Original Article

A retrospective cohort study of geographic differences in the semen of 1,012 sperm donors in China

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Background: Male reproductive health has become a serious public health concern, and semen quality is essential to male reproduction. We aimed to investigate geographical differences in the semen quality of sperm donors from northern and southern China by enrolling donors across the country.

Methods: A total of 1,012 sperm donors were enrolled in this study between 2015 and 2019. Donors were first divided into two parts based on their birthplace according to the "Qinling-Huaihe" line, and secondly, by their residential latitude. Finally, donors were re-classified into two groups (typically north and south) which contained 667 samples.

Results: Statistically significant differences in sperm concentration were observed among men from different latitudes in China (P=0.04). The sperm concentrations of males from 18° to 27° north latitude were significantly lower than those from 36° to 45° and 45° to 54° [median 131, 134, and 146, respectively, P=0.021 (18° to 27° *vs.* 36° to 45°) and P=0.01 (18° to 27° *vs.* 45° to 54°)].

Conclusion: We hypothesize environmental pollution and mental stress due to the increased population size may be the main factors underlying differences in the sperm quality of men in northern and southern China.

Keywords: Geographic differences; semen quality; sperm donor; sperm concentration

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Introduction

Male reproductive health and seminal quality have become a serious concern for public health (1). In a recent study, researchers found the sperm quality of Estonian men was better than that of Finnish men in terms of sperm number and normal sperm count (2), although the limited geographical distribution of the results were not sufficient to explain whether they were related to geographical

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differences. Furthermore, these results may have been influenced by different factors such as the recruitment of volunteers, semen analysis, and data processing methods (3). A study performed on semen quality among different provinces and municipalities in Spain showed that even in a particular country with uniform processing methods, there were still geographic differences in semen quality (4). This finding supported the hypothesis that there are regional differences in semen quality. Similarly, a study from Qatar also indicated that even within a specific recruitment agency, differences in semen quality between two regions still existed (5).

However, little has been done to investigate variations in semen quality in Asian settings. Preliminary comparisons conducted by reproductive centers across China highlighted some abnormalities in semen parameters and suggested there was a difference between northern and southern geographical regions. Specifically, abnormal sperm parameters were more frequent in the south than the north, possibly indicating a link with regional pollution (6-8), although this may have been be caused by sampling procedures (9).

Therefore, this study focused on differences in male semen quality in northern and southern China by comparing different latitudes and regional divisions. We present the following article in accordance with the STROBE reporting checklist (available at https://tau.amegroups.com/article/view/10.21037/tau-22-578/rc).

Highlight box

Key findings

- We found sperm concentration was significantly higher in sperm donors living in northern China than in southern China.

What is known and what is new?

- Over the years, there have been many debates about whether there are differences in semen quality between different regions.
- This study focused on differences in male semen quality in northern and southern China by comparing different latitudes and different regional divisions.

What is the implication, and what should change now?

- We hypothesize environmental pollution and mental stress due to increased population size may be the main factors underlying the difference in sperm quality between the northern and southern regions of China.
- We suggest future research should focus on the effects of changes in lifestyle habits, environmental pollution, and psychological factors on male reproductive health.

Methods

Recruitment

Based on the standard protocol for human sperm banking revised by the Ministry of Health of the People's Republic of China, eligible donors from 27 provinces and cities whose sperm was kept in a large sperm bank in Beijing from October 2015 to May 2019 were selected. The inclusion criteria were as follows: (I) aged between 22 and 44 years; (II) no noticeable baldness or hair loss; (III) no color weakness and no color blindness; (IV) the donor was in good health, on the basis of both physical examinations and psychological evaluations, including an examination of the reproductive system; (V) following sperm bank regulations, volunteers had abstained from smoking and alcohol for 3 months prior to official sperm donation; (VI) a rigorous medical screening was conducted, and those with a family history of hereditary disease were excluded; (VII) potential donors had undergone laboratory tests to exclude sexually transmitted infections and high-risk genetic diseases, including human immunodeficiency virus 1 and 2, hepatitis B and C, syphilis, gonorrhea, mycoplasma, chlamydia, cytomegalovirus, toxoplasma gondii, rubella virus, herpes simplex virus types 1 and 2, and karyotype analysis; (VIII) as polymorphisms of the human chromosomes have certain clinical effects associated with abortion, stillbirth, and infertility, normal males with chromosome karyotype 46, XY were chosen.

Semen quality was analyzed by trained technicians in accordance with the standardized methods described in the World Health Organization Laboratory Manual (10). Screening criteria for donor semen quality were based on basic human sperm bank standards and technical specifications, and internal quality control was used to ensure there were no significant differences between the research results derived from different technicians. Semen quality needed to meet the following criteria: (I) all semen samples were fresh and drained into a sterile container following masturbation; (II) the liquefaction time was less than 60 minutes and the sperm concentration was greater than or equal to 60×10^6 mL, the semen volume was more than 2 mL, the proportion of sperm with normal morphology was more than 9%, the proportion of sperm with forward motility was more than 60%, the total motile sperm count was greater than or equal to 12×10^6, and the freeze-thaw survival rate was more than 60%; and (III) the abstinence period of each sperm donor volunteer was 3 days. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013) and approved by the Ethics Committee of Peking University Third Hospital (No.2020SZ-002). Individual consent for this retrospective analysis was waived.

Data collection

Trained sperm bank doctors used structured questionnaires to collect demographic and lifestyle information from each donor. The survey covered date of birth, ethnicity, level of education, marriage status, childbearing history, profession, place of birth, year of semen examination, and abstinence period. Each sperm donor was graded by physical examination, including height and weight. According to World Health Organization (WHO) guidelines, body mass index (BMI) was calculated as weight (kg) divided by height squared (m^2) (11).

Semen collection and semen analysis

All semen samples were collected into a sterile cup by masturbation in a private room at the sperm bank. Samples were then immediately delivered to the sperm bank laboratory through a delivery window in the semen collection room. Aseptic conditions were guaranteed throughout the process, and samples were analyzed by trained clinical technicians in the same laboratory. Liquefied samples were placed in a water bath at 37 ℃ within an hour of collection. Sperm analysis was performed using a computer-aided sperm analysis (CASA, SuiJia Software, Beijing, China) to determine sperm concentration and progressive motility within 1 hour, in accordance with World Health Organization guidelines (fifth edition) (12).

Several semen quality parameters were assessed, including appearance, volume, viscosity, agglutination, liquefaction time, pH value, sperm concentration, sperm morphology, sperm motility, and the proportion of motile sperm. According to a protocol formulated by the World Health Organization (13), sperm volume was determined by drawing up the entire sample into a wide-mouthed (~2 mm) 5 mL disposable calibrated serological pipette (non-pyrogenic) by means of a mechanical device. To assess sperm concentration and motility, 10 µl of mixed semen was placed in a clean Makler chamber (at 37 ℃) and covered gently with a cover glass (14). The sample was then analyzed at a magnification of ×200, with ten of the 100 squares in the microscopic field randomly scanned and the sperm count recorded by a cytometer. Sperm pH was determined by pH test paper then compared with calibration tape. An ocular grid was used to calculate progressive motility and non-progressive motility, and total motility determined. Finally, the total sperm number as semen volume multiplied by sperm concentration was calculated and the total motile sperm count as total sperm number multiplied by total motility was determined (15).

Each semen sample was analyzed twice. An internal quality control was adopted to ensure there were no significant deviations from the results derived from different clinical technicians.

Statistical analysis

All statistical analyses were performed with SPSS 25.0 (SPSS, Chicago, IL, USA). Median and interquartile range (IQR) was used to describe continuous variables, and frequency and percentage described discrete variables. Pearson's correlation coefficient was used to evaluate the relationships between different sperm parameters, and for differences between regions or latitudes, the Mann-Whitney U test was used to test for statistical significance (16). For non-normal distribution, median and interquartile range (IQR) and the Kruskal-Wallis H test were used as appropriate (17). The extensive experimental data set obtained from this study was used to build multiple linear regression models so the effect of confounding variables that might affect semen parameters could be excluded. All analyses involved two-sided tests and $P<0.05$ denoted statistical significance.

Results

A total of 1,012 participants were enrolled between 2015 and 2019. The spatial distribution of participants from 27 provinces in China is shown in *Figure 1*, which shows the provinces with more than 100 participants were Beijing Municipality [177], Hebei [146], and Shandong [104].

The "Qinling Mountain–Huaihe River" line is an essential geological boundary dividing northern and southern regions of China, and this was used to divide participants according to their location (*Table 1*). The mean (IQR) age of all participants was 26 years (23–31 years). Of the 1,012 participants, 752 were from northern China with a median (IQR) age of 27 years (23–31 years) and 360 were from southern China with a median (IQR) age of 24 years (22–29 years). In total, 94% of participants were from the Han ethnic group.

As shown in *Table 2*, progressive motility was significantly

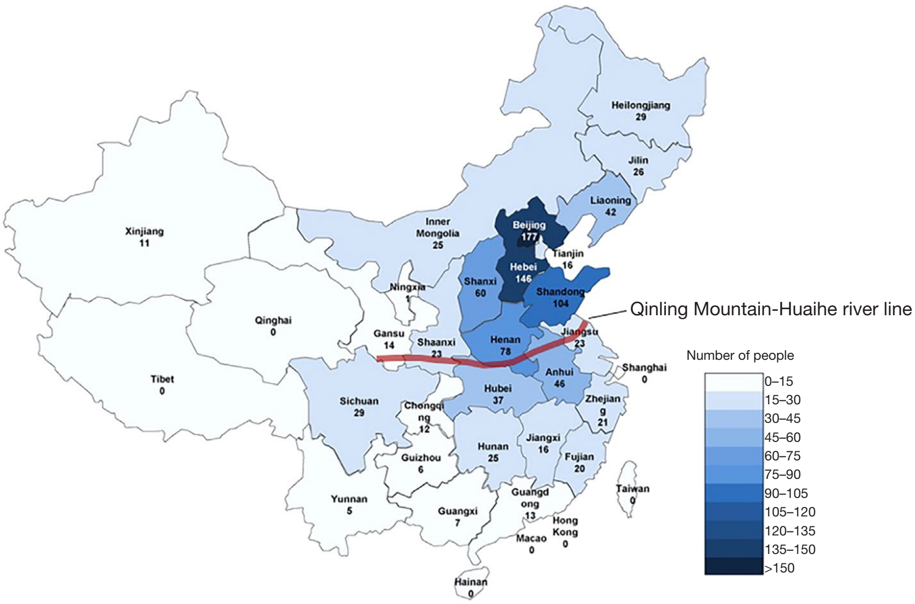

Figure 1 Birthplace of 1,012 donors from 2015 to 2019 in China. Provinces with more than 100 participants included Beijing Municipality [177], Hebei [146] and Shandong [104]. Some provinces had fewer participants [<15], such as Gansu [14], Chongqing [12], Guizhou [6], Yunnan [5], Guangxi [7], Xinjiang [11], and Guangdong [13]. To highlight the main subject, the latitude and longitude of China has been intercepted, resulting in a seemingly incomplete map of the country.

Table 1 Characteristics of the study subjects

Characteristic	Group	Number of subjects			P
		China	Northern China	Southern China	
Age (year), n (%)	22–24	126 (12.5)	85 (11.3)	41 (15.8)	<0.001
	25–29	403 (39.8)	270 (35.9)	133 (51.2)	
	30–39	417 (41.2)	345 (45.9)	72 (27.7)	
	40–44	66 (6.5)	52 (6.9)	14 (5.3)	
	Median (IQR)	26 (23.0–31.0)	27 (23.0–31.0)	24 (22.0–29.0)	
Ethnicity, n (%)	Han	955 (94.4)	703 (93.5)	252 (96.9)	0.038
	Others	57 (5.6)	49 (6.5)	8 (3.1)	
Education, n (%)	Junior college and lower	167 (16.5)	114 (15.2)	25 (9.6)	<0.001
	Undergraduate	612 (60.5)	456 (60.6)	156 (60)	
	Postgraduate and higher	233 (23.0)	182 (24.2)	79 (30.4)	
Marriage, n (%)	Unmarried	769 (76.0)	546 (72.6)	223 (85.8)	<0.001
	Married	243 (24.0)	206 (27.4)	37(14.2)	
Volunteer status, n (%)	Students	399 (39.4)	253 (33.6)	146 (56.2)	<0.001
	Others	613 (60.6)	499 (66.4)	114 (43.8)	
BMI, n (%)	<18.5 kg/m^2	8 (0.8)	5 (0.7)	3 (1.2)	0.009
	18.5–24.9 kg/m^2	812 (80.2)	588 (78.2)	224 (86.2)	
	25–29.9 kg/m^2	192 (19.0)	159 (21.1)	33 (12.6)	

IQR, inter quartile range; BMI, body mass index.

Table 2 Multiple linear regression was used to estimate 95% confidence interval (CI) with adjustment for age, ethnicity, education, smoking, and marital status

Characteristic	Semen parameters (β)					
	SV	SC	PM	FSC	FPM	PSR
Age	−0.017 (−0.227, 0.146)	0.034 (−4.778, 12.148)	−0.022 (−1.420, 0.791)	0.064 (−0.453, 4.681)	−0.024 (−2.719, 1.437)	0.039 (−0.007, 0.022)
Marital status	0.078[b] (0.010, 0.460)	−0.067 (−19.290, 1.077)	−0.053 (−2.274, 0.386)	−0.040 (−4.722, 1.457)	0.006 (−2.299, 2.704)	0.027 (−0.011, 0.024)
Volunteer status	0.008 (−0.179, 0.220)	0.004 (−8.632, 9.478)	−0.106[a] (−2.833, −0.472)	0.017 (−2.135, 3.349)	0.032 (−1.294, 3.146)	0.073 (−0.001, 0.030)
BMI	0.013 (−0.156, 0.239)	−0.005 (−9.726, 8.208)	0.086[a] (0.440, 2.778)	0.001 (−2.661, 2.769)	0.021 (−1.482, 2.915)	−0.019 (−0.020, 0.011)
Ethnicity	0.035 (−0.150, 0.535)	0.028 (−8.467, 22.555)	−0.018 (−2.608, 1.445)	0.021 (−3.138, 6.276)	−0.002 (−3.918, 3.704)	0.034 (−0.012, 0.041)
Education	0.019 (−0.097, 0.175)	0.051 (−1.466, 10.816)	0.032 (−0.419, 1.185)	0.051 (−0.442, 3.285)	0.013 (−1.206, 1.812)	−0.012 (−0.014, 0.007)

[a] and [b] indicate that the analysis of variance was significant at the P value <0.01 and <0.05 level, respectively. SV, semen volume, mL; SC, sperm concentration, 10^6/mL; PM, progressive motility, %; FSC, frozen-thawed sperm concentration, 10^6/mL; FPM, frozen-thawed progressive motility, %; PSR, post-thaw survival rate, %; BMI, body mass index.

and negatively associated with volunteer status and positively associated with BMI, while semen volume was significantly and positively associated with marital status. Statistical analysis showed no significant difference in semen quality between the two regions (*Table 3*, P>0.05). Although most parameters did not differ between the two regions, some showed a strong correlation with each other, and *Table 4* shows the spearman correlation coefficients for these relationships. Semen volume exhibited a convincing negative correlation with sperm concentration (r=−0.239, P<0.001), whereas progressive motility exhibited a convincing positive correlation with sperm concentration (r=0.169, P<0.001).

To further illustrate the regional difference in semen quality, we refined the north and south by excluding remote areas while setting provinces in south-eastern China (including Jiangsu, Shanghai, Zhejiang, Fujian, Jiangxi, and Anhui) and southern China (including Guangdong, Guangxi, Hainan, Hongkong, Macao, and Taiwan) as the south, and provinces in northern China (including Beijing, Tianjin, Hebei, Shanxi, and Inner Mongolia) and northeast (including Heilongjiang, Jilin, and Liaoning) China as the north. This reduced the number of samples to 667 and led to a significant difference in sperm concentration between the two regions (*Table 5*, P=0.015). No significant differences were observed between the two regions in terms of semen volume, progressive motility, post-thaw survival rate, frozen-thawed sperm concentration, and frozen-thawed progressive motility (P>0.05). To further verify this result, we then divided China into four regions by latitude, including regions between 18° to 27° north latitude, 27° to 36° north latitude, 36° to 45° north latitude, and 45° to 54° north latitude, and performed statistical analysis. As shown in *Table 6*, there were statistically significant differences in sperm concentration among men from different latitudes (P=0.04), but no statistically significant differences in semen volume, progressive motility, frozen-thawed sperm concentration, frozen-thawed progressive motility, and post-thaw survival rate. The sperm concentrations of those from 18° to 27° north latitude were significantly lower than those from 36° to 45° and 45° to 54° [median 131, 134 and 146, respectively, P=0.021 (18° to 27° *vs.* 36° to 45°) and P=0.01 (18° to 27° *vs.* 45° to 54°)].

Discussion

Male reproductive health is an area of increasing concern, and whether differences in semen quality exist between different regions remains unclear. In this study, we performed a quantitative correlation analysis of semen quality in 667 sperm donors between northern and southern China. Our data revealed donors living in northern China had significantly higher sperm concentrations than those in southern China. Similarly, there were statistically significant differences in sperm concentration among men from different latitudes. Sperm concentration is an important parameter of semen and is considered an indicator of fertility. Our results are the first to comprehensively describe differences in semen quality between the north and south of China, and these regional differences may have biological significance.

A previous study with a population of healthy Chinese men showed age-related changes were not evident in terms of sperm concentration among different age groups (18). These findings were consistent with a previous meta-analysis performed with a stronger methodology, which

Table 3 Distribution of semen quality parameters (N=1,012)

Characteristic	Northern China (N=752)	Southern China (N=260)	P
Semen volume, mL	3.0 (2.4–4.0)	3.2 (2.4–4.4)	0.142
Sperm concentration, 10^6/mL	134.0 (106.0–168.0)	128.2 (102.3–163.0)	0.155
Progressive motility, %	65.0 (60.0–71.0)	65.0 (60.0–71.0)	0.964
Frozen-thawed sperm concentration, 10^6/mL	71.9 (63.0–84.9)	70.0 (62.0–82.0)	0.379
Frozen-thawed progressive motility, %	53.0 (48.0–58.0)	52.0 (48.0–58.0)	0.223
Post-thaw survival rate, %	0.8 (0.8–0.9)	0.8 (0.7–0.9)	0.256

Data are shown as median (IQR). IQR, inter quartile range.

Table 4 Spearman rank coefficients between semen parameters

	SV	SC	PM	FSC	FPM	PSR
SV						
Spearman Corr.	1	–0.239*	0.019	–0.214*	–0.055	–0.079*
P value	–	<0.001	0.556	<0.001	0.081	0.012
SC						
Spearman Corr.	–0.239*	1	0.169*	0.861*	0.078	–0.056
P value	<0.001	–	<0.001	<0.001	0.013	0.073
PM						
Spearman Corr.	0.019	0.169*	1	0.094*	0.515*	–0.367*
P value	0.556	<0.001	–	0.003	<0.001	<0.001
FSC						
Spearman Corr.	–0.214*	0.861*	0.094*	1	0.067*	0.005
P value	<0.001	<0.001	0.003	–	0.033	0.881
FPM						
Spearman Corr.	–0.055*	0.078*	0.515*	0.067*	1	0.555*
P value	0.081	0.013	<0.001	0.033	–	<0.001
PSR						
Spearman Corr.	–0.079*	–0.056	–0.367*	0.005	0.555*	1
P value	0.012	0.073	<0.001	0.881	<0.001	–

* P value <0.05. Spearman Corr., Spearman correlation coefficient; SV, semen volume, mL; SC, sperm concentration, 10^6/mL; PM, progressive motility, %; FSC, frozen-thawed sperm concentration, 10^6/mL; FPM, frozen-thawed progressive motility, %; PSR, post-thaw survival rate, %.

also found no relationship between male age and sperm concentration, although other important parameters for male fertility, including motility and morphology, were affected by age (19). Our present analysis concurred with these earlier findings in that we found no significant differences between male age and sperm concentration. According to relevant statistics, the distribution of the Chinese population has undergone tremendous changes. In recent years, the population of the north has been on a decline, whereas the south shows an opposite trend (20). This is believed to be related to the migration caused by different distributions of the regional economy (21) and has

Table 5 Distribution of semen quality parameters (N=667)

Characteristic	Northern China (N=521)	Southern China (N=146)	P
Semen volume, mL	3.0 (2.2–4.0)	3.1 (2.4–4.2)	0.189
Sperm concentration, 10^6/mL	134.0 (106.7–170.0)	125.0 (99.5–156.3)	0.015
Progressive motility, %	65.0 (60.0–70.0)	64.0 (60.0–70.0)	0.620
Frozen-thawed sperm concentration, 10^6/mL	71.7 (63.0–86.8)	69.7 (62.0–79.3)	0.090
Frozen-thawed progressive motility, %	53.0 (48.0–58.0)	52.0 (47.8–58.0)	0.140
Post-thaw survival rate, %	0.8 (0.8–0.9)	0.8 (0.8–0.9)	0.248

Data are shown as median (IQR). IQR, inter quartile range.

Table 6 Distribution of semen parameters by different latitude strata (N=1,012)

Characteristic	Latitude (°N)				P
	45–54 (N=29)	36–45 (N=622)	27–36 (N=310)	18–27 (N=51)	
Semen volume, mL	2.6 (2.2–3.6)	3.0 (2.2–4.0)	3.2 (2.4–4.2)	3.4 (2.4–4.6)	0.084
Sperm concentration, 10^6/mL	146.9 (121.5–177.0)[b]	134.0 (105.7–170.0)[b]	131.0 (104.5–163.2)	120.0 (95.1–158.0)	0.040
Progressive motility, %	64.0 (60.0–73.5)	65.0 (60.0–70.0)	65.0 (61.0–71.0)	65.0 (59.0–71.0)	0.758
Frozen-thawed sperm concentration, 10^6/mL	73.0 (61.7–84.8)	72.0 (63.0–86.1)	71.6 (62.0–82.0)	65.0 (60.0–75.3)	0.108
Frozen-thawed progressive motility, %	50.0 (48.0–56.5)	53.0 (48.0–58.3)	52.0 (48.0–58.3)	53.0 (47.0–58.0)	0.464
Post-thaw survival rate, %	0.8 (0.7–0.9)	0.8 (0.8–0.9)	0.8 (0.7–0.9)	0.8 (0.7–0.9)	0.080

Data are shown as median (IQR). [b], Compared with 18-27 °N, P value <0.05. IQR, inter quartile range.

led to a heavier level of pollution. Consistent with previous studies demonstrating the negative impact of pollution on semen quality (22), we observed a higher concentration of sperm in the semen samples from donors living in the north, which is in accordance with the milder levels of pollution in that region. This may also explain the better semen quality in another study of volunteers who lived in rural areas (23), where population density is low. In addition, people tend to suffer from greater psychological burdens in more densely populated areas. The association between mental stress and reduced semen quality is supported by a study that demonstrated biological plausibility (24). Under mental stress, the male endocrine system is interrupted, which manifests as low levels of testosterone and luteinizing hormone, interfering with sperm production and reducing fertility (25). Therefore, mental stress may also account for the regional differences in sperm concentration between northern and southern China. The influence of environmental factors and lifestyle on semen quality may be explained by the changes in fertility patterns brought about by migration (26).

In addition to the factors mentioned above, others such as seasons, ethnicity, dietary patterns, and obesity may also affect semen quality. A previous assessment of male infertility data in the United States showed that compared with Asian men, white males had a higher total sperm number but lower sperm concentration (27). In the present study, only 5.6% of our sperm donors were from ethnic minorities, with most from the Han ethnic group. Thus, due to limited data, we were unable to determine whether the difference in semen quality between northern and southern China was related to ethnicity.

In terms of diet, a cross-sectional study in Spain previously assessed the relationship between dietary patterns and fertility and suggested adherence to a healthy diet was positively correlated with sperm concentration and sperm motility (28). A Chinese study also suggested healthy dietary patterns have beneficial effects on sperm concentration, total sperm count, and progressive sperm motility in males (29). In contrast to the western diet, the traditional Chinese diet is based on grains, and due to differences in latitude and climate, people in southern

China tend to eat rice while those in the north eat flour. At present, there is no literature relating to the effect of rice and flour on semen quality. However, a previous study used reliable blood glucose index (GI) data sources and showed sperm concentration was negatively correlated with the frequency and quantity of grain consumption (30), because grains are rich in starch. When dietary intake leads to an increase in blood sugar, hyperglycemia will increase the risk of inflammation, leading to a decline in semen quality (31). Data relating to the relationship between diet and semen quality or factors related to male infertility, remains limited, and additional prospective studies, including those exploring specific biological mechanisms, are required (32). Therefore, further investigation is needed to identify how differences in the diet structure of southern and northern Chinese men can influence semen quality.

With regard to seasonality, a previous report found the sperm concentration and total sperm count of outdoor workers in summer were significantly lower than from the same workers collected in winter (33). Previous study has also shown spring is associated with higher sperm motility and morphologically normal sperm in comparison to other seasons (34). In our present study, we did not record the season in which donors provided samples and will incorporate this data in future studies.

With regards to obesity, the reduction of fertility in obese men is thought to be linked to changes in the levels of hormones that help form sperm (35). Since obese sperm donors were excluded from the current study, our results are independent of the influence of obesity. A high BMI is known to be associated with negative effects on sperm quality (36), and a previous study indicated differences in the distribution of BMI between adolescents in northern and southern China. Adolescents in the northern regions of China generally have a bulky figure, a phenomenon that is independent of the level of socio-economic development (37). Previous study suggested adaptation to the surrounding environment may cause regional differences in human growth and development (38). Moreover, sufficient sunshine, low annual average temperature, and the seasonal variability in temperature in northern China are all beneficial to the accumulation of body fat (39,40), which may be the reason for the higher BMI of sperm donors in that region.

This was a preliminary study relating to regional differences in the semen quality of sperm donors in northern and southern China. Future investigations are required to validate the geographic variations in larger regions with a more accurate study design to address the issues listed above. More participants from southern China should also be included in future analyses to verify our conclusions and validate the generalizability of our findings. Due to technical limitations, we currently do not have enough data to show the distribution of air pollution on a map.

Although the reasons for regional differences in semen quality remain unclear, lifestyle and environmental factors have been implicated in the decline of sperm quality (41-45). We hypothesize that environmental pollution and mental stress due to an increased population size may be the main factors underlying the difference in sperm quality in men living in northern and southern China.

Conclusions

We identified differences in sperm quality between men living in northern and southern China. Our data revealed sperm donors living in the north of China have significantly higher sperm concentrations than those in the south.

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Footnote

Reporting Checklist: The authors have completed the STROBE reporting checklist. Available at https://tau.amegroups.com/article/view/10.21037/tau-22-578/rc

Data Sharing Statement: Available at https://tau.amegroups.com/article/view/10.21037/tau-22-578/dss

Conflicts of Interest: All authors have completed the ICMJE uniform disclosure form (available at https://tau.amegroups.com/article/view/10.21037/tau-22-578/coif). The authors have no conflicts of interest to declare.

Ethical Statement: The authors are accountable for all aspects of the work in ensuring that questions related to the accuracy or integrity of any part of the work are appropriately investigated and resolved. The study was conducted in accordance with the Declaration of Helsinki (as revised in 2013). The study was approved by the Ethics Committee of Peking University Third Hospital (No.2020SZ-002) and individual consent for this retrospective analysis was waived.

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