Semen characteristic profiles of men of different ages and duration of infertility

Kamala Gopalkrishnan†, Varsha Padwal, Donta Balaiah, P. Meherji, J. Gokral and R. Shah*

Department of Electron Microscopy, Institute for Research in Reproduction (ICMR), Parel, Mumbai 400 012, India
*Bhatia General Hospital, Mumbai 400 007, India

Semen characteristics were evaluated in men (n = 660) attending the infertility clinics, *vis-à-vis* age and duration of infertility. Men were divided into three groups according to age: 20–29 (n = 148), 30–39 (n = 437), > 40 (n = 75). Duration of infertility was grouped as follows: 1–5 years (n = 317), 6–10 years (n = 223), > 10 years (n = 120). The routine semen analysis was done according to WHO manual. Various semen parameters like semen volume, sperm count, motility, morphology and viability were evaluated. Values showed a decrease with increase in age and duration of infertility. It appears from the study that age and duration of infertility contributed to the decline in semen quality in men over the age of 40 years in the population studied. Thus these two parameters may possibly act as indicators to the quality of semen, specially in infertile men.

DATA on the reproductive function of men of different ages, specially of older men are rare. Regarding the reproductive and endocrine functions in ageing males, many questions still remain unanswered. Representative data in semen parameters are very few for ageing men. The study of Nieschlag *et al.*[1] have shown a significant decrease in sperm motility in older men along with ejaculate volume and fructose concentration. Only a small percentage of patients attending the infertility clinics are older than 40 years. It is well established that very few children are born to marriages in which husband is beyond the age of 60 years. This might be interpreted as an indication of reduced reproductive capacity with increasing age not only for the female partner but also for the male. Relationship between men's age and fecundity is difficult to approach directly due to many confounding factors like contraception, conception, abstinence, infection, degree of sexual activity, etc. It is well documented that in women with increasing age, there is a decrease in fertility[2]. An indirect but useful contribution regarding the relationship between age and fecundity as well as duration of infertility of the couple can be made by evaluating the influence of these on semen parameters. Thus retrospective analysis of the

†For correspondence. (e-mail: dirirr@vsnl.com)

data was done with respect to age and duration of infertility *vis-à-vis* semen characteristics. The objective was to evaluate semen characteristics profile in men with respect to age and duration of infertility.

This is a retrospective study done on the semen analysis data obtained from men ($n = 660$) enrolled for different programmes of our Institute *vis-à-vis* age and duration of infertility from 1986 to 1998. Only cases assessed for all five semen parameters, viz. volume, count, motility, morphology and viability were selected for the study. Men were divided into three groups according to age [group I (20–29 yrs), II (30–39 yrs) and III (> 40 yrs)] and according to duration of infertility [group I (1–5 yrs), II (6–10 yrs) and III (> 10 yrs)]. Apart from this, from the same study population, semen parameters were compared in age groups of < 40 yrs ($n = 585$) > 40 yrs ($n = 75$).

Ejaculates were obtained by masturbation after 3–5 days of abstinence. The ejaculates were allowed to liquify at room temperature and the analysis was done according to WHO manual[3]. The following parameters were evaluated: Semen volume, sperm count, motility, morphology and viability. Sperm morphology was evaluated on Papanicolaou stained smears with critical attention to all parameters including actual dimensions. Viability of spermatozoa was assessed by using trypan blue dye exclusion test.

Student's *t* test for testing the difference between the two groups and one-way analysis of variance with Scheffe's multiple range test were used for testing the difference between the three groups. Correlation analysis has been carried out to understand the strength of association between groups.

The distribution of men according to age and duration of infertility shows that majority of cases were between the ages of 30 and 39 years (67.7%). The number of cases ($n = 223$) with duration of infertility 6–10 yrs were approximately 34% in the population of men studied. 48% men had duration of infertility between 1–5 years.

Age and duration of infertility were significantly correlated in group II and III as compared to group I (Table 1). With respect to seminal parameters, volume and viability were not significantly different in all the groups. Significant differences were found in sperm count in group I as compared to the other groups. Differences in motility and morphology were significant ($P < 0.05$) when group I was compared with groups II and III. It was obvious to find significant increase in duration of infertility with increase in age.

Significant correlation was found in sperm count between groups II and III as compared to group I ($P < 0.05$). Sperm motility and morphology were significantly correlated in different groups, i.e. group I with II and III. The number and percent of cases showing normal semen parameters in different groups of age and duration of infertility was analysed.

Duration of infertility and age were significantly correlated in groups II and III as compared to group I ($P < 0.05$) (Table 2). When analysed according to dura-

Table 1. Mean (± SD) values of semen parameters with respect to different age groups

Parameters	20–29 yrs Group I $n = 148$	30–39 yrs Group II $n = 437$	> 40 yrs Group III $n = 75$
Volume (ml)	2.9 ± 1.36	2.95 ± 1.45	3.16 ± 2.00
Count (10^6/ml)	29.95 ± 21.96	44.30 ± 40.83*	66.95 ± 56.92*
Motility (%)	50.75 ± 13.71	52.95 ± 16.56*	56.38 ± 15.08*
Morphology (%)	9.95 ± 9.98	13.54 ± 11.33*	16.29 ± 12.48*
Viability (%)	65.91 ± 16.97	62.06 ± 19.81	62.94 ± 18.84
Duration of infertility (years)	4.31 ± 2.31	7.37 ± 3.66*	12.25 ± 4.75*

*$P < 0.05$.

Table 2. Mean (± SD) values of semen parameters for different groups with varying duration of infertility

Parameters	1–5 yrs Group I $n = 317$	6–10 yrs Group II $n = 223$	>10 yrs Group III $n = 120$
Volume (ml)	2.72 ± 1.33	3.09 ± 1.51*	3.18 ± 1.83*
Count (10^6/ml)	36.94 ± 28.57	48.56 ± 47.67*	54.27 ± 50.31*
Motility (%)	53.11 ± 15.53	52.89 ± 16.67	53.50 ± 15.64
Morphology (% normal)	12.08 ± 10.33	13.82 ± 12.29	14.76 ± 11.49
Viability (%)	64.60 ± 18.90	62.31 ± 18.54	60.79 ± 20.86
Age (years) Mean (SD)	31.62 ± 4.01	34.88 ± 4.24*	38.61 ± 4.43*

*$P < 0.05$.

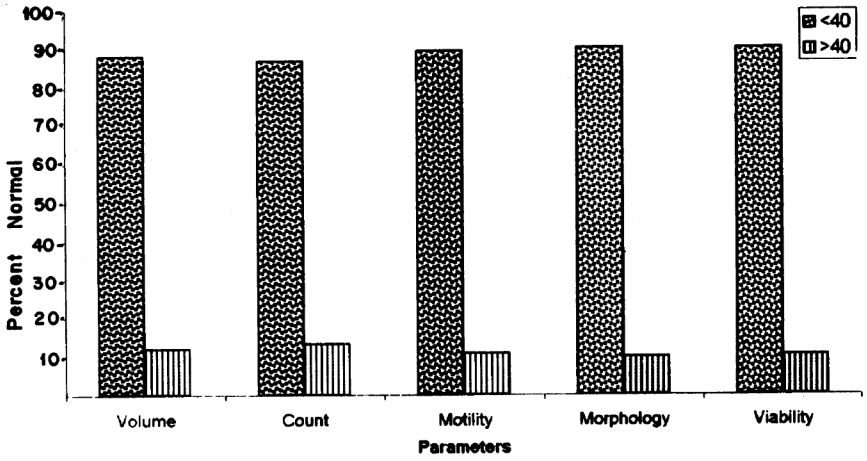

Figure 1. Percent normal of different semen parameters in the men of age groups of < 40 and > 40 years.

Table 3. Correlation of age and duration of infertility with various seminal parameters (n = 660)

Parameters	Age (r value)	Duration of infertility (r value)
Age	1.00	0.59**
Duration of infertility	0.59**	1.00
Volume	0.01	0.13**
Count	0.30**	0.20**
Motility	0.17**	0.11*
Morphology	0.14**	0.11*
Viability	0.05	0.04

*P < 0.01; **P < 0.001.

tion of infertility with respect to seminal parameter, the results showed that mean volume and sperm count were significantly different in the groups II and III as compared to group I. Sperm motility, morphology and viability were not significantly different. No changes were noticed in the mean percent normal forms and percentage of viability.

The number and cases showing percent normal of semen parameters in different group of duration of infertility was analysed. The percent normal for different semen parameters was on the basis of normal values for the population, viz. sperm count ≥ 20 mill/ml, motility ≥ 50%, morphology ≥ 14% and viability ≥ 50%. The percent normal for each parameter with respect to age shows that in the age group of 30–39 years, the number of subjects were highest. Whereas with respect to duration of infertility, percent normals in semen volume were highest in group III. Except for the sperm count the percent normals with respect to motility, morphology and viability were higher in group I with duration of infertility.

Correlation of age and duration of infertility with other seminal parameters is given in Table 3, significant correlation (r value) was seen between age and the seminal parameters like sperm count, motility and morphology along with duration of infertility.

Percentage of cases in two different age groups (< 40 and > 40 years) and two different durations of infertility (< 5 and > 5 years) shows that 585 cases were < 40 years in which majority, i.e. 95.71% had duration of infertility < 5 years. There were 75 men of > 40 years of age and majority of these men had duration of infertility more than 5 years (90.24%).

The seminal parameters in the group of < 40 years (n = 585) and > 40 years (n = 75) showed significant difference only for sperm count and motility (P < 0.01). The percent normal of semen parameters showed significant lower values in > 40 years group (Figure 1).

The study was undertaken to evaluate the possible age-dependent influences on male infertility and seminal parameters. In this study, the majority population belonged to the age group of 30–39 years, of which 95.7% had duration of infertility less than five years. This probably is due to the age at marriage and trying time. There was a good correlation between age and duration of infertility. The present results are in accordance with the study of Merino and Carranza-Lira *et al.*[4] in which only small percentage of men attending the infertility clinics are older than 40 years. It is known that many of the andrological parameters are age related[5–7]. Controversial reports exist on the changes in semen parameters with reference to age[6,8–10]. Robertson *et al.*[11] have reported a gradual decline of male fertility associated with age.

Since the number of males > 40 years referred to infertility clinics are lower, the analysis grouped the findings into two major groups, of > 40 and < 40 years. The percent normal of all seminal parameters were significantly higher in < 40 years age group as compared to > 40 years, indicating the possible effect of age apart from duration of infertility, the age of the partner and other compounding factors. Large epidemiological studies have shown a general mean decline in serum testosterone levels in older men but the variation is large[12,13]. Whether this in turn will affect the reproductive health parameters needs a prospective study.

The changes in semen parameters can be explained on the basis of possible lower ejaculatory frequency. In this study, information on frequency of sexual intercourse was not available. Whether changes in ejaculate volume is associated with reduced testosterone level or related to secretory activity is not known. The changes observed in the semen parameters with increasing age may, however also reflect the natural ageing of gonad. An interesting observation has been made by Takizawa and Hatakeyama[14] regarding important microvascular changes at testicular level appearing often during the 5th decade. The consequence of these vascular modifications is a reduction in blood supply and alteration in the function of seminiferous tubule and epididymis[15]. The present study indicates significant alteration in the semen parameters. Similar findings have been reported[16,17]. Further, Nieschlag and Michel[18] have reported no change in sperm function using hamster oocyte penetration test in men from older group. Since there were no changes in sperm morphology and viability, the above results are explainable. With the lower general health in older group, the reproductive health also goes down and specially in cases of older infertile men, the semen volume and fructose concentration were also found to be lower. Schwartz *et al.*[10] have reported a gradual decrease in sperm concentration with increasing age. Andersen[19] observed significant drop in fertility from the age of 42 years. This is the age group (> 40 years) where most noticeable changes have been seen in the present study. These may also be influenced/accelerated by environmental factors.

Longer period of abstinence can lead to higher output and lower motility[20,21]. In the present study, this variable could not be affecting the parameters since abstinence interval was laid down as 3–5 days as per WHO criteria. The mean (± SD) values of seminal parameters show a significant increase in sperm count and motility in > 40 years group ($p < 0.01$). The other parameters like volume, morphology and motility did not show much difference. Since there is a large variation between values, the data was analysed on the basis of percent normal. This gives much more information as to how many were within 'normal' or *vice versa*. Similarly, for the semen parameters in different groups of age, group II (between 30–39 years) had majority of cases falling under normal. The percentage normals in group III was the least which shows the possible age as one of the factors. In duration of infertility, there were no major changes between groups with regard to percent normal of semen parameters. It must be taken into consideration that even with the reduced semen parameters, the possibility of fathering a child still remains[22]. Reduced semen parameters only lessen the possbility of achieving pregnancy. This has more significance in an infertile population. Along with this, the age of female partner also progresses which is a prognostic factor for unexplained infertility[23–25].

In conclusion, the observed changes in fertility are not as marked as in the female, yet the results of the study suggest that age and duration of infertility may contribute to quality of semen specially in the infertile group. These two parameters may act as possible pointers towards semen quality.

1. Nieschlag, E., Lammers, U., Freischemc-W., Langer, K. and Wickings, E. J., *J. Clin. Endocrinol. Metabol.*, 1982, **55**, 676–681.
2. Ashkenazi, J., Orvieto, R., Gold-Dentch, R., Feldberg, D., Dicker, D., Volioviitch, I. and Ben-Rafael, Z., *Eur. J. Obstet. Gynecol. Reprod. Biol.*, 1997, **66**, 155–159.
3. World Health Organization, *Who Laboratory Manual for the Examination of Human Semen and Semen-cervical Mucus Interaction*, Cambridge University Press, Cambridge, 1992, 3rd edn.
4. Merino, G. and Carranza-Lira, S., *Arch. Androl.*, 1995, **35**, 219–224.
5. Macleod, J. and Gold, R. Z., *Fertil. Steril.*, 1953, **4**, 194–207.
6. Bujan, L., Miensset, R., Mondinat, C., Mansat, A. and Pontonnier, F., *Andrologia*, 1998, **20**, 121–128.
7. Jouannet, P., Ducot, B., Feneux, D. and Spira, A., *Int. J. Androl.*, 1988, **11**, 379–394.
8. Chukudebelu, W. O., *Int. J. Fertil.*, 1978, **23**, 238–239.
9. Singer, W. G., Armitage, J. O. and Schmidt, M. A., *JAMA*, 1980, **244**, 789–790.
10. Schwartz, D., Mayaux, M. J., Spira, A., Moscato, M. L., Jouannet, P., Czygliu, F. and David, G., *Fertil. Steril.*, 1983, **39**, 530–535.
11. Robertson, S. B., Birell, W., Itzkowie, D. and Tischler, G., An Australian non-resource paper presented at the Tenth World Congress of Gynecology and Obstetrics, San Francisco, USA, 1982.
12. Belanger, A., Candas, B., Dupont, A., Cusan, L., Diamond, P., Gomez, J. L. and Labrie, F., *J. Clin. Endocrinol. Metabol.*, 1994, **79**, 1086–1090.
13. Simon, D., Preziosi, P., Barret-Connor, E., Roger, M., Saint-Paul, M., Nahoul, K. and Papoz, L., *Am. J. Epidemiol.*, 1992, **135**, 783–791.
14. Takizawa, T. and Hatakeyama, S., *Acta Pathol. Jpn*, 1978, **28**, 541–544.
15. Pirke, K. M. and Doerr, P., *Acta Endocrinol.*, 1975, **80**, 1971–1977.
16. Rolf, C., Behre, H. M. and Nieschlag, E., *Int. J. Androl.*, 1996, **19**, 135–142.
17. Homonnai, Z. T., Fainman, N., David, M. P. and Paz, G. F., *Andrologia*, 1982, **14**, 164–170.
18. Nieschlag, E. and Michel, E., in *Aging, Reproduction and the Climacteric* (eds Mastroianni, L. and Paulsen, C. A.), Plenum, New York, 1986, pp. 59–71.
19. Andersen, B. A., *Pop. Index*, 1975, **41**, 561.
20. Cooper, T. G., Keck, C., Oberdieck, U. and Nieschlag, E., *Hum. Reprod.*, 1993, **8**, 1251–1258.
21. Johnson, L., *J. Androl.*, 1986, **7**, 331–354.
22. Hargreave, T. B. and Elltori, R. A., *Br. J. Urol.*, 1986, **58**, 194–197.
23. Schwartz, D. and Mayaux, M. J., *New. Engl. J. Med.*, 1982, **306**, 404–406.
24. Collins, J. A. and Rowe, T. C., *Fertil. Steril.*, 1989, **54**, 774–779.
25. Bostofte, E., Bagger, P., Michael, A. and Stakemann, G., *Fertil. Steril.*, 1993, **59**, 102–107.

ACKNOWLEDGEMENTS. We thank Dr H. S. Juneja for help and encouragement.

Received 10 April 2000; revised accepted 19 June 2000