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The Importance of Discordant Follicle Stimulating Hormone and Inhibin B Levels in Primary Infertile Men: Findings from a Cross-Sectional Study

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Purpose: We aimed to investigate the relationship between follicle stimulating hormone (FSH) and inhibin B (InhB).
Materials and Methods: Data from 1,230 consecutive men presenting for primary couple's infertility were analyzed. Health-significant comorbidities were scored with Charlson comorbidity index. Quartiles of FSH and InhB were considered to determine threshold values. Descriptive statistics and logistic regression models tested association between FSH and InhB values.
Results: Overall, 1,080 (87.8%) men had concordant FSH and InhB values. Conversely, 150 patients (12.2%) had discrepancies in FSH and InhB, with 78 (6.3%) and 72 (5.9%) men reporting both low and high FSH and InhB values, respectively. Infertile men with discordant values were younger (median [interquartile range] 38.0 years [34–41 years] *vs*. 36.0 years [31–40 years]); had smaller testicular volume (TV) (12 mL [10–15 mL] *vs*. 15 mL [12–20 mL]); and, had more frequently a sperm DNA fragmentation test >30% (179 [59.1%] *vs*. 40 [78.4%]) than those with concordant values (all p<0.05). Moreover, a higher frequency of previous cryptorchidism (27.3% *vs*. 11.9%), lower sperm concentration (3.0 million/mL [0.9–11.0 million/mL] *vs*. 13.8 million/mL [3.1–36.0 million/mL]), lower progressive sperm motility rates (12.0% [5.0%–25.3%] *vs*. 20.0% [7.0%–36.0%]), and greater rates of non-obstructive azoospermia (36.4% *vs*. 23.9%) were found in men with discordant FSH and InhB values (all p≤0.005). At multivariable logistic regression analysis, higher body mass index (odds ratio [OR], 1.08; p=0.001), smaller TV (OR, 0.91; p<0.001), and a history of cryptorchidism (OR, 2.49; p<0.001) were associated with discordant FSH and InhB values.
Conclusions: More than one out of ten infertile men had discordant FSH and InhB values in the real-life setting showing worse clinical profiles than those with concordant levels. Smaller TV and history of cryptorchidism could be used as clinical markers to better tailor the need to test InhB.

Keywords: Follicle stimulating hormone; Infertility, male; Inhibin B; Reproductive health; Semen analysis

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The World Journal of MEN's HEALTH

INTRODUCTION

Spermatogenesis is a multi-stage process depending on a complex interplay of endocrine and paracrine signals. In human beings, this process occurs in seminiferous tubules and ends with the production of mature gamete in approximately 74 days. A leading role in the spermatogenesis is played by Sertoli cells, which constitute about 20% of the seminiferous tubules in adult men [1]. Moreover, various hormones are involved in this process, thus including follicle stimulating hormone (FSH) (reference range: 1.4–9.0 mUI/mL), luteinizing hormone (LH; reference range: 1.7–12.0 mUI/mL), testosterone (reference range: 2–10 ng/mL), Inhibin B (InhB) (reference range: 25–325 pg/mL) and anti-Mullerian hormone (reference range: 0.77–14.5 ng/mL). In particular, in adult men FSH stimulates the production of InhB in the testis, a glycoprotein member of the transforming growth factor-β (TGF-β) superfamily, which in turn activates a negative feedback on the pituitary secretion of FSH itself. Moreover, the release of InhB is regulated by other factors over FSH such as the amount for sperm count, and its levels could reflect the functional status of seminiferous epithelium [2].

Indeed, InhB is considered a direct marker of testicular function and cellular density, and an indirect marker of spermatogenesis, showing temporal changes in its secretion from infancy to adulthood [3]. As such, both InhB and FSH are indirect clinical markers of the global exocrine function of the testis.

Since the functional state of the seminiferous epithelium is expressed by the levels of both InhB and FSH, sperm quality and fecundability outcomes have been associated with these hormones. Nevertheless, neither FSH nor InhB alone can predict alterations of the spermatogenesis, and the role of both hormones in the diagnostic work-up of couples seeking medical help for infertility has been critically discussed throughout the literature [4]. Besides the opposite correlation between FSH and InhB which is expected, current literature is poor of data regarding the discordance between these hormones. Consequently, in this cross-sectional study we aimed to: i) investigate the relationship between circulating FSH and InhB, with particular focus on discordant values; and, ii) to assess the impact of the discordance toward clinical and semen characteristics in a cohort of white-European non-Finnish primary infertile men.

MATERIALS AND METHODS

1. Study cohort, variables, and outcome definition

The analyses of this cross-sectional study were conducted on a cohort of the latest 1,230 consecutive men assessed at a single academic tertiary-center for primary couple's infertility. Infertility was defined according to the World Health Organization (WHO) guidelines as the failure to achieve a pregnancy after 12 months or more of regular, unprotected sexual intercourse [5]. We included only men with pure male factor infertility (MFI), diagnosed after a comprehensive diagnostic evaluation of the female partners excluding any female infertility factor. All patients were assessed with a thorough self-reported medical and sexual history including age and any prevalent comorbidities. Our evaluation of medical history examined potential risk factors that may have an adverse impact on male fertility, with a particular emphasis on lifestyle factors, familial history (inclusive of testicular cancer), comorbidities (as scored with the Charlson comorbidity index [CCI] [6]), and previous testicular surgical procedures. Likewise, our focus was directed towards identifying any congenital or acquired anomalies that might compromise the integrity or functionality of the testicles, such as history of uni- or bilateral cryptorchidism, history of testicular torsion and/or trauma. We also considered any known exposure to gonadotoxins, thus including potential iatrogenic causes, i.e., gonadotoxic medications or prior radiation exposure, recreational and illicit drug use, as well as environmental exposure. Body mass index (BMI), defined as weight in kilograms by height in square meters, was measured for each patient. Testicular volume (TV) was assessed using a Prader's orchidometer (PO) by the same urologist [7]. For the specific purpose of this study, we recorded the volume of each testicle and the median value between the two sides.

2. Blood, hormonal, semen and genetic analyses

Venous blood samples were drawn from each patient between 7 AM and 11 AM after an overnight fast. More in details, FSH, LH, TSH and prolactin (PRL) were measured using a heterogeneous competitive magnetic separation assay. InhB was measured with an enzyme-linked immunosorbent assay. Total

testosterone levels were measured via a direct chemiluminescence immunoassay, and sex hormone–binding globulin levels were measured via a solid-phase chemiluminescent immunometric assay. Likewise, karyotype analysis was performed [8]. The same laboratory was used for all patients (IRCCS Ospedale San Raffaele).

All patients underwent at least one semen analysis [4], with semen parameters assessed according to 2010 WHO reference criteria; hence, we considered sperm concentration, progressive sperm motility and normal sperm morphology. In particular, azoospermia was defined as the absence of any sperm cell in the ejaculate in at least two consecutive semen analyses; oligozoospermia with having a sperm concentration lower than 15 million/mL; asthenozoospermia with a sperm progressive motility <32%; teratozoospermia with <4% of normal morphology spermatozoa. Total motile sperm count (TMSC) was calculated for every patient by multiplying the volume of the ejaculate in milliliters by the sperm concentration and the proportion of motile sperms divided by 100. A threshold of TMSC $<20\times10^6$ was considered pathological [9].

According to our internal protocol for male infertility, all primary infertile patients had their sperm DNA fragmentation index (SDF) tested according to sperm chromatin structure assay (SCSA) [10]. Pathologic SDF was defined with a threshold of >30% [11].

3. Statistical analysis

For the specific purpose of this study, we evaluated the quartile values of serum FSH and InhB levels. In this regard, concordant values were categorized when FSH level exceeded the 3rd quartile of 12.5 mUI/mL synchronously with an InhB level falling below the 1st quartile of 41.5 pg/mL, and discordant values for the inverse scenario. To further clarify, this meant that concordant values were recognized when there was a high FSH level coupled with a low InhB level, while discordant values when a low FSH level was found alongside a high InhB level. This categorization allowed to identify a "concordant" relationship between these two key hormones in relation to their involvement in spermatogenesis. Thereof, the term "discordance" was used to describe a condition where FSH and InhB levels did not follow the expected inverse relationship, i.e., if FSH levels were high, InhB levels were not correspondingly low, or vice versa.

Patients' characteristics are presented as medians and interquartile ranges (IQRs) or frequencies and proportions for continuous or categorical variables, respectively. The Mann–Whitney and the chi-squared tests were used to investigate potential differences in the distribution of continuous or categorical variables among patients without and with discrepancy between FSH and InhB serum level, respectively. The same tests were used to compare patients with both low or high FSH and InhB serum levels. Univariable and multivariable logistic regression models were fitted to predict which clinical markers were able to identify discordance between hormones.

Statistical analyses were performed using IBM SPSS v.26 (IBM Corp.). All tests were two sided, and statistical significance level was determined at $p<0.05$.

4. Ethics statement

Data collection followed the principles outlined in the Declaration of Helsinki; all patients had signed an informed consent agreeing to deliver their own anonymous information along with blood and seminal fluid. The study was approved by the IRCCS San Raffaele Hospital Ethical Committee, Milan, Italy (Prot. 2014 – Pazienti Ambulatoriali).

RESULTS

Table 1 details descriptive statistics for the whole cohort of 1230 patients and after further segregation according to FSH and InhB levels. Of all, 150 (12.2%) patients presented discordant FSH and InhB serum levels. Infertile men with discordant FSH and InhB levels were younger ($p=0.03$), had lower TV ($p<0.001$) and higher serum LH levels ($p<0.001$) compared to those with concordant values. Sperm concentration ($p<0.001$) and rates of progressive sperm motility ($p=0.004$) were lower in men with discordant hormone levels; moreover, higher rates of azoospermia ($p=0.001$), oligozoospermia ($p<0.001$), asthenozoospermia ($p=0.005$) and a greater prevalence of history of cryptorchidism ($p<0.001$) were found in infertile men with discordant *vs.* concordant FSH and InhB values. The proportion of patients with SDF>30% was higher among men with discordant compared to those with concordant hormonal levels ($p=0.01$). No further differences were found between these two groups (Table 1).

Table 2 depicts the whole cohort of infertile patients with discordant values of FSH and InhB furtherly

The World Journal of MEN's HEALTH

Table 1. Socio-demographic and clinical characteristics of the whole cohort (n=1,230) of infertile patients with concordant *vs*. discordant FSH and InhB serum levels

Variable	Whole Cohort	Infertile men with concordant FSH and InhB	Infertile men with discordant FSH and InhB	p-value
Socio-demographic and clinical characteristics				
Number of patients	1,230 (100)	1,080 (87.8)	150 (12.2)	
Age (y)	37.5 (34–41)	38.0 (34–41)	36.0 (33–41)	0.03
BMI (kg/m^2)	25.1 (23.2–27.2)	25.0 (23.2–27.1)	25.7 (23.5–27.6)	0.10
Cigarette smoking	364 (29.6)	320 (29.7)	44 (29.3)	0.94
CCI ≥1	94 (7.6)	81 (7.5)	13 (8.7)	0.61
Mean testicular volume (mL)	15 (12–20)	15 (12–20)	12 (10–15)	<0.001
Infertility length (mo)	20 (12–32.3)	19 (12–30)	24 (12–36)	0.22
Cryptorchidism	170 (13.8)	129 (11.9)	41 (27.3)	<0.001
Varicocele	710 (57.7)	630 (58.3)	80 (53.3)	0.25
Karyotype abnormalities	46 (3.7)	44 (4.1)	2 (1.3)	0.10
Serum hormone				
TSH (mUI/L)	1.7 (1.2–2.3)	1.7 (1.2–2.3)	1.7 (1.2–2.3)	0.97
LH (mUI/L)	4.2 (2.9–6.1)	4.2 (2.8–5.9)	5.3 (3.8–7.0)	<0.001
FSH (mUI/L)	6.0 (3.4–12.5)	7.0 (3.9–15.0)	4.4 (3.0–9.5)	<0.001
InhB (pg/mL)	103.6 (41.5–163.4)	99.0 (31.7–150.0)	120.0 (57.2–178.0)	<0.001
SHBG (nmol/L)	32.0 (24.0–42.0)	32.1 (24.0–42.0)	32.0 (23.0–41.5)	0.70
Total testosterone (ng/mL)	4.4 (3.3–5.7)	4.5 (3.4–5.7)	4.4 (3.3–5.6)	0.83
Prolactin (ng/mL)	8.4 (6.3–12.0)	8.3 (6.2–11.9)	9.5 (7.0–12.1)	0.15
SDF (%)	36.4 (22.2–52.1)	34.8 (20.5–51.4)	47.1 (31.6–53.8)	0.14
SDF >30%	219 (61.9)	179 (59.1)	40 (78.4)	0.01
Semen characteristic				
Concentration (million/mL)	12.0 (2.8–32.9)	13.8 (3.1–36.0)	3.0 (0.9–11.0)	<0.001
Volume (mL)	3 (2–4)	3 (2–4)	3 (2–4)	0.67
Progressive motility (%)	20.0 (7.0–35.0)	20 (7.0–36.0)	12.0 (5.0–25.3)	0.004
Normal morphology (%)	2.0 (1.0–10.0)	2.0 (1.0–10.0)	2.0 (1.0–16.0)	0.59
Azoospermia	296 (25.4)	245 (23.9)	51 (36.4)	0.001
Oligozoospermia	469 (53.9)	400 (51.2)	69 (77.5)	<0.001
Astenozoospermia	602 (70.6)	530 (69.1)	72 (83.7)	0.005
Teratozoospermia	475 (56.4)	430 (56.6)	45 (54.9)	0.77

Values are presented as number (%) or median (interquartile range).
BMI: body mass index, CCI: Charlson comorbidity index, TSH: thyroid-stimulating hormone, LH: luteinizing hormone, FSH: follicle stimulating hormone, InhB: inhibin B, SHBG: sex hormone-binding globulin, SDF: sperm DNA fragmentation index.

segregated into low values *vs*. high values. Infertile men presenting low levels of both FSH and InhB had higher proportion of SDF>30% (p=0.03) and azoospermia (p=0.002); conversely, the group of infertile men with high serum levels of both FSH and InhB depicted higher counts of oligozoospermia (p<0.001), asthenozoospermia (p=0.02) and of history of cryptorchidism (p<0.001). Groups did not differ in terms of other variables (Table 2).

Table 3 depicts the univariable and multivariable logistic regression models predicting the discordance of FSH and InhB values. At univariable logistic regression analysis, men with higher BMI (odds ratio [OR], 1.06; p=0.01), lower TV (OR, 0.90; p<0.001) and higher rates of history of cryptorchidism (OR, 2.77; p<0.001) were more likely to have discordant serum values of FSH and InhB. Accordingly, at multivariable logistic regression analysis, higher BMI (OR, 1.08; p=0.001), lower TV (OR, 0.91; p<0.001) and the history of cryptorchidism (OR, 2.49; p<0.001) were found to be independent predictors for discordant serum values of FSH and InhB values, after accounting for age, CCI and karyotype (Table 3).

Fig. 1 graphically displays rates of semen parameters

between concordant and discordant FSH and InhB groups.

DISCUSSION

We retrospectively analyzed data from a cohort of 1,230 consecutive white-European non Finnish men presenting for primary couple's infertility associated with pure male factor at a single tertiary outpatient clinic. Overall, more than one in ten patients had discordant FSH and InhB serum levels. These patients showed worse clinical, hormonal and semen characteristics with respect to men without discordant values. Of note, patient's BMI, TV and history of cryptorchidism emerged as predictors of discordant FSH/InhB ratio.

Testicular hypotrophy has been associated with spermatogenic dysfunction; indeed, various studies have shown that mean PO-derived TV in infertile men is smaller compared to age-comparable fertile controls [12]. For instance, Boeri et al [7] proposed TV as a reliable clinical marker of both hormonal and spermatogenic activity. Notwithstanding, Tunc et al [13] reported that TV was not a predictable marker of an altered spermatogenesis in men submitted to testicular sperm extraction (TESE). In addition, Kumanov et al [14] analyzed InhB as a marker of spermatogenesis in a cohort of 75 infertile men and found a positive correlation between TV and InhB/FSH ratio (r [correlation coefficient]=0.36, p=0.0001 and r=0.35, p=0.002, respectively for right and left TV). However, this correlation was not confirmed in the sub-group of idiopathic infertile men. In this context, current findings depicted that infertile men with a disrupted balance between circulating FSH and InhB levels had lower TV than those with concordant values, thus partially confirming previous findings regarding the relationship between

Table 2. Socio-demographic and clinical characteristics of whole cohort of infertile patients (n=150) with discordant values of FSH and InhB furtherly segregated into both low values *vs*. both high values

Variable	Both low values	Both high values	p-value
Number of patients	78 (6.3)	72 (5.9)	
Cigarette smoking	23 (29.5)	21 (29.2)	0.10
CCI ≥1	7 (9.0)	6 (8.3)	0.87
SDF >30%	15 (83.3)	25 (75.8)	0.03
Azoospermia	30 (41.1)	21 (31.3)	0.002
Oligospermia	30 (69.8)	39 (84.8)	<0.001
Astenozoospermia	33 (80.5)	39 (86.7)	0.02
Teratozoospermia	20 (50.0)	25 (59.5)	0.66
Cryptorchidism	18 (23.1)	23 (31.9)	<0.001
Varicocele	43 (55.1)	37 (51.4)	0.46
Karyotype abnormalities	1 (1.3)	1 (1.4)	0.26

Values are presented as number (%).
FSH: follicle stimulating hormone, InhB: inhibin B, CCI: Charlson comorbidity index, SDF: sperm DNA fragmentation index.

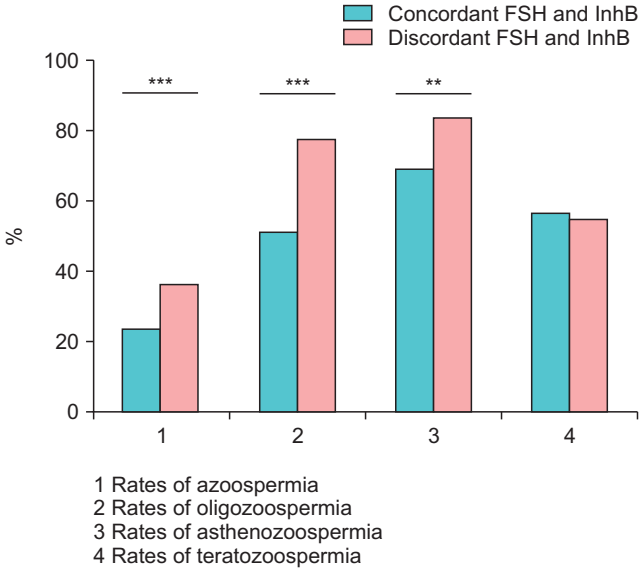

Fig. 1. Differences in semen parameters between concordant and discordant FSH and InhB groups. FSH: follicle stimulating hormone, InhB: inhibin B. Statistically significant (**p≤0.01, ***p≤0.001).

Table 3. Univariable (UVA) and multivariable (MVA) logistic regression analysis showing predictors of discordant FSH and InhB values

Variable	UVA		MVA	
	OR (95% CI)	p-value	OR (95% CI)	p-value
Age (y)	0.98 (0.95–1.01)	0.11	0.98 (0.95–1.00)	0.10
BMI (kg/m^2)	1.06 (1.02–1.11)	0.01	1.08 (1.03–1.13)	0.001
CCI ≥1	1.03 (0.77–1.37)	0.85	1.43 (0.73–2.81)	0.30
Testicular volume (mL)	0.90 (0.87–0.93)	<0.001	0.91 (0.88–0.94)	<0.001
Cryptorchidism	2.77 (1.85–4.15)	<0.001	2.49 (1.59–3.89)	<0.001

FSH: follicle stimulating hormone, InhB: inhibin B, OR: odds ratio, CI: confidence interval, BMI: body mass index, CCI: Charlson comorbidity index.

impaired hormonal values and TV in infertile men.

Oligozoospermia is one of the most common scenario at semen analyses, being observed in up to 35% of infertile men. Kong et al [15] showed that the diagnosis of idiopathic oligozoospermia was highly correlated to serum levels of FSH and InhB as represented by high area under the curve (0.781 and 0.802, respectively). Furthermore, focusing on a population of men with unknown fertility status, Jensen et al [16] depicted a predictive power of 100% in detecting oligozoospermia among men whose InhB and FSH were below 80 pg/mL and above 10 mIU/mL, respectively. Findings from the current study only partially corroborated published data, since infertile men with both high levels of InhB and FSH were more likely to have oligozoospermia. Similarly, Jankowska et al [17] showed that sperm count was positively correlated to InhB levels ($r=0.75$, $p<0.001$) and negatively to FSH levels ($r=-0.46$, $p<0.001$).

Similar findings were found in case of asthenozoospermia. Petrozzi et al [18] showed that sperm progressive motility was negatively correlated to FSH ($r=-0.155$) and positively to InhB ($r=0.095$). Here we observed that patients with discordant FSH and InhB values had a higher proportion of asthenozoospermia compared to those with hormonal concordance.

Azoospermia is considered the most severe form of MFI, accounting for 15% of all cases. In this context, Foresta et al [19] found a significant inverse correlation between FSH and InhB plasma levels in 89 azoospermic subjects ($r=-0.503$, $p<0.0001$). Nonetheless, they reported that InhB serum levels per se did not specifically allow to discriminate between obstructive and non-obstructive forms of azoospermia. Of note, the same inverse correlation between these hormones was observed also in normozoospermic men. In contrast, current findings showed that azoospermia cases were more prevalent in infertile men with discordant FSH and InhB values (Fig. 1), and mostly in infertile men with low levels of both hormones.

In particular, InhB would reflect a complex interaction between FSH, Sertoli, Leydig and germ cells, playing an autocrine or paracrine role in the testis. As such, our findings could reflect a pituitary secretion deficit of FSH associated to a testis with diminished ability to respond. In addition, available literature showed that high InhB levels are related to higher sperm retrieval rates during TESE [20]. Our findings would corroborate even more the importance of considering the balance between FSH and InhB serum levels, more than standalone hormonal levels. Indeed, infertile men with synchronous low FSH and InhB levels depicted higher count of azoospermia, while infertile men with both high hormonal levels had higher count of asthenozoospermia and oligozoospermia.

A high BMI has been associated with male subfertility. Obesity has been linked to abnormal semen characteristics, in particular hormonal abnormalities associated with obesity is likely to play a vital role [21]. Previous data showed that InhB levels are decreased in obese men [22]. As a result of obesity, inflammatory cytokines secreted from fat tissue inhibit testosterone release and the increased leptin level disrupts gonadotropins release. Our findings confirm BMI as a predictive parameter for infertile men also in presence of discordant InhB and FSH values.

SDF is a relatively new predictor of longer time-to-pregnancy and reduced success rate in natural and *in vitro* fertilization [23]. Improper packaging during sperm maturation, post-meiosis defective apoptosis and oxidative stress are usually involved in the etiology of sperm DNA strand breaks [24]. FSH regulates testis development and function, and it exerts anti-apoptotic actions on germ cells [25]. In this context, Garolla et al [26] suggested that FSH treatment improves SDF with subsequent increased pregnancy rates after *in vitro* assisted reproductive techniques. Our results showed a higher rate of SDF ≥30% in men with discordant low values of FSH and InhB, further confirming that hormonal imbalance could impact sperm integrity. As a whole, further research is needed to emphasize the influence of InhB on SDF, and its balance with FSH.

Cryptorchidism is a well-known condition associated with testicular failure which could decrease the number of testicular germ cells if not corrected during the first year of life [27]. Similarly, Gracia et al [28] showed a significant difference in FSH levels between unilateral *vs.* bilateral cryptorchidism (5.9 *vs.* 9 mIU/mL respectively, $p<0.001$). Furthermore, Lee and Coughlin [29] showed that patients with bilateral cryptorchidism had higher FSH and lower InhB levels than men with unilateral disease and healthy controls (17.4 *vs.* 6.7 *vs.* 4.0 U/L and 59.8 *vs.* 112.5 *vs.* 152.5 U/L, respectively, all $p<0.001$). Notably, in our analysis a history of cryptorchidism emerged as an independent predictor for discordant FSH and InhB values. Given the negative association between discordant values and seminal

outcomes, a history of cryptorchidism could be a clinical marker indicating the need to test InhB during the diagnostic work-up of men presenting for primary couple's infertility associated with pure male factors.

Our study is certainly not devoid of limitations. First, even though we examined a homogeneous, same-ethnicity cohort of men presenting for primary infertility, this was a single-center cross-sectional study, thus raising the possibility of a number of selection biases. Second, although having homogeneous data from white-European non Finnish men may only represent a further strength of the analyses, different geographical areas and ethnicity groups might have produced different results; thereof, larger studies across different centers and cohorts are needed to externally validate our findings. Third, given its cross-sectional design, our study did not include a control group of normal fertile men. Fourth, we measured TV through means of an estimate using PO as it is firstly assessed in clinical practice [30]. In order to acquire homogenous data, the same highly-experienced urologist performed all physical examinations. However, we may further validate our data performing testicular ultrasound.

Nevertheless, our results indicate that an accurate investigation of both FSH and InhB levels may be important throughout a better tailored diagnostic work-up of infertile men, mostly in primary infertile couples with severe pure male factors.

CONCLUSIONS

One out of ten men seeking first medical help for primary infertility showed discordant values of circulating FSH and InhB. Men with discordant values were younger, with smaller TV and more frequently reported history of cryptorchidism than those with normal values. Infertile men with discordant FSH and InhB values had worse clinical profile compared to those with concordant ones. Smaller TV and a history of cryptorchidism could be used as further clinical markers to indicate the need to test InhB during the diagnostic work-up of primary infertile men.

Conflict of Interest

The authors have nothing to disclose.

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Author Contribution

Conceptualization: FN, LB. Data curation: FN, LB, SC, EP. Formal analysis: FN, LB. Funding acquisition: FM, AS. Investigation: LB, GF, EV, LC, MR, AS. Methodology: LB, SC, EP, FB, CC. Project administration: AD. Supervision: LB, SC, FM, AS. Validation: LB, SC. Visualization: FM, AS. Writing – original draft: FN, LB, SC. Writing – review & editing: FM, AS.

Data Sharing Statement

The data required to reproduce these findings cannot be shared at this time due to legal and ethical reasons.

REFERENCES

1. Neto FT, Bach PV, Najari BB, Li PS, Goldstein M. Spermatogenesis in humans and its affecting factors. Semin Cell Dev Biol 2016;59:10-26.
2. Meachem SJ, Nieschlag E, Simoni M. Inhibin B in male reproduction: pathophysiology and clinical relevance. Eur J Endocrinol 2001;145:561-71.
3. Radicioni AF, Anzuini A, De Marco E, Nofroni I, Castracane VD, Lenzi A. Changes in serum inhibin B during normal male puberty. Eur J Endocrinol 2005;152:403-9.
4. Minhas S, Bettocchi C, Boeri L, Capogrosso P, Carvalho J, Cilesiz NC, et al.; EAU Working Group on Male Sexual and Reproductive Health. European Association of Urology guidelines on male sexual and reproductive health: 2021 update on male infertility. Eur Urol 2021;80:603-20.
5. World Health Organization (WHO). Human reproduction programme. Sexual and reproductive health: infertility definitions and terminology [Internet]. WHO; c2017 [cited 2014 Dec 31]. Available from: http://www.who.int/reproductivehealth/topics/infertility/definitions/en
6. Salonia A, Matloob R, Gallina A, Abdollah F, Saccà A, Briganti A, et al. Are infertile men less healthy than fertile men? Results of a prospective case-control survey. Eur Urol 2009;56:1025-31.

7. Boeri L, Capogrosso P, Ventimiglia E, Cazzaniga W, Pozzi E, Belladelli F, et al. Testicular volume in infertile versus fertile white-European men: a case-control investigation in the real-life setting. Asian J Androl 2021;23:501-9.
8. Ventimiglia E, Capogrosso P, Boeri L, Pederzoli F, Cazzaniga W, Scano R, et al. When to perform karyotype analysis in infertile men? Validation of the European Association of Urology guidelines with the proposal of a new predictive model. Eur Urol 2016;70:920-3.
9. Hamilton JA, Cissen M, Brandes M, Smeenk JM, de Bruin JP, Kremer JA, et al. Total motile sperm count: a better indicator for the severity of male factor infertility than the WHO sperm classification system. Hum Reprod 2015;30:1110-21.
10. Evenson DP. Sperm chromatin structure assay (SCSA®). Methods Mol Biol 2013;927:147-64.
11. Jerre E, Bungum M, Evenson D, Giwercman A. Sperm chromatin structure assay high DNA stainability sperm as a marker of early miscarriage after intracytoplasmic sperm injection. Fertil Steril 2019;112:46-53.e2.
12. Condorelli R, Calogero AE, La Vignera S. Relationship between testicular volume and conventional or nonconventional sperm parameters. Int J Endocrinol 2013;2013:145792.
13. Tunc L, Kirac M, Gurocak S, Yucel A, Kupeli B, Alkibay T, et al. Can serum inhibin B and FSH levels, testicular histology and volume predict the outcome of testicular sperm extraction in patients with non-obstructive azoospermia? Int Urol Nephrol 2006;38:629-35.
14. Kumanov P, Nandipati K, Tomova A, Agarwal A. Inhibin B is a better marker of spermatogenesis than other hormones in the evaluation of male factor infertility. Fertil Steril 2006;86:332-8.
15. Kong X, Ye Z, Chen Y, Zhao H, Tu J, Meng T, et al. Clinical application value of inhibin B alone or in combination with other hormone indicators in subfertile men with different spermatogenesis status: a study of 324 Chinese men. J Clin Lab Anal 2021;35:e23882.
16. Jensen TK, Andersson AM, Hjollund NH, Scheike T, Kolstad H, Giwercman A, et al. Inhibin B as a serum marker of spermatogenesis: correlation to differences in sperm concentration and follicle-stimulating hormone levels. A study of 349 Danish men. J Clin Endocrinol Metab 1997;82:4059-63.
17. Jankowska K, Suszczewicz N, Rabijewski M, Dudek P, Zgliczyński W, Maksym RB. Inhibin-B and FSH are good indicators of spermatogenesis but not the best indicators of fertility. Life (Basel) 2022;12:511.
18. Petrozzi A, Pallotti F, Pelloni M, Anzuini A, Radicioni AF, Lenzi A, et al. Inhibin B: are modified ranges needed for orchiectomised testicular cancer patients? Asian J Androl 2019;21:332-6.
19. Foresta C, Bettella A, Petraglia F, Pistorello M, Luisi S, Rossato M. Inhibin B levels in azoospermic subjects with cytologically characterized testicular pathology. Clin Endocrinol (Oxf) 1999;50:695-701.
20. Datta AK, Nayini K, Eapen A, Barlow S, Lockwood G. Can we predict the chance of successful epididymal or testicular sperm aspiration following vasectomy? Hum Fertil (Camb) 2016;19:120-6.
21. Incedal Irgat S, Bakirhan H. The effect of obesity on human reproductive health and foetal life. Hum Fertil (Camb) 2022;25:860-71.
22. Aggerholm AS, Thulstrup AM, Toft G, Ramlau-Hansen CH, Bonde JP. Is overweight a risk factor for reduced semen quality and altered serum sex hormone profile? Fertil Steril 2008;90:619-26.
23. Ribas-Maynou J, Yeste M, Becerra-Tomás N, Aston KI, James ER, Salas-Huetos A. Clinical implications of sperm DNA damage in IVF and ICSI: updated systematic review and meta-analysis. Biol Rev Camb Philos Soc 2021;96:1284-300.
24. Aitken RJ, Bronson R, Smith TB, De Iuliis GN. The source and significance of DNA damage in human spermatozoa; a commentary on diagnostic strategies and straw man fallacies. Mol Hum Reprod 2013;19:475-85.
25. Muratori M, Baldi E. Effects of FSH on sperm DNA fragmentation: review of clinical studies and possible mechanisms of action. Front Endocrinol (Lausanne) 2018;9:734.
26. Garolla A, Ghezzi M, Cosci I, Sartini B, Bottacin A, Engl B, et al. FSH treatment in infertile males candidate to assisted reproduction improved sperm DNA fragmentation and pregnancy rate. Endocrine 2017;56:416-25.
27. Rodprasert W, Virtanen HE, Mäkelä JA, Toppari J. Hypogonadism and cryptorchidism. Front Endocrinol (Lausanne) 2020;10:906.
28. Gracia J, Sánchez Zalabardo J, Sánchez García J, García C, Ferrández A. Clinical, physical, sperm and hormonal data in 251 adults operated on for cryptorchidism in childhood. BJU Int 2000;85:1100-3.
29. Lee PA, Coughlin MT. Fertility after bilateral cryptorchidism. Evaluation by paternity, hormone, and semen data. Horm Res 2001;55:28-32.
30. Nieschlag E, Behre HM. Anamnesis and physical examination. In: Nieschlag E, Behre HM, Nieschlag S, editors. Andrology: male reproductive health and dysfunction. 3rd ed. Springer; 2010;93-100.