clelland CD, Choi M, Romberg C, et al. A functional role for adult hippocampal neurogenesis in spatial pattern separation. Science. 2009;325:210-213.
[138] Creer DJ, Romberg C, Saksida LM, et al. Running enhances spatial pattern separation in mice. Proc Natl Acad Sci U S A. 2010;107:2367-2372.
[139] Holden HM, Hoebel C, Loftis K, et al. Spatial pattern separation in cognitively normal young and older adults. Hippocampus. 2012.
[140] Yassa MA, Mattfeld AT, Stark SM, et al. Age-related memory deficits linked to circuit-specific disruptions in the hippocampus. Proc Natl Acad Sci U S A. 2011;108: 8873-8878.

[141] Duman RS, Malberg J, Thome J. Neural plasticity to stress and antidepressant treatment. Biol Psychiatry. 1999;46:1181-1191.
[142] Seidman SN. The aging male: androgens, erectile dysfunction, and depression. J Clin Psychiatry. 2003;64 Suppl 10:31-37.
[143] Wilhelm K, Mitchell P, Slade T, et al. Prevalence and correlates of DSM-IV major depression in an Australian national survey. J Affect Disord. 2003;75:155-162.
[144] Shores MM, Moceri VM, Sloan KL, et al. Low testosterone levels predict incident depressive illness in older men: effects of age and medical morbidity. J Clin Psychiatry. 2005;66:7-14.
[145] Zitzmann M, Faber S, Nieschlag E. Association of specific symptoms and metabolic risks with serum testosterone in older men. J Clin Endocrinol Metab. 2006;91:4335-4343.
[146] Almeida OP, Yeap BB, Hankey GJ, et al. Low free testosterone concentration as a potentially treatable cause of depressive symptoms in older men. Arch Gen Psychiatry. 2008;65:283-289.
[147] Moser MB, Moser EI. Functional differentiation in the hippocampus. Hippocampus. 1998;8:608-619.
[148] Fanselow MS, Dong HW. Are the dorsal and ventral hippocampus functionally distinct structures? Neuron. 2010;65:7-19.
[149] Snyder JS, Radik R, Wojtowicz JM, et al. Anatomical gradients of adult neurogenesis and activity: young neurons in the ventral dentate gyrus are activated by water maze training. Hippocampus. 2009;19:360-370.
[150] Boldrini M, Underwood MD, Hen R, et al. Antidepressants increase neural progenitor cells in the human hippocampus. Neuropsychopharmacology. 2009;34: 2376-2389.
[151] Sapolsky RM. Is impaired neurogenesis relevant to the affective symptoms of depression? Biol Psychiatry. 2004;56:137-139.
[152] Krause W, Mueller U, Mazur A. Testosterone supplementation in the aging male: which questions have been answered? Aging Male. 2005;8:31-38.
[153] Bassil N. Late-onset hypogonadism. Med Clin North Am. 2011;95:507-23, x.
[154] Emmelot-Vonk MH, Verhaar HJ, Nakhai Pour HR, et al. Effect of testosterone supplementation on functional mobility, cognition, and other parameters in older men: a randomized controlled trial. JAMA. 2008;299:39-52.
[155] Gooren L. Testosterone and the brain. JMH. 2007;4: 344-351.
[156] Carruthers M. The paradox dividing testosterone deficiency symptoms and androgen assays: a closer look at the cellular and molecular mechanisms of androgen action. J Sex Med. 2008;5:998-1012.
[157] Nieschlag E, Swerdloff R, Behre HM, et al. Investigation, treatment and monitoring of late-onset hypogonadism in males. ISA, ISSAM, and EAU recommendations. Eur Urol. 2005;48:1-4.
[158] van Praag H, Kempermann G, Gage FH. Running increases cell proliferation and neurogenesis in the adult mouse dentate gyrus. Nat Neurosc. 1999;2:266-270.
[159] Chigurupati S, Son TG, Hyun DH, et al. Lifelong running reduces oxidative stress and degenerative changes in the testes of mice. J Endocrinol. 2008;199:333-341.
[160] Yeap BB, Almeida OP, Hyde Z, et al. Healthier lifestyle predicts higher circulating testosterone in older men: the Health In Men Study. Clin Endocrinol (Oxf). 2009;70: 455-463.
[161] Lou SJ, Liu JY, Chang H, et al. Hippocampal neurogenesis and gene expression depend on exercise intensity in juvenile rats. Brain Res. 2008;1210:48-55.
[162] Di Luigi L, Romanelli F, Sgrò P, et al. Andrological aspects of physical exercise and sport medicine. Endocrine. 2012.
[163] Voss MW, Nagamatsu LS, Liu-Ambrose T, et al. Exercise, brain, and cognition across the life span. J Appl Physiol. 2011;111:1505-1513.
[164] Cassilhas RC, Viana VA, Grassmann V, et al. The impact of resistance exercise on the cognitive function of the elderly. Med Sci Sports Exerc. 2007;39:1401-1407.
[165] Liu-Ambrose T, Nagamatsu LS, Voss MW, et al. Resistance training and functional plasticity of the aging brain: a 12-month randomized controlled trial. Neurobiol Aging. 2012;33:1690-1698.
[166] Nagamatsu LS, Handy TC, Hsu CL, et al. Resistance training promotes cognitive and functional brain plasticity in seniors with probable mild cognitive impairment. Arch Intern Med. 2012;172:666-668.
[167] Morgenstern NA, Lombardi G, Schinder AF. Newborn granule cells in the ageing dentate gyrus. J Physiol. 2008;586:3751-3757.
[168] Shepherd JE. Effects of estrogen on congnition mood, and degenerative brain diseases. J Am Pharm Assoc (Wash). 2001;41:221-228.
[169] Ferrini RL, Barrett-Connor E. Sex hormones and age: a cross-sectional study of testosterone and estradiol and their bioavailable fractions in community-dwelling men. Am J Epidemiol. 1998;147:750-754.
[170] Fester L, Prange-Kiel J, Jarry H, et al. Estrogen synthesis in the hippocampus. Cell Tissue Res. 2011;345:285-294.
[171] Mak GK, Weiss S. Paternal recognition of adult offspring mediated by newly generated CNS neurons. Nat Neurosci. 2010;13:753-758.
[172] Curtis MA, Low VF, Faull RL. Neurogenesis and progenitor cells in the adult human brain: a comparison between hippocampal and subventricular progenitor proliferation. Dev Neurobiol. 2012;72:990-1005.

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