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Chapter 47

# State of the Art Anti-Aging Practice: Diagnostic Difficulties of the Andropause

*Robert S Tan, M.D.*
*Associate Professor, Department of Family Medicine at the University of Texas/Houston*

## ABSTRACT

Even before contemplating hormonal replacement for men, practitioners should be certain of a diagnosis of low testosterone causing andropause-related symptoms. This is partly because accurate diagnose may be confounded due to factors such as clinical depression, hypothyroidism, anemia, alcoholism, psychological states, and side effects of medications.

There is undeniable evidence that bioavailable testosterone declines with age. However, there are variations in testosterone measures based on timing and method used for quantification, and this will be explored in this chapter. There are limitations of blood and saliva measurements of testosterone, and andropause should be diagnosed only in conjunction with a clinical assessment, as it is a complex syndrome.

The decline of testosterone leads to hypogonadism, which can present with symptoms in older men. However, like in the menopause, not all older men present with transitional symptoms as they enter the andropause. The long-term effects of andropause in men may be similar to menopause, as decline in androgens can affect libido, musculoskeletal, cardiac, and cognitive functions. In addition, in contrast to menopause, men in andropause do not necessarily suffer from infertility as sperm production continues. The evidence of the impact of low testosterone on the aging male's physiology will be examined.

Androgen supplementation in the form of testosterone may be effective for some individuals who are symptomatic. At the present time, is no routine recommendation for androgen replacement in all males. The practitioner should select his patient carefully based on symptoms and biochemical evidence of hypogonadism. Monitoring of side effects of androgens, including changes in prostate, hemoglobin levels, etc., is mandatory.

**Keywords:** Testosterone; Andropause; Hypogonadism; Symptoms; Libido

## INTRODUCTION

As a physician, it is not unusual to come across male patients in their forties and beyond, complaining of loss of libido, erectile dysfunction, fatigue and depression. Psychological problems and medical illness are often confounders to the andropause. Knowledge of the patient's

history and a careful examination combined with laboratory tests are keys to an accurate diagnosis of symptomatic hypogonadism, or the andropause syndrome.[1]

Androgens are a group of hormones that include testosterone, dihydroepiandrosterone (DHEA), androstenedione, and others. It is a misnomer to classify them as these "male hormones" as they are present in both males and females, albeit in different amounts. There is undeniable evidence that aging results in a lowering of androgens. When total testosterone is measured, 20% of men above 55 years are hypogonadal.[2,3] However, when bioavailable testosterone is measured, 50% of men above 50 years are defined as hypogonadal.[4] Ninety-eight percent of circulating testosterone is bound to plasma proteins, the remaining 2% of free testosterone is responsible for biological activity. Approximately 40% of the bound testosterone is bound to sex hormone binding globulin (SHBG). The rest is weakly bound to albumin and is readily available to tissue when needed. Bioavailable testosterone includes free testosterone and that loosely bound to albumin.[5]

## *DEFINITIONAL ASPECTS OF ANDROPAUSE*

As the decline in androgens is gradual, the alternative term of "Androgen Decline in Aging Males" (ADAM) has been used. "Partial Androgen Decline in Aging Males" (PADAM) has also been suggested, because the androgen deficiency in older men is generally moderate and not a complete deficiency. There is often confusion that andropause is a symptomatic state. It must be stressed that like the menopause, there could be the presence or absence of symptoms. Transitory symptoms can include changes in mood and sexuality. The long-term effects of hypogonadism can result in osteoporosis, muscle atrophy and cognitive changes. Symptomatic hypogonadism is sometimes referred to as the "andropause syndrome."[6]

Symptoms can develop with the andropause syndrome. Our previous study of 302 male subjects revealed that loss of libido and erectile dysfunction (46%), fatigue (41%), and memory loss (36%) were dominant symptoms in that order.[1] The correlation of symptoms to levels of testosterone is very variable and is the subject of on going investigation by us. Most laboratories give a normal range of 260-1000 ng/dl for total testosterone, 50-210 pg/dl for free testosterone, and 66-417 ng/dl for bioavailable testosterone.[7] The SI conversion factor is approximately 35. The range often is not age-adjusted and poses dilemmas for physicians. Patients can have low normal levels and yet display symptoms, which are reversed after androgen supplementation. This suggests the possibility of "relative hypogonadism" in which levels are appropriate for each individual.[8] Another dilemma is that testosterone levels can vary in the course of a day. Frequent sampling of testosterone in a study of 20 normal men revealed ranges between 105 to 1,316 ng/dl between subjects.[9]

## *CLINICAL ASSESSMENT, SCREENING QUESTIONNAIRES, AND CONFOUNDING FACTORS FOR DIAGNOSIS OF ANDROPAUSE*

It is important for the physician to take a careful history and physical examination when assessing a patient for hypogonadism. The physician should inquire about symptoms of loss of libido, and distinguish it from erectile dysfunction. A loss of early morning erection can be indicative of hypogonadism. Fertility issues should be explored.[10] Stress and chronic illness can depress testosterone levels; and so can medications including cimetidine, digoxin, and spironolactone. Diabetes, insulin resistance and obesity have been associated with hypogonadism.[11,12] As such, one should look for symptoms and signs associated with diabetes. A

history of chronic alcoholism must be ascertained as it can suppress the production of androgens. Rare conditions can be associated with hypogonadism, including Prader Willi syndrome, Klinefleters syndrome, and Kallmann's syndrome, and they have to be excluded in the clinical examination. The physical examination should include weight, body mass index, waist/hip ratio, and a measurement of body fat. The skin should be examined for evidence of hyperestrogenism such as spider telangiectasia. The face, axilla, and groin should be inspected for hair loss. Testicular size can be measured using an orchidometer. A prostate examination should also be done as part of the screening.

Screening tools like the ADAM questionnaire can be helpful.[13] Unfortunately, the ADAM questionnaire is sensitive but not specific. Patients with depression and even anemia have been noted to score high on the ADAM questionnaire in the author's practice. Screening for depression can be done with the Geriatric Depression Scale.[14] The Folstein Mini-Mental State Examination can be used to screen for cognitive problems.[15] Unfortunately, memory loss in the andropause can be very subtle and more sophisticated neuropsychological tools may sometimes be needed.[16]

## Different Tests to Measure Testosterone

Many clinicians and laboratories are confused as the correct test to use when determining androgen status in aging men. Although opinion leaders have agreements in some areas as to which test to use, there are also areas of disagreement. This is in part because symptoms of androgen deficiency are not proportional to androgen levels, and also because of the pulsatile nature of secretion of hormones. In any event, it is important for the practitioner to decipher which test to order based on the clinical assessment of the patient.

Most laboratories measure the three domains of testosterone: total testosterone, free testosterone, and bioavailable testosterone. Total testosterone refers to all the testosterone that is measurable, including those bound and unbound portions. Testosterone is bound to proteins like albumin and SHBG. As mentioned, changes in protein concentrations can alter levels true levels and give false impressions. Testosterone is loosely bound to SHBG, and as such comes off easily, making it "free." The actual free amount and that bound to SHBG is referred to as bioavailable testosterone. Laboratories can measure free testosterone using analog ligand radiommunoassay methods or they can sometimes calculate it based on a formula.

In older men, the binding of testosterone to SHBG is increased, making it less likely for it to be released and become free testosterone. As such, total testosterone in older men is much less reliable, and bioavailable testosterone is recommended instead. Bioavailable testosterone represents the "active form" of testosterone, and has a satisfactory correlation with androgenicity. It is also reduced more rapidly than total testosterone and as such approaches a real time view of the androgen status of the patient. Unfortunately, most laboratories charge more for this test, as it is more difficult to perform.

### *Total Testosterone*

Most laboratories use automated machines to determine total amounts of testosterone. To measure total testosterone, these instruments must first displace bound testosterone from SHBG and albumin. Usually low pH buffers, surfactants, salicylates, or a competing steroid that does not bind to anti-testosterone antibody is used in the immonoassay. However, the testosterone antisera used in commercial preparations often cross-react with other steroids, including dihydrotestosterone (DHT). Solvent extraction and chromatography have been used to remove these interfering compounds prior to testosterone measurement. Unfortunately, these cannot be incorporated into the methods used by automated analyzers. Fortunately, the plasma levels of

DHT are only about 10% of testosterone levels and cross reactivity is usually less than 5%. Thus, in clinical application, the impact of DHT is minimal in most instances.

### *"Free" Testosterone*

It has been generally agreed that free testosterone, rather than total testosterone, yields a better measure of androgenicity.[17] However, different laboratories may report free testosterone using various methodologies. It is prudent that the clinician be aware of the different methods of obtaining a free testosterone level, and interpret it in the context of the patient. Testosterone circulates in plasma and binds to SHBG and albumin. Testosterone binding to transcortin and orosomucoid is negligible. Albumin bound testosterone is released into the plasma easily as compared to those bound to SHBG. There are several measures of free testosterone.

**Apparent Free Testosterone as Measured by Equilibrium Dialysis (AFTC):** AFTC or testosterone as determined by equilibrium dialysis at 37°C is arguably the method of choice for measuring free testosterone in vivo. The measurement of free testosterone in the serum is technically demanding, as the free testosterone concentration is very low (2%). Routinely available assays are not sensitive enough to quantify free testosterone directly. Usually, free testosterone is estimated by indirect methods. In these indirect methods, titrated testosterone is added to the sample and allowed to come to equilibrium with testosterone in the serum at a physiological temperature (37°C). The amount of added radiolabeled testosterone must be low enough to guarantee that addition will not significantly increase the total testosterone concentration. Once equilibrium is achieved, the free testosterone is separated from the bound by filtration through a membrane. The filtration is accomplished by equilibrium dialysis or centrifugal ultrafiltration. The radioactivity of the protein-free ultrafiltrate is measured and used to calculate the percentage of free testosterone. The concentration of free testosterone can then be calculated by multiplying the percentage of free testosterone by the total testosterone concentration. Measurement of free testosterone by these methods is not available in most clinical laboratories due to the complicated nature of the testing and the requirement of a scintillation counter to measure the titrated testosterone concentration. Overall, the results of equilibrium dialysis and centrifugal ultrafiltration methods have been shown to be quite comparable. While equilibrium dialysis is often considered the "gold standard," centrifugal ultrafiltration is somewhat simpler to perform and may theoretically be more accurate because the equilibrated sample is not diluted with dialysis buffer.

**Free Androgen Index (FAI):** The concentration of testosterone in the different free and bound forms is really a function of total testosterone concentration and the relative concentrations of SHBG and albumin. It can be predicted that increased SHBG will decrease the concentration of both free and bioavailable testosterone for a given total testosterone concentration. Many clinicians use a calculated free androgen index to estimate physiologically active testosterone. This index is typically calculated as the ratio of total testosterone divided by SHBG and multiplied by 100 to yield numerical results comparable in free testosterone concentration. Otherwise, more complicated mathematical algorithms can be used to approximate the percentage of free testosterone from the SHBG concentration alone or in combination with albumin concentration. The precision of these algorithms is subject to the combined errors of the individual tests carried out.

**Direct Immunoassay of Free Testosterone With a Labeled Testosterone Analog (aFT):** There are several commercial kits available for the direct estimation of free testosterone in serum. These kits use a labeled testosterone analogue that has a low binding affinity for both

SHBG and albumin, but is bound by anti-testosterone antibody. Since the analogue is unbound in the plasma, it competes with free testosterone for binding sites on an anti-testosterone antibody that is immobilized on the surface of the well or assay tube. The first kits developed used a radiolabeled testosterone analogue to compete with free testosterone for binding sites on an antibody-coated polypropylene tube. More recently developed kits employ an enzyme-labeled analogue that can be measured after competitive binding to anti-testosterone antibodies coated to microtiter wells. These analogue methods are technically less demanding than equilibrium dialysis or centrifugal ultrafiltration, and require substantially less blood samples. The analogue methods also offer the benefit of direct estimation of free testosterone concentration without the need to measure total testosterone. Many laboratories can readily perform the enzymatic methods because they are nonisotopic.

Recently, Winters and colleagues have found the analogue method to correlate better with total testosterone levels than with bioavailable testosterone determined by the ammonium sulfate precipitation method.[18] They suggested that the analogue free testosterone results might be misleading in men with low SHBG concentration. Ooi suggested that the problems observed by Winters might, in large part, be resolved simply by using a more appropriate population-based reference interval.[19] Vermeulen and colleagues found that the analogue-free testosterone method correlated well with free testosterone by equilibrium dialysis but did not correspond with a free testosterone calculated from total testosterone and SHBG.[20]

Free testosterone can also be calculated from total testosterone and immunoassayed SHBG (FT).

### *Bioavailable Testosterone (BT)*

BT is the fraction of serum testosterone not precipitated by 50% ammonium sulfate concentration. As in the free testosterone methods described above, titrated testosterone is added to serum that is then allowed to come to equilibrium at physiologic temperature. Testosterone bound to SHBG is then selectively precipitated with 59% ammonium sulfate, leaving free and albumin bound testosterone in the solution. The percentage of titrated label not bound to SHBG is multiplied by the total testosterone to produce the bioavailable testosterone. Another method of measuring bioavailable testosterone is by direct radioimmunoassay in the supernatant after solvent extraction.

Care must be taken to ensure that the labeled tracer testosterone used for measuring the FT fraction is highly purified. In a study by Vermeulen et al, AFTC was correlated against the other measures of testosterone. A coefficient of correlation of 1.0 would mean a perfect correlation. In that study with men, they found that the correlation of AFTC with FT (calculated free testosterone) = 0.987, aFT (immonoassayed free testosterone) = 0.937, FAI (free androgen index) = 0.848. In other words, calculated free testosterone approaches the accuracy of measuring testosterone by dialysis equilibrium. It has to be noted that conditions that alter SHBG may alter the results of not only total testosterone but also FT. In men, conditions like obesity, hypothyroidsim, and acromegaly can lead to lowered levels of SHBG, and as such confound the results of FT. Incidentally, pregnancy also leads to altered levels of SHBG, and as a result leads to false levels of FT as well. Otherwise, calculated free testosterone (FT) may be a practical means for the clinician to measure free testosterone, as it is less time consuming and expensive than testosterone by equilibrium dialysis (AFTC). Bioavailable testosterone is a more expensive test, but will be more useful and accurate in older patients as SHBG binding increases with age, and BT measures only the free amounts and those loosely bound to albumin. In older patients, one often finds normal levels of total testosterone, but BT is often significantly depressed.

### *Salivary Testosterone*

This form of testing is novel, especially for the patient, for whom this method is often preferred as it avoids a needle stick. It is also convenient for the patient, as an in-office appointment is not necessary. The patient spits into a bottle and mails the sample to the laboratory. This test may be useful for screening for hypogonadism, but is not suitable for diagnosis of the andropause syndrome. The history and physical examination should be weighted more than salivary tests, or any other blood tests.

Saliva testosterone does not give a real-time assessment of the androgen status, and as such, patients on androgen therapy often have levels of testosterone in the thousands. It gives a picture of accumulated testosterone. However, when done properly, saliva testosterone has good correlation to free testosterone (r=0.90), less for total testosterone (r=0.85). The correlation for DHEA is less at 0.70, and androstenedione 0.74.

The patient has to rinse his mouth 5 minutes before testing, and avoid food and tooth brushing 30 minutes. He chews on a gum, spits into a bottle, and mails the specimen to the laboratory.

### *Dynamic Testing for Testosterone*

- **HCG Stimulation Test** – HCGor Human chorionic gonadotrophin is a glycoprotein with similar physiological actions like LH. After an intramuscular injection of HCG, the hormone binds to the LH receptors in the Leydig cells, and stimulates the production and secretion of testosterone. Typically, the test dose is 4000 IU for 4 days. This test is used to assess the viability of the axis and whether there is a gonadal disease. A positive response usually results in doubling of testosterone levels and improvement in symptoms. An alternate dosing is 5000 IU and measuring testosterone levels 3 days later. Obviously, if there is no response, it is indicative of testicular failure, and if there is a response, it is indicative of pituitary-hypothalamic failure.[21]
- **Clomiphene Citrate Test** – Clomiphene is a non-steroid oral compound with estrogenic effects. It binds to estrogenic receptors in the body, and the hypothalamus responds by secreting LH. The test dose is 50-100mg bid for 10 days, and both testosterone and LH levels are measured. By and large, healthy men have a 50-200% increase in LH and testosterone levels. Obviously, if there is no response, it is indicative of testicular failure and if there is a response, it is indicative of pituitary-hypothalamic failure. [21]

## Measuring LH and FSH in Aging Men

Luteinizing hormone (LH) and Follicular Stimulating Hormone (FSH) are glycoprotein gonadotrophins composed of alpha and beta subunits secreted by the same cell. The biologic activity of HCG, which is a glycoprotein from the placenta, resembles that of LH. LH and FSH bind to receptors in the testes and regulate gonadal function by promoting sex steroid production and gametogenesis. LH stimulates testosterone production from the interstitial cells of the testes (Leydig cells). Maturation of spermatozoa requires both LH and FSH. The secretion of LH and FSH is episodic with secretory bursts every hour and are mediated by a concomitant episodic gonadotrophin releasing hormone (GnRH) release. Testosterone is not the sole inhibitor of gonadotrophin secretion in men as selective destruction of the testes by chemotherapy results in azospermia and a rise in FSH only. Inhibin, which is secreted by the Sertoli cells of the seminiferous tubules, is the major factor that inhibits FSH secretion by negative feedback.

Typically, LH rises with aging and there is a concomitant drop in bioavailable testosterone levels. However, it is to be noted that the rise in LH seems to be inconsistent in aging men. In younger men, gonadal failure leads to fairly consistent rises in LH, partly because the hypothalamic-piutitary-testicular axis is intact. With aging, this system is impaired and results in inconsistent rises. The disparity of low bioavailable testosterone and yet normal or low LH is consistent with a hypothesis of a relative hypogonatropism, often seen in the andropause syndrome.

Urban et al reported that some healthy older men exhibited evidence of neuroendocrine dysfunction, reflected by irregular bursts of bioactive LH release followed by transiently low plasma bioactive/immunoreactive (B/I) ratios.[22] It has been reported that mean basal plasma bioactive LH concentrations, B/I ratios, and spontaneous LH pulse properties (peak, frequency, amplitude, duration, and enhanced B/I ratios within LH peaks) were not altered in older men. On the other hand, augmentation of bioactive LH secretion and enhancement of bioactive LH and enhancement of plasma B/I ratios by pulsed injections of exogenous GnRH were significantly reduced or absent in older men. In that study, authors conclude that bioactive LH reserve is markedly attenuated in older men challenged with either exogenous GnRH or antiestrogens. In another study by Veldhius et al, they explored an andropause state using ketoconazole, which is an anti androgen.[23] This study found a discrepancy between young and older men. In young men, the chemically induced andropausal state resulted in an elevated LH peak frequency, whereas there was a reduced incremental LH pulse area in older men. This study also supported the earlier study in that older men had an impoverished augmentation of LH pulse mass, impaired orderliness of LH release, and diminished 24 hour rhythmic LH secretion. Mitchell et al also reported that there were age related changes in the pituitary-testicular axis in normal men.[24] However, they pointed out in their study that immunoreactive LH remained unchanged despite finding that levels of total testosterone and bioactive LH fell with age. The important lesson from this study is that as men age, there is a hypothalamo-pituitary defect, which in turn leads to lower bioactive LH levels, which in turn is responsible for diminished gonadal steroidogenesis. In yet another study by Veldhius et al, they found that age was a negative determinant of LH secretory burst amplitude and a positive predictor of LH secretory burst frequency as well as basal LH secretory rates.[25] They suggested that the attenuation of LH secretory burst amplitude as an approximate basis for hypoandrogenism of healthy aging in older men.

Should the clinician measure LH levels to determine if the patient has indeed undergone andropause? The LH surge in women undergoing menopause is not seen in men. If the LH levels were very high in older men, a pituitary cause for hypogonadism must be excluded. Often, men with andropausal symptoms have LH levels within the normal range (0.5 -15 ng/dl), but have low bioavailable testosterone. As such, the use of LH is more useful in excluding diagnosis of other pathological states. When ordering LH, the clinician should distinguish between bioavailable and immunoreactive LH. Some laboratories are not able to provide both tests, and do only the immunoreactive LH. Overall, LH tends to be secreted in spurts as well, and it is difficult to rely on single values. FSH is the gonadotrophin that stimulates spermatogenesis in the Sertoli cells. It has been suggested that FSH is less pulsatile, and that it may be a better measure of gonadal failure as a result of aging in men. This is yet to be verified in clinical trials.

## Measuring Estrogens in Men

In recent years, the role of estrogens in men have become more important because of the discovery of human models of estrogen deficiency such as estrogen resistance or aromatase deficiency.[26] In men, testosterone remains the major source of plasma estradiol. The main biologically active estrogen is estradiol. 20% of men's estradiol is secreted by the testes. On the other hand, plasma estrone (5% of which is converted to plasma estradiol), originates from tissue aromatization of, mainly adrenal, androstenedione. The plasma concentration of estradiol in older men is about 20-30 pg/ml and its production rate in blood is 25-40 micrograms/24 h. It is interesting to note that both of these values are actually significantly higher than in postmenopausal women. Plasma levels of estradiol do not necessarily reflect tissue-level activity as peripherally formed estradiol is partially metabolized *in situ*. Not all enters the general circulation, with a fraction remaining only locally active. Of the factors influencing plasma estradiol levels, plasma testosterone remains most important. However, the age-associated decrease in testosterone levels is scarcely reflected in plasma estradiol levels, as a result of increasing aromatase activity with age and the age-associated increase in fat mass. Free and bioavailable estradiol levels do decrease modestly with age, as does the ratio of free testosterone to free estradiol. Estradiol levels are positively related to body fat mass and more specifically to subcutaneous abdominal fat, but not to visceral (omental) fat. Indeed, aromatase activity in omental fat is only one-tenth of the activity in gluteal fat. Estrogens in men play an important role in the regulation of the gonadotropin feedback, cognitive functions, bone maturation, regulation of bone resorption and also lipid metabolism. Estrogens also affect skin metabolism and are important determinants of sexual interest in men.

## *CONCLUSIONS*

It is important for the practitioner to be certain of a diagnosis of low testosterone causing symptoms before instituting replacement therapy. Without question, androgen supplementation in the form of testosterone may serve to be an effective for some individuals who are symptomatic. Prohormones like DHEA, androstenedione, and progesterone are weaker androgens and have been studied much less than testosterone. In fact, most of the knowledge of these androgens is based on animal models. At this point, there is no routine recommendation for androgen replacement in all males. The practitioner should select his patient carefully based on symptoms and biochemical evidence of hypogonadism. Sometimes, a therapeutic trial may be needed for diagnosis. Alternatively, the more cumbersome 24-hour urinary androgens may give a better picture of a true androgen state. After starting therapy, monitoring of side effects of androgens including prostate, hemoglobin etc. is mandatory.

## *REFERENCES*

1. Tan RS, Philip P. Perception and risk factors for the andropause. *Arch Androl.* 1999;43:97-103.
2. Smith KW, Feldman HA, McKinlay JB. Construction and field validation of a self-administered screener for testosterone deficiency (hypogonadism) in aging men. *Clin Endocrinol*. 2000;53:703-711.
3. Tenover JS. Androgen administration to aging men. *Endocrinol Metab Clin North Am.* 1994;23:877-892.
4. Sih R, Morley JE, Kaiser FE, et al. Testosterone replacement in older hypogonadal men: a 12-month randomized controlled trial. *J Clin Endocrinol*. 1997;82:1661-1667.
5. Basaria S, Dobs AS. Hypogonadism and androgen replacement therapy in elderly men. *Am J Med*. 2001;110:563-572.
6. Morales A, Heaton JP, Carson CC. Andropause: a misnomer for a true clinical entity. *J Urol*. 2000;163:705-712.
7. DeGroot LJ, Jameson JL. *Endocrinology*. 4th edition, New York: WB Saunders; 2001.
8. Tan RS. Andropause: introducing the concept of "relative hypogonadism" in aging males. *Int J Impot Res*. 2002;14:319.
9. Spratt DI, O'Dea LS, Crowley WF et al. Neuroendocrine-gonadal axis in men: frequent sampling of LH, FSH and testosterone. *Am J Physiol.* 1998;254:658-666.
10. Gallardo E, Guanes PP, Simon C, et al. Effect of age on sperm fertility potential: oocyte donation as a model. *Fertil Steril.* 1996;66:260-264.
11. Marin P, Holmang S, Jonsson L, et al. The effects of testosterone treatment on body composition and metabolism in middle-aged obese men. *Int J Obe*. 1992;16:991-997.
12. Ukkola O, Gagnon J, Rankinen T, et al. Age, body mass index, race and other determinants of steroid hormone variability: the HERITAGE Family Study. *Eur J Endocrinol*. 2001;145:1-9.
13. Morley JE, Charlton E, Patrick P, et al. Validation of screening questionnaire for androgen deficiency in aging males. *Metabolism* 2000;49:1239-1242.
14. Sheikh JI, Yesavage JA. Geriatric Depression Scale: recent evidence and development of a shorter version. *Clin Gerontol*. 1986;5:165-172.
15. Folstein MF, Folstein SE, McHugh PR. Mini-mental state: a practical method for grading the cognitive state of patients for clinician. *J Psychiat.* 1975;12:189-198.
16. Tan RS, Pu SJ. The andropause and memory loss: is there a link between androgen decline and dementia in the aging male? *Asian J Androl.* 2001;9:169-174.
17. Valcour A. Testosterone: Free T or not Free T. *Advance Laboratory* 2001;11:3-10.
18. Winters SJ, Kelly DE, Goodpaster B. The analog free testosterone assay: Are the results in men clinically useful? *Clin Chem*. 1998;44:2178-2182.
19. Ooi DS, Donnely JG. More on the analog free testosterone assay. *Clin Chem*. 1999;45:715.
20. Vermeulen A, Verdonck L, Kaufmann JM. A critical evaluation of simple metods for the estimation of free testosterone in serum. *J Clin Endocrinol Metab*. 2001;86:2903.
21. Greenspan FS, Gardener DG. In: *Basic and Clinical Endocrinology*. 6th edition. New York: Lange Medical Books; 2001.
22. Spratt DI, O'Dea LS, Schoenfiled D, et al. Neuroendocrine-gonadal axis in men: frequent sampling of LH, FSH and testosterone. *Am J Physiol*. 1988;254:E658-666.
23. Morley JE, Patrick P, Perry HM. Evaluation of assays to measure free testosterone. *Metabolism* 2002;51:554-559.
24. Urban RJ, Veldhius JD, Blizzard RM, et al. Attenuated release of biologically active luteinizing hormone in healthy aging men. *J Clin Invest.* 1988;81:1020-1029.
25. Veldius JD, Urban RJ, Lizarralde G, et al. Attenuation of luteinizing hormone secretory burst amplitude as a proximate basis for the hypoandrogenism of healthy aging men. *J Clin Endocr Metab.* 1992;75:704-706.
26. Vermeulen A, Kaufman JM, Goemaere S, et al. Estradiol in elderly men. *Aging Male*. 2002;5:98-102.

### Author/Article Information

Dr Robert S Tan is Geriatrician and Associate Professor in the Department of Family Medicine at the University of Texas in Houston, and Medical Director of Garden Terrace Alzheimer's Center. He is Fellow of the American Geriatrics Society and Founding Board Member of International Society for Men's Health. He has published more than 80 scientific articles and is author of the book "*The Andropause Mystery.*" Peers selected him to "*Best Doctors in America.*"

*Correspondence*: Phone: (+1) 713-500-7580 E-mail: robert.s.tan@uth.tmc.edu.