American Journal of Men's Health
November-December 1–13

DOI: 10.1177/1557988320977991
journals.sagepub.com/home/jmh
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The Epidemiological Characteristics of Late-Onset Hypogonadism in Chinese Middle-Aged and Elderly Men: Two Cross-Sectional Studies in the Same Community

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Abstract

The purpose of this study was to investigate the prevalence and epidemiological characteristics of late-onset hypogonadism (LOH) in middle-aged and elderly Chinese men. Two cross-sectional studies were conducted at 5-year intervals in community-dwelling men living in the same area. A total of 1472 (Study 1, S1) and 944 (Study 2, S2) men aged 40–69 years old were recruited as subjects. Subjects were evaluated through combining serum reproductive hormone levels with the Androgen Deficiency in Aging Males (ADAM) questionnaire and the Aging Males' Symptoms (AMS) scale. A significant difference was found in mean testosterone deficiency (TD) prevalence between S1 and S2, using either serum total testosterone (TT; 14.02% vs. 6.36%) or serum calculated free testosterone (cFT; 43.69% vs. 16.53%) cutoff values. According to the S1 or S2 data, the mean prevalence of LOH was 37.85%/15.47% in the positive ADAM test and 15.42%/9.43% in the positive AMS test ($p < .01$). According to classifications of TD based on gonadal status, the prevalence of secondary TD (27.34%) was higher than the primary (16.36%) and compensated (15.42%) TD in S1 ($p < .01$). However, there were significant differences among the prevalence of primary (6.89%), secondary (9.64%), and compensated (27.65%) TD in S2 ($p < .05$). Different types of testosterone levels, TD cutoff values, and questionnaires influenced the prevalence of TD and LOH. The serum FT cutoff value was an optimal threshold for evaluating and diagnosing TD and LOH, whose prevalence increased gradually with male aging.

Keywords

aging, hypogonadism, epidemiology, male, prevalence, testosterone

Received May 5, 2020; revised November 7, 2020; accepted November 11, 2020

Typically, cross-sectional studies have been used to investigate late-onset hypogonadism (LOH). However, only a few studies have used long-term, regular follow-up, longitudinal, or cohort studies either domestically or overseas (Feldman et al., 2002; Harman et al., 2001; Wu et al., 2010). Several studies on LOH administrated by European and American scholars frequently focused on a particular disease, or involved interdisciplinary studies moreover focused more on epidemiological data featuring a unique experimental design, including distinct characteristics of populations, assessment methods of symptoms, levels of reproductive hormones, specific cutoff values of testosterone deficiency (TD), TD prevalence, and prevalence and incidence of LOH (Araujo et al., 2004; Harman et al., 2001; Wu et al., 2010).

The Baltimore Longitudinal Study on Aging (BLSA) recruited 890 men aged 22.5–91.3 years (58.8 ± 15.8 years). This was the first international report of TD cutoff value, TD prevalence, and descent rate of serum testosterone (T) concentration. This landmark study put forward a new method and direction on intensive studies of LOH (Harman et al., 2001). The Massachusetts Male Aging Study (MMAS) was one of most representative epidemiological investigations on the health status of middle-aged and elderly male populations aged 40–70

years and was one of several longitudinal cohort studies on LOH that formulated a diagnostic standard of LOH based on related symptoms and TD. Researchers calculated the prevalence and incidence of LOH in both the baseline data and the follow-up data. This approach had the advantages of detailed and reliable data, lacking in other studies (Araujo et al., 2004; Feldman et al., 2002; O'Donnell et al., 2004). The European Male Aging Study (EMAS) was another longitudinal clinical study conducted after the MMAS study incorporating a large sample size (Wu et al., 2010). This study employed age-stratified and random sampling to investigate subjects in the general population. In this study, there were 3369 men aged 40–79 years (mean age, 59.7 years) at eight European centers. The LOH prevalence was 2.1% along with a prevalence presented ascent trend with aging in the EMAS. The EMAS was an evidence-based and general population cohort study on LOH, whose aim was to identify the thresholds of serum testosterone levels assumed to be specific for TD and symptoms, and to define essential criteria for LOH (Corona et al., 2013).

An innovative classification of TD was introduced on the basis of the EMAS (Tajar et al., 2010). The method of classification was adopted and recommended by the guidelines of the European Association of Urology (EAU; Salonia et al., 2020). Among male subjects aged 40–79 years old, different categories were evaluated by combining luteinizing hormone (LH) with T and without regard to LOH symptoms, such that 11.8%, 2.0%, and 9.5% were classified into the secondary, primary, and compensated TD categories, respectively.

The number of Chinese LOH patients may be prevalent due to the highly abundant aging population; therefore, it was required to conduct a series of detailed and accurate studies on the status of LOH prevalence in Chinese men. However, several studies on LOH in Chinese subjects over the past decade still had some shortcomings. For example, statistical analyses of data were lacking as well as reports on classification of TD (He et al., 2012; Li et al., 2005; Liu et al., 2016; Sun et al., 2012; Wu et al., 2013; Zhou et al., 2010).

Two community population-based cross-sectional studies were performed over the same area during a 5-year interval, in order to investigate reproductive health status, and prevalence of TD and LOH on Chinese middle-aged and elderly males. The first study (Study 1, S1) was a preliminary trial concerning an epidemiological investigation of LOH. This study investigated the prevalence of LOH in the local area while validating research methods. The second study (Study 2, S2) was a part of a nationwide, multicenter trial on the reproductive health status of middle-aged and elderly men. The two studies performed in the same area by skilled investigators in S1 allowed for a smooth implementation of S2. Data were analyzed between the two studies. The strength of this report is that the two studies were carried out on TD and LOH from the same area in order to provide important information, as well as advantages and disadvantages for future studies.

Materials and Methods

Study Design

Two studies were conducted in the same area at 5-year intervals. These studies were cross-sectional surveys of 1560 (Study 1, S1) and 1200 (Study 2, S2) community-dwelling Chinese men aged 20–69 years and 20–89 years respectively, selected via cluster and age-stratified sampling, and residing in Fucheng County, Hebei provinces. Each survey identified a local population register to provide a sampling frame from which participants

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were randomly selected. Participants were recruited for each study between August 2007 and November 2008 (S1) and between July 2013 and January 2014 (S2) after obtaining written informed consent. The two groups of participants were representative for the regional population in each year.

Participants

Participants were invited to attend a local reproductive health service facility to complete interviewer-assisted questionnaires and to undergo a general physical examination, including height and weight measurements, and blood tests for biochemical and hormone levels. Since some diseases or treatments may influence reproductive hormone levels, the exclusion criteria were defined as follows: (1) previously or currently diagnosed malignancies, corticosteroid use, or presence of liver cirrhosis; (2) testosterone supplement, or androgen-deprivation therapy use, 5-α reductase inhibitor treatment, or history of orchiectomy; and (3) current hypothalamus-pituitary disease, acute cardiovascular disease, or acute or chronic renal failure. In order to compare the data of same age groups and after screening, 1472 (S1) and 944 (S2) participants aged 40–69 years were included in the data analysis and taken as subjects who filled out valid questionnaires. Fifty-nine (S1) and 98 (S2) participants aged 20–39 years were taken as control groups after one (S1) and two (S2) unqualified participants were excluded.

Questionnaires

Each participant filled in a questionnaire that included information concerning sociodemographic and general health status, lifestyle, medical conditions, medications, and two assessment questionnaires for LOH (ADAM—the Androgen Deficiency in Aging Males questionnaire and AMS—the Aging Males' Symptoms scale). When a positive result of ADAM was obtained or an AMS score of ≥ 27, the LOH symptoms were considered present. The positive rates of LOH symptoms assessment were evaluated by counting the positive results of questionnaires of subjects aged 40–69 years. Alcohol consumption was defined as one or more alcoholic drinks per week, including beer, wine, and spirits. Smoking status was classified as never/ex-smokers or current smokers.

Clinical and Laboratory Measurements

A single fasting venous blood sample was obtained from each participant in the morning (before 9 am), and the serum was stored in aliquots at −70 °C until the time of the assay. Serum samples were measured together in batches in the central laboratory of the Beijing Coordinating Center (China).

Serum Measurements of S1. Only 428 sequential men of 1472 subjects and 59 participants taken as controls were recruited to measure the concentration of serum reproductive hormones, because the budget for purchasing kits was limited during S1 put into practice. The concentration of TT and LH were measured with the MPAIA kit from Beijing Bio-Ekon Biotechnology Co. Ltd (Beijing, China). The sensitivity of the TT and LH kits was 0.3 nmol/L, and 0.2 IU/L, respectively. The coefficients of variation (CV) of intra-assay were 1.8%, and 4.9%, respectively. The inter-assay CV was less than 8.6%. Serum sex hormone-binding globulin (SHBG) was measured with an ELISA kit from Diagnostic Systems Laboratories, Inc (Dallas, Texas, USA). The sensitivity of the kit was 0.61 nmol/L, and the intra- and inter-assay CVs were 6.67%, and 9.78%, respectively. The proportion of serum calculated free testosterone (cFT) was calculated using the equation described by Vermeulen et al (Vermeulen et al., 1999).

Serum Measurements of S2. Serum TT, SHBG, and LH concentrations in 944 subjects and 98 participants taken as controls were measured using a Beckman UniCel DXI800 automatic chemiluminescence immune analyzer (Beckman Coulter, Fullerton, California, USA). The calculation method of cFT was the same as for S1. The lower limits of the TT, SHBG, and LH levels were 0.35 nmol/L, 0.017 nmol/L, and 0.2 IU/L, respectively. The intra-assay CVs for TT, SHBG, and LH were 2.7%, 4.8%, and 3.8%, respectively. The mean inter-assay CVs for TT, SHBG, and LH were 5.6%, 5.3%, and 6.4%, respectively.

The Definitions of TD or LOH

TD was defined in our two studies as serum T concentration of subjects lower than the cutoff value, irrespective of subjects with or without LOH symptoms. LOH was defined as the presence of LOH symptoms (i.e., positive results of ADAM or AMS) in combination with the serum cFT levels measuring lower than the cFT cutoff value.

Classification Definitions of TD Based on Gonadal Status

Based on published literature, four classifications of gonadal status were defined as eugonadal (normal T and normal LH), secondary (low T and low/normal LH), primary (low T and elevated LH), and compensated (normal T and elevated LH) TD (Tajar et al., 2010). The setting methods of T and LH cutoff values were similar to TD and LOH diagnosis.

The Definitions for True Positive and True Negative of the Diagnostic Test Used to Calculate the Sensitivity and Specificity of the Questionnaires

The sensitivity and specificity of ADAM and AMS were validated using the TT and cFT cutoff values to evaluate the accuracy of questionnaires assessed subjects with or without TD. Setting the serum T levels as golden standard, true positive of the diagnostic test was defined as serum T concentration of subjects lower than the cutoff value, and with the positive results of questionnaires as well. True negative of the diagnostic test was defined as serum T concentration of subjects equal or higher than the cutoff value, and with the negative results of questionnaires as well.

Statistical Analyses

The data obtained from the two studies were analyzed using SPSS21.0 (International Business Machines Corp., Armonk, New York, USA). Due to the skewed distributions of hormones, the data were analyzed using nonparametric statistics. Data representing the characteristics of the subjects were represented using percentile. Hormone levels of the two groups were compared using the Mann–Whitney U test on two sets of data. Positive rates of LOH symptoms assessment, TD prevalence, LOH prevalence, and other rates of different age groups and different evaluation methods were compared using the Chi-square (χ^2) test. Results were considered statistically significant if null hypotheses could be rejected at the .05 level.

Results

Subject Characteristics

Characteristics of the subjects and controls are presented in Table 1. Regarding the subjects, the median age, waist circumference (WC), and waist to hip ratio (WHR) of the subjects were significantly lower in S1 compared to S2. However, median height and weight were notably higher in S1 compared to S2. There was a significant difference in hormone levels between 428 subjects of S1 and 944 subjects of S2. For example, TT, cFT, and LH levels were significantly lower, while SHBG levels was significantly higher in S1 compared to S2. Regarding the controls, the median age, TT, and SHBG levels of the controls were significantly lower in S1 compared to S2.

Serum Testosterone Cutoff Values of TD and LOH Diagnosis

The detailed data of serum reproductive hormones levels were published in previous literature (Zhou et al., 2020). The serum reproductive hormone levels [50% percentile (10%–90% percentile)] of controls (20–39 years group) in S1 and S2 showed 17.11 (9.13–29.34) versus 13.83 (9.35–20.55) nmol/L for TT, 286.00 (169.00–490.00) versus 310.00 (213.90–454.30) pmol/L for cFT, and 2.82 (1.55–5.08) versus 2.98 (1.74–6.18) IU/L for LH, respectively. The data are presented in Table 1. Mann–Whitney U tests identified that, except for serum cFT and LH levels in the control group, there was significant difference in serum TT and SHBG levels between S1 and S2 in controls ($p < .01$). Setting 10% percentile of the serum hormone levels of controls (20–39 years group) as the cutoff value (Li et al., 2005), the relative cutoff values of TD in S1 and S2 were 9.13 nmol/L and 9.35 nmol/L for serum TT and 169.00 pmol/L and 213.90 pmol/L for serum cFT, respectively.

Positive Rates of LOH Symptoms Assessment

The questionnaires (ADAM or AMS) were used to assess subjects. The positive rates ranged from 55.73% to 97.05% in S1, and from 78.57% to 96.93% in S2 when the ADAM was used to assess subjects. Positive rates ranged from 9.51% to 63.71% in S1, and from 29.05% to 63.43% in S2 when the AMS was used to assess subjects. The rates increased gradually with male aging ($p < .01$). Using the ADAM questionnaire, the mean positive rates of LOH symptoms assessment were 80.77% in S1 and 90.89% in S2, respectively. Using the AMS questionnaire, the mean positive rates were 32.34% in S1 and 48.83% in S2, respectively. The mean positive rates of ADAM assessment were higher than the mean rates of AMS assessment, either in S1 ($\chi^2 = 702.80$, $p < .01$) or in S2 ($\chi^2 = 396.48$, $p < .01$). The data are presented in Table 2.

The Prevalence of TD

When the serum cFT cutoff value was used, the TD prevalence increased gradually with aging, ranging from 30.30% to 57.35% in S1 and from 5.24% to 21.23% in S2. However, when the serum TT cutoff value was used, the TD prevalence did not show an increasing trend with aging. χ^2 tests indicated that there were significant differences in the prevalence of TD among the three age groups examined using the serum cFT cutoff value ($p < .01$). The mean TD prevalence that used the cFT cutoff was higher than the TT cutoff (43.69% vs. 14.02% in S1, $\chi^2 = 91.78$, $p < .01$; 16.53% vs. 6.36% in S2, $\chi^2 = 48.18$, $p < .01$). In addition, a significant difference was found in the mean TD prevalence between S1 and S2, using either serum TT (14.02% vs. 6.36%, $p < .01$) or serum cFT (43.69% vs. 16.53%, $p < .01$) cutoff values. The TD prevalence is presented in Table 2.

Table 1. Characteristics of the Subjects and Controls.

	Subjects				Controls		
	S1 (n_1 = 1472)#	S1 (n_2 = 428)#	S2 (n = 944)		S1 (n = 59)	S2 (n = 98)	
Variables	50% (10%, 90%)	50% (10%, 90%)	50% (10%, 90%)	*p*	50% (10%, 90%)	50% (10%, 90%)	*p*
Age (years)	55.00 (40.00, 69.00)	55.00 (41.00, 68.00)	58.00 (45.00, 67.00)	.000*	29.00 (25.00, 36.00)	30.50 (26.00, 38.00)	.035
Height (cm)	169.00 (161.00,175.00)	168.00 (160.00, 175.00)	166.00 (159.00, 174.00)	.000*	170.00 (161.00, 176.00)	170.00 (161.60, 178.80)	.687
Weight (kg)	70.00 (58.00,85.00)	70.00 (59.00, 85.00)	69.00 (56.00, 84.00)	.001*	71.00 (61.00, 85.00)	70.00 (55.00, 85.40)	.425
BMI (kg m^{-2})	24.75 (20.96, 29.07)	25.15 (21.26, 29.05)	24.97 (20.76, 29.41)	.291*	24.66 (21.19, 30.47)	24.16 (19.23, 29.54)	.332
WC (cm)	90.00 (77.00, 102.00)	90.00 (77.00, 103.00)	91.00 (76.00, 103.00)	.047*	90.00 (74.80, 103.40)	87.00 (70.60, 97.40)	.468
HC (cm)	97.00 (87.00, 107.00)	98.00 (88.00, 109.00)	97.00 (89.00, 106.00)	.958*	99.00 (89.40,111.40)	96.00 (87.00, 106.00)	.132
WHR (cm/cm)	0.93 (0.85, 0.98)	0.92 (0.83, 0.98)	0.93 (0.85, 1.01)	.035*	0.89 (0.80, 0.97)	0.90 (0.80, 0.97)	.511
TT (nmol/L)	–	13.24 (8.50, 24.18)	15.32 (10.20, 22.89)	.000**	17.11 (9.13, 29.34)	13.83 (9.35, 20.55)	.006
cFT (pmol/L)	–	177.14 (101.84, 332.08)	390.00 (260.00, 610.00)	.000**	286.00 (169.00, 490.00)	310.00 (213.90, 454.30)	.147
SHBG (nmol/L)	–	62.91 (31.24, 123.77)	39.45 (21.70, 67.50)	.000**	41.92 (23.36, 71.98)	26.45 (12.78, 49.00)	.000
LH (IU/L)	–	3.99 (1.77, 7.82)	5.03 (2.59, 9.46)	.000**	2.82 (1.55, 5.08)	2.98 (1.74, 6.18)	.162

Note. BMI = body mass index; WC = waist circumference; HC = hip circumference; WHR = waist to hip ratio; TT = total testosterone; cFT = calculated free testosterone; SHBG = sex hormone-binding globulin; LH = luteinizing hormone.

#Only 428 sequential men (n_2) of 1472 subjects (n_1) were recruited to measure the concentration of serum reproductive hormones in S1.

*The difference between S1 (n_1) and S2.

**The difference between S1 (n_2) and S2.

Table 2. The Prevalence of LOH Symptoms Assessment, TD, and LOH in Different Age Groups, *n*/*N* (%).

Variables	Study	Age Group of Subjects				*p*	
		40–49 Years	50–59 Years	60–69 Years	Mean	Age Groups*	Mean**
Positive rate of LOH symptoms assessment (ADAM)	S1	287/515 (55.73)	442/483 (91.51)	460/474 (97.05)	1189/1472 (80.77)	$\chi^2 = 324.71$, $p < .01$	$\chi^2 = 45.48$, $p < .01$
	S2	165/210 (78.57)	314/343 (91.55)	379/391 (96.93)	858/944 (90.89)	$\chi^2 = 55.90$, $p < .01$	
Positive rate of LOH symptoms assessment (AMS)	S1	49/515 (9.51)	125/483 (25.88)	302/474 (63.71)	476/1472 (32.34)	$\chi^2 = 345.07$, $p < .01$	$\chi^2 = 65.94$, $p < .01$
	S2	61/210 (29.05)	152/343 (44.31)	248/391 (63.43)	461/944 (48.83)	$\chi^2 = 69.03$, $p < .01$	
The prevalence of TD (TT cutoff)	S1	17/132 (12.88)	24/160 (15.00)	19/136 (13.97)	60/428 (14.02)	$\chi^2 = 0.27$, $p > .05$	$\chi^2 = 21.66$, $p < .01$
	S2	14/210 (6.67)	27/343 (7.87)	19/391 (4.86)	60/944 (6.36)	$\chi^2 = 2.83$, $p > .05$	
The prevalence of TD (cFT cutoff)	S1	40/132 (30.30)	69/160 (43.13)	78/136 (57.35)	187/428 (43.69)	$\chi^2 = 19.96$, $p < .01$	$\chi^2 = 115.91$, $p < .01$
	S2	11/210 (5.24)	62/343 (18.08)	83/391 (21.23)	156/944 (16.53)	$\chi^2 = 26.26$, $p < .01$	
The prevalence of LOH (ADAM+, cFT cutoff)	S1	25/132 (18.94)	61/160 (38.13)	76/136 (55.88)	162/428 (37.85)	$\chi^2 = 38.87$, $p < .01$	$\chi^2 = 84.76$, $p < .01$
	S2	8/210 (3.81)	57/343 (16.62)	81/391 (20.72)	146/944 (15.47)	$\chi^2 = 30.42$, $p < .01$	
The prevalence of LOH (AMS+, cFT cutoff)	S1	6/132 (4.55)	11/160 (6.88)	49/136 (36.03)	66/428 (15.42)	$\chi^2 = 65.22$, $p < .01$	$\chi^2 = 10.55$, $p < .01$
	S2	3/210 (1.43)	30/343 (8.75)	56/391 (14.32)	89/944 (9.43)	$\chi^2 = 26.89$, $p < .01$	

Note. LOH = late-onset hypogonadism; TD = testosterone deficiency; ADAM = the Androgen Deficiency in Aging Males questionnaire; AMS = the Aging Males' Symptoms scale; TT = total testosterone; cFT = calculated free testosterone.

*The difference among the rates of three age groups.

**The difference between the mean rate of S1 and the mean rate of S2.

Table 3. The Classifications and Rates of TD Based on Gonadal Status, *n*/*N* (%).

Variables of Gonadal Status	Studies		
	S1	S2	*p**
The rates of normal or eugonadal	175/428 (40.89)	527/944 (55.83)	$\chi^2 = 26.31$, $p < .01$
The rates of secondary TD	117/428 (27.34)	91/944 (9.64)	$\chi^2 = 71.73$, $p < .01$
The rates of primary TD	70/428 (16.36)	65/944 (6.89)	$\chi^2 = 29.76$, $p < .01$
The rates of compensated TD	66/428 (15.42)	261/944 (27.65)	$\chi^2 = 24.26$, $p < .01$

Note. TD = testosterone deficiency.
*The difference between the rate of S1 and the rate of S2.

The Prevalence of LOH

According to subjects' positive results of the ADAM, the prevalence of LOH ranged from 18.94% to 55.88% in S1, and from 3.81% to 20.72% in S2. According to subjects' positive results of the AMS, the prevalence of LOH ranged from 4.55% to 36.03% in S1, and from 1.43% to 14.32% in S2. The prevalence of LOH increased gradually with male aging ($p < .01$), and the mean rates of LOH in S1 were higher than the mean rates in S2 ($p < .01$). According to S1, the mean prevalence of LOH was 37.85% in positive for the ADAM questionnaire and 15.42% in positive for the AMS questionnaire ($\chi^2 = 55.10$, $p < .01$). According to S2, the mean prevalence of LOH was 15.47% in positive for the ADAM questionnaire and 9.43% in positive for the AMS questionnaire ($\chi^2 = 15.79$, $p < .01$). The data are presented in Table 2.

Classifications of TD Based on Gonadal Status

The chosen cFT cutoff value of 169.00 (S1) and 213.90 (S2) pmol/L was similar to that used in the TD and LOH diagnosis. The LH cutoff, corresponding to the 90% percentile (the upper limit of normal) value in the control group (20–39 years old), was 5.08 IU/L (S1) and 6.18 IU/L (S2). Subjects were differentiated into the four classifications of normal or eugonadal (S1: cFT $\geq$ 169.00 pmol/L and LH $\leq$ 5.08 IU/L; S2: cFT $\geq$ 213.90 pmol/L and LH $\leq$ 6.18 IU/L), secondary TD (S1: cFT $<$ 169.00 pmol/L and LH $\leq$ 5.08 IU/L; S2: cFT $<$ 213.90 pmol/L and LH $\leq$ 6.18 IU/L), primary TD (S1: cFT $<$ 169.00 pmol/L and LH $>$ 5.08 IU/L; S2: cFT $<$ 213.90 pmol/L and LH $>$ 6.18 IU/L), and compensated TD (S1: cFT $\geq$ 169.00 pmol/L and LH $>$ 5.08 IU/L; S2: cFT $\geq$ 213.90 pmol/L and LH $>$ 6.18 IU/L). The rates of the four classifications of the gonadal status are presented in Table 3. The relationship between cFT and LH in subjects is presented in Figure 1. The prevalence of secondary TD was higher than primary and compensated TD in S1 ($p < .01$); however, there were significant differences among the prevalence of primary, secondary, and compensated TD in S2 ($p < .05$).

The Sensitivity and Specificity of Questionnaires (ADAM and AMS)

The characteristics of the ADAM questionnaire had high sensitivity and low specificity in S1 and S2, while the AMS had low sensitivity and high specificity in S1; however, both variables were similar in S2 ($p > .05$). The sensitivity and specificity of questionnaires are presented in Table 4.

Discussion

Our two cross-sectional S1 and S2 studies were conducted in Chinese community-dwelling men in the same area. The data on the prevalence of TD and LOH were collected, and the applicability, sensitivity, and specificity of the ADAM and AMS questionnaires to Chinese population were validated. The diagnostic criteria of LOH were set up and compared with previous studies. The studies presented here had epidemiological characteristics of LOH in Chinese men.

The Positive Rates of LOH Symptoms Assessment

Using admitted questionnaires to investigate and assess the incidence and prevalence of LOH symptoms in community-dwelling population improved the accuracy and comparability of the trial results. Generally, positive results represent subjects that had related symptoms, but cannot diagnose subjects that suffered from TD or LOH. In our two studies, the mean rates of S2 were higher than the mean rates of S1 when the ADAM or AMS assessments ($p < .01$) was used. Although the ADAM had brief, user-friendly characteristics, the positive rates were too high to reduce the accuracy and specificity. The positive rates increased gradually with male aging ($p < .01$); therefore, age should be one of the main factors related to the symptoms of LOH.

The positive assessment rates in this study were closely related to the reports from Chinese study (84.56% in the ADAM, and 59.88% in the AMS), which also showed

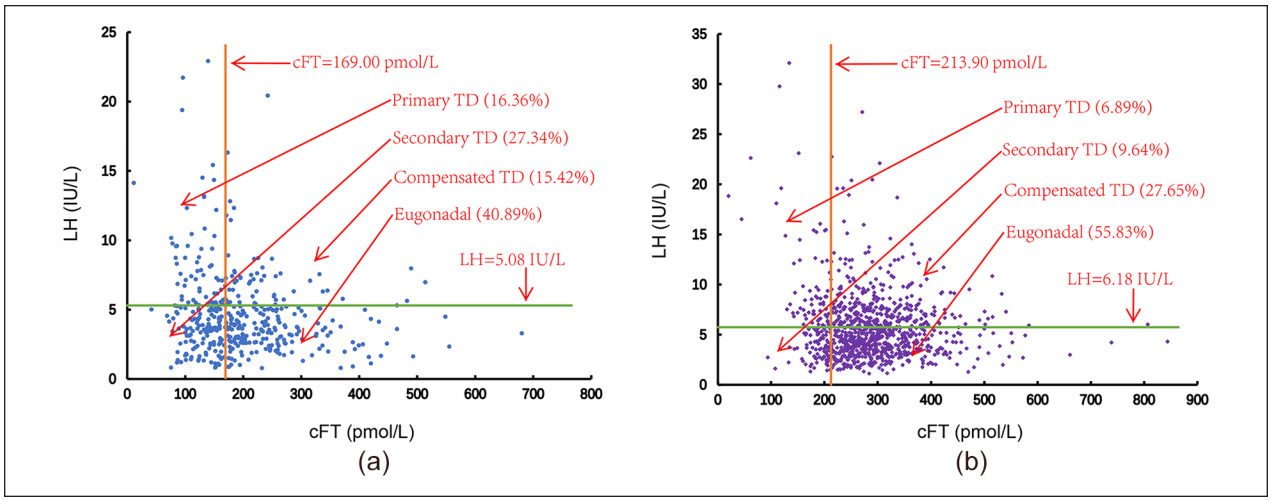

Figure 1. The relationship between cFT and LH in subjects. The four classifications of gonadal status were differentiated according to the cFT and LH cutoff. (a) S1: The *vertical line* corresponds to cFT = 169.0 pmol/L, and the *horizontal line* corresponds to LH = 5.08 IU/L. The majority of subjects (40.89%) were eugonadal, and the highest rate (27.34%) was secondary TD in three classifications of TD. (b) S2: The *vertical line* corresponds to cFT = 213.9 pmol/L, and the *horizontal line* corresponds to LH = 6.18 IU/L. The majority of subjects (55.83%) were eugonadal, and the highest rate (27.65%) was compensated TD in three classifications of TD.
LH = luteinizing hormone; TD = testosterone deficiency; cFT = calculated free testosterone.

Table 4. Sensitivity and Specificity of the ADAM and AMS Questionnaires.

		TT Cutoff		cFT Cutoff	
Variables	Study	ADAM	AMS	ADAM	AMS
Sensitivity (%)	S1	88.33	38.33	86.63	35.29
	S2	89.83	49.15	93.59	57.05
Specificity (%)	S1	20.92	64.40	24.48	63.49
	S2	9.04	51.19	9.64	52.79

Note. ADAM = the Androgen Deficiency in Aging Males questionnaire; AMS = the Aging Males' Symptoms scale; TT = total testosterone; cFT = calculated free testosterone.

positive rates that increased with male aging (Sun et al., 2012). The high positive rates of the questionnaire assessment were in part due to the frequent response of Chinese middle-aged and elderly men reporting degrees of decline in sexual function and libido. Participants tended to answer "yes" to question 1 (Do you have a decrease in libido [sex drive]?) and question 7 (Are your erections less strong?) in the ADAM questionnaire. In addition, two Chinese studies indicated that the assessment positive rates (62.86% in the ADAM, and 23.05%/10.7% in the AMS) were lower than the rates reported in our results (He et al., 2012; Wu et al., 2013).

The Prevalence of TD

The results presented here indicated that there were significant differences in serum TT, cFT, LH, and SHBG levels between S1 and S2 ($p < .001$; Zhou et al., 2020). It was clear that the cutoff values of serum TT and cFT in Chinese men, especially in S1 subjects, were notably lower than those of the EMAS study (11 nmol/L for TT, 220 pmol/L for free testosterone [FT]) and the EAU guidelines (12 nmol/L for TT, 225 pmol/L for cFT; Salonia et al., 2020; Wu et al., 2010). Comparing these data with our results, only the cutoff value of serum cFT in S2 was close to the EMAS and EAU values. Our results indicated that the TT and cFT levels of S2 controls were lower than the older groups. It is postulated that the limited sample numbers of the controls led to a bias in the data and might not have been representative of the accurate testosterone levels in young male adults. Regarding the two studies, optimal measures should be taken to ensure that the control numbers were equal to or close to the numbers of a subjects' subgroup. In addition, the sampling bias should contribute to lower testosterone levels in S2 controls.

We found that the serum cFT cutoff value was close to optimal for screening, evaluating, and diagnosing TD and LOH. In addition, it was suitable for different age groups due to the ladder-like change patterns of serum hormones. TD prevalence increased gradually with male aging, which showed a real and potential prevalence trend when using the serum cFT cutoff value. Although EAU guidelines suggest using serum TT to diagnose TD and reserving use of serum cFT only for men with borderline serum TT, the literature (Antonio et al., 2016) demonstrated that low serum cFT, even in the presence of normal serum TT, was associated with TD-related symptoms. However,

normal serum cFT, despite low serum TT, was not associated with cognate symptoms. In brief, it should be more precise and reliable to use serum FT levels to evaluate the TD and LOH status in aging males. The serum FT cutoff value is more valuable and more significant than the TT cutoff value.

A systematic review (Millar et al., 2016) identified that the prevalence of low testosterone ranged between 2% and 77%. The threshold testosterone levels used for reference standards also varied substantially, and a weak correlation was found between signs, symptoms, and testosterone levels. Additionally, there was some uncertainty about what threshold testosterone levels should be considered low for aging men. The BLSA (Harman et al., 2001) reported that the serum TT cutoff (11.3 nmol/L) was used to calculate the TD prevalence resulting in 12%–49% from 50- to 80-year-old groups. Regarding Chinese middle-aged and elderly men from different areas, the cFT cutoff value (0.3 nmol/L) was used to calculate TD prevalence, which showed 13.0%–46.7% from 40–49 to ≥70-year-old groups (Li et al., 2005). Another published study reported using the TT cutoff value, the TD prevalence was 21.3% (<12 nmol/L) (He et al., 2012). Obviously, there was a difference in the TD prevalence between the abovementioned results and the S1 and S2 results presented here.

The results presented here indicate that the TD prevalence that used the cFT cutoff was higher than the TT cutoff ($p < .01$). Therefore, different types of cutoff influenced the results. The data showed that the prevalence of TD was significantly different between S1 and S2 when different cutoff values were used. The reasons for the significant difference found in TD prevalence between S1 and S2 are presented in the limitations section.

The Prevalence of LOH

The diagnostic criteria of LOH are not unified worldwide. A recent study result reported no significant association between the total score of hypogonadism symptoms (AMS questionnaire) and serum TT and FT levels. However, there was a significant association with most symptoms related to psychological diagnoses (Samipoor et al., 2018). Another study (Antonio et al., 2016) reported that low cFT was associated with TD-related symptoms. In addition, results from the Turku Male Ageing Study (Huhtaniemi et al., 2008) identified that the poor correlation of low T levels and LOH symptoms is unclear, while increased incidence of sexual problems in aging men may be the most sensitive symptom for LOH. In general, it was difficult to accurately distinguish between erectile dysfunction (ED) and sexual symptoms of LOH. The highest accuracy for TT and cFT in detecting ED subjects with two symptoms was observed for reduced morning erections and low libido. The addition of a third symptom, ED, further improved the accuracy of the study. The simultaneous presence of reduced morning erections and low libido was the cluster of symptoms that, along with TT <10.4 nmol/L or cFT <225 pmol/L, defines LOH in a specific, evidence-based manner (Rastrelli et al., 2016).

Several studies reported the diagnostic criteria and prevalence of LOH for over 20 years. The MMAS (Araujo et al., 2004; Feldman et al., 2002; O'Donnell et al., 2004) has data available on eight TD-related signs/symptoms including: (1) loss of libido; (2) ED; (3) depression; (4) lethargy; (5) inability to concentrate; (6) sleep disturbance; (7) irritability; and (8) depressed mood. Men were considered to have LOH if they met one of the following two conditions: (1) at least three signs/symptoms and TT less than 6.94 nmol/L; or (2) at least three signs/symptoms and TT 6.94–13.88 nmol/Land FT less than 0.3092 nmol/L. Estimates of the crude prevalence of LOH at baseline and follow-up were 6.0% and 12.3%, respectively. Prevalence increased significantly with age where the crude incidence rate of LOH was 12.3 per 1000 person-years, and the rate increased significantly ($p < .0001$) with age. The Boston Area Community Health Survey (Araujo et al., 2007) recommended that LOH was defined as low TT (<300 ng/dL) and FT (<5 ng/dL) plus the presence of low libido, ED, osteoporosis or fracture, or two or more of the following symptoms: sleep disturbance, depressed mood, lethargy, or diminished physical performance. Approximately 24% of the subjects had TT less than 300 ng/dL and 11% had FT less than 5 ng/dL. Low testosterone levels were associated with symptoms, but many men with low testosterone levels were asymptomatic (e.g., in men ≥50 years, 47.6%), and prevalence of LOH was 5.6% (95% confidence interval, CI: [3.6%, 8.6%]). The EMAS (Tajar et al., 2012; Wu et al., 2010) suggested that LOH can be defined by the presence of at least three sexual symptoms (poor morning erection, low sexual desire, ED) associated with TT level less than 11 nmol/L and FT level less than 220 pmol/L; 4.1% of subjects had TT level less than 8.0 nmol/L and 17.0% had TT level less than 11 nmol/L. The overall prevalence of LOH in the EMAS study population would be 2.1% (1.2% moderate LOH and 0.9% severe LOH) and increased with age from 0.1% to 5.1%. A study (Liu et al., 2016) identified that TT levels <13.21 nmol/L or cFT <268.89 pmol/L were associated with an increase in the three sexual symptoms (decreased ability/frequency of sexual activity, decreased number of morning erections, and decreased libido). The prevalence of LOH was 9.1% under these criteria, including all three sexual symptoms with the aforementioned TT and cFT cutoff. Another study (Liu et al., 2009) reported that the

prevalence of TD was 24.1% based on the criterion of TT levels <300 ng/dL, and 16.6% based on the criterion of both TT levels <300 ng/dL and FT levels <5 ng/dL. The prevalence of LOH (ADAM positive) was 12.0%. The prevalence of LOH increased with older age, obesity, and diabetes mellitus, which served as independent risk factors for TD and LOH.

In comparison with older literature, the diagnostic criteria and prevalence of LOH focused primarily on sexual symptoms and other LOH-related symptoms. In order to ensure accuracy, we combined serum FT cutoff values with questionnaires (e.g., ADAM or AMS) to evaluate the prevalence of LOH. According to S1 and S2, the mean prevalence of LOH was 37.85% and 15.47% in positive for the ADAM questionnaire, and 15.42% and 9.43% in positive for the AMS questionnaire, respectively. Different assessment questionnaires influenced the rates of LOH prevalence. LOH prevalence in S1 was higher than the rates reported in the literatures, but the prevalence of S2 was similar.

Classifications of TD Based on Gonadal Status

The published literature (Tajar et al., 2010) reported the secondary, primary, and compensated TD categories to be 11.8%, 2.0%, and 9.5%, respectively. Secondary TD was associated with obesity and primary TD predominately with age. Compensated TD could be considered a distinct clinical state associated with aging. Another classification of TD was defined as the aging men with T levels <10.5 nmol/L and LH levels >9.4 IU/L as biochemical primary hypogonadism (PHG; Ahern et al., 2016). Subjects were classified as persistent eugonadal (pEUG, 97.5%), incident (i) PHG (1.1%), pPHG (1.1%), and reversed PHG (0.3%), and the incidence of PHG was 0.2%/year. Higher age (>70 years) and chronic illnesses were predictors of iPHG. Upon transition from EUG to PHG, erectile function, physical vigor, and hemoglobin worsened significantly. Men with pPHG had decreased morning erections, sexual thoughts, and hemoglobin with increased insulin resistance. The two kinds of classifications on TD from the literatures paid close attention to etiology and risk factors, the longitudinal change of hormones, and prognosis of TD. These data provide significant insight on TD with the hope of improving the diagnosis and management of LOH, while providing more information for future researchers to design treatment methods.

In the results presented here, the majority of subjects were eugonadal (40.89% in S1 vs. 55.83% in S2). According to three classifications of TD, the highest rates were secondary TD (27.34%) in S1 and compensated TD (27.65%) in S2, respectively. The rates of secondary TD in S1 or compensated TD in S2 were higher than the rates reported in the literatures.

The Sensitivity and Specificity of Questionnaires

The ADAM questionnaire was introduced initially with a sensitivity of 88% and a specificity of 60% and was regarded as a screening test of LOH (Morley et al., 2000). Similarly, other reports (Kratzik et al., 2004; Morley et al., 2006; Tancredi et al., 2004) suggested that the sensitivity of the ADAM and AMS questionnaires may be as high as 97%/81% and 83%/75%, and the specificity was 30%/21.6% and 39%/71%, respectively. Despite a high sensitivity, the ADAM questionnaire was reported to have a specificity of only 24%–60%, which suggested that its use beyond an initial assessment may be limited (Martinez-Jabaloyas et al., 2007; Rabah & Arafa, 2009). Moreover, the ADAM perhaps overestimated LOH occurrence, because its sensitivity was the highest (0.878) but it had the lowest specificity (0.099) (Lu et al., 2016). Another report stated that the ADAM questionnaire rendered sensitivity of 88.1%, specificity of 44.7%, and accuracy of 61.4%, and low libido alone had better specificity (75.5%) and accuracy (73.2%) than the entire questionnaire (Ugwu & Ikem, 2018). In addition, the ADAM questionnaire showed adequate sensitivity (73.6%) in diagnosing male patients with low levels of cFT. However, due to the lack of specificity (31.9%), the ADAM could not replace cFT assessments in men aged 40 years and above (Cabral et al., 2014). The specificity of ADAM in our S1 (20.92%/24.48%) and S2 (9.04%/9.64%), and the sensitivity of AMS in our S1 (38.33%/35.29%) were mildly lower compared to literatures.

The majority of effectiveness validity of questionnaires indicated that the sensitivity of the ADAM was higher than that of the AMS, but the specificity of the ADAM was lower than that of the AMS. The effectiveness of both questionnaires generally met the requirement of LOH screening or assessment. The ADAM has the characteristics of high sensitivity and ease of operation, is efficient in saving time, is beneficial for the assessment of LOH-related symptoms, and results in a decrease of missed diagnosis. Furthermore, The AMS has the characteristics of high specificity and can contribute to assessing therapeutic effect and the state of recovery.

Limitations

In comparison with American and European studies, the two studies presented here still pose some shortcomings. For example, the laboratory measurements of serum TT, SHBG, and LH used different methods in the two studies. This was due in large part to the fact that the two studies were conducted at 5-year intervals. In addition, the central laboratory of the Beijing Coordinating Center (China) changed the instruments and kits before S2 was carried

out. The use of different measuring methods was one of several reasons that led to significant differences in serum TT, cFT, LH, and SHBG levels between S1 and S2. The differences of hormone levels could influence the TD cutoff values and thus the prevalence of TD and LOH. In addition, there was an inconsistency in the sample size and the significant differences of age, lifestyle, and medical conditions between participants in the two groups. These limitations may affect research findings, which will be taken into consideration in designing and conducting further studies. Even with these limitations, the results presented here provide valuable knowledge for future researchers.

Conclusion

The results presented in this study showed that the positive rates of LOH symptoms assessment were closed to other published reports from Chinese studies. The TD prevalence reported in the literature fell between the S1 and S2 results presented here. We found that the LOH prevalence of S1 was higher than the rates reported in the literature, but the prevalence of S2 was found to be similar. Our results indicated that the serum cFT cutoff value was an optimal threshold for screening, evaluating, and diagnosing TD and LOH. Prevalence increased gradually with male aging excluding TD prevalence reported in the TT cutoff. Based on our results, we do not recommend either the use of questionnaires such as ADAM or AMS alone, or the use of TD alone to diagnose and monitor LOH. Support of the diagnosis criteria of LOH should at least include the presence of LOH symptoms (i.e., positive questionnaire results of ADAM or AMS) in combination with the serum FT levels measuring lower than the FT cutoff value.

Acknowledgments

The authors wish to thank all participants and their families for participating in this study. We gratefully acknowledge the help of Dr. Ru-Ming Shu, Dr. Can-Gang Wang, Dr. Li-Hua Zhuang, and other staff of Fucheng Technical Service Center of Family Planning for their excellent technical assistance in participants recruitment, questionnaire management, and the collection, storage, and transport of blood samples. And we gratefully acknowledge the help of Dr. Dian He for the statistical analysis, Dr. He was from Department of Epidemiology and Health Statistics, School of Public Health, Capital Medical University, Beijing, China. We also thank International Science Editing (http://www.internationalscienceediting.com) for editing this manuscript.

Declaration of Conflicting Interests

The author(s) declared no potential conflicts of interest with respect to the research, authorship, and/or publication of this article.

Funding

The author(s) disclosed receipt of the following financial support for the research, authorship, and/or publication of this article: This work was supported by the State-Level Special Commonweal Research Project (2007JZ01), the National "Twelfth Five-Year" Plan for Science and Technology Support (2012BAI32B03) and the Peking University International Hospital Research Funds (YN2016QN06).

Ethical Statement

Both studies were approved by the Ethics Committee and Institutional Review Board of National Research Institute for (Blinded For Review). Signed informed consent was taken from all participants prior to data collection. Anonymity and confidentiality of data were assured.

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References

Ahern, T., Swiecicka, A., Eendebak, R. J., Carter, E. L., Finn, J. D., Pye, S. R., O'Neill, T. W., Antonio, L., Keevil, B., Bartfai, G., Casanueva, F. F., Forti, G., Giwercman, A., Han, T. S., Kula, K., Lean, M. E. J., Pendleton, N., Punab, M., Rastrelli, G., . . . Wu, F. C. (2016). Natural history, risk factors and clinical features of primary hypogonadism in ageing men: Longitudinal data from the European Male Ageing Study. *Clinical Endocrinology*, *85*(6), 891–901. doi: 10.1111/cen.13152 [doi].

Antonio, L., Wu, F. C., O'Neill, T. W., Pye, S. R., Ahern, T. B., Laurent, M. R., Huhtaniemi, I. T., Lean, M. E. J., Keevil, B. G., Rastrelli, G., Forti, G., Bartfai, G., Casanueva, F. F., Kula, K., Punab, M., Giwercman, A., Claessens, F., Decallonne, B., & Vanderschueren, D. (2016). Low free testosterone is associated with hypogonadal signs and symptoms in men with normal total testosterone. *Journal of Clinical Endocrinology and Metabolism*, *101*(7), 2647–2657. doi: 10.1210/jc.2015-4106 [doi].

Araujo, A. B., Esche, G. R., Kupelian, V., O'Donnell, A. B., Travison, T. G., Williams, R. E., Clark, R. V., & McKinlay, J. B. (2007). Prevalence of symptomatic androgen deficiency in men. *Journal of Clinical Endocrinology and Metabolism*, *92*(11), 4241–4247. doi: jc.2007-1245 [pii] 10.1210/jc.2007-1245 [doi].

Araujo, A. B., O'Donnell, A. B., Brambilla, D. J., Simpson, W. B., Longcope, C., Matsumoto, A. M., & McKinlay, J. B. (2004). Prevalence and incidence of androgen deficiency in middle-aged and older men: Estimates from the Massachusetts Male Aging Study. *Journal of Clinical Endocrinology and Metabolism*, *89*(12), 5920–5926. doi: 89/12/5920 [pii] 10.1210/jc.2003-031719 [doi].

Cabral, R. D., Busin, L., Rosito, T. E., & Koff, W. J. (2014). Performance of Massachusetts Male Aging Study (MMAS) and androgen deficiency in the aging male (ADAM) questionnaires in the prediction of free testosterone in patients aged 40 years or older treated in outpatient regimen. *Aging*

Male, *17*(3), 147–154. doi: 10.3109/13685538.2014.908460 [doi].

Corona, G., Rastrelli, G., & Maggi, M. (2013). Diagnosis and treatment of late-onset hypogonadism: Systematic review and meta-analysis of TRT outcomes. *Best Practice & Research: Clinical Endocrinology & Metabolism*, *27*(4), 557–579. doi: S1521-690X(13)00048-1 [pii] 10.1016/j.beem.2013.05.002 [doi].

Feldman, H. A., Longcope, C., Derby, C. A., Johannes, C. B., Araujo, A. B., Coviello, A. D., Bremner, W. J., & McKinlay, J. B. (2002). Age trends in the level of serum testosterone and other hormones in middle-aged men: Longitudinal results from the Massachusetts male aging study. *Journal of Clinical Endocrinology and Metabolism*, *87*(2), 589–598. doi: 10.1210/jcem.87.2.8201 [doi].

Harman, S. M., Metter, E. J., Tobin, J. D., Pearson, J., & Blackman, M. R. (2001). Longitudinal effects of aging on serum total and free testosterone levels in healthy men. Baltimore Longitudinal Study of Aging. *Journal of Clinical Endocrinology and Metabolism*, *86*(2), 724–731. doi: 10.1210/jcem.86.2.7219 [doi].

He, L. J., Wang, Y., Zhou, Z., & Na, Y. Q. (2012). Urologic and reproductive health status of 30–60 years old males: Investigation among 1006 men in Shijingshan District of Beijing. *Zhonghua Nan Ke Xue*, *18*(4), 356–358.

Huhtaniemi, I., Makinen, J. I., Perheentupa, A., & Raitakari, O. T. (2008). Late-onset hypogonadism in men. Experience from the Turku Male Ageing Study (TuMAS). *Hormones (Athens)*, *7*(1), 36–45. doi: 10.14310/horm.2002.1111036 [doi].

Kratzik, C. W., Reiter, W. J., Riedl, A. M., Lunglmayr, G., Brandstatter, N., Rucklinger, E., Metka, M., & Huber, J. (2004). Hormone profiles, body mass index and aging male symptoms: Results of the Androx Vienna Municipality study. *Aging Male*, *7*(3), 188–196. doi: 10.1080/13685530412331284650 [doi].

Li, J. Y., Li, X. Y., Li, M., Zhang, G. K., Ma, F. L., Liu, Z. M., Zhang, N-Y., & Meng, P. (2005). Decline of serum levels of free testosterone in aging healthy Chinese men. *Aging Male*, *8*(3–4), 203–206. doi: R27T8X0323772V27 [pii] 10.1080/13685530500356010 [doi].

Liu, C. C., Wu, W. J., Lee, Y. C., Wang, C. J., Ke, H. L., Li, W. M., Hsiao, H-L., Yeh, H-C., Li, C-C., Chou, Y-H., Huang, C-H., & Huang, S. P. (2009). The prevalence of and risk factors for androgen deficiency in aging Taiwanese men. *Journal of Sexual Medicine*, *6*(4), 936–946. doi: S1743-6095(15)32479-6 [pii] 10.1111/j.1743-6109.2008.01171.x [doi].

Liu, Z. Y., Zhou, R. Y., Lu, X., Zeng, Q. S., Wang, H. Q., Li, Z., & Sun, Y. H. (2016). Identification of late-onset hypogonadism in middle-aged and elderly men from a community of China. *Asian Journal of Andrology*, *18*(5), 747–753. doi: 160883 [pii] 10.4103/1008-682X.160883 [doi].

Lu, T., Hu, Y. H., Tsai, C. F., Liu, S. P., & Chen, P. L. (2016). Applying machine learning techniques to the identification of late-onset hypogonadism in elderly men. *Springerplus*, *5*(1), 729. doi: 10.1186/s40064-016-2531-8 [doi] 2531 [pii].

Martinez-Jabaloyas, J. M., Queipo-Zaragoza, A., Rodriguez-Navarro, R., Queipo-Zaragoza, J. A., Gil-Salom, M., & Chuan-Nuez, P. (2007). Relationship between the Saint Louis University ADAM questionnaire and sexual hormonal levels in a male outpatient population over 50 years of age. *European Urology*, *52*(6), 1760–1767. doi: S0302-2838(07)00755-5 [pii] 10.1016/j.eururo.2007.05.021 [doi].

Millar, A. C., Lau, A. N. C., Tomlinson, G., Kraguljac, A., Simel, D. L., Detsky, A. S., & Lipscombe, L. L. (2016). Predicting low testosterone in aging men: A systematic review. *CMAJ: Canadian Medical Association Journal*, *188*(13), E321–E330. doi: cmaj.150262 [pii] 10.1503/cmaj.150262 [doi].

Morley, J. E., Charlton, E., Patrick, P., Kaiser, F. E., Cadeau, P., McCready, D., & Perry, H. M., III. (2000). Validation of a screening questionnaire for androgen deficiency in aging males. *Metabolism: Clinical and Experimental*, *49*(9), 1239–1242. doi: S0026-0495(00)25964-7 [pii] 10.1053/meta.2000.8625 [doi].

Morley, J. E., Perry, H. M., III, Kevorkian, R. T., & Patrick, P. (2006). Comparison of screening questionnaires for the diagnosis of hypogonadism. *Maturitas*, *53*(4), 424–429. doi: S0378-5122(05)00182-9 [pii] 10.1016/j.maturitas.2005.07.004 [doi].

O'Donnell, A. B., Araujo, A. B., & McKinlay, J. B. (2004). The health of normally aging men: The Massachusetts Male Aging Study (1987–2004). *Experimental Gerontology*, *39*(7), 975–984. doi: 10.1016/j.exger.2004.03.023 [doi] S0531556504001275 [pii].

Rabah, D. M., & Arafa, M. A. (2009). Validation of an Arabic ADAM questionnaire for androgen deficiency screening in the Arab community. *Aging Male*, *12*(4), 95–99. doi: 10.3109/13685530903265065 [pii] 10.3109/13685530903265065 [doi].

Rastrelli, G., Corona, G., Tarocchi, M., Mannucci, E., & Maggi, M. (2016). How to define hypogonadism? Results from a population of men consulting for sexual dysfunction. *Journal of Endocrinological Investigation*, *39*(4), 473–484. doi: 10.1007/s40618-015-0425-1 [doi] 10.1007/s40618-015-0425-1 [pii].

Salonia, A., Bettocchi, C., Carvalho, J., Corona, G., Jones, T. H., Kadioglu, A., Martinez-Salamanca, I., Minhas, S., Serefoğlu, E. C., & Verze, P. (2020). *Sexual and reproductive health*. https://uroweb.org/guideline/sexual-and-reproductive-health/#3

Samipoor, F., Pakseresht, S., Rezasoltani, P., & Mehrdad, M. (2018). The association between hypogonadism symptoms with serum testosterone, FSH and LH in men. *Aging Male*, *21*(1), 1–8. doi: 10.1080/13685538.2017.1382468 [doi].

Sun, K., Liang, G. Q., Chen, X. F., Ping, P., Yao, W. L., Zhang, S. J., Wang, B., Sun, Y-H., & Li, Z. (2012). Survey for late-onset hypogonadism among old and middle-aged males in Shanghai communities. *Asian Journal of Andrology*, *14*(2), 338–340. doi: aja2011171 [pii] 10.1038/aja.2011.171 [doi].

Tajar, A., Forti, G., O'Neill, T. W., Lee, D. M., Silman, A. J., Finn, J. D., Bartfai, G., Boonen, S., Casanueva, F. F., Giwercman, A., Han, T. S., Kula, K., Labrie, F., Lean, M. E. J., Pendleton, N., Punab, M., Vanderschueren, D., Huhtaniemi, I. T., & Wu, F. C. (2010). Characteristics of secondary, primary,

and compensated hypogonadism in aging men: Evidence from the European Male Ageing Study. *Journal of Clinical Endocrinology and Metabolism*, *95*(4), 1810–1818. doi: jc.2009-1796 [pii] 10.1210/jc.2009-1796 [doi].

Tajar, A., Huhtaniemi, I. T., O'Neill, T. W., Finn, J. D., Pye, S. R., Lee, D. M., Bartfai, G., Boonen, S., Casanueva, F. F. F., Forti, G., Giwercman, A., Han, T. S., Kula, K., Labrie, F., Lean, M. E. J., Pendleton, N., Punab, M., Vanderschueren, D., & Wu, F. C. (2012). Characteristics of androgen deficiency in late-onset hypogonadism: Results from the European Male Aging Study (EMAS). *Journal of Clinical Endocrinology and Metabolism*, *97*(5), 1508–1516. doi: jc.2011-2513 [pii] 10.1210/jc.2011-2513 [doi].

Tancredi, A., Reginster, J. Y., Schleich, F., Pire, G., Maassen, P., Luyckx, F., & Legros, J. J. (2004). Interest of the androgen deficiency in aging males (ADAM) questionnaire for the identification of hypogonadism in elderly community-dwelling male volunteers. *European Journal of Endocrinology of the European Federation of Endocrine Societies*, *151*(3), 355–360. doi: 10.1530/eje.0.1510355 [doi].

Ugwu, E. T., & Ikem, R. T. (2018). Androgen deficiency in aging male questionnaire for the clinical detection of testosterone feficiency in a population of black Sub-Saharan African men with type 2 diabetes mellitus: Is it a reliable tool? *Current Diabetes Reviews*, *14*(3), 280–285. doi: 10.2174/1573399812666161228152036 [doi] CDR-EPUB-80647 [pii].

Vermeulen, A., Verdonck, L., & Kaufman, J. M. (1999). A critical evaluation of simple methods for the estimation of free testosterone in serum. *Journal of Clinical Endocrinology and Metabolism*, *84*(10), 3666–3672. doi: 10.1210/jcem.84.10.6079 [doi].

Wu, F. C., Tajar, A., Beynon, J. M., Pye, S. R., Silman, A. J., Finn, J. D., O'Neill, T. W., Bartfai, G., Casanueva, F. F., Forti, G., Giwercman, A., Han, T. S., Kula, K., Lean, M. E. J., Pendleton, N., Punab, M., Boonen, S., Vanderschueren, D., Labrie, F., . . . Huhtaniemi, I. T. (2010). Identification of late-onset hypogonadism in middle-aged and elderly men. *New England Journal of Medicine*, *363*(2), 123–135. doi: NEJMoa0911101 [pii] 10.1056/NEJMoa0911101 [doi].

Wu, M., Li, J. H., Yu, X. H., Liang, G. Q., Li, P., Liu, Z. Y., Huang, Y-R., Sun, Y-H., & Li, Z. (2013). Late-onset hypogonadism among old and middle-aged males in a rural community of Zhejiang Province. *Zhonghua Nan Ke Xue*, *19*(6), 522–526.

Zhou, S. J., Lu, W. H., Yuan, D., Li, H., Shu, R. M., Di, G., & Gu, Y. Q. (2010). Clinical validation of screening scales for late onset of hypogonadism in Chinese males. *Zhonghua Nan Ke Xue*, *16*(2), 106–111.

Zhou, S. J., Zhao, M. J., Yang, Y. H., Guan, D., Li, Z. G., Ji, Y. D., Zhang, B-L., Shang, X-J., Xiong, C-L., & Gu, Y. Q. (2020). Age-related changes in serum reproductive hormone levels and prevalence of androgen deficiency in Chinese community-dwelling middle-aged and aging men: Two cross-sectional studies in the same population. *Medicine (Baltimore)*, *99*(1), e18605. doi: 10.1097/MD.0000000000018605 [doi] 00005792-202001030-00048 [pii].