

Semen alterations in HIV-1 infected men

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BACKGROUND: Couples in whom the man is infected by human immunodeficiency virus (HIV) increasingly request assisted reproductive technology (ART) to allow safe procreation. Semen quality is critical in such situations. **METHODS:** Semen characteristics were evaluated in 189 HIV-infected men requesting ART. At the time of semen analysis all men were healthy and 177 were receiving anti-retroviral therapy. Comparisons were made with HIV-seronegative men, partners of women requiring IVF because of tubal infertility, after matching for age and sexual abstinence delay. **RESULTS:** The most significant semen alterations found in the HIV-infected men were reduced percentages of rapidly progressive sperm [median (range), 10% (0–30%) compared with 15% (5–30%) in the controls, $P < 0.001$], and increased concentrations of non-spermatic cells [$3 \times 10^6/\text{ml}$ ($0.2\text{--}16 \times 10^6/\text{ml}$) compared with $1.1 \times 10^6/\text{ml}$ ($0.1\text{--}14 \times 10^6/\text{ml}$) in the controls, $P < 0.001$]. HIV-infected men also showed lower ejaculate volumes [2.8 ml ($0.6\text{--}9.3$ ml) compared with 3.6 ml ($1.1\text{--}11$ ml), $P < 0.05$] and total sperm counts [262.5×10^6 ($0\text{--}1003 \times 10^6$) compared with 310.5×10^6 ($48.3\text{--}1679 \times 10^6$), $P < 0.05$]. **CONCLUSIONS:** Semen evaluation in a large population of HIV-infected men requesting ART evidenced several alterations. Some of these anomalies might be related to anti-retroviral treatments.

Key words: HIV-1/semen analysis/sperm motility

Introduction

Couples in whom only the man is infected by human immunodeficiency virus type 1 (HIV-1) increasingly request assisted reproductive technology (ART) in order to procreate with a reduced risk of transmitting the virus to the uninfected woman and child. Indeed several studies have demonstrated that HIV-1 is present in the semen of most infected men (Gupta *et al.*, 1997; Dulioust *et al.*, 1998; Tachet *et al.*, 1999), even when it is not detected in blood (Zhang *et al.*, 1998; Dornadula *et al.*, 1999; Mayer *et al.*, 1999). Infectious HIV-1 is found in seminal plasma and in seminal leukocytes (Quayle *et al.*, 1997; Coombs *et al.*, 1998; Zhang *et al.*, 1998), whereas the possibility of close association between HIV-1 and sperm is debated (Dussaix *et al.*, 1993; Baccetti *et al.*, 1994; Bagasra *et al.*, 1994; Pudney *et al.*, 1999). HIV-RNA and HIV-DNA can be detected with high sensitivity in the different semen components (Leruez-Ville *et al.*, 2002). Therefore, ART can be performed with a high level of safety using sperm populations previously validated by virus undetectability (Marina *et al.*,

1998), although the requests of serodifferent couples still raise medical, technical and ethical questions (Jouannet *et al.*, 1998).

The technical requirements for ART when the man is infected by HIV, and especially the need for testing large enough samples of selected sperm to ensure high virological safety, raise the question of semen characteristics in HIV-infected men. Until now, only a few studies have focused on this issue and conflicting results have been published (Krieger *et al.*, 1991; Crittenden *et al.*, 1992; Politch *et al.*, 1994; Dondero *et al.*, 1996; Lasheeb *et al.*, 1997; Muller *et al.*, 1998). Furthermore, an increasing number of data show that antiretroviral drugs can induce toxic effects (Brinkman *et al.*, 1998; Carr *et al.*, 2000) so the possible consequences of these treatments upon reproductive function deserve investigation. Here we report on the semen characteristics of 189 HIV-infected men who were free from AIDS symptoms and requested medical assistance to procreate. The semen samples were analysed with strictly standardized methodology, and the results were compared with those of a control population of

men, partners of women requesting IVF because of tubal infertility.

Patients and methods

Patients

Semen samples were obtained from two populations of men attending the Laboratoire de Biologie de la Reproduction at the Cochin Hospital. The study population consisted of HIV-1-seropositive men, partners of HIV-seronegative women, who were free from AIDS symptoms and wanted to have a child. These men and their partners were volunteers participating in a study initiated to allow procreation with a minimal risk of virus transmission to the woman and to the fetus. A total of 189 men provided a semen sample between January 1999 and June 2000. The control population consisted of healthy HIV-seronegative men, partners of women with tubal infertility who had provided a semen sample in the laboratory during the study period before an IVF attempt. All men gave their informed consent to participate in the study.

Ages of HIV-1-infected men ranged from 24–58 years (median 36.8 years). Seventy men (37%) had acquired HIV infection through sexual intercourse, 52 (27%) were former i.v. drug users and 20 (11%) had received contaminated blood or blood-derivative transfusions. The mode of infection was unknown or not documented in 47 (25%) cases. The known duration of HIV infection before semen analysis, as estimated by each man, ranged from 2–19 years (median 10 years). At the time of semen collection, 177 men (94%) were receiving antiretroviral therapy (monotherapy: 1; bitherapy: 22; tritherapy: 129; quadritherapy: 16; pentatherapy: 1; not documented: 8). All treated men had been receiving antiretroviral drugs for >1 year. All men were free from AIDS symptoms at the time of semen collection. CD4 cell counts ranged from 110–1298/mm³ (median 479/mm³). HIV-RNA concentration in blood plasma ranged from undetectable to 25 000 copies/ml (median 500 copies/ml) and was <200 copies/ml in 73% of the men. Ages of the HIV-seronegative men ranged from 22–50 years (median 35.9 years). All men included in the study were free from symptomatic urogenital infection at the time of semen analysis.

Patients aged >40 years were more frequent in the HIV-infected population than in the controls and mean (\pm SD) sexual abstinence delay at semen collection was significantly higher in the former (4.9 ± 2.7 days versus 4.0 ± 1.3 days, $P < 0.01$). As both factors can influence semen characteristics (Auger *et al.*, 1995), HIV-infected and control men were matched on sexual abstinence delay (± 0.5 day) and age (± 2 years) in order to compare semen characteristics. Seventy-nine pairs were obtained. The 79 matched HIV-infected men were similar to the whole HIV-infected population with respect to age (27–52 years, median 37.6), estimated duration of HIV-infection (2–19 years, median 10 years), CD4 cell count (110–1200/mm³, median 486/mm³) and plasma HIV-1 RNA concentration (undetectable to 160 000 copies/ml, median 500 copies/ml). Ninety-five percent of them were receiving antiretroviral therapy.

Semen analysis

Semen samples were collected at the laboratory by masturbation into a sterile container after passing urine and washing hands and penis with chlorhexidine. Semen samples were kept at 37°C for 15–30 min for liquefaction before being analysed according to the World Health Organization (WHO) guidelines (World Health Organization, 1992). The volume of the ejaculate was measured by aspiration into a graduated pipette. Semen pH was read onto a pH paper (range 6.1–10.0). Consistency was graded normal, low or high according to the

drop of semen by gravity from a 5 ml pipette. Sperm motility was evaluated by examination of at least 200 sperm in a 10 μ l drop of semen covered with a 20 mm \times 20 mm coverslip under 100 \times and 400 \times phase contrast magnification at 37°C. Motility was graded (a) rapid progressive motility; (b) slow progressive motility, (c) non progressive motility and (d) no motility according to WHO guidelines. Sperm vitality was assessed on a smear after staining with eosin-nigrosin. Sperm and non-sperm cell (NSC) concentrations were assessed in duplicate with an haemocytometer on formalin diluted 1/10–1/40 specimens. Sperm morphology was not evaluated according to WHO guidelines but to modified David's classification (Auger *et al.*, 2001) on Shorr stained smears under 1000 \times magnification. The values for motility, vitality and normal morphology were expressed as percentages. Semen bacterial analysis was carried out for all men. It included the usual bacterial and mycoplasmas cultures and chlamydial detection by direct immunofluorescence.

Statistical analyses

Non-parametric methods were used for all comparisons of semen characteristics. Distributions in matched HIV-infected and control men were compared by the Kolmogorov–Smirnov test and mean values were compared by the matched Wilcoxon-test. Correlations were estimated by the Spearman rank test. When HIV-RNA was undetectable, the attributed value was half of the threshold value. Semen characteristics according to categories of duration of HIV-infection, CD4 cell count and plasma HIV-1 RNA were compared among the HIV-infected population using the Mann–Whitney test and association between these indicators and semen characteristics was verified after adjustment on age and sexual abstinence delay using multivariate linear regression. The chosen level of significance was 0.05. Data were analysed using SPSS release 10.0 for Mac OS.

Results

Semen characteristics in HIV-seropositive men compared with control men

Semen characteristics in the whole population of HIV-infected men and in the subgroups of HIV-infected and control men matched by age and sexual abstinence delay are presented in Table I. Comparison between matched HIV-infected and control men showed no significant difference with regard to semen consistency (not shown) or pH, sperm concentration, percentages of total progressively motile (a type + b type), percentages of live or morphologically normal sperm.

Ejaculate volumes were significantly lower in the HIV-infected men, both when comparing mean values (3.2 versus 3.7 ml in controls, $P < 0.05$, matched Wilcoxon-test) and distributions (medians 2.8 and 3.6 ml respectively, $P < 0.05$, Kolmogorov–Smirnov test). Figure 1a shows the distributions of ejaculate volume in both groups and the higher incidence of low volume in the HIV-infected men.

Mean total sperm counts were also decreased in HIV-infected men (296.9×10^6 versus 417.6×10^6 in the controls, $P = 0.021$, matched Wilcoxon test) although the distributions of this parameter did not significantly differ between the two groups (Kolmogorov–Smirnov test). Non spermatic cell (NSC) concentrations were significantly higher in the HIV-infected men (mean values 3.9×10^6 /ml versus 1.6×10^6 /ml in controls, $P < 0.001$, matched Wilcoxon test) and displayed significantly different distributions (median 3×10^6 /ml in the HIV-infected

Table I. Semen parameters in HIV-infected and control men

	HIV-infected men <i>n</i> = 189	Matched groups		
		HIV-infected <i>n</i> = 79	Control <i>n</i> = 79	<i>P</i>
Volume (ml)	3.3 (1.5) ^a 1.4/3.1/5.2 ^b	3.2 (1.7) ^a 1.3/2.8/5.1 ^b	3.7 (1.5) ^a 1.9/3.6/5 ^b	0.036 ⁽¹⁾ 0.021 ⁽²⁾
pH	7.9 (0.3) 7.7/7.9/8.3	7.9 (0.3) 7.7/7.9/8.3	7.8 (0.2) 7.7/7.9/8.1	NS NS
Sperm conc. (10 ⁶ /ml)	96.7 (84.3) 12/75/199	99.2 (73.0) 15/87/178	117.7 (93.3) 34/92/228	NS NS
Total sperm count (10 ⁶)	298.3 (251.8) 35.4/232/645.5	296.9 (235.2) 51/262/608	417.6 (347.9) 96/310/851	0.021 NS
NSC ^c concentration (10 ⁶ /ml)	5.2 (6.0) 0.9/4/10	3.9 (3.1) 0.7/3/8	1.6 (2.0) 0.3/1.1/3.5	< 0.001 < 0.001
Total NSC count (10 ⁶)	15.6 (16.7) 2.2/9.9/35.3	11.5 (11.8) 1.6/7.8/25.6	5.7 (6.5) 0.6/3.8/12.1	< 0.001 < 0.001
a+b motility (%)	38.4 (11.4) 25/40/53	40.1 (9.9) 30/40/55	38 (7.9) 25/35/50	NS NS
a motility (%)	10.3 (7.7) 3/10/20	11.7 (8.2) 4/10/25	17.4 (7.4) 10/15/30	< 0.001 < 0.001
b motility (%)	27.8 (9.6) 15/30/40	27.9 (8.5) 20/30/37	20.6 (5.5) 15/20/30	< 0.001 < 0.001
Vitality (%)	73.0 (15.3) 54/75/90	72.7 (14.4) 57/74/90	70.5 (11.7) 55/72/83	NS NS
Normal morphology (%)	40.4 (16.7) 20/40/60	40.0 (17.7) 18/39/62	41.2 (11.8) 27/43/55	NS NS

^aMean values (SD).^b10th percentile/median/90th percentile values.^cNon spermatic cells.⁽¹⁾Matched Wilcoxon test.⁽²⁾Kolmogorov-Smirnov test.

NS = no significant difference.

men versus 1.1×10^6 /ml in the controls, $P < 0.001$, Kolmogorov–Smirnov test) as shown in Figure 1b. Similarly, and despite lower ejaculate volumes, total NSC counts were increased in the HIV-infected men both when comparing mean values (11.5×10^6 versus 5.7×10^6 in controls, $P < 0.001$, matched Wilcoxon-test) and distributions (median 7.8×10^6 versus 3.8×10^6 in controls, $P < 0.001$, Kolmogorov–Smirnov test). Morphological analysis indicated that in most cases these NSC were not polymorphonuclear leukocytes.

The detailed evaluation of sperm motility evidenced striking differences in sperm motility patterns. The HIV-infected men displayed significantly lower percentages of rapidly progressive (a type) sperm than the controls, both when comparing mean values (11.7 and 17.4% respectively, $P < 0.001$, matched Wilcoxon test) and distributions (medians 10 and 15% respectively, $P < 0.001$, Kolmogorov–Smirnov test). Figure 1c shows the downward shift in the proportions of rapid progressive sperm in the HIV-infected group. Conversely, the percentages of slow progressive (b type motile) sperm were significantly higher in the HIV-infected men both when comparing mean values (27.9 versus 20.6% in the controls, $P < 0.001$, matched Wilcoxon test) and distributions (median 30 versus 20% in the controls, $P < 0.001$, Kolmogorov–Smirnov test). There was no correlation between percentages of rapid or slow progressive sperm and NSC concentrations.

The results of semen bacterial culture were similar in the HIV-infected and in the control population. In the HIV-infected population, 17 (9%) of semen cultures contained at least 10^4

colony-forming units (CFU) of one predominant bacterial species. The isolated bacteria were mainly intestinal ones (*E. coli*, *E. faecalis*, *S. faecalis*, *P. mirabilis*, *P. vulgaris*) except for two men (1%) in whom *U. urealyticum* was found. In the control population, eight (9%) of cultures showed at least 10^4 CFU of various intestinal bacteria, and *U. urealyticum* was found in three cultures (3.1%).

Relationship between semen characteristics and HIV-1 infection

We observed no relationship between semen characteristics and the mode of infection. Correlation analysis (Spearman rank test) showed a significant negative correlation between the estimated duration of HIV infection and ejaculate volume ($r = -0.185$, $P = 0.016$), whereas a significant positive correlation between this duration and sperm concentration ($r = 0.234$, $P = 0.002$) was found. Men in whom estimated duration of HIV infection was >10 years ($n = 79$) had significantly lower ejaculate volumes than men ($n = 91$) in whom estimated duration of HIV infection was <10 years, [median (min–max) 2.7 ml (0.6–1.1 ml) versus 3.5 ml (0.9–9.3 ml) $P = 0.005$, Mann–Whitney test].

In contrast, higher sperm concentrations were observed when the estimated duration of HIV infection was >10 years [median (min–max) $10^4 \times 10^6$ /ml ($0–680 \times 10^6$ /ml) versus 60×10^6 /ml ($0–273 \times 10^6$ /ml), $P = 0.002$, Mann–Whitney test]. As a result, total sperm count was not affected by the duration of HIV infection. The observed differences were still present

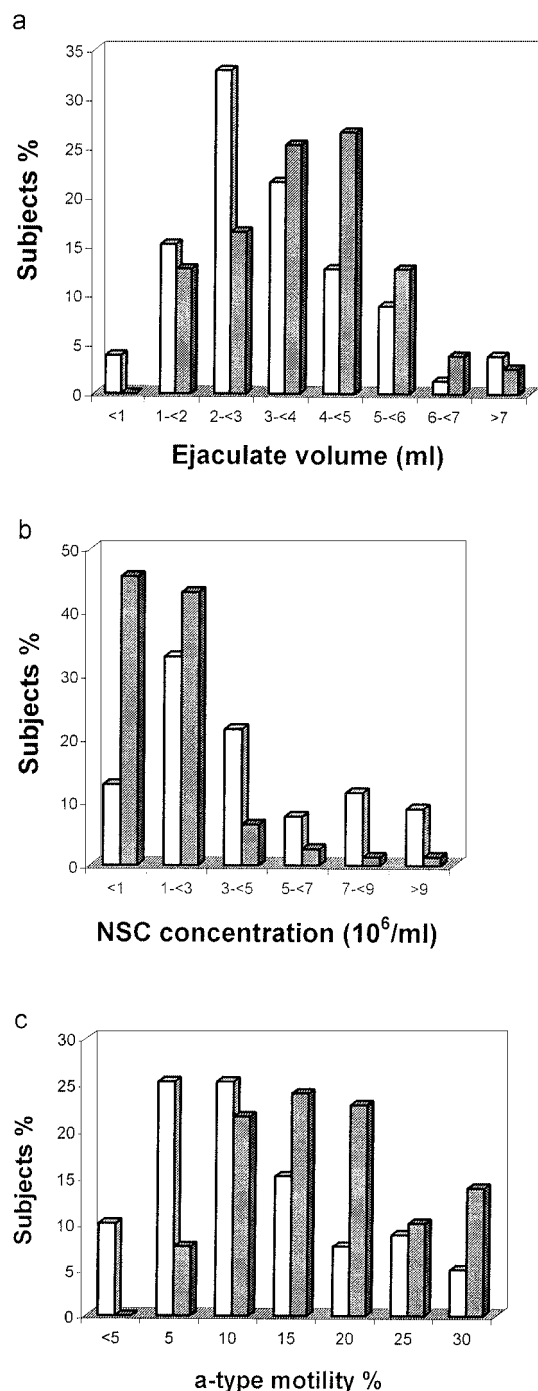


Figure 1. Distribution of ejaculate volume (a), non spermatic cells concentration (NSC) (b) and percentage of rapidly progressive sperm (WHO grade a) (c) in 79 HIV-seropositive (□) and 79 HIV-seronegative (■) men, matched on age and sexual abstinence delay.

after adjustment for age and sexual abstinence delay and were not associated with significant changes in CD4 cell count or HIV-RNA plasmatic concentration. We found no relation between the duration of HIV infection and other semen parameters. Comparisons according to the type of antiretroviral regimen (bi-, tri- or quadritherapy) received at the time of semen collection did not reveal clear differences in semen characteristics.

Table II summarizes the relationship between CD4 cell

count or blood plasma HIV-RNA concentration and semen characteristics. Ejaculate volume was the only semen parameter significantly correlated with CD4 cell count ($r = 0.186$, $P = 0.015$). Comparison of semen characteristics according to CD4 cell count (≥ 200 CD4 cells/mm³, $n = 156$, versus < 200 /mm³, $n = 12$) showed lower sperm concentrations [median (min-max) 33.3×10^6 /ml (0.3 – 165.0×10^6 /ml) versus 83.0×10^6 /ml (0.0 – 680.0×10^6 /ml), $P = 0.042$, Mann-Whitney test] and lower total sperm counts [median (min-max) 126.3×10^6 (1.9 – 429.0×10^6 /ml) versus 250.1×10^6 (0.0 – 1292.0×10^6 /ml), $P = 0.042$, Mann-Whitney test] when CD4 cell count was < 200 /mm³. These differences were still present after adjustment for age and sexual abstinence delay. The percentage of slow progressive sperm (WHO grade b) and the percentage of morphologically normal sperm were negatively correlated with plasmatic HIV-RNA concentration ($r = -0.205$, $P = 0.008$ and $r = -0.175$, $P = 0.025$, respectively). A significantly reduced sperm vitality was found in the men whose plasmatic HIV-RNA concentration was ≥ 1000 copies/ml ($n = 23$) when compared with the men ($n = 148$) with < 1000 HIV-RNA copies/ml (medians 75 and 65% respectively, $P = 0.004$, Mann-Whitney test).

Discussion

This study provides a reliable description of the main semen characteristics in a large population of HIV-infected men free of AIDS symptoms; 94% of the population were receiving antiretroviral therapy, and all were requesting medical assistance for safe procreation. Comparison with control men showed that several semen characteristics were altered in the HIV-infected group. The most striking finding was a slowing down of sperm motion, revealed by an altered pattern of sperm progressive motility as the result of a decreased rapid (a type) motility and an increased slow (b type) motility in more than one third of the HIV-infected men. Increased concentration and total count of non-spermatic cells were the other most obvious anomalies found in the HIV-infected population. Significant decreases of ejaculate volume and total sperm count were also found in the HIV-infected men but there were no changes in semen pH, sperm concentration, total motility, vitality and morphology.

Previous studies on semen characteristics in HIV-infected men (Krieger *et al.*, 1991; Crittenden *et al.*, 1992; Politch *et al.*, 1994; Dondero *et al.*, 1996; Lasheeb *et al.*, 1997; Muller *et al.*, 1998) have reported various anomalies, which often differed from one study to another. Differences in health and fertility status of the HIV-seropositive and -seronegative populations recruited, methodological variations in semen analysis and low numbers of subjects probably explained these discrepancies. A correlation between CD4 cell count and sperm motility and a trend to a general semen deterioration in men at advanced stages of HIV infection were reported by several authors (Crittenden *et al.*, 1992; Politch *et al.*, 1994; Dondero *et al.*, 1996; Muller *et al.*, 1998) but, as variable proportions of men at advanced stages of HIV-infection were included in most studies, specific conclusions regarding the semen characteristics in healthy HIV-infected men could not be drawn.

Table II. Relations between CD4 cell count or plasmatic HIV-RNA load and semen parameters

	CD4 cell count		HIV-RNA load	
	Correlation <i>P</i> (<i>r</i> value)	<200 versus $\geq 200^b$ <i>P</i> (medians)	Correlation <i>P</i> (<i>r</i> value)	<1000 versus $\geq 1000^c$ <i>P</i> (medians)
Volume (ml)	0.015 (-0.185)	NS	NS	NS
pH	NS	NS	NS	NS
Sperm concentration (10^6 /ml)	NS	0.04 (33.3 versus 83.0)	NS	NS
Total sperm count (10^6)	NS	0.04 (126 versus 250)	NS	NS
NSC ^a concentration (10^6 /ml)	NS	NS	NS	NS
Total NSC count (10^6)	NS	NS	NS	NS
a+b motility (%)	NS	NS	NS	NS
a motility (%)	NS	NS	NS	NS
b motility (%)	NS	NS	0.008 (-0.205)	NS
Vitality (%)	NS	NS	NS	0.004 (75 versus 65)
Normal morphology (%)	NS	NS	0.025 (-0.175)	NS

^aNon-spermatic cells.^bCD4⁺ cells/mm³.^cHIV-RNA copies/ml.

The present study avoided several of these biases. The HIV-infected group represented the largest sample of healthy HIV-infected men in whom semen characteristics have been evaluated to date. The homogeneity of these men with respect to clinical and immunological status reduced the risk of non-specific semen alterations due to poor clinical conditions. All the control HIV-seronegative men were healthy partners of women with tubal infertility and their semen analysis was carried out as a systematic investigation before IVF. This avoided the possible bias toward better semen characteristics due to recruitment of men on the basis of recent fatherhood as well as the inverse bias that may occur when recruiting infertile men requesting semen analysis. Furthermore, all semen specimens were collected and evaluated in the same laboratory, during the same period and according to standardized procedures. Finally, as age and sexual abstinence delay can influence semen parameters (Auger *et al.* 1995), comparisons were made on subgroups of HIV-seropositive and -seronegative subjects with identical distributions for these two confounding factors.

Relationships between estimated length of HIV-infection, CD4 cell count or HIV-RNA plasmatic concentration and semen characteristics were investigated by correlation analysis and comparisons between categories. Both methods showed that duration of HIV-infection was significantly associated with a reduced ejaculate volume and an increased sperm concentration, which was the consequence of the reduced volume since total sperm count was unchanged. The results were less concordant with respect to CD4 cell count or HIV-RNA concentration. Only few semen parameters were significantly correlated with these indicators of HIV-infection. This could be due to the clinical and biological homogeneity of the HIV-infected population investigated in this study, since only 12 men had <200 CD4 cells/mm³ and 23 had >1000 HIV-RNA copies/ml in blood plasma. Moreover, HIV-RNA was often undetectable and the same arbitrary value (half the detection threshold) was attributed to many men. However, the important decreases of sperm concentration and total sperm

count observed in the men with a CD4 cell count below 200/mm³ are in agreement with earlier reports (Crittenden *et al.*, 1992; Politch *et al.*, 1994; Dondero *et al.*, 1996; Muller *et al.*, 1998).

What could be the causes of the semen alterations observed in the present population of HIV-infected men? Currently no clear evidence supports the hypothesis of direct effects of HIV itself on germ cells, sperm or the genital tract. Furthermore, we did not observe, in our HIV-infected patients, increased frequencies of alcohol intake or smoking, and no man was taking illicit drugs at the time of semen collection. Among other hypotheses, the existence of silent genital tract inflammation might be proposed to explain both the altered motility pattern and the increased NSC concentrations, although only 17 of the 189 HIV-infected men had a positive bacterial semen culture, without greater semen alterations than the men with negative culture (data not shown). Indeed, an association between reduced sperm motility and increased leukocyte concentration in semen has been reported and attributed to an increased production of reactive oxygen species (Kovalski *et al.*, 1992; Baker *et al.*, 1996; Armstrong *et al.*, 1999). However, in such situations, all motility types were reduced and the overall percentage of motile sperm was decreased. The pattern observed here, associating reduced percentages of rapid sperm and increased percentages of slow sperm, was different. Furthermore, we did not observe high quantities of polymorphonuclear leukocytes and the percentages of rapidly progressive sperm were not related to NSC concentration. However, as the routine semen analyses performed in this study did not allow precise identification of the non-sperm cells, we cannot rule out the possibility of an increased presence of macrophages or lymphocytes in the HIV-infected men.

Impaired motility could also be the consequence of a dysfunction of accessory glands since decreased ejaculate volumes were observed in our series. However, decreased volumes were mainly observed in men who were reported to be infected for >10 years and were not associated with different percentages of rapid or slow motile sperm. Finally,

as most men in our study had been receiving an antiretroviral therapy for >1 year at the time of semen analysis, a third hypothesis is that some semen alterations could be the consequence of antiretroviral treatments. Antiretroviral drugs are currently implicated in various metabolic and endocrine dysfunctions (Carr *et al.*, 2000) that could in turn also affect testis, reproductive tract and gamete functions. In our study, only a few men were not on antiretroviral therapy, so it was not possible to compare semen characteristics between treated and untreated men. The men were treated for their HIV-infection in many different centres and we had not enough information to pertinently compare their semen characteristics according to the different therapeutic histories. Nevertheless, as sperm motility is highly ATP-consuming (Ford *et al.*, 1990), the sperm motion anomalies that we observed might be related to the mitochondrial toxicity of nucleoside analogues (Brinkman *et al.*, 1998). The recent report of an increased frequency of multiple mitochondrial DNA deletions in the sperm of patients treated with highly active antiretroviral therapy for >12 months (White *et al.*, 2001) supports this hypothesis. On the other hand, a study evaluating semen parameters in 20 men at initiation of antiretroviral therapy and 4–12 weeks later concluded that such treatments may improve semen quality (Robbins *et al.*, 2001) but evolution after treatment onset depended on CD4 cell count at study entry. Long term prospective studies in men initiating antiretroviral therapy are needed to better evaluate the effects of antiretroviral drugs on semen.

The semen alterations observed in HIV-infected men are probably not marked enough to severely impair fecundity. However, they may have practical consequences regarding the management of serodifferent couples in whom the man is infected by HIV and who request ART to procreate. These alterations also raise questions about their causes and about their long-term evolution.

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