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SHORT PAPER

Genital ulcers in a primary health clinic in Rwanda: impact of HIV infection on diagnosis and ulcer healing (1986–1992)

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Summary: During 1986–88 and 1990–92, 1025 (97%) out of 1057 genital ulcer patients in Kigali, Rwanda, were tested for HIV antibodies and for infection with *Treponema pallidum*, *Haemophilus ducreyi* and herpes simplex virus. Overall, 57% of men and 80% of women had antibodies to HIV-1. The most frequent laboratory diagnoses were chancroid (27%), syphilis (19%) and genital herpes (19%) among men and syphilis (35%), genital herpes (23%) and chancroid (20%) among women. HIV-1 seroprevalence increased sharply over time among men but not among women. The clinical presentation of ulcers as well as laboratory diagnoses were similar in the HIV-1 seropositive and seronegative groups. The relative frequency of all laboratory diagnoses remained unchanged over time. HIV-1 seropositivity had no impact on ulcer healing. Advanced immunodeficiency was diagnosed among 12% of the HIV-1 seropositive patients and was significantly associated with increasing age and genital herpes.

Keywords: HIV-1, genital ulcer, genital herpes, syphilis, chancroid

INTRODUCTION

In Kigali, capital of Rwanda, infection with HIV-1 was diagnosed for the first time in 1983 and spread rapidly across the country[1,2]. A national serosurvey conducted in December 1986 showed a 20% HIV-1 seroprevalence in the urban population between 20 and 40 years old, versus 2% in the rural sample[3]. Among women attending the antenatal clinics of the Centre Hospitalier de Kigali (CHK) the HIV-1 seroprevalence was 29% in 1986–87 and 34% in 1992–93[4,5]. We report here on the increasing HIV-1 seroprevalence among genital ulcer disease (GUD) patients and describe clinical and laboratory features in relation to HIV-1 serostatus and degree of immunodeficiency.

PATIENTS AND METHODS

The study was carried out at the Laboratory of the CHK, Kigali, capital of Rwanda, and at the Centre Médico-Social de Bilyogo (CMS), Nyamirambo, Kigali. The CMS is a primary health clinic in an area of the city where prostitution is widespread. It serves mainly the lower socioeconomic strata of the population.

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Study design and patients

The study comprised 2 parts. The first part (1986–88) intended to document the relative frequencies of the causes of GUD. The second part (1990–92) put emphasis on the history of patients, clinical presentation of ulcers and follow-up. All patients were seen at the CMS, in a consecutive order, during September 1986–March 1988 (n=638) and September 1990–June 1992 (n=419). Overall, 1025 (97%) of the 1057 patients were tested for HIV antibodies: 674 (65.8%) showed a positive, 343 (33.4%) a negative and 8 (0.8%) an undetermined result. HIV-2 antibodies were not detected. Baseline data of HIV-1 seropositivity were obtained during a preliminary study in 1985 among 120 consecutive GUD patients recruited among the similar population as seen in the present study. The HIV-1 seroprevalence was 46% (36/78) among men and 60% (25/42) among women.

Diagnostic procedures and treatment regimens

Demographic, epidemiological and clinical information were obtained in a standard interview and physical examination of the external genitals and inguinal regions was performed. During the second part a clinical aetiological diagnosis was

made prior to any knowledge of the laboratory results. Criteria for clinical and laboratory diagnosis, including HIV infection, have been described earlier[6,7]. Laboratory methods for the diagnosis of syphilis, chancroid and genital herpes remained unchanged over time. A breakdown of the freezing system prevented preservation of specimens for herpes simplex virus (HSV) culture in 1987. T lymphocyte subset analysis and assessment of ulcer healing were performed during 1990–92[7,8]. Treatment regimens used during the second period have been published earlier[6,7]. Specific treatment for herpes infection was not given. Ulcer healing was assessed on days 7 and 14 after the initial visit. Among HIV-1 seropositive patients absolute CD4+ counts of $<200/mm^3$, $\geqslant 200 <500$, and $\geqslant 500/mm^3$ were considered as indicating advanced, moderate and minimal immunosuppression respectively[9].

Statistical analysis

The Yates' corrected Chi-square test (when appropriate) and the two-tailed Fisher's exact test were applied to assess differences in proportions for statistical significance. The odds ratio (OR) was used for measuring associations. The Mann–Whitney U (M–W U) test was performed to compare results of 2 patient groups and the Kruskal–Wallis H (K–W H) test to compare data from multiple groups. The Mantel–Haenszel method (M–H summary χ^2 and M–H adjusted odds ratio) was used for comparing 2 patient groups after stratification for a qualitative factor. The χ^2 for trend was used for demonstration of a linear trend. Statistical analysis was performed using the Epi-Info software, Version 6, CDC, Atlanta.

RESULTS

Background data

Only 20 (3%) of 640 men and no women were circumcised. Thirty (19%) of the 162 women who visited the clinic during the second period were self-identified sex workers (SWs), the number of SWs in the first period remains unknown. HIV-1 seropositive and seronegative men and women were comparable with respect to number of ulcers, delay between ulcer onset and presentation to the clinic (ulcer onset <8 days, 35%; 8–14 days, 33%; 14 days–4 weeks, 15%; >4 weeks, 17%) and previous use of antibiotics (28%). Overall, men were older than women (mean ages: 25.8 ± 5.1 *vs* 24.1 ± 5.3 years; $P<0.001$) and men examined in 1990–92 were older than men in 1986–88 (26.5 ± 5.0 *vs* 25.7 ± 5.0 years; $P<0.001$). The ages of women were comparable between both periods ($P=0.38$). In contrast to findings obtained from HIV-1 seropositive men, a positive trend between ulcer onset and a reactive syphilis serology was observed among HIV-1 seronegative men (χ^2_{trend}, 5.7; $P=0.02$). Furthermore, only one (3%) of the 32 HIV-1 seronegative versus 15 (28%) of the 53 HIV-1 seropositive men with an ulcer onset of <8 days had a reactive syphilis serology ($P=0.009$), suggesting that a pre-existing reactive syphilis serology, unrelated to the present ulcer, was more frequent in the latter group. In 1990–92, HIV-1 seronegative men were more frequently positive for *H. ducreyi* than seropositive ones and buboes more frequently associated with *H. ducreyi* than with the remaining laboratory diagnoses (22% or 17/78 *vs* 8% or 13/169; OR, 3.34; $P=0.003$). After stratification for *H. ducreyi* no association between buboes and HIV-1 status of men was observed (M–H summary χ^2, 2.65; M–H weighted OR, 2.65; $P=0.10$). After exclusion of herpes infections, HIV-1 seropositive men with buboes ($n=12$) had higher CD4+ cell counts/ml than those without ($n=120$), (mean CD4+ cells/mm^3, 717 ± 302 *vs* 581 ± 392; $P=0.07$). Similarly, HIV-1 seronegative men with buboes ($n=14$) had a higher CD4+ cell counts/ml than those without buboes ($n=61$) (mean CD4+ cells/mm^3, 1273 ± 435 *vs* 947 ± 421; $P=0.009$). None of the laboratory diagnoses was associated with the age of the patients (Tables 1a and 1b).

HIV-1 seroprevalence over time

An increasing prevalence of HIV-1 antibodies was observed in all age groups of men (Figure 1). Among women <30 years old, a steep increase of HIV-1 seropositivity was observed between 1985 and 1986–88 (Figure 2).

CD4+ T lymphocyte depletion and laboratory diagnosis among HIV-1 seropositive patients

Of the 270 HIV-1 seropositive patients tested for CD4+ and CD8+ lymphocyte subpopulations, 31 (12%) showed advanced, 104 (38%) moderate and 135 (50%) minimal immunodeficiency; of the 100 tested HIV-1 seronegative patients, only 8 (8%)

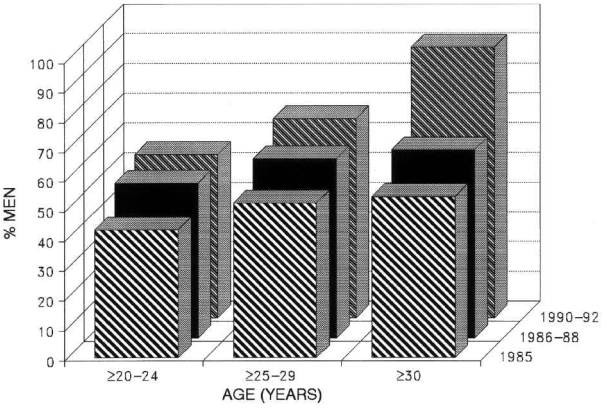

Figure 1. Prevalence of HIV-1 antibodies among men according to age and study period

Table 1a. Association of HIV infection with selected clinical and diagnostic features among men with genital ulcer disease

	1986–1988					1990–1992				
Features	HIV+ n=200	(%)	HIV− n=185	(%)	P	HIV+ n=166	(%)	HIV− n=81	(%)	P
Age (years)	25.6±4.3		25.1±5.8		0.004	27.8±5.3		24.1±3.7		<0.001
History of STD*										
Ulcers	28/61†	(46)	5/23†	(22)	0.08	72	(43)	12	(15)	<0.001
Urethral discharge						97	(58)	27	(33)	<0.003
Genital warts						37	(22)	13	(16)	NS
No history						40	(24)	46	(57)	<0.001
Inguinal lymph nodes	35/108§	(32)	31/85§	(37)	NS	72	(43)	43	(53)	NS
Bubo	7/108§	(7)	10/85§	(12)	NS	15	(9)	15	(19)	0.05
Clinical presentation										
Genital herpes						56	(34)	19	(24)	NS
Syphilis						19	(11)	10	(12)	NS
Chancroid						64	(39)	40	(49)	NS
Undetermined						27	(16)	12	(15)	NS
Laboratory diagnosis‡										
RPR≥1:2+TPHA≥1:80	32	(16)	32	(17)	NS	42	(25)	14	(17)	NS
Haemophilus ducreyi	49	(25)	41	(22)	NS	47	(28)	31	(38)	NS
Herpes simplex virus	12/108§	(11)	17/85§	(20)	NS	34	(21)	17	(21)	NS
No features	55/108§	(51)	33/85§	(39)	NS	64	(39)	23	(28)	NS
CD4+%**						18±10		32±9.4		<0.001
CD4+/mm^3						581±395		1018±444		<0.001
CD4+/CD8+						0.45±0.41		1.14±0.51		<0.001

Results are given as proportions (%) and as mean values±SD
*Some men had more than one STD. †Men observed in 1988. §Men observed in 1986 and 1988. ‡Some men were positive for more than one feature. **CD4+ counts were available for 156 HIV+ and 76 HIV− men
NS=non significant; RPR=rapid plasma reagin test; TPHA=*Treponema pallidum* haemagglutination test

had CD4+ cells <500/mm^3. Genital herpes as single laboratory diagnosis was significantly associated with advanced immunodeficiency since it was diagnosed among 10 (32%), 13 (13%) and 21 (16%) HIV-1 seropositive patients with advanced, moderate and minimal immunodeficiency (P=0.03). Advanced immunodeficiency was not associated with any of the remaining laboratory diagnoses. CD4+ cells/mm^3 and CD4+/CD8+ ratios declined significantly with increasing age of HIV-1 seropositive but not of HIV-1 seronegative patients (data not shown). Advanced immunodeficiency was observed among 6% (6/102), 9% (9/99) and 23% (16/69) of HIV-1 seropositive patients aged <25, ≥25–29 and ≥30 years respectively (P<0.01).

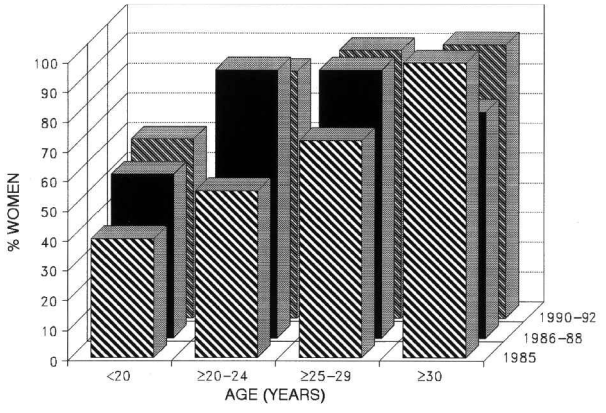

Figure 2. Prevalence of HIV-1 antibodies among women according to age and study period

Association of clinical and laboratory diagnosis in relation to HIV-1 status

Only results from men were analysed because the number of HIV-1 seronegative women was too small for analysis. Since clinical diagnosis did not intend to identify ulcers of mixed aetiology, the sensitivity of the clinical diagnosis, which reflects the clinical expression of the respective diseases, was calculated for single infections (Table 2).

Ulcer healing in relation to HIV-1 serostatus and degree of immunodeficiency

Fourteen out of 395 patients, tested for HIV-1 antibodies during 1990–92, were lost to follow-up because of severe civil unrest. Among the remaining group loss to follow-up was 22% (85/381) on day 7 and 35% (104/296) on day 14 and was comparable between HIV-1 seropositive and seronegative patients, irrespective of laboratory diagnosis. Ulcer healing was similar among the HIV-1 seropositive and negative groups (Table 3).

DISCUSSION

This study shows the increasing prevalence of HIV-1 infection among genital ulcer patients in Rwanda.

Table 1b. Association of HIV infection with selected clinical and diagnostic features among women with genital ulcer disease

	1986–1988					1990–1992				
Features	HIV+ *n*=185	(%)	HIV− *n*=52	(%)	*P*	HIV+ *n*=123	(%)	HIV− *n*=25	(%)	*P*
Age (years)	24.3± 4.8		22.8± 6.2		NS	25.3± 5.7		21.6± 4.4		<0.001
History of STD*										
Ulcers	13/37†	(35)	1/6†	(17)	NS	44	(36)	4	(16)	0.09
Vaginal discharge						69	(56)	7	(28)	0.02
Genital warts						20	(16)	1	(4)	NS
No history						37	(30)	16	(64)	0.003
Inguinal lymph nodes	11/114§	(10)	5/29§	(17)	NS	50	(41)	9	(36)	NS
Bubo	–		–			5	(4)	1	(4)	NS
Clinical presentation										
Genital herpes						45	(37)	10	(40)	NS
Syphilis						16	(13)	6	(24)	NS
Chancroid						31	(25)	6	(24)	NS
Undetermined						31	(25)	3	(12)	NS
Laboratory diagnosis‡										
RPR⩾1:2+TPHA⩾1:80	68	(37)	13	(25)	NS	47	(38)	7	(28)	NS
Haemophilus ducreyi	33	(18)	6	(12)	NS	33	(27)	4	(16)	NS
Herpes simplex virus	23/114§	(20)	6/29§	(21)	NS	29	(24)	9	(36)	NS
No features	38/114§	(33)	14/29§	(48)	NS	35	(29)	5	(20)	NS
CD4+%**						18± 10.3		34± 9.3		<0.001
CD4+/mm^3						641± 436		1121± 481		<0.001
CD4+/CD8+						0.43± 0.37		1.26± 0.66		<0.001

Results are given as proportions (%) and as mean values± SD
*Some women had more than one STD. †Women observed in 1988. §Women observed in 1986 and 1988. ‡Some women were positive for more than one feature. **CD4+ counts were available for 114 HIV+ and 24 HIV− women
NS=non significant; RPR=rapid plasma reagin test; TPHA=*Treponema pallidum* haemagglutination test

The relative frequency of syphilis, chancroid and genital herpes remained unchanged over time. Laboratory features were comparable among the HIV-1 seropositive and negative groups. No appreciable effect of HIV-1 seropositivity on the clinical presentation of ulcers or sensitivity of the clinical diagnosis was observed among men. The lower sensitivity of the clinical diagnosis of syphilis among HIV-1 seropositive men may reflect the higher proportion of reactive tests not related to the present lesion. The diagnosis of syphilis by the detection of serum antibody to *T. pallidum* is problematic in a population with a high background of STD. The number of syphilis cases in the present study is undoubtedly overestimated by including infections treated previously but still showing reactive serological tests. Our data confirm the lack of accuracy of the clinical aetiologic diagnosis of genital ulcers[10–13].

It has been postulated that HIV infection delays ulcer healing, enhances the recrudescence of genital herpes and increases the susceptibility for acquiring genital ulcers[14]. Treatment failure of chancroid was associated with HIV-1 seropositivity in Kenya but not in South Africa[15–18]. Delay in ulcer healing was associated with HIV seropositivity in

Table 2. Sensitivity and positive predictive value (PPV) of the clinical diagnosis of genital ulceration according to HIV-1 serostatus of men

		HIV-1 serostatus		
		Positive	Negative	*P*
Syphilis	Se	11% (3/27)	42% (5/12)	0.08
	PPV	21% (4/19)	50% (5/10)	NS
Chancroid	Se	65% (20/31)	82% (22/27)	NS
	PPV	47% (30/64)	60% (24/40)	NS
Herpes	Se	42% (10/24)	40% (6/15)	NS
	PPV	21% (12/56)	32% (6/19)	NS

Se=sensitivity; NS=non significant

Table 3. Ulcer healing according to laboratory diagnosis and HIV-1 serostatus

		Ulcer healing/improvement			
Laboratory diagnosis		Day 7 *n*/*n**	(%)	Day 14 *n*/*n**	(%)
Syphilis	HIV+	30/36	(83)	18/20	(90)
	–	10/13	(77)	10/11	(91)
Genital herpes	HIV+	27/40	(67)	16/19	(84)
	–	15/21	(71)	12/14	(86)
Chancroid	HIV+	20/34	(59)	18/24	(75)
	–	13/19	(68)	12/13	(92)
Mixed aetiology†	HIV+	20/29	(69)	19/22	(86)
	–	1/3	(33)	1/1	(100)
Undetermined	HIV+	55/75	(73)	50/55	(91)
	–	21/26	(81)	13/13	(100)
All diagnoses	HIV+	152/214	(71)	121/140	(86)
	–	60/82	(73)	48/52	(92)

*Proportion of patients who presented for follow-up
†Including 28 chancroid, 10 herpes and 27 syphilis infections

Malawi[19]. In the present study HIV-1 infected individuals with moderate or minimal immunodeficiency had a similar ulcer healing than normal individuals and treatment failure of chancroid was related to resistance of *H. ducreyi* to trimethoprim/sulphamethoxazole, as reported earlier[7]. The reasons for the discrepancy between our findings and those obtained in Kenya and Malawi remain unclear. Since the spread of HIV-1 started earlier in Rwanda than in Kenya or Southern Africa, it is unlikely that a higher proportion of GUD patients in these countries would have a more advanced degree of immunodeficiency than in Kigali during the period under review[20,21].

There exists a debate whether circumcision protects men from HIV infection or other STDs[22]. The 3% (20/640) frequency of circumcision contrasts strongly to the 29% (243/837) observed among men married to women who were recruited in a cohort study on the natural history of HIV infection in Kigali ($P<0.001$)[23]. This high rate of circumcision is very surprising since neonatal or adolescent circumcision is not performed routinely in Rwanda[24,25]. It is a common belief that men suffering from GUD or warts will be cured definitively by removing the foreskin, even in the presence of active lesions. Therefore, in Rwanda, the circumcised state reflects a higher exposure to sexually transmitted infections, including HIV, than the uncircumcised state[23]. It is to be feared that the proportion of patients with advanced immunodeficiency will increase over time, causing serious diagnostic and therapeutic problems since severe mucocutaneous herpes lesions will become more frequent[26]. As a consequence of the civil war in 1994, a high proportion of the population served by the CMS succumbed or fled to neighbouring countries. Future research cannot be linked to the population observed in the present study.

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