the screening of semen donors for donor insemination. Hum Reprod 1993;8:1521-3.

- 20 Irvine DS, Cawood EHH, Richardson DW, MacDonald E, Aitken RJ. A survey of semen donation; phase II—the view of the donors. *Hum Reprod* 1995;10:2752-3.
- 21 Aitken RJ, Best FSM, Richardson DW, Djahanbakch O, Lees MM. The correlates of fertilizing ability in normal fertile men. *Fertil Steril* 1982;38: 68-76.
- World Health Organisation. Laboratory manual for the examination of human semen and semen-cervical mucus interaction. Singapore: Press Concern, 1980.
 World Health Organisation. WHO laboratory manual for the examination of
- 23 World Health Organisation. WHO laboratory manual for the examination of human semen and semen-cervical mucus interaction. Cambridge: Cambridge University Press, 1987.
- 24 World Health Organisation. WHO laboratory manual for the examination of human semen and sperm-cervical mucus interaction. Cambridge: Cambridge University Press, 1992.
- 25 Johnson L. Spermatogenesis and ageing in the human. J Androl 1986;7: 331-54.

26 Mortimer D, Templeton AA, Lenton EA, Coleman RA. Influence of

- abstinence and ejaculation-to-analysis delay on semen analysis parameters of suspected infertile men. Arch Androl 1982;8:251-6.
 27 MacLeod J, Gold RZ. The male factor in fertility and infertility. II.
- 27 MacLeod J, Gold KZ. The male factor in fertility and inferdity. If Spermatozoon counts in 1000 men of known fertility and in 1000 cases o infertile marriage. J Urol 1951;66:436-49.
- 28 MacLeod J, Wang Y. Male fertility potential in terms of semen quality: a review of the past, a study of the present. *Fertil Steril* 1979;31:103-16.
- 29 Irvine DS, Aitken RJ. Seminal fluid analysis and sperm function testing. Endocrinol Metab Clim North Am 1994;23:725-48.
- 30 Aitken RJ, Irvine DS, Wu FC. Prospective analysis of sperm-oocyte fusion and reactive oxygen species generation as criteria for the diagnosis of infertility. Am J Obstet Gymecol 1991;164:542-51.
- infertility. Am J Obstet Gynecol 1991;164:542-51.
 Templeton A, Fraser C, Thompson B. The epidemiology of infertility in Aberdeen. BMJ 1990;301:148-52.
 Sharpe RM, Fisher JS, Millar MM, Joblling S, Sumpter JP. Gestational
- exposure of rats to xenoestrogens results in reduced testicular size and sperm production. *Environ Health Perspect* 1995;103:2-9.

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Time series analysis of sperm concentration in fertile men in Toulouse, France between 1977 and 1992

L Bujan, A Mansat, F Pontonnier, R Mieusset

See editorial, pp 467, 506

Objectives—To investigate whether sperm production has changed during the past 16 years in the Toulouse area of France.

Design—Time series analysis of sperm donors' specimens between 1977 and 1992.

Setting—Sperm bank of university hospital in Toulouse, France.

Subjects—302 healthy fertile men candidate sperm donors more than 20 and up to 45 years old and without any infertile brothers.

Main outcome measure—Spermatozoa concentration.

Results—Donors' mean age at time of donation was 34.05 (SD 5.13), but this increased significantly (P<0.001) during the study, from 32.4 in 1977 to 36 in 1992. Mean sperm count of samples was $83.12\times10^{\circ}$ /ml (SD $68.42\times10^{\circ}$ /ml). Sperm concentration was positively linked to the year of donation (Pearson's coefficient r=0.12, P<0.05), but this correlation disappeared after adjustment for age of donors (r=0.09, P>0.05).

Conclusion—Sperm concentration has not changed with time in the Toulouse area.

Introduction

Abstract

Several studies have suggested that the sperm count of healthy men has declined in the past few decades. Carlsen *et al* recently reported a decrease in sperm count and volume in the past 50 years.¹ This decrease was confirmed by Auger *et al* in the Paris area of France and was associated with qualitative alterations of sperm—that is, decreased motility of spermatozoa and fewer normally shaped spermatozoa.² Moreover, other. studies have reported increases in the incidence of cryptorchidism³ and testicular cancer.⁴⁵

Several hypotheses have been suggested to explain this decrease in sperm quality—for example, environmental exposure to harmful compounds⁶ such as oestrogens or compounds with oestrogen-like activity.⁷ In order to investigate potential environmental factors, we analysed the quality of semen supplied by donors to our sperm bank in south west France, a less populated area than Paris and one with different water supplies and air quality.

Methods

We studied the first ejaculates from healthy unpaid candidate sperm donors that were collected between 1977 and 1992 in our centre (Centre d'Etude et de Conservation des Oeufs et du Sperme Humain Midi-Pyrénées). All the donors had previously fathered at least one child. We excluded donors aged less than 20 and over 45 as age can affect the characteristics of sperm⁸ and excluded donors with an infertile brother.²

Donors provided semen samples by masturbation at the laboratory after a recommended period of sexual abstinence of three to five days. The samples were analysed as described previously.⁹ Sperm counts underwent logarithmic (base 10) transformation before statistical analysis, which was done with the PCSM package (Delta Soft, Meylan, France).

Results

We included 302 candidate donors in the study: 113 lived in the Toulouse conurbation, 64 lived in smaller cities, 115 lived in small towns or rural areas, and 10 came from other parts of France. The donors' mean age at the time of donation was 34.05 (SD 5.13, range 21-44), but this increased significantly (P<0.001) during the study from 32.4 in 1977 to 36 in 1992 (table 1).

The mean sperm count of the samples was $83 \cdot 12 \times 10^{\circ}$ /ml (SD $68 \cdot 42 \times 10^{\circ}$ /ml). Figure 1 shows the sperm counts according to the year of donation. Linear regression analysis between sperm count and year of donation showed a positive relation (Pearson's coefficient r=0.12, P < 0.05). However, when adjustment was made for the donor's age the relation between sperm count and year of donation was no longer sig-



Fig 1—Sperm count of semen samples by year when sample donated (box plots represent median and first and third quartile; bars represent 10th and 90th centiles)

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Table 1—Age of spermdonors by year whensample donated

		Age (years)	
Year	No of donors	Mean	Range
1977	11	32.4	25-42
1978	22	32.2	21-43
1979	27	31.7	24-39
1980	23	31.4	22-44
1981	25	33.4	26-41
1982	27	32.3	24-44
1983	15	35.7	28-43
1984	26	36.5	26-44
1985	17	35-2	27-41
1986	-17	37.2	30-44
1987	14	35-2	21-44
1988	14	34.6	27-39
1989	21	34.4	27-43
1990	17	35-5	27-44
1991	15	34.5	25-43
1992	11	36.0	30-43

CECOS Midi-Pyrénées and Centre de Stérilité Masculine, Centre Hospitalier Universitaire La Grave, 31052 Toulouse, France L Bujan, associate professor of reproductive biology A Mansat, associate professor of reproductive biology F Pontonnier, professor of urology R Mieusset, associate professor of histology

Correspondence to: Dr Bujan.

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nificant (r=0.09, P>0.05). As the variable representing the year of donation was composite (a combination of each donor's age and his year of birth), we performed a multiple regression analysis including these variables (table 2). Only the donor's age at the time of donation contributed significantly to sperm concentration (an increase of 3.3% for each year increase in age).

 Table 2—Effects of age and year of birth on sperm concentration of semen samples from 302 fertile men studied by multiple regression analysis

	Percentage change in regression coefficient (95% confidence interval)*	P value	
Increased age (by 1 year)	3-3 (0-7 to 6-1)		
Later birth year (by 1 year)	1.9 (-0.3 to 4.3)	0.09	
*Converted from the antile regression coefficients.	og of the logarithmic valu	e of the	

Discussion

Contrary to the results of Auger *et al*, we did not observe a decrease in the sperm counts of semen collected between 1977 and 1992. As the conditions for recruitment of donors were identical in the two studies, both study populations were similar in age and fertility status. Donors' age at the time of donation increased during Auger *et al*'s study just as it did in ours.

The duration of sexual abstinence before donation has been shown to affect the sperm count: the longer the abstinence, the higher the sperm count.¹⁰ Auger et al reported that the duration of sexual abstinence increased with donor's age.² We did not measure duration of abstinence in our study, and this might explain the different trends in sperm counts in ours and the Parisian study. However, when this parameter was excluded from the multiple regression analysis of the Parisian data there was still a decrease in sperm count with increasing age of donor (-2.5%) (95% confidence interval -3.0% to -1.9%) per year, P<0.001) or with year of birth (-2.0% (-2.9% to -1.1%)) per year, P < 0.001) (J Auger, personal communication), though this was less pronounced than when the parameter was included (-3.3%) with increasing age, -2.6% with later year of birth).2

The persistent differences in trends in sperm counts observed in these two studies (decreasing in the Parisian area and steady in the Toulouse area) might be due to environmental differences. In this way, differences in sperm count among men living in the London area were found to reflect differences in the water supply." The Toulouse and Paris areas differed in air quality, water supply, and matters of lifestyle (such as time spent commuting and stress factors). Population density was much higher in Paris than in Toulouse (20421 inhabitants/km² v 3032 in the city centre and 2901 v 491 in the suburbs), as was the density of cars and the concentration of nitrogen dioxide in the air (45 mg/m³ v 30 mg/m³ in summer and 47 mg/m³ v 37 mg/m³ in winter¹²). Industrial pollution was higher in Paris than Toulouse with regard to the output of registered waste, emission of oxidizable waste into the water, and production of sulphur dioxide.13 Water pollution was also greater in Paris with regard to concentrations of nitrates and phos-

Key messages

• Several reports have suggested that sperm production is declining in men

• This decline in sperm count was recently confirmed in the Paris area of France

• We studied sperm production of healthy fertile men in the Toulouse area of south west France

• The men were recruited according to the same selection criteria as in the Parisian study, but, contrary to the Parisian results, the sperm count of the semen samples had remained constant during the past 16 years

• These discrepant findings could be explained by different environmental conditions noted between the two areas

phates as well as the oxygen requirement of the organic matter present.¹⁴

However, sperm counts could also be affected by many other environmental factors⁶¹⁵ as well as behavioural factors.¹⁶¹⁷ Our findings indicate the need for further studies on environmental conditions and male reproductive function.

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- Carlsen E, Giwercman A, Keiding N, Skakkebaek NE. Evidence for decreasing quality of semen during past 50 years. *BMJ* 1992;305:609-13.
- 2 Auger J, Kunstmann JM, Czyglik F, Jouannet P. Decline in semen quality among fertile men in Paris area during the past 20 years. N Engl J Med 1995;332:281-5.
- 3 John Radcliffe Hospital Cryptorchidism Study Group. A prospective study of 7500 consecutive male births, 1984-8. Arch Dis Child 1992;67:892-9.
- Moller H. Clues to the aetiology of testicular germ cell tumours from descriptive epidemiology. *Eur Urol* 1993;23:8-15.
 Adami HO, Berstrom R, Mohner M, Zatonski W, Storn H, Ekbon A, et al.
- 5 Adami HO, Berstrom R, Mohner M, Zatonski W, Storn H, Ekbon A, et al. Testicular cancer in nine northern European countries. Int J Cancer 1994;59:33-8.
- 6 Michal F, Grigor KM, Negro-Vilar A, Skakkebaek NE. Impact of the environment on reproductive health: executive. *Environ Health Perspect* 1993;101(suppl 2):159-69.
- Sharpe RM, Skakkebaek NE. Are oestrogens involved in falling sperm counts and disorders of the male reproductive tract? *Lancet* 1993;341:1392-5.
 Schwartz D, Mayaux MJ, Spira A, Moscato ML, Jouannet P, Czyglick F, et al.
- 5 Schwartz D, Mayaux MJ, Spira A, Moscato ML, Jouannet P, Czyglick F, et al. Semen characteristics as a function of age in 833 fertile men. Fertil Steril 1983;39:530-5.
- Mieusset R, Bujan L, Mondinat C, Mansat A, Pontonnier F, Grandjean H. Association of scrotal hyperthermia with impaired spermatogenesis in infertile men. *Fertil Steril* 1987;48:1006-11.
 Jouannet P, Czyglik F, David G, Mayaux MJ, Spira A, Moscato ML, et al.
- 0 Jouannet P, Czyglik F, David G, Mayaux MJ, Spira A, Moscato ML, et al. Study of a group of 484 fertile men. I: Distribution of semen characteristics. Int J Androl 1981;4:440-9.
- 11 Ginsburg J, Okolo S, Prelevic G, Hardiman P. Residence in the London area and sperm density. *Lancet* 1994;343:230.
- 12 Della Massa JP, Gerboles M, Payrissat M. Etude de la distribution du dioxyde d'azote dans l'agglomération Toulousaine par la méthode des échantilloneurs passifs. In: DG 13 télécommunication industrie de l'information et innovation CE, eds. Rapport de la Commission Européenne CK CECA-CEEA. Bruxelles: Luxembourg Office EC, 1994:5-25.
- Information et innovation CE, eds. Kapport at la Commission Europeanne CK CECA-CEEA. Bruxelles: Luxembourg Office EC, 1994:5-25.
 Lavoux T, Theys J. L'industrie. In: IFEN, ed. L'environmement en France. Edition 1994-1995. Paris: Dunod and IFEN, 1994:216-23, 368.
 Couzet P. Les eaux continentales. In: IFEN, ed. L'environmement en France.
- Couzet P. Les eaux continentales. In: IFEN, ed. L'environnement en France. Edition 1994-1995. Paris: Dunod and IFEN, 1994:9-24, 354-6.
 Toppari J, Larsen JC, Christiansen P, Giwercman A, Grandjean P, Guillette
- 15 Toppari J, Larsen JC, Christiansen P, Giwerman A, Grandjean P, Guillette LJ, et al. In: Male reproductive health and environmental chemicals. Denmark: Ministry of Environmental and Energy, 1995. (Miljoprojekt No 290.)
- 16 Steeno OP, Pangkahila A. Occupational influences on male fertility and sexuality. Andrologia 1984;16:5-22.
- 17 Mieusset R, Bujan L. Testicular heating and its possible contributions to male infertility: a review. Int J Androl 1995;18:169-84.

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