INTRODUCTION

Infertility has been a serious social problem in many countries and regions. According to the National Institutes of Health, of all infertility cases, 1/3 are caused by male reproductive problems, and approximately 10%-15% of men who are infertile have a complete lack of spermatozoa. Among infertility couples, the rate of male infertility with or without female infertility factor has reached as high as 50%, and it is likely to continue to rise owing to pathological factors, environmental pollution, and poor lifestyles. At present, the overall trend of semen quality in men worldwide is still controversial, but quality decline in local areas has been recognized. From the perspective of regional disease prevention and control, it is of great significance to grasp the distribution characteristics and trend of semen quality of infertile male patients. There are many epidemiological studies on semen quality in male infertility patients, mainly focusing on semen quality and factors influencing it. However, there are relatively few studies on the long-term trend of semen quality in patients with male infertility because of a lack of many samples and long series of data.

Wenzhou is a prefecture-level city having 9 million people in the south of Zhejiang province, eastern China. It is one of the economically developed areas in China that ranks 49th in comprehensive competitiveness Ranking of China cities. It is also one of the typical areas where environmental protection has lagged behind economic development for a long time. With the continuous advancement of urbanization and industrialization, a trend of low fertility and aging is intertwined. According to the Wenzhou Statistical Yearbook, the birth rate in 2018 decreased by 2.2/1000 compared with that in 2017. One of the underlying reasons is the increase in the incidence of infertility.

The main goal of this article was to analyze the distribution of infertile male patients, identify the population differences of semen quality and changing trends, and provide a basis for local governments and health management departments to improve residents’ reproductive health.

PATIENTS AND METHODS

The Center for Reproductive Health of The First Affiliated Hospital of Wenzhou Medical University began to establish a database of male infertility patient samples and medical records from 2006 and has accumulated a long-term series of datasets. Infertile male patient, is defined as the wife possesses normal reproductive capability but is not becoming pregnant after 1 year's continual unprotected intercourse. In this study, datasets from January 2008 to November 2016 were used. Patients whose cohabitation life and family history were normal after...
marriage were excluded from the study, as were those with organic diseases and female infertility. In general, a total of 38 905 male infertile patients were screened, with an average age of 32.3 years (standard deviation [s.d.]: 6.3). All the data from The First Affiliated Hospital of Wenzhou Medical University database were de-identified, and the extracted data did not require informed consent. Besides, the study was conducted in accordance with the Declaration of Helsinki (as revised in 2013) and was approved by the Ethics Committee of The First Affiliated Hospital of Wenzhou Medical University (issuing No. 2020160).

All patients were required to abstain from sexual intercourse for 3–7 days before semen provision. Semen was collected by masturbation in a clean cup in a specific semen collection room of the hospital and placed in a 37°C incubator for liquefaction. The degree of liquefaction was checked every 10 min, and the test was performed immediately after the specimen was liquefied. If the semen had not liquefied after 60 min, the sample was mechanically pipetted and mixed before analysis. Semen volume, color, viscosity, and liquefaction time were assessed according to the method of the WHO Laboratory Manual for the Examination and Processing of Human Semen (4th and 5th Edition;12,13 there is no difference between the two editions in methodology). The computer-assisted semen quality analysis system was SCA–H–01 (MICROPTIC S.L. Company, Viladomat, Barcelona, Spain). In order to make a unified judgment standard, we used the WHO 5th reference values.13 In this study, semen is defined as normal if it meets all the following requirements: semen volume ≥1.5 ml, sperm concentration ≥15 × 10⁶ ml⁻¹, total forward motile spermatozoa ≥40% or fast forward motile spermatozoa ≥32%, spermatozoa with normal morphology ≥4%, and liquefaction time ≤60 min, whereas semen is defined as abnormal when it did not meet one or more of these requirements. The characteristics of abnormal semen were classified as low semen volume (semen volume <1.5 ml), asthenozoospermia (total forward motile spermatozoa <40% or fast forward motile spermatozoa <32%), and teratozoospermia (normal sperm morphology <4%). We defined the abnormality of two or more of the above-stated four criteria as multi-abnormal sperm disease (MSD).

Statistical analysis was performed in R programming language for statistical computation (the R foundation, version 3.4.3, St. Louis, MO, USA). The comparison of measurement data between groups used single-factor analysis of variance and the Student–Newman–Keuls (SNK) post-hoc test, while the variance was tested as homogeneity; otherwise, Kruskal–Wallis multiple comparison was used. Time trends of semen quality and semen abnormalities were analyzed by means of linear regression models.

RESULTS

Distribution of the patients

We collected four common semen quality parameters of the 38 905 patients and made a descriptive statistic (Supplementary Table 1). Among the 38 905 infertile male patients, those with normal semen quality accounted for 24.9%. For age distribution, the proportions of age ≤25 years, 25 years < age ≤30 years, 30 years < age ≤35 years, 35 years < age ≤40 years, and age >40 years were 6.4%, 32.9%, 33.5%, 18.3%, and 9.0%, respectively. For occupational distribution, workers, peasants, intellectuals, businessmen, and others accounted for 87.6%, 2.0%, 6.5%, 3.4%, and 0.4%, respectively. For spatial distribution, peasants, intellectuals, businessmen, and others accounted for 87.6%, 5.8%, 3.4%, and 3.2%, respectively. For time distribution, the number of patients showed a linear upward trend (r² = 0.96, P < 0.001) during 2008–2016, with an average annual increase of about 647 cases (Supplementary Figure 1).

Overall situation and group differences in semen quality

Among the different age groups, the mean values of percentage of fast forward motile spermatozoa, total forward motile spermatozoa, spermatozoa with normal morphology, and semen volume for patients over 40 years old were significantly different from those of the other age groups (Table 1). Among the different occupational groups, the mean values of percentage of fast forward motile spermatozoa, total forward motile spermatozoa, and spermatozoa with normal morphology for workers were significantly different from those of the other occupational groups (Table 2).

Changes of semen quality with time

Figure 1 shows the semen quality of the 38 905 patients during 2008–2016. The annual mean percentage of fast forward motile spermatozoa and percentage of total forward motile spermatozoa for peasants continued to decline from 44.7% to 58.9% in 2008 to 31.3% and 45.0% in 2016, respectively. The annual mean percentage of spermatozoa with normal morphology decreased from 10.1% in 2008 to 4.2% in 2011, and then decreased to 3.4% in 2016. The annual mean semen volume increased from 2.9 ml in 2008 to 3.1 ml in 2016. Univariate linear regressions were performed with the annual average of each parameter as the dependent variable and year as the independent variable. During 2008–2016, the annual mean percentage of fast forward motile spermatozoa, percentage of total forward motile spermatozoa, and percentage of spermatozoa with normal morphology decreased linearly with slopes of −2.11, −2.59, and −0.70, respectively (Figure 2a–2c). However, the annual mean semen volume had no significant linear time trend (P = 0.39; Figure 2d).

Symptoms of the patients

For all the patients, low semen volume, asthenozoospermia, and teratozoospermia accounted for 8.4%, 50.5%, and 54.1%, respectively. The proportion of patients with teratozoospermia increased from 9.9% in 2008 to 64.9% in 2016 (Figure 3). The proportion of patients with asthenozoospermia and MSD increased from 26.7% and 8.9% in 2008 to 61.7% and 47.8% in 2016, respectively. The proportion of patients with low semen volume had a linear downward trend, from 12.4% in 2008 to 8.5% in 2016.

Similarly, a univariate linear regression equation was established with the proportion of patients with each symptom as the dependent variable and time as the independent variable. All the models showed significant linear relationships in results, and the r² values ranged from 0.73 to 0.97 (Figure 4). The proportion of patients with asthenozoospermia and MSD increased from 2008 to 2016 with
Table 1: Comparison of semen quality in infertile male patients of different age groups

| Parameter                                      | Age ≤ 25 years (n=2472) | 25 years < age ≤ 30 years (n=12 783) | 30 years < age ≤ 35 years (n=13 047) | 35 years < age ≤ 40 years (n=7116) | Age > 40 years (n=3487) |
|------------------------------------------------|-------------------------|--------------------------------------|--------------------------------------|-----------------------------------|------------------------|
| Percentage of fast forward motile spermatozoa (%) | 36.00* (24.70, 46.10)   | 36.40* (25.30, 46.80)                | 36.80* (26.00, 47.00)               | 35.60* (24.60, 45.70)             | 33.4* (21.25, 43.65)   |
| Percentage of total forward motile spermatozoa (%) | 50.80* (39.30, 40.80)   | 51.80* (63.40, 60.30)                | 52.10* (61.30, 60.70)               | 50.60* (62.10, 59.40)             | 47.7* (54.80, 59.40)   |
| Percentage of sperm with normal morphology (%)   | 3.40* (1.60, 5.60)      | 3.50* (1.60, 6.10)                   | 3.80* (1.80, 6.50)                  | 3.60* (1.80, 6.20)                | 3.40* (1.60, 5.70)     |
| Semen volume (ml), mean (s.d.)                   | 3.17* (0.35)            | 3.10* (1.79)                         | 2.97* (1.43)                        | 2.89* (1.87)                    | 2.71* (1.31)           |

Table 2: Comparison of semen quality in infertile male patients of different occupational groups

| Parameter                                      | Worker (n=34 094) | Peasant (n=778) | Intellectual (n=2529) | Businessman (n=1319) | Others (n=175) |
|------------------------------------------------|-------------------|----------------|----------------------|----------------------|---------------|
| Percentage of fast forward motile spermatozoa (%) | 35.90* (24.70, 46.20) | 38.60* (28.57, 49.17) | 34.80* (24.00, 45.00) | 40.00* (29.70, 51.95) | 40.00* (28.05, 49.00) |
| Percentage of total forward motile spermatozoa (%) | 51.20* (40.0, 62.70) | 54.64* (43.42, 63.33) | 49.40* (38.30, 61.00) | 56.40* (44.70, 68.45) | 54.80* (41.95, 66.60) |
| Percentage of sperm with normal morphology (%)   | 3.5* (1.70, 6.10)  | 4.00* (2.10, 7.00) | 3.50* (1.70, 6.00) | 5.4* (2.55, 8.90) | 5.40* (2.30, 7.90) |
| Semen volume (ml), mean (s.d.)                   | 2.99* (1.6)       | 3.00* (1.43)     | 3.00* (2.39)         | 2.80* (1.29)       | 2.80* (1.25)    |

Figure 2: Linear models between the annual mean values of the four semen quality parameters and time. (a) The percentage of fast forward motile spermatozoa, (b) the percentage of total forward motile spermatozoa, and (c) the percentage of spermatozoa with normal morphology were declining year by year and showed a linear trend. (d) Semen volume was increasing year by year and showed a linear trend.

slopes of 4.70 and 4.87, respectively. The proportion of patients with teratozoospermia increased from 2008 to 2011 and from 2011 to 2016 with slopes of 17.10 and 2.09, respectively, while for low-semen disease, it decreased with a slope of −0.47 in the same time period.

DISCUSSION

Semen quality is an important reference index for evaluating male fertility, which is of great significance for the diagnosis, treatment, and prevention of male infertility. There are many factors that affect male semen quality, such as occupation (polluting workshops, long-term driving, etc.), environment (air pollution, environmental endocrine disruption, etc.), and lifestyle (smoking, alcohol abuse, staying up late, etc.). The semen quality of male workers who were infertile in Wenzhou was significantly lower than that of the rest, especially for sperm motility. The workers in this study are mainly drivers, renovation workers, stone practitioners, and general staff and a majority are shoe factory workers. Owing to the nature of the work, these people are often exposed to the reported risk factors of infertility. The current articles have reported that ionizing radiation, radiant heat, inorganic lead, the fungicide ethylene dibromide, and ethylene glycol ethers have become established male reproductive toxicants in humans in the last 30 years. Wang et al. revealed the adverse effects of formaldehyde exposure on semen quality, especially on sperm motion parameters. The shoe-making industry is flourishing in Wenzhou, and a large...
The main influencing factors of the health can be divided into three categories of genetic factors, environmental factors, and lifestyle habits, that also influence semen quality. Genetic factors are congenital and are impossible to change, but we can start with acquired factors. Among the different populations in Wenzhou, the semen quality of male infertile patients in the worker group was significantly worse than that of other occupational groups, and the semen quality of male infertile patients in the 40-year-old group was significantly worse than that of other age groups. For government departments, it is necessary to focus on the worker group for occupational disease prevention and control, to manage industrial enterprises in accordance with laws and regulations, and to create a good working environment for workers. For male infertility patients, they should actively create a good working and living environment and cultivate good living habits. This is the most significant both to them and to their family and future generations.

Reproductive health is closely related to improving the quality of life. It is a prerequisite for a happy life, and it is the basis for social stability and economic development. The main purpose of this article is not to study the influencing factors of semen quality of male infertility patients, but to focus on revealing the population differences and time trends of semen quality, and to provide decision-making basis for regional reproductive health management. Wenzhou has been intertwined with a low birth rate and an aging population, and the semen quality of male infertility patients has shown a significant decline overall. Reversing this situation requires the joint efforts of the government, health and disease control departments, academia, business, and individuals, which will be a long and arduous task.

Substantial limitations apply to our study, which suffer from the lack of specific information on occupation, especially the history of poison exposure.

AUTHOR CONTRIBUTIONS
ZGW, WKC, and QJF drafted the manuscript. ZGW, YLL, and XDL collected data and performed the statistical analysis. XJS and HH conceived the study and participated in its design and coordination and helped to draft the manuscript. All authors read and approved the final manuscript.

COMPETING INTERESTS
All authors declare no competing interests.

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REFERENCES
1. Sharpe RM. Lifestyle and environmental contribution to male infertility. Br Med Bull 2000; 56: 630-42.
2. Chen WH, Fang XW, Chen J. (Quality analysis of semen in infertility men in Zhongshan area). Chin J Birth Health Hered 2013; 21: 137-8. [Article in Chinese].
3. Andolz P, Bielsa MA, Vila J. Evolution of semen quality in North-eastern Spain: a study in 22 759 infertile men over a 36 years period. Hum Reprod 1999; 14: 731-5.
4. Pal PC, Rajalakshmi M, Manocha M, Sharma RS, Mittal S, et al. Sperm quality and sperm functional parameters in fertile Indian men. Andrologia 2006; 38: 20-5.
5. Gu YQ. Variation trend of male fertility and semen parameters. Zhonghua Nan Ke Xue 2014; 20: 1059-62. [Article in Chinese].
6. Zhang YL, Wu QY, Shi SY. (2794 cases of infertility semen analysis in Changsha). Chin J Modern Med 2012; 12: 56-8. [Article in Chinese].
7. Chen XX, Cun JT, Li LH. (Investigation on the sperm quality of 1661 males in Kunming). Med Pharm Yunnan 2015; 36: 25-7. [Article in Chinese].
8 Laven JS, Haverkorn MJ, Bots RS. Influence of occupation and living habits on semen quality in men (scrotal insulation and semen quality). *Eur J Obstet Gynecol Reprod Biol* 1988; 29: 137–41.

9 Fu L, Zhang HB, Mao XG. [Correlation between age and semen parameters analyses results of 5405 infertile males in Southern Sichuan]. *Sichuan Med J* 2015; 36: 14–7. [Article in Chinese].

10 Yang HJ, Hui Y, Shi SP. [Influencing factors of male infertility in rural areas in Zunyi: a case-control study]. *J Environ Health* 2016; 33: 223–6. [Article in Chinese].

11 Mo JY, Zhao GL, Li SH. A single center study on male semen quality in infertility clinic during 2005-2017. *J Reprod Med* 2018; 27: 368–71.

12 World Health Organization. WHO Laboratory Manual for the Examination of Human Semen and Semen-Cervical Mucus Interaction. 4th ed. Cambridge: Cambridge University Press; 1999.

13 World Health Organization. WHO Laboratory Manual for the Examination and Processing of Human Semen. 5th ed. Geneva: WHO Press; 2010.

14 Jensen TK, Bonde JP, Joffe M. The influence of occupational exposure on male reproductive function. *Occup Med (Lond)* 2006; 56: 544–53.

15 Wang HX, Li HC, Lv MQ, Zhou DX, Bai LZ, et al. Associations between occupation exposure to formaldehyde and semen quality, a primary study. *Sci Rep* 2015; 5: 15874.

16 Katukam V, Kulakami M, Syed R, Alharbi K, Naik J. Effect of benzene exposure on fertility of male workers employed in bulk drug industries. *Genet Test Mol Biomarkers* 2012; 16: 592–7.

17 Li CJ, Tzeng CR, Chen RY. [Decline in semen quality in men in Northern Taiwan between 2001 and 2010]. *Chin J Physiol* 2016; 59: 355–65. [Article in Chinese].

18 Jurewicz J, Hanke W, Radwan M, Bonde JP. Environmental factors and semen quality. *Int J Occup Med Environ Health* 2009; 22: 305–29.

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Supplementary Table 1: Descriptive statistics on the four common semen quality parameters

| Group                                      | Normal group (n=9686) | Abnormal group (n=29218) | Total (n=38905) |
|--------------------------------------------|-----------------------|--------------------------|-----------------|
| Percentage of fast forward motile spermatozoa (%) | 48.10 (41.90, 55.10)  | 31.30 (25.00, 46.40)     | 36.10 (25.00, 46.40) |
| Percentage of total forward motile spermatozoa (%) | 64.80 (57.80, 72.05)  | 45.90 (34.40, 56.20)     | 51.30 (40.10, 62.90) |
| Percentage of sperm with normal morphology (%) | 6.90 (5.30, 9.50)     | 2.60 (1.30, 4.30)        | 3.60 (1.70, 6.20)  |
| Semen volume (ml)                          | 3.11 (1.28)           | 2.95 (1.75)              | 2.99 (1.65)      |

Data of percentage of fast forward motile spermatozoa, percentage of total forward motile spermatozoa, and percentage of spermatozoa with normal morphology presented as median (first quartile, third quartile), while for semen volume presented as mean (s.d.). s.d.: standard deviation

Supplementary Figure 1: The number of patients (2008–2016).