Genital ulcers and concomitant complaints in men attending a STI clinic: implications for STI management

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ABSTRACT:

Background:

Although genital herpes has emerged as the most common cause of genital ulcers in Southern Africa, treatment for herpes is not available routinely in the region. This study was done to determine the aetiology of genital ulcers in men in Durban and assess other STI –related symptoms, presentation and treatment patterns in this group.

Methods:

Polymerase chain reaction (PCR) tests were performed on specimens from consecutive male patients with genital ulcers to detect sexually transmitted pathogens. PCR was also performed for the detection of *Neisseria gonorrhoeae*, *Chlamydia trachomatis*, and *Trichomonas vaginalis* on urethral specimens from consecutive subjects with dysuria and/or urethral discharge. Antibody tests for syphilis and Herpes simplex virus type-2 (HSV-2) and HIV antibodies were performed.

Results:

Of 162 patients enrolled with genital ulcers, 77.7% were HIV positive and 84.6% had antibodies to HSV-2. PCR results showed the following prevalences: HSV-2 53.7%, lymphogranuloma venereum 13.6%, *Treponema pallidum* 3.7%, *Hemophilus ducreyi* 1.2%, mixed infections 6.2%, no pathogens identified 33.3%. One case of donovanosis was diagnosed clinically. In men with HSV-2 ulcers, delay before attendance recorded for 68 men was: 1-3 days (24%), 4-7 days (47%), 8-14 days (12%), 15-30 days (12%), >30 days (6%). History taking using prompting increased the sensitivity but decreased the specificity and positive predictive value of reported genital ulceration when assessed against ulcers seen on examination.

Conclusions:

Men at risk of genital ulcers should be asked about relevant symptoms with and without prompting and examined clinically to maximize the likelihood of correct diagnosis and treatment. The finding of a high prevalence of HSV-2 and associated dysuria cautions against providing empiric treatment for gonorrhoea and chlamydia in ulcer patients with dysuria but without urethral discharge. Innovative strategies to limit the burden of HSV-2 infection in this population are required.

Short summary

In men with genital ulcers, prompting about other symptoms and clinical examination increased the likelihood of correct diagnosis. Syndromic management strategies are challenged when the prevalence of genital herpes is high.

INTRODUCTION

Genital ulcer disease (GUD) is a significant risk factor both for acquisition and onward transmission of HIV [1]. Early diagnosis and correct treatment of GUD is therefore an important element of any HIV control programme. In Durban, South Africa, a significant association between GUD and HIV was identified early on [2]. Syphilis and chancroid were then common in Durban as elsewhere in other parts of Africa [3, 4] but these conditions have both declined whilst genital herpes has emerged as the most frequent cause of GUD in Southern Africa [5-9].

Current WHO STI treatment guidelines for GUD [10] now recommend treatment for chancroid and syphilis, and herpes if the prevalence is 30% or greater and, if episodic therapy is chosen, treatment should be started during the prodrome or within one day after onset of lesions. However, there is a need to validate these guidelines that include counselling and education in Africa where the association between genital herpes and HIV is strong [11]. Treatment for genital herpes is still considered not to be feasible in the public sector due to lack of education with respect to symptom recognition and cost of therapy. We therefore sought to determine the aetiology of genital ulcers in men in Durban and assess other STI-related presentation and treatment patterns in this group that has a high prevalence of HIV.

METHODS

Study population

Consecutive men attending the Prince Cyril Zulu STI clinic in Durban for a new STI related problem between January-March 2004 were enrolled. Inclusion criteria for subjects were being sexually active, heterosexual, aged 16-75 and with the potential ability to return in two weeks. Subjects were included even though they may have had antibiotics in the previous 14 days. Signed consent was obtained from all subjects. At enrolment, sociodemographic data were obtained and a sexual behaviour questionnaire was administered by a trained interviewer in a face-to-face interview as reported previously [12]. Subjects were initially asked about their presenting complaint(s) as an open question. They were then asked if they had any additional complaints relating to genital ulceration, urethral discharge or dysuria. Information about duration of symptoms of the main initial unprompted complaint was collected. Clinical examination of the genital area was performed by a physician and information recorded on circumcision status. For this study, a genital ulcer was defined as an area with mucosal discontinuity and included healing lesions. Patients with vesicles only were also included. Additional information about symptoms and clinical signs was elicited during the examination. Cases were managed according to National STI syndromic guidelines.

Specimen collection and laboratory methodology

Ulcers were cleaned with a sterile dry gauze and impression smears made onto glass slides for the detection of *Calymmatobacterium granulomatis* by RapiDiff staining [7]. A specimen was then obtained by rolling a Probetec® cervical sponge over the base of the ulcer. The material collected was suspended in 1.0 mL PBS, pH 7.6 and placed in an ice-containing cooler box that was transported to the laboratory within 4 hours. DNA was extracted immediately on arrival using the Qiagen Blood and Tissue Mini Kit (Qiagen, USA). DNA was stored at -70° C until use.

Polymerase chain reaction (PCR) was used for the detection of Herpes Simplex virus type 2 (HSV-2), *Treponema pallidum* (TP), *Chlamydia trachomatis* lymphogranuloma

venereum (LGV) biovars and *Haemophilus ducreyi* (HD) as described previously [7, 13]. LGV was diagnosed using a nucleic acid amplification test [14]: a PCR targeting the 60-kDa cysteine-rich outer membrane protein was followed by digestion with *AccI*. *C. Trachomatis* L2 (ATCC VR-902B), *C. trachomatis* trachoma biovar (a laboratory isolate from a cervical specimen) and water were included as controls in each run. Detection of *Neisseria gonorrhoeae* and *Chlamydia trachomatis* on urethral specimens from subjects with dysuria and/or urethral discharge was done by means of the Strand Displacement Test (ProbeTec®) while an in-house PCR was used for *Trichomonas vaginalis* (TV) [15]. Specimens negative in all in-house PCR were subjected to a β -globin PCR. Those testing negative were excluded.

HIV was diagnosed using Determine HIV 1/2 (Abbott Laboratories, Chicago, IL). Positive results were confirmed with a second serological test (Capillus HIV1/2, Trinity Biotech, USA). Serological testing for HSV-2 antibodies was by Focus HSV-2 (Focus Diagnostics, Cypress, CA) with a positive test defined as a ratio of 1.10 or more of the optical density reading over that of the cut-off calibrator provided with the kit. Serological testing for syphilis was with the rapid plasma reagin (RPR) test (Becton Dickinson, Cockeysville, MD), and *Treponema pallidum* haemagglutination assay (TPHA) (Omega Diagnostics, UK). All laboratory testing was carried out unlinked and anonymised. Data from the questionnaires were also anonymised and linked to the results of laboratory testing. Participants were offered voluntary counseling and testing for HIV at the clinic site.

Swabs were taken from the subpreputial space in an additional nine patients without genital ulcers, five of whom had balanitis diagnosed clinically and four with marked inguinal lymphadenopathy. Fine needle biopsies from lymph nodes were obtained for PCR to diagnose herpes, syphilis, chancroid and LGV infections in four of these nine cases, three of whom had marked inguinal lymphadenopathy alone and one with both balanitis and inguinal lymphadenopathy.

Analysis and informed consent

Questionnaire data were double-entered and validated using EpiData v3.0 [16]. Laboratory data were entered into an Excel spreadsheet (Microsoft). The sensitivity, specificity, positive and negative predictive values of self reported and prompted symptoms of genital ulceration in men with genital ulceration on clinical examination were assessed. Ethical approval for the study was obtained from the ethics committees of the Nelson Mandela Medical School, University of KwaZulu- Natal, Ethekewini Municipality Health Department Research Committee, Durban and the London School of Hygiene & Tropical Medicine.

RESULTS

Six hundred and fifty men were asked to participate and none declined. Of these, 165 had genital ulcers confirmed on clinical examination and completed questionnaires. Laboratory results were available for 162. Table 1 shows some of the presenting features and microbial aetiology of patients with genital ulcers amongst STI clinic attenders.

Demographic characteristics

Data was available for 164/165 patients. The mean age was 29.9, standard deviation 7.8. Seventy six (46.3%) were new patients, 35 (21.2%) had no education or primary education only, 12 (7.3%) were married, 50 (30.3%) were living with a partner and 158 (95.8%) were uncircumcised. Fourteen (8.5%) had received antibiotics in the last 14 days.

Analysis of symptoms

Of the 161 men that complained of a genital ulcer spontaneously, 128 (79.5%) had ulcers on clinical examination (Table 1). An additional 30 complained of genital ulcers on direct questioning of whom 18 had ulcers confirmed on examination. Only one case presented with vesicles typical of genital herpes. Nineteen of those with ulcers on clinical examination did not complain of ulcers. Table 2 shows the sensitivity, specificity, positive and negative predictive values of self reported and prompted symptoms of genital ulcers compared to ulcers observed on clinical examination. History taking using prompting increased the sensitivity of reported genital ulceration from 77.6% to 88.5% but reduced both the specificity from 93.2% to 90.7% and the positive predictive value (PPV) from 79.5% to 76.4%.

Microbiology and serology

Of the 162 patients with genital ulcers confirmed by clinical examination and with samples for microbiological analysis, 126 (77.7%) were HIV positive and 137/160 (84.6%) had antibodies to HSV-2. PCR results showed the following prevalences: HSV-2 87 (53.7%), LGV 22 (13.6%), syphilis six (3.7%), chancroid two (1.2%), mixed 10

(6.2%) including seven with genital herpes and LGV, two with syphilis and LGV and one with chancroid and LGV. The prevalence of HIV in those with HSV-2 and LGV infections detected in ulcers was 77% and 86.4% respectively. One case of donovanosis was diagnosed clinically. No pathogens were identified in 54 (33.3%) in whom eight (14.8%) had both positive TPHA and RPR tests, 46 (85.2%) had antibodies to HSV-2 and 40 (74.1%) were HIV positive. LGV was confirmed in an additional four patients, all HIV positive, without ulcers, three from material obtained by fine needle aspiration from inguinal lymph nodes and one by swabbing the subpreputial space.

Duration of ulcers before presentation

The duration of delay before presentation to the clinic in all patients with genital ulcers (n=120) was 0-3 days in 20 (16.7%), 4-7 days in 55 (45.8%), 8-14 days in 19 (15.8%), 15-30 days in 13 (10.8%) and > 30 days in 18 (10.8%). The duration of ulceration was available for 68 of the men with genital herpes confirmed by PCR testing in whom the delay before attending was 0-3 days in 16 (23.5%), 4-7 days in 32 (47.1%), 8-14 days in eight (11.8%), 15-30 days in eight (11.8%) and > 30 days in four (5.9%); only one man presented within one day; HSV-2 antibodies were not detected in 11 of the 68 men, six of whom presented between 1-3 days and five between 4-7 days after onset of symptoms. The delay before attending in the 39 subjects (overall = 54, data for 39) with GUD and no pathogens identified was: 1-3 days in four (10.3%), 4-7 days in 17 (43.6%), 8-14 days in eight (20.5%), 15-30 days in three (7.7%) and > 30 days in seven (17.9%). There was little difference in the delay before attending between those with either genital herpes or no pathogens identified.

Genital ulcers, dysuria, urethral discharge

Seventy nine of the 165 (47.8%) patients with confirmed ulcers on examination complained either initially or during the clinical examination and further questioning of either urethral discharge and/or dysuria or were found to have urethral discharge. In this group, the following STIs were identified- gonorrhoea and/or chlamydia in 19 and

trichomoniasis in 13 including three with both gonorrhoea and chlamydia, one each with gonorrhoea or chlamydia. Of the 87 genital herpes cases confirmed by PCR, 44 (50.6%) complained of either dysuria or urethral discharge although only 15 complained of urethral discharge, eight had gonorrhoea, an additional three had chlamydia and three had trichomoniasis (one with gonorrhoea and one with chlamydia). All the patients with both ulcers confirmed on examination and urethral discharge/dysuria received treatment for chancroid, syphilis, gonorrhoea and chlamydia although only five additional co-infections with syphilis, all with low RPR titres were identified.

DISCUSSION

We found a notable inconsistency between patients' symptoms and what was found on clinical examination. The addition of prompting to patients' spontaneous complaints increased the sensitivity but decreased the specificity and PPV of history taking. Of note, we also found 45 patients who reported ulcers that did not have ulcers reinforcing the need both to spend time with patients to probe about other complaints and conduct a clinical genital examination to include retracting the foreskin in uncircumcised men.

The high prevalence of HSV-2 identified from genital ulcers and associated dysuria cautions against providing empiric treatment for gonorrhea and chlamydia in ulcer patients with dysuria but without urethral discharge. Our study found that about half of the 165 subjects with a genital ulcer confirmed on examination were also treated for urethritis although only 19 (11.5%) had either gonorrhoea or chlamydia, 15 (78.9%) of whom complained of urethral discharge. A previous study in mineworkers from South Africa concluded that because of the significant prevalence of urethritis in patients with GUD, syndromic management based on the presence of ulcers alone was inappropriate [17]. We found much lower prevalences of concomitant infections with gonorrhoea and chlamydia which may reflect the fact that contact with sex workers with high prevalence rates of gonorrhoea and chlamydia was much higher in the mineworker group than in Durban [12]. We did find TV in the urethra of 13 ulcer cases that had tests for TV and there could be a case for giving metronidazole for urethritis symptoms as in Malawi [18].

Although the emergence of herpes in Africa was identified some time ago, strategies to deal with this have been slow to emerge. This is partly due to the cost of anti-herpes medication and also the self-limiting nature of the condition. We found that about 6% of our patients with genital HSV-2 infection presented either within 1 day of symptoms or greater than 30 days, outside of which time treatment is usually not indicated other than in primary cases or HIV positive subjects. However, 77% of our subjects with HSV-2 positive ulcers were also HIV positive. Amongst STI clinic attenders with GUD in Malawi, an unweighted algorithm with a HSV-2 prevalence of 35% and chancroid of 30%, concluded that men aged > 25 years with ulcers, symptoms > one week, shallow or

deep ulcers with or without lymph nodes could be treated for herpes, chancroid and syphilis although herpes treatment was not given routinely [19]. Although suppressive therapy is more effective than patient-initiated episodic treatment, the latter is a much cheaper option than and could be tried in our population [20, 21]. However, this would require specific education about recurrent episodes of genital ulceration and would need the provision of adequate levels of motivated trained staff in STI clinics.

Our study identified LGV as the cause of ulceration in 13.6% of cases. We also identified LGV in a few cases without ulcers but with inguinal lymphadenopathy and one case with balanitis. This raises the question about onward transmission from individuals without an obvious source of infective material. The reasons for the significant LGV prevalence are unclear but improved diagnostics have undoubtedly been a factor. Eighty six per cent of our LGV cases were HIV positive, a similar level to those with either herpes or ulcers with no cause identified. The clinical diagnosis of LGV is difficult and local syndromic management may not be adequate in that treatment with erythromycin was given for five days whereas WHO advise two weeks or even longer for definitive treatment [10]. However, good responses to treatment have been reported previously in Durban after five days of erythromycin and we would recommend that patients are followed up after 7 days if they are not improving significantly [7].

Despite technological advances, a significant number of ulcers cases were seen in whom no definitive diagnosis could be made. We believe that a large proportion of these are due to genital herpes. Our study showed that the most frequent delay in those with HSV-2 PCR positive ulcers was 4-7 days. Given this delay it may be that the PCR sensitivity would be below that expected both in this group and those with older HSV ulcers that were resolving spontaneously as reported elsewhere [22]. Also, we enrolled patients who had received antibiotics in the previous two weeks and some of these may have had ulcers due to chancroid, LGV or syphilis infections not identified by PCR.

Chancroid, syphilis and donovanosis have diminished considerably in Durban over the last 15 years. The reasons for this are probably multifactorial. Firstly, it could reflect

expanded access and higher quality syndromic management than that prior to the 1990's when tetracycline rather than erythromycin was prescribed widely to cover chancroid and benzathine penicillin administration for syphilis was patchy. The long delays in seeking health care seen previously with donovanosis appear to have been resolved by increased awareness of and willingness to treat at the primary care level. In addition condom use has increased although levels of use are still quite low [12].

The management of possible cases of syphilis still presents problems. Syphilis has decreased significantly to less than 5% of ulcers but treatment is still recommended for syphilis in all cases of GUD. This means that those with genital herpes and recurrent herpes are treated for syphilis every time they attend with genital ulcers and this impacts considerably on staff costs and time. One way of resolving this issue might be to do an on-site RPR test in those with ulcer symptoms of more than 14 days duration when the RPR test has a high sensitivity for detecting early syphilis. The downside of this option would be that a very small number of syphilis ulcers might be missed. However, this would obviate the need for unnecessary benzathine penicillin injections in the vast majority and would free up time for the RPR test. Furthermore, those with positive RPR results could be treated with three injections of benzathine penicillin to cover latent syphilis rather than the one injection that they receive currently.

Although algorithms for GUD are generally held in higher regard than those for other STI syndromes conditions [23], this situation has changed with the emergence of genital herpes and there must be real concerns about syndromic management of GUD and the mixed messages of giving treatment for syphilis and chancroid but not herpes in those with genital herpes particularly in patients with a high prevalence of HIV.

References

1. Dickerson MC, Johnstone J, Delea TE et al. The causal role for genital ulcer disease as a risk factor for transmission of human immunodeficiency virus. *Sex Transm Dis* 1996;23: 429-440
2. O'Farrell N, Windsor I, Becker P. Risk factors for HIV-1 in heterosexual attenders at a sexually transmitted diseases clinic in Durban. *S Afr Med J* 1991; 80: 17-20
3. Bogaerts J, Vuylsteke B, Martinez Tello W et al. Simple algorithms for the management of genital ulcers: evaluation in a primary health care centre in Kigali, Rwanda. *Bull WHO* 1995; 73: 761-767
4. Ndinya- Achola JO, Kihara AN, Fisher LD et al. Presumptive specific clinical diagnosis of genital ulcer disease in a primary health care setting in Nairobi. *Int J STD AIDS* 1996;7: 201-5
5. O'Farrell N. Increasing prevalence of genital herpes in developing countries: implications for heterosexual HIV transmission and STI control programmes. *Sex Trans Inf* 1999;75:377-384
6. Chen CY, Ballard RC, Beck-Sague CM et al. Human immunodeficiency virus infection and genital ulcer disease in South Africa: the herpetic connection. *Sex Transm Dis* 2000;27;21-9
7. Moodley P, Sturm PJ, Vanmali T et al. Association between HIV-1 infection, the etiology of genital ulcer disease, and response to syndromic management. *Sex Transm Dis* 2003;30: 241-245
8. Lai W, Chen CY, Morse SA et al. Increasing relative prevalence of HSV-2 infection among men with genital ulcers from a mining community in South Africa. *Sex Trans Infect* 2003;79: 202-7
9. Paz- Bailey G, Rahman M, Chen CY et al. Changes in the aetiology of sexually transmitted diseases in Botswana between 1993 and 2002: implications for the management of genital ulcer disease. *Clin Infect Dis* 2005;41: 1304-1312.
10. WHO. Guidelines for the management of Sexually Transmitted Infections. 2003
11. Wald A, Link K. Risk of Human Immunodeficiency Virus infection in herpes simplex virus type 2 seropositive persons: a meta analysis. *J Infect Dis* 2002; 185-45-52
12. O'Farrell N, Morison L, Moodley P et al. Association between HIV and subpreputial penile wetness in uncircumcised men in South Africa. *JAIDS* 2006;43: 69-77

13. Sturm PD, Moodley P, Govender K et al. Molecular diagnosis of lymphogranuloma venereum in patients with genital ulcer disease. *J Clin Micro* 2005;43: 2973-2975
14. Watson MW, Lambden PR, Clarke IN. Genetic diversity and identification of human infection by amplification of the chlamydial 60-kilodalton cysteine-rich outer membrane protein gene. *J Clin Microbiol* 1991;29: 1188-1193
15. Sturm PD, Moodley P, Khan M et al. Aetiology of male urethritis in patients recruited from a population with a high HIV prevalence. *Int J Antimicrobial Agents* 2004;24 (Suppl 1) : S8- 14
16. Lauritsen JM & Bruus M. EpiData (version 3). A comprehensive tool for validated entry and documentation of data. The EpiData Association, Odense Denmark, 2003
17. Ballard RC, Fehler HG, Htun Y et al. Coexistence of urethritis with genital ulcer disease in South Africa: influence on provision of syndromic management. *Sex Trans Infect* 2002;78:274-277
18. Price MA, Zimba D, Hoffman IR et al. Addition of treatment for trichomoniasis to syndromic management of urethritis in Malawi. *Sex Transm Dis* 2003;30: 516-522
19. Hoyo C, Hoffman I, Moser BK et al. Improving the accuracy of syndromic diagnosis of genital ulcer disease in Malawi. *Sex Transm Dis* 2005;32: 231-237
20. Fife KH, Almekinder J, Ofner S. A comparison of one year of episodic or suppressive treatment of recurrent genital herpes with valacyclovir. *Sex Transm Dis* 2007;34:297-301
21. O'Farrell N, Moodley P, Sturm AW. Genital herpes in Africa: time to rethink treatment. *Lancet* (in press)
22. Scoular A, Gillespie G, Carman WF. Polymerase chain reaction for diagnosis of genital herpes in a genitourinary medicine clinic. *Sex Trans Inf* 2002;78:21-25
23. Trollope-Kumar K, Guyatt G. Syndromic approach for treatment of STIs: time for a change. *Lancet* 2006; 367: 1380-1

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Table 1: Presentation and microbial aetiology of patients with genital ulcers amongst STI clinic attenders.

				Aetiology of GUD (n=161) ¹				
Genital ulcer symptom	Number of men N=650	genital ulcers seen on examination N=165	Number swabbed N=162	HSV ² n=87	LGV ³ n=22	TP ⁴ n=6	HD ⁵ n=2	No pathogens isolated n=54
Duration of spontaneously reported genital ulceration: 1-3 4-7 8-14 15-30 <30 Missing				16 32 8 8 4 19	1 6 3 4 2 6	- 3 0 1 - 2	- 2 0 0 - -	4 17 8 3 7 15
Total reporting genital ulceration spontaneously	161 (24.8)	128 (79.5)	128	65	20	5	1	47
Additional reporting genital ulceration on direct questioning	30 (4.6)	18 (60.0)	18	11	1	1	1	4
Total genital ulceration(spontaneous and direct questioning)	191 (29.4)	146 (76.4)	145	76	21	6	2	51
No genital ulcers reported	459 (70.6)	19 (4.1)	17	11	3	0	0	3

1: 1 case of clinical donovanosis; 2: Herpes Simplex virus (HSV), includes 7 mixed infections with lymphogranuloma venereum (LGV); 3: LGV, includes 7 mixed infections with HSV, 1 with Treponema Pallidum (TP), 1 with Hemophilus ducreyi (HD); 4: TP, includes 1 mixed infection with LGV; 5: HD, includes 1 mixed infection with LGV

Table 2: Validity of self reported and prompted symptoms of genital ulcers in men with genital ulceration on clinical examination

Genital ulceration symptoms	Genital ulcers seen on clinical examination			
	Sensitivity (%)	Specificity (%)	Positive predictive value (%)	Negative predictive value (%)
Unprompted	128/165 (77.6)	452/485 (93.2)	128/161 (79.5)	452/489 (93.2)
Prompted	146/165 (88.5)	440/485 (90.7)	146/191 (76.4)	440/459 (95.9)