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Semen characteristics of fertile and subfertile men in a fertility clinic and correlation with age

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KEYWORDS

Age; Asthenozoospermia; Azoospermia; Male infertility; Saudi population; Semen; Spermatozoa **Abstract** *Background:* The characteristics and semen quality in the men of different populations have been reported, though such data are lacking in Saudis.

Objectives: (i) To characterize the semen parameters of fertile and subfertile men, (ii) To study the prevalence of abnormality of semen parameters in the subfertile group, and (iii) To identify the relationship between semen parameters and age.

Methods: This study included 49 fertile and 160 subfertile men and 76 men with unproven fertility attending a fertility clinic in Riyadh. Their semen parameters were estimated, statistically analyzed, characterized, and correlation studies were conducted.

Results: The median age of the fertile and subfertile groups was quite similar. Significant differences were demonstrated in the median values of sperm concentration $(98.6 \times 10^6/\text{ml vs } 14.5 \times 10^6/\text{ml})$, P < 0.001, progressive sperm motility (58% vs 40%, P < 0.001), and abnormal sperm morphology (55% vs 75%, P < 0.001) between fertile and subfertile men. The percentage of normal semen viscosity was higher in fertile men, whereas the median semen volume values were nearly similar in the fertile and subfertile men (2.5 vs 2.75 ml). The prevalence of asthenozoospermia (36%) and azoospermia (26%) among subfertile men was the highest among other semen abnormality categories. There was an inverse correlation between the age and both sperm motility and semen volume in the investigated groups.

Conclusion: The main semen parameters in the fertile and subfertile subjects in this study differ significantly and the age was demonstrated to be correlated inversely with sperm motility and semen volume. Further studies in other regions of Saudi Arabia are needed.

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1. Introduction

Semen quality is one of the most valuable indications of male reproductive health where semen analysis plays a critical role in the diagnosis and treatment of male infertility. Semen analysis is widely undertaken applying the reference values for normal semen measurements published by the World Health Organization (WHO, 1999).

Several studies during the last decades have highlighted the concern of a time-related decrease in the semen quality world-

Table 1 Social and semen parameters in	n different groups of Sau	di men.						
	Pooled data	Fe	ertile		Subfertile		Unproven fertility	
	<i>n</i> (Mean \pm S.D.) Medi	an (25–75) <i>n</i>	(Mean \pm S.D.)	Median (25–75)	<i>n</i> (Mean \pm S.D.)	Median (25–75)	<i>n</i> (Mean \pm S.D.)	Median (25–75)
Age(years)	$285 \ 33.79 \pm 8.257 \ 32(28)$	(-39) 49	37 ± 7.056	37(31.5-40.5)	$160 \ 35.65 \pm 8.67$	34(29-40)	$76\ 27.81\ \pm\ 4.05$	27(25–30)
Duration of unprotected intercourse (years, Semen volume (ml)	$20/9.83 \pm 8.03$ $7(3-1)$ 283 3.122 ± 1.69 $3(2-4)$	(c) (48 (f) (f)	10.89 ± 7.06 3.03 ± 1.61	10(4.25–15) 2.5(1.75–4.125)	$158 \ 2.94 \ \pm \ 1.6$	6(3-13) 2.75(2-4)	$-$ - 76 3.56 \pm 1.87	- 3(2.5-4.5)
Sperm Concentration (10 ⁶ /ml)	$283 \ 60.48 \pm 63.42 \ 48.6($	5-93) 49	116.40 ± 57.97	98.6**(80-151.5)	$158 \ 39.38 \pm 49.54$	$14.5^{**}(1-75)$	$76\ 68.28\ \pm\ 69.22$	55.5(11.95–91.75)
Sperm motility (%)	$234 \ 45.01 \pm 20.54 \ 46(30$.75–59) 49	57.43 ± 15.97	58**(49-67.5)	$118 \ 39 \pm 19.8$	$40^{**}(25-50)$	$67 \ 46.64 \pm \ 20.61$	48(30-60)
Sperm abnormal morphology (%)	$221 \ 65.56 \pm 16.54 \ 65 \ (5$	5-80) 49	55.20 ± 12.78	55**(45-65)	$107 \ 71.74 \pm 15.66$	75**(60–85)	$65\ 63.18\ \pm\ 16.14$	65(50-77.5)
Semen viscosity:								
Normal	283 246 (86.9%)	49	0 46 (93.88%)		158 133 (84.2%)		76 67 (88.16%)	
Increase	24 (8.5%)		2 (4.1%)		17 (10.76%)		5 (6.69%)	
Highincrease	13 (4.6%)		1 (2.05%)		8 (5.06%)		4 (5.26%)	
Occupation:								
Civilian	278 207 (74.5%)	49	36 (73.5%)		154 116 (75.3%)		75 55 (73.33%)	
Military	71 (25.5%)		13 (26.5%)		38 (24.7%)		20 (26.7%)	
n = number of the subjects; S.D. = standa The difference between fertile and subfertile	rd deviation $(25-75) = 25t$. e groups is significant at * <i>H</i>	h–75th percen $2 \leq 0.05$ or **	ttile. $P \leqslant 0.001.$					

wide (Miyamoto et al., 2012). They provided the evidences of a decreasing trend in sperm count and percentage of sperm motility or normal sperm morphology over the last decades, from France (Auger et al., 1995), Scotland (Irvine et al., 1996), Italy (Bilotta et al., 1999), Denmark (Andersen et al., 2000; Jensen et al., 2002), India (Adiga et al., 2008), and Tunisia (Feki et al., 2009).

These findings are of an important concern since men with sperm count $<40 \times 10^6$ /ml were indicated to experience reduced fecundity (Bonde et al., 1998). However, in contradiction, other studies reported nonsignificant change in human semen quality (Bujan et al., 1996; Fisch et al., 1996; Rasmussen et al., 1997; Andolz et al., 1999; Acacio et al., 2000; Swan et al., 2000; Marimuthu et al., 2003; Axelsson et al., 2011). Hence, the global temporal trend in semen quality is still on debate.

Regional differences in semen quality have been reported for some areas in the USA (Fisch et al., 1996), Europe (Jørgensen et al., 2001; Jørgensen et al., 2002), Japan (Iwamoto et al., 2006), India (Adiga et al., 2008), and China (Gao et al., 2007). The European study of fertile men showed that sperm concentration of Danish men was 74% of that of Finnish men and 82% of the Scottish men (Jørgensen et al., 2001). In Southwest China, Li et al. (2009) showed that the semen parameters' values of men were markedly different from those reported for the other Chinese, USA or Europeans. Japanese fertile men had a semen quality level similar to Danish men that reported to have the lowest values among the investigated men in Europe (Iwamoto et al., 2006).

Nevertheless, some studies suggested that a decline in semen parameters is associated with increased age (Kidd et al., 2001; Chen et al., 2003; Eskenazi et al., 2003; Maya et al., 2009).

However, others showed no such association (Chen et al., 2004; Seo et al., 2000; Li et al., 2009).

In Saudi Arabia, the WHO reference values have been used to assess the reproductive health of Saudi men and there are no available data for the semen parameters in different cities, neither in other Arabic countries, except those reported for subfertile Tunisian men by Feki et al. (2009).

This study aimed to evaluate the semen characteristics of Saudi fertile, subfertile men, and men of unproven fertility in a fertility clinic in Riyadh city and to investigate the relationship between age and semen parameters.

2. Methods

2.1. Subjects

The study samples were male partners from couples attending a fertility clinic in Riyadh city, including 49 fertile, 160 subfertile men, and 76 men with unproven fertility. Fertile men included individuals of proven fertility whose wives achieved a full term pregnancy within the last two years. Subfertile men were men whose female partners failed to conceive but had no diagnosed fertility disorder after one year of unprotected intercourse. Men of unproven fertility were those visiting the clinic for reasons other than the fertility issue.

Each individual completed an extensive questionnaire regarding age, social status, occupation, and the reproductive history. As the occupations of most of the participants were either military or office jobs, the study population was grouped

Semen quality	n (%)	Sperm concentration	on (10 ⁶ /ml)	Sperm motility	
		Mean \pm S.D.	Median (25-75)	Mean \pm S.D.	Median (25-75)
Fertile	49	116.40 ± 57.97	98.6(80-151.5)	57.43 ± 15.8	58(49-67.5)
Subfertile	158				
Normal ^a	18 (11.4%)	99.27 ± 39.8	97.1(79.95-116.5)	66 ± 11.2	65(55-75.25)
Azoospermia	41(26%)	-		-	
Oligozoospermia	9(5.7%)	$8.97~\pm~4.78$	8.6**(5.25-13.15)	60 ± 8.2	58(53.5-67)
Asthenozoospermia	56 (35.5%)	73.28 ± 47.1	67.45**(42.75-93.75)	36.03 ± 12.5	40**(29.25-43.75)
Oligoasthenozoospermia	34 (21.5%)	$7.14~\pm~4.45$	5.3**(4.5-9.475)	26.47 ± 15.4	24.5**(12-40.75)

 Table 2
 Incidence of semen abnormal categories and sperm parameters in subfertile men.

The difference between fertile and subfertile groups is significant at $*P \le 0.05$ or $**P \le 0.001$.

(25-75) = 25th-75th percentile.

^a Normal: refers to the subfertile men with normal values of sperm concentration and motility according to WHO values.



Figure 1 Scatter plots with regression lines of semen volume versus age for the pooled data and the different study groups.



Figure 2 Scatter plots with regression lines of sperm motility versus age for the pooled data and the different study groups.

Table 3 Ave	srage val	lues of semen para	umeters according	to the	occupations (pool	led data).						
Occupation	Semen	volume (ml)		Sperm	1 concentration (10	(6/ml)	Sperm	1 motility (%)		Spern	1 abnormal morphe	ology (%)
	и	(Mean \pm S.D.)	Median(25–75)	и	(Mean \pm S.D.)	Median(25–75)	и	(Mean \pm S.D.)	Median(25–75)	и	(Mean \pm S.D.)	Median(25–75)
Civilian	213	3.07 ± 1.67	3(2-4)	213	58.31 ± 64.9	42.3(3.8–91.95)	170	43.4 ± 20.3	45(28.5–58)	159	65.50 ± 16.77	65(55–80)
Militaries	70	3.27 ± 1.74	3(2-4)	70	67.07 ± 58.65	57(17.97–96.35)	64	49.3 ± 20.73	48.5(34.25–62)	62	65.7 ± 16.07	65(55–80)
(25-75) = 25ti	h-75th pe	ercentile.										

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according to occupation, and categorized as civilian and military.

2.2. Semen collection and analysis

Semen samples were collected by masturbation after 3–5 days of sexual abstinence in clean metal-free plastic containers. After liquefaction, an aliquot of semen was centrifuged at 1400g for 10 min. Subsequently, semen analysis was carried out according to WHO guidelines (1999) including liquefaction time, pH, odor, volume, viscosity, the presence of pus/epithelial cells, sperm motility, sperm concentration, and abnormal morphology. Sperm motility was assessed as either motile (WHO motility classes A + B + C) or immotile (class D).

Semen findings in subfertile men were categorized as normal (normal semen values according to WHO standards); azoospermia (no spermatozoa in the ejaculate); oligozoospermia (sperm concentration $< 20 \times 10^6$ /ml); asthenzoospermia (< 50% motile sperm); oligoasthenozoospermia (including both criteria).

2.3. Statistical analysis

Statistical analyses were performed by using statistical software package (SAS) version 9.1.3 (SAS Institute Inc. Cary, NC, USA). Because semen parameters follow markedly skewed (non-normal) distributions, the 25th-75th percentiles, medians, means and standard deviations were calculated. The data in the different groups were compared by Mann-Whitney, a non-parametric test, or the Chi-square test as appropriate. Logarithmic transformation of the age and the semen volume of fertile group and square root transformation of age and sperm concentration of fertile group yielded normal distributions, therefore Pearson's correlation coefficient was calculated. In contrast, no suitable transformations for the other variable groups yield normal distributions; hence Spearman's correlation coefficient was calculated to correlate age and semen parameters. Only the semen parameters that significantly correlated were plotted versus age for the pooled data and the different status along with the regression line. ANO-VA test was used to determine mean differences according to occupation. Differences were considered statistically significant at P < 0.05.

3. Results

3.1. Social parameters

There was nonsignificant difference either in the median age (37 and 34 years) or in the duration of marriage (10 and 6 years) in fertile or subfertile groups, respectively. Chi-Square test showed nonsignificant difference in the percentage of each of the occupation categories (civilian and military personnel) between the fertile and subfertile groups (civilian: 73.5% and 75.3%; militaries: 26.5% and 24.7%, respectively) (Table 1).

3.2. Sperm characteristics

There was a significant difference in the median values of sperm concentration, sperm motility, and sperm abnormal morphology between the fertile and subfertile men. Sperm con-

able 4 Sellien parallie		allerent	populations.						
kegion	Selected subjects	и	Period of study	Age Year)	Semen volume (ml)	Sperm concentration (10 ⁶ /ml)	Sperm motility (%)	Sperm normal morphology (%)	Reference
Liyadh (present tudy)	Fertile men	49	2010	37 ± 7	3.03	116	57	45	Present study
apan	Fertile	324		32.5 ± 4.5	3.7	53	62	42	Iwamoto et al. (2006)
outhwest China	Healthy men	1346	2007	20-40	2.3	77.8	70.9	I	Li et al. (2009)
ISA	Fertile	969	I	33.5 ± 5	I	67 ± 50	54 ± 13	14 ± 5	Guzick et al. (2001)
Cleveland (USA)	Fertile	56	I	I	1	6.69	72.5	37.7	Nallella et al. (2006)
Openhagen (Denmark)	Fertile	349	1996 - 1998	31.5 ± 4.3	3.8	77	60	49	Jørgensen et al. (2001)
aris (France)	Fertile men	207	1997-1998	32 ± 4.4	4.2	94	56	50	Jørgensen et al. (2001)
urku (Finland)	Fertile men	275	1996 - 1998	30 ± 4.5	4.1	105	66	52	Jørgensen et al. (2001)
Idinburgh (Scotland)	Fertile men	251	1996-1997	32.5 ± 4.2	3.9	92	67	50	Jørgensen et al. (2001)
cotland	Unknown fertility	577	1984 - 1995	27	3.4	104.5	61.3	I	Irvine et al. (1996)
taly	Fertile	1068	1981-1995		I	61	66	63	Bilotta et al. (1999)
aris	Fertile	1351	1973-1992	32–36	3.8	60	66	61	Auger et al. (1995)

centration and sperm motility were higher in the fertile, while abnormalities were more frequent in the subfertile group.

Semen volume, showed nonsignificant difference between the two groups although the percentage of men with normal viscosity was higher in the fertile than in the subfertile men, and the percentage of men with increased viscosity was higher in subfertile men.

3.3. Sperm quality of subfertile men

11.4% of the subfertile group had sperm concentration and sperm motility within normal range being nonsignificantly different from that of the fertile men. Interestingly, the prevalence of asthenozoospermia (35.5%) and azoospermia (26%) were the highest among other categories of subfertile group (Table 2).

3.4. Correlation of semen parameters with age

The results of age correlation with each of the semen parameters in the different groups (Figs. 1 and 2) were as follow:

3.4.1. Fertile men

The age range of men in this group 23–53 years, the median was 37 (31.5–40.5) years. The age was positively correlated with most of the semen parameters but was only significant with the sperm concentration (r = 0.304, P = 034).

3.4.2. Subfertile men

The age range was 21–74 years, the median was 34 (29–40) years. There was an inverse correlation between age and each of the semen parameters but there was significant negative correlation with sperm motility (r = -0.235, P = 0.01).

3.4.3. Unproven fertility men

The age range was 20–44 years; the median was 27 (25–30) years. There was negative correlation with sperm motility (r = -0.318, P = 0.009).

3.4.4. Pooled data

The age range of all men was 20–74 years, the median was 32 (28–39) years. There was a significant negative correlation between age and semen volume (r = -0.152, P = 0.011) and sperm motility (r = -0.149, P = 0.023).

3.5. Correlation of sperm quality with the occupation

ANOVA test showed nonsignificant correlation between the occupation categories and semen parameters. Semen volume, sperm concentration and sperm motility were nonsignificantly higher in the military group (Table 3).

4. Discussion

This is the first study conducted, as a pilot one, to characterize the semen parameters in Saudi men in Riyadh city. The number of men of proven fertility was lower than the subfertile men due to the difficulty in collecting semen samples from men of general population as it is an embarrassing process unless they have to collect semen for analysis in the fertility clinics.

Table 5 Semen paramete	rs in subfertile men in different pol	pulation	s.						
Country	Selected subjects	и	Period of study	Age (year)	Semen volume (ml)	Sperm concentration (10 ⁶ /ml)	1 Sperm motility (%)	Normal sperm morphology (%)	Reference
Riyadh, (present study)	Subfertile men	160	2010	35.65 ± 8.67	2.97 ± 1.61	39.38 ± 49.54	39.28 ± 19.58	29	Present study
Northeast of Scotland	Infertile couples $< 20 \times 10^6$	4832	1994-2005	34 ± 6	1	61	49	I	Sripada et al., 2007
Munirka, New Delhi, India	Subjects attending the Fertility Clini	c 1176	1990-2000	31.2	2.6 ± 0.1	60.6 ± 0.9	I	1	Marimuthu et al., 2003
South India	Infertile individuals	1610	2004-2005	I	2.64	26.61	47.14	19.75	Adiga et al., 2008
USA	Infertile couples	765	I	34.7 ± 4.9	1	52 ± 42	49 ± 15	11 ± 6	Guzick et al., 2001
Veina, Austria	Infertile men	7,780	1986-2003	31.6	1	10.25	21	15	Lackner et al., 2005
Korea	Healthy men with infertility	22,249	1989-1998	(32)21 - 40	I	60.5	I	I	Seo et al., 2000
Southern Tunisia, Sfax area	Men in infertile relationships	2940	1996 and 2007	$7\ 36.0\pm 6.9$	3.2 ± 1.6	96.1 ± 88.2	$44.2~\pm~12.7$	25.0 ± 16.5	Feki et al., 2009

The social parameters of the fertile and subfertile groups were in the same range regarding age, duration of unprotected intercourse, and the occupation in each group. Hence the two groups were well matched, the social parameters of the unproven fertility men were also in the same range as the other two groups except age. Age was lower in the last group compared with other groups, as most of these men were single, due to the social customs and traditions of the Saudi society that encourage men to get married at young age.

The differences in the median values of the main semen characteristics between the fertile and subfertile groups were in consistence with those reported by Nallella et al. (2006) and Guzick et al. (2001). The values of semen parameters of each of fertile and subfertile men were compared with those reported in other populations; USA (Acacio et al., 2000; Guzick et al., 2001; Nallella et al., 2006), Europe (Auger et al., 1995; Bilotta et al., 1999; Andolz et al., 1999; Jørgensen et al., 2001; Lackner et al., 2005; Sripada et al., 2007), China (Li et al., 2009), Japan (Iwamoto et al., 2006), india (Marimuthu et al., 2003; Adiga et al., 2008) and Korea (Seo et al., 2000) (Tables 4 and 5).

Sperm concentration of the fertile men was nearly two times higher than that recorded for the Americans or Danish fertile men and was also higher than the values reported for other populations, such as French, Scottish, Finnish, Italian, and Chinese men; whereas, Japanese men had the lowest sperm concentration.

Sperm motility was lower than that reported in different populations, i.e. American fertile men, Danish, Scottish and Finnish, Japanese and Italian men, but was similar to the results reported in French.

The percentage of sperm normal morphology was lower than that reported for the European populations, but higher than Japanese and in men in Cleveland (USA).

These differences could be due to endocrine, ethnic, geographical, environmental, nutritional, or life style variations. Specifically, the higher temperatures during most of the year in Riyadh city may affect sperm motility. In addition, genetic factors may be a factor where differences are due to different polymorphisms in the genes involved in influencing these parameters.

In subfertile men; sperm concentration was significantly higher than that the WHO (1999) reference ($<20 \times 10^6$ /ml) that could be due to the high percentage of the subfertile men ($\sim 50\%$) categorized as subfertile for reasons other than the sperm concentration (normal: 99.27×10^6 /ml and asthenospermia: 73.28×10^6 /ml). Nallella et al. (2006) reported a large group of patients with male factor infertility that presented higher sperm concentration. This average value was also higher than that reported for subfertile men in South India and Vienna (Austria), and lower than that of American subfertile men.

Sperm motility of subfertile Saudis was lower than reported for subfertile men of other population such as in South India, Northeast of Scotland and USA, but not that reported in Vienna (Austria). These variations could be due to the relatively lower sperm motility in the general population of our study compared with other populations.

Around one third of the subfertile men were asthenozoospermic, and one fifth were oligoasthenozoospermic. This shows a relatively high percentage ($\sim 57\%$) of Saudi subfertile men with abnormal sperm motility that was similar to the per-

Study	Region	Period of study (years)	Study subjects	Number of subjects and Age (year)	Oligozoospermia (%)	Astheno- zoospermia (%)	Oligoastheno- zoospermia (%)	Sperm abnormal morphology (%)	Azoospermia (%)
Present study	Riyadh, Saudi Arabia	2010	Subfertile men	$160\ 35.65\ \pm\ 8.67$	5.7	35.5	21.5	-	26
Acacio et al. (2000)	Los Angeles, USA	1994–1997	Male partners of women presenting for an infertility evaluation	1,385	18	51	-	14	4
Adiga et al. (2008)	South India	2005	Infertile individuals	1610	-	-	-	-	7.2
Marimuthu et al. (2003)	Munirka, New Delhi India	1990–2000	Subjects attending the Fertility Clinic	1176 31.2	1.8 (severe oligospermic were excluded)	63	-	_	Excluded
Andolz et al. (1999)	Northeast, Spain	1960–1996	Studied because of infertility	22,759	-	-	-	-	6
Seo et al. (2000)	Korea	1989–1998	Healthy men with infertility	22,249 32 (range 21–40 year)	-	-	-	_	19

)8)			individuals						
rimuthu	Munirka,	1990-2000	Subjects attending the	1176	1.8 (severe	63	-	-	
1. (2003)	New Delhi India		Fertility Clinic	31.2	oligospermic were excluded)				
lolz 1. (1999)	Northeast, Spain	1960–1996	Studied because of infertility	22,759	-	_	-	-	
et al. 00)	Korea	1989–1998	Healthy men with infertility	22,249 32 (range 21–40 year)	-	-	-	_	
ble 7 Imp	act of age on	semen parameters	in previous studies.						
dy		Region	Period of study	Selected su	ıbjects	Number	Age average (age ran	ige)	Impact of semen par
sent study		Riyadh	2010	Fertile and	l subfertile men	209	35.9 ± 8.3		Inverse im and sperm
et al. (2000))	Korea	1989–1998	Healthy m	en with infertility	22,249	32 (21-40)		None

Study	Region	Period of study	Selected subjects	Number	Age average (age range)	Impact of age on semen parameters
Present study	Riyadh	2010	Fertile and subfertile men	209	35.9 ± 8.3	Inverse impact on volume and sperm motility
Seo et al. (2000)	Korea	1989–1998	Healthy men with infertility	22,249	32 (21–40)	None
Kidd et al. (2001)	Review study of the literature	1980–1999	-	-	30 and 50	Inverse impact, but not with sperm concentration
Chen et al. (2003)	Massachusetts, USA	1989–2000	From andrology clinic	551	-	Inverse impact on all parameters
Chen et al. (2004)	Massachusetts, USA	2000-2002	From andrology clinic	306	18–54 years (35.9 \pm 5.6)	None
Cavalcante et al.(2008)	Northeast of Brazil	2002-2004	Men of conjugal infertility	531	37 ± 7.9	Inversely only with volume
Maya et al. (2009)	Medellin, Colombia	-	Men attending an andrology center	1364	\leq to 30 years; between 31 and 39 years; and \geq to 40 years	Inversely with all
Li et al. (2009)	Southwest, China	2007	Healthy men	1346	20-40	None
Mukhopadhyay et al. (2010)	Kolkata, India	1981-85 and 2000-2006	Men with infertility problems	3729	33.24 ± 6.13 and	A decline was seen
			and normal sperm count		35.17 ± 5.043 (22–62)	in sperm motility with increasing age in both decades

Table 6 Incidence of a zoospermia and other subfertile semen categories in different population.

centage reported in Los Angeles and Munirka (India). That percentage, followed by around 27% of the Saudi subfertile men who had sperm concentration $< 20 \times 10^6$ /ml (5.7% oligo-zoospermic and 21.5% oligoasthenozoospermic). This percentage was higher than that reported for men in Los Angeles.

In comparison with other studies (Table 6), the prevalence of azoospermia (26%) was higher than that reported for subfertile men in South India, Los Angeles (USA), Northeast Spain, and Korea. Acacio et al. (2000), and Andolz et al. (1999) demonstrated that 4% and 6%, respectively, of the subjects were azoospermic. The subjects of their studies were of subfertile relationship so they were not diagnosed as subfertile men, and were also of unknown age. Adiga et al. (2008) recruited a population of subfertile men of undefined age of whom 7.2% were azoospermic. Seo et al. (2000) investigated a population that is similar to that in our study (subfertile men; 32 years) reporting that 19% of the population was azoospermic. This value was closer to the value reported in the present study.

There was a significant correlation between age and the sperm concentration only in fertile men group that was not observed in other groups. This could be due to the lower variability in the age range and the small sample size of fertile group, of whom 57% fell in the age range of 30-40 years old, 13% were >45 years, and the oldest man was 53 years. The sample size of the subfertile group was higher, the age range was wider (50% were in the 30–40 years range), 13% were >45 years and the oldest man was 74 year. In the group of unproven fertility men, the sample size was higher than the fertile group, and they were younger with 26% in the age range 30-39 year old and the oldest was 39 years. Other studies (Table 7) agreed with the finding of this study suggesting no impact of age on sperm concentration (Cavalcante et al., 2008; Li et al., 2009; Seo et al., 2000). However, others were in disagreement with our results (Maya et al., 2009). Chen et al. (2004), reported nonsignificant impact of age on semen parameters in a sample size of 306, whereas Chen et al. (2003) reported an inverse relationship between age and semen parameters when the sample size and the age range were larger.

There was an inverse correlation between age and sperm motility and semen volume in almost all groups in agreement with Kidd et al. (2001) suggesting that advanced age was associated with a decrease in semen volume, sperm motility, and sperm morphology but not sperm concentration. Cavalcante et al. (2008) showed an inverse effect of age on semen volume but not on other semen parameters. Maya et al. (2009) showed an inverse relationship between age and the main semen parameters. In contrast, some studies reported no impact of age on semen parameters (Li et al., 2009; Chen et al., 2004).

The contradiction between these studies could be due to differences in the age-range and the number of participated individuals, in addition, to other confounder factors such as ethnics, genetics, geographical location and the surrounding environment.

The relatively low sperm motility in the general population and the relatively high prevalence of azoospermia are of important implications with respect to infertility and further studies using large number of fertile and subfertile subjects with additional information on their smoking, socioeconomic condition, and life- style related factors are recommended.

In conclusion, our findings suggest that the values of sperm parameters were, in agreement with WHO criteria, significantly different in normal fertile from that of the subfertile Saudi men. Age has no effect on sperm concentration, viscosity, and morphology, but an inverse effect of age was observed on sperm motility and semen volume. As Riyadh city has the highest population (18.5%) among other Saudi cities, due to diverse socioeconomic factors, it is important to assess the semen quality in its different parts for further validation of the statement. In addition, further studies in other regions of Saudi Arabia are also needed.

5. Ethical statement

After IRB approval, all participants were asked to sign an informed consent form if they agreed to take part in this study.

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