Update on the Royal Perth Hospital
Anogenital Wart Database

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1. Introduction

Genital warts are still thought to be a benign cosmetic problem despite increasing evidence that they may harbour high-grade intra-epithelial neoplasia (IN). We have previously published high rates of high-grade IN in surgically excised warts from December 1995 to December 2004 in a retrospective review of 115 men and 38 women, 29 of whom had HIV infection. The rates of IN were 78% (52% high-grade) in men with HIV, and 33% (20% high-grade) in men without HIV. In women the IN rate was 8.3% (2.8% high-grade).(McCloskey et al. 2007) Since reporting the data initially, the database has continued to accrue cases and we have analysed the data to see if the previous findings have held and to see if there has been a change in the frequency of reported IN over time.

2. Materials and methods

The patient population, surgical and specimen handling process has been previously reported.(McCloskey et al. 2007) The population consists of patients referred to the Sexual Health Clinic at Royal Perth Hospital from December 1995 to December 2010. Anal or perianal warts were treated by scissor excision by a single sexual health physician if they were large or multiple or located within the anal canal by scissor excision. Patients who
were immunosuppressed or who had only mapping biopsies performed or whose HIV status was unknown were excluded from the main analysis.

3. Data analysis

Descriptive analysis used percentages for categorical and ordinal variables and means for continuous variables. Viral load and CD4 counts and percentages for HIV positive patients were found to follow a skewed log-normal distribution (Shapiro-Wilk’s test, p > 0.05 for all variables) and the geometric mean was reported as an unbiased estimate of the mean. These variables were transformed using a natural logarithm for further analysis that was based upon an assumption of normality. For bivariable analysis of categorical and ordinal variables the likelihood ratio \( \chi^2 \) test was used. For these analyses with continuous variables the Wilcoxon rank-sum test was used. Multivariable linear logistic regression analysis was used to examine associations with HIV status and other dichotomous variables and ordinal logistic regression was used to examine associations with the degree of IN. The validity of these latter statistical models depends upon an assumption of proportional odds across the outcome categories and this was assessed using the Brant test (Brant 1990). In every case the analysis was found to be valid. The assumption of linearity for continuous independent variables was assessed using restricted cubic splines and where non-linearity was found a spline regression was used to model the association (Harrell 2001). Analysis of the degree of IN used operation as the unit of investigation and because operations are clustered within patients the assumption of independent sampling is violated. Estimates of standard error and p values were based upon a robust estimation of the clustered data. A p value less than 0.05 was regarded as statistically significant for all analysis. The analysis was conducted using the Stata statistical package (Version 11.1). (Corp 2009).

4. Results

4.1 Patient characteristics and operative procedures

Removal of anal and/or perianal warts was performed during 461 operations in 343 patients, 255 (74%) men and 88 (26%) women of whom 278 (81%) of patients and 378 (82%) of operations were eligible to be included in the analysis. Reasons for exclusion included lack of histology results because of insufficient surgical material (total 44 patients, 30 men and 14 women), unknown HIV status (9 patients), operations performed for mapping biopsies (19 operations) and immunosuppression other than HIV (16 patients). In some instances there were multiple reasons for exclusion. Table 1.1 shows demographic data HIV antibody status for males, and Table 1.2 shows demographic data by HIV antibody status for females. Sixty-one eligible patients had HIV infection (59 men and two women). Men with HIV were significantly older than men without HIV (mean 8.7 years, 95% CI: 7.7-9.67, p<0.00001 rank sum test) (see Figure 1: Age distribution by HIV status).

A spline regression analysis showed that the likelihood of being HIV positive increased non-linearly with age in this population. The proportion positive increased at a significantly greater rate with each increasing year of age until about 30 years after which the proportion increases at a statistically significant but slower rate of increase (see Figure 2: HIV risk for men by age).
Geometric mean age = 30.5 yrs

Geometric mean age = 40.4 yrs

Graphs by HIV status, p < 0.00001, rank-sum test.

Fig. 1. Age distribution by HIV status.

Note: Shaded area shows the 95% confidence interval.

Fig. 2. HIV risk for men by age, results of a non-linear spline regression
Seventy-three percent of male patients were homosexual/bisexual, 27% heterosexual and 47% current smokers. At the time of the first operation, the average duration of HIV infection was 3.4 years (95% CI: 2.4-5.0, geometric mean); the average CD4 T-cell count at first surgery was 329 x10^6/L (95% CI: 258-420 x10^6/L, geometric mean) and the average nadir CD4 T-cell count prior to surgery was 96 x10^6/L (95% CI: 66-139 x10^6/L, geometric mean). The HIV viral loads ranged from 40 to 135,000 viral copies/mL with 50% having a viral load less than 395, 25%, 395–20,400 and 15%, 20,400 –38,000). Sixty-four% of men without HIV were homosexual/bisexual, 36% heterosexual and 43.5% current smokers. The number of operations performed ranged from one to five for both HIV -positive men and HIV-negative men.

4.2 Change over time
Differences between the period December 1995-December 2004 and January 2005-December 2010 shows that there has been significant change in the patient population in the period December 2004 to Jan 2005 compared to the previously reported period of 1995 to 2004 (Table 2). Variables that show the change are age (3.0 years older, p=0.033), almost half as many (57% more) HIV infected patients (p =0.032), fewer homosexual (7% less) but more bisexual men (39% more), the number of lifetime partners is increased in males but not females, a greater proportion of patients have a history of chlamydia, there is less perianal disease and more anal disease. Importantly however between the two periods there has been no significant change in AIN or perianal intraepithelial neoplasia (PAIN).

4.3 Rates of IN were highest in patients with HIV infection
When all the operations are considered men with HIV infection and for whom histological data was available, 45 (44.1%) had AIN 2–3 (high-grade AIN) and 19 (58.1%) had PAIN 2–3 (Table 3). Overall, men with HIV had some form of high-grade IN in 68 (57.8%), with 85 (78%) having any grade of abnormality (AIN or PAIN 1-3). HIV negative men had high-grade IN in 49 (24.7%), 25 (5.7%) had high-grade PAIN, and 32 (21.4%) had high-grade AIN. The risk of IN was 2.9 times higher for HIV positive men than those without HIV for AIN (p<0.0001, ordinal logistic regression analysis) and 4.0 times higher than men without HIV for PAIN (p<0.0001, ordinal logistic regression analysis). The rate of AIN 2-3 was 10.7% for HIV-negative women and 1.8% for PAIN. Except for a few perianal lesions with classical features of Bowen’s disease, none of the IN was evident at operation and was only discovered with histopathological examination.

The rate of PAIN or AIN 2–3 was significantly higher 64.7% (66/36) for homosexual/bisexual men with HIV and 33.3% (45/90) in those without HIV infection (P<0.0001). The rate of IN was consistently higher in the anal canal compared with the perianal area (p < 0.00001) (Table 3.1 and 3.2). Table 4 shows this difference for all operations, the samples would fall diagonally in the table if there were no difference between anal and perianal samples (see Table 4). Of the patients with PAIN or AIN, there is a statistically significant interaction between HIV status and current smoking, with those who are HIV positive and current smokers being 6.0 times more likely to have a higher grade of IN than those who are HIV negative and have never smoked (P=0.010, ordinal logistic regression).
### Table 1.1. Demographic data by HIV antibody status for males

|                          | HIV positive n = 64 (%) | HIV negative n = 153 (%) | P    |
|--------------------------|------------------------|--------------------------|------|
| Mean age (years)         | 40.4                   | 30.1                     | <0.0001 |
| Range                    | 22.9 – 71.8            | 13.7 – 71.7              |      |
| **Sexual preference**    |                        |                          |      |
| Heterosexual             | 3 (4.7)                | 55 (36.0)                | <0.001 |
| Homosexual               | 50 (78.1)              | 77 (50.3)                |      |
| Bisexual                 | 11 (17.2)              | 21 (13.7)                |      |
| **Lifetime Number of Sexual Partners** |          |                          |      |
| 1 - 10                   | 4 (8.2)                | 32 (30.2)                | 0.002 |
| 11 - 50                  | 17 (34.7)              | 39 (36.8)                |      |
| >50                      | 28 (57.1)              | 35 (33.0)                |      |
| **Smoking status**       |                        |                          | 0.445 |
| Current                  | 34 (53.1)              | 67 (43.8)                |      |
| Ex-smoker                | 5 (7.8)                | 13 (8.5)                 |      |
| Never smoked             | 25 (39.1)              | 73 (47.7)                |      |
| **History of STDs**      |                        |                          |      |
| Syphilis                 | 8 (12.5)               | 4 (2.6)                  | 0.006 |
| Gonorrhoea               | 20 (31.3)              | 14 (9.2)                 | <0.001 |
| Chlamydia                | 9 (14.1)               | 18 (11.8)                | 0.643 |
| NSU                      | 5 (7.8)                | 9 (5.9)                  | 0.604 |
| History of genital herpes| 17 (26.6)              | 12 (8.0)                 | <0.001 |
| HSV-2 seropositive       | 22 (42.3)              | 20 (18.2)                | 0.001 |
| Hepatitis C antibody positive | 5 (8.8)              | 4 (3.1)                  | 0.107 |
| **Wart site first operation** | 3 (4.7)                | 34 (22.2)                | <0.001 |
| Perianal only            | 21 (32.8)              | 35 (22.98)               |      |
| Anal only                | 38 (59.4)              | 74 (48.4)                |      |
| Perianal & anal          | 0                      | 9 (5.9)                  |      |
| Perianal & penile        | 1 (0.7)                | 2 (3.1)                  |      |
| Perianal, anal & penile  |                        |                          |      |
| **Number of operations** | 38 (59.4)              | 118 (77.1)               | 0.029 |
| 1                        | 13 (20.3)              | 26 (17.0)                |      |
| 2                        | 8 (12.5)               | 6 (3.9)                  |      |
| 4                        | 4 (6.3)                | 2 (1.3)                  |      |
| 5                        | 1 (1.6)                | 1 (0.7)                  |      |

Table 1.1. Demographic data by HIV antibody status for males
**Table 1.2. Demographic data by sex and HIV antibody status for females**

| **Women** | **n = 2** | **n = 59** | **p** |
|-----------|----------|-----------|------|
| Mean age (years) | 22.2 | 27.4 | 0.352 |
| Range | 19.5-25.4 | 16.9-65.0 |  |
| **Sexual preference** | | | 0.723 |
| Heterosexual | 2 (100) | 57 (96.6) |  |
| Homosexual/bisexual | 0 | 2 (3.4) |  |
| **Lifetime number of sexual partners** | | | 0.562 |
| 1 - 10 | 2 (100) | 32 (74.4) |  |
| 11 - 50 | 0 | 6 (14.0) |  |
| >50 | 0 | 5 (11.6) |  |
| **Smoking status** | | | 0.160 |
| Current | 0 | 31 (52.5) |  |
| Ex-smoker | 0 | 5 (8.5) |  |
| Never smoked | 2 (100) | 23 (39.0) |  |
| **History of STDs** | | |  |
| Syphilis | 0 | 0 |  |
| Gonorrhoea | 1 (50.0) | 3 (5.1) | 0.081 |
| Chlamydia | 1 (50.0) | 5 (8.5) | 0.138 |
| NSU | 0 | 1 (1.7) | 0.795 |
| History of genital herpes | 0 | 3 (5.4) | 0.642 |
| HSV-2 seropositive | 0 | 4 (9.5) |  |
| Hepatitis C Antibody positive | 0 | 0 |  |
| **Wart site first operation** | | | 0.732 |
| Perianal only | 0 | 11 (18.6) |  |
| Anal only | 0 | 4 (6.8) |  |
| Perianal and anal | 0 | 11 (18.6) |  |
| Perianal, anal and vulval | 1 (50.0) | 13 (22.0) |  |
| Perianal and vulval | 1 (50.0) | 16 (27.1) |  |
| **Number of operations** | | | 0.138 |
| 1 | 1 (50.0) | 54 (91.5) |  |
| 2 | 1 (50.0) | 5 (8.5) |  |
| Variable                          | Coefficient | 95% confidence interval | p       |
|----------------------------------|-------------|-------------------------|---------|
|                                  |             | LCL | UCL          |         |
| Age                              | 3.04        | 0.25 | 5.84        | 0.033   |
| HIV status                       | 0.57        | 0.01 | 1.12        | 0.046   |
| Sexual preference (male)         |             |     |             |         |
| Heterosexual                     | 0 (reference)|     |             |         |
| Homosexual                       | 0.93        | 0.26 | 1.59        | 0.006   |
| Bisexual                         | 1.39        | 0.48 | 2.30        | 0.003   |
| Sexual preference (female)       |             |     |             |         |
| Heterosexual                     | 0 (reference)|     |             |         |
| Lesbian                          | 0.45        | -2.37 | 3.27        | 0.756   |
| Lifetime sexual partners         |             |     |             |         |
| Male                             | 0.73        | 0.11 | 1.34        | 0.020   |
| Female                           | -0.02       | -1.37 | 1.33        | 0.976   |
| Smoking status                   |             |     |             |         |
| Never                            | 0 (reference)|     |             |         |
| Current                          | -0.41       | -0.91 | 0.08        | 0.102   |
| Ex-smoker                        | 0.28        | -0.62 | 1.18        | 0.543   |
| History of:                      |             |     |             |         |
| Syphilis                         | 1.33        | 0.005 | 2.66        | 0.049   |
| Gonorrhoea                       | 0.34        | -0.35 | 1.03        | 0.332   |
| Chlamydia                        | 1.00        | 0.23 | 1.76        | 0.011   |
| HSV2                             | -0.12       | -.086 | 0.63        | 0.756   |
| NSU                              | 0.06        | -0.99 | 1.10        | 0.914   |
| Warts                            | -1.90       | -2.76 | -1.04       | <0.001  |
| HAV                              | 0.26        | -0.29 | 0.82        | 0.349   |
| HCV                              | -0.33       | -1.67 | 1.02        | 0.634   |
| HSV serology                     | -0.50       | -1.16 | 0.16        | 0.140   |
| Prior wart treatment             | -0.93       | -1.41 | -0.44       | <0.001  |
| Number of operations             | -0.20       | -0.41 | 0.00        | 0.051   |
| Site perianal                    | -0.86       | -1.38 | -0.35       | 0.001   |
| Site anal                        | 1.48        | 0.89 | 2.08        | <0.001  |
| PAIN                             | -0.23       | -0.93 | 0.47        | 0.528   |
| AIN                              | 0.17        | -0.31 | 0.65        | 0.488   |
| IN                               | 0.28        | -0.19 | 0.74        | 0.246   |
| CD4 count at surgery (ln)        | -0.28       | -0.76 | 0.21        | 0.262   |
| Viral load at surgery (ln)       | -1.63       | -2.85 | -0.41       | 0.010   |
| CD4 count at nadir (ln)          | -0.06       | -0.35 | 0.23        | 0.681   |

Table 2. Differences between the period December 1995-December 2004 and January 2005-December 2010
5. Discussion

The data is consistent with our earlier observations of high rates of any IN men with HIV infection. Approximately 25% of HIV negative men had high-grade IN, indicating that anal/perianal warts in HIV negative men cannot be assumed to be benign lesions. Conversely anogenital warts in men with HIV have a high likelihood of containing some degree of IN with 62% having high-grade. The consistency of the data over the longer time period it highlights the importance of treating anal/perianal warts to remove IN that may eventually progress to invasive cancer as has previously been reported in the literature. (Scholefield, Castle and Watson 2005, Siegel 1962, Sturm et al. 1975) In particular the rates of anal cancer are rapidly rising in men with HIV (Crum-Cianflone et al. 2010) and it may be that men with warts represent a population of men at increased risk especially given the relatively common findings of anal cancer in men with warts (8.8%). Genital warts are recognised as a risk factor for the development of IN in renal transplant recipients. (Patel et al. 2010) AIN has been reported to progress to anal cancer with 10% progression at 5 years with higher rates in immunosuppressed individuals or those with multifocal disease. (Scholefield, Harris and Radcliffe 2011) Unlike other authors (Schlect et al. 2010) we did not detect anal cancer in any of the anal warts excised, but in our population, men with warts are referred early for surgery soon after the warts are diagnosed in the clinic. Schlect et al. (Schlect et al. 2010) have similarly found high rates of IN in anal condylomata in 75/159 (47%) HIV positive men. In particular, given the increasing rates of anal cancer in men (Daling et al. 1982, Goedert et al. 1998) with HIV, further studies to explore the role of ‘low-risk’ HPV genotypes associated with warts and their possible role in anal cancer development should be undertaken. The fact that genotyping studies of anal cancer are

|                | HIV positive | HIV negative | P     |
|----------------|--------------|--------------|-------|
| Anal and/or perianal IN | n= 109       | n= 199       |       |
| None           | 24 (22.0)    | 112 (56.3)   | <0.001|
| IN 1           | 17 (15.6)    | 38 (19.1)    |       |
| IN 2           | 20 (18.4)    | 27 (13.6)    |       |
| IN 3           | 48 (44.0)    | 22 (11.1)    |       |
| Total IN       | 85 (78.0)    | 87 (43.7)    |       |
| Anal           | n = 102      | n = 150      |       |
| None           | 36 (35.3)    | 87 (58.0)    | <0.001|
| IN 1           | 21 (20.6)    | 31 (20.7)    |       |
| IN 2           | 20 (19.6)    | 22 (14.7)    |       |
| IN 3           | 25 (24.5)    | 10 (6.7)     |       |
| Total AIN      | 66 (64.7)    | 63 (42.0)    |       |
| Perianal       | n=67         | n=140        | <0.001|
| None           | 42 (61.2)    | 119 (85.0)   |       |
| IN 1           | 7 (10.5)     | 13 (9.3)     |       |
| IN 2           | 4 (6.0)      | 2 (1.4)      |       |
| IN 3           | 15 (22.4)    | 6 (4.3)      |       |
| Total IN       | 26 (38.8)    | 21 (15.0)    |       |

Table 3.1 IN by HIV antibody status , and anatomical site-perianal/anal for all operations for males
sometimes finding only HPV 6 indicate this ‘low –risk’ HPV genotype may not in fact be ‘low risk’ in the anal canal in men. (Hillman et al. 2010, Abramowitz et al. 2010) Vaccination of boys to prevent infection of HPV with both low and high-risk HPV types should be promoted.

**Anal and/or perianal IN**

|               | n = 3       | n = 62       |       |
|---------------|-------------|--------------|-------|
| None          | 2 (66.7)    | 45 (72.6)    | 0.308 |
| IN 1          | 0           | 12 (19.4)    |       |
| IN 2          | 0           | 1 (1.6)      |       |
| IN 3          | 1 (33.3)    | 4 (6.5)      |       |
| Total IN      | 1 (33.3)    | 17 (27.5)    |       |

|               | n = 1       | n = 33       |       |
|---------------|-------------|--------------|-------|
| None          | 0           | 17 (60.7)    | 0.298 |
| IN 1          | 0           | 8 (28.6)     |       |
| IN 2          | 0           | 0            |       |
| IN 3          | 1 (100)     | 3 (10.7)     |       |
| Total IN      | 1 (100)     | 11 (33.3)    |       |

|               | n = 3       | n = 56       |       |
|---------------|-------------|--------------|-------|
| None          | 2 (66.7)    | 53 (94.6)    | 0.088 |
| IN 1          | 0           | 2 (3.6)      |       |
| IN 2          | 0           | 1 (1.8)      |       |
| IN 3          | 1 (50.0)    | 0            |       |
| Total IN      | 1 (50.0)    | 3 (5.4)      |       |

Table 3.2 IN by HIV antibody status, and anatomical site- perianal/anal for all operations for females

| PAIN | PAIN | PAIN | PAIN | PAIN | PAIN |
|------|------|------|------|------|------|
| AIN  | Negative | IN 1 | IN 2 | IN 3 | Total |
|------|-----------|------|------|------|-------|
| Negative | 86 | 3 | 1 | 1 | 91 |
| %   | 49.7 | 1.7 | 0.6 | 0.6 | 52.6 |
| IN 1 | 29 | 6 | 1 | 2 | 38 |
| %   | 16.8 | 3.5 | 0.6 | 1.2 | 22.0 |
| IN 2 | 17 | 6 | 3 | 0 | 26 |
| %   | 9.8 | 3.5 | 1.7 | 0.0 | 15.0 |
| IN 3 | 7 | 0 | 1 | 10 | 18 |
| %   | 4.1 | 0.0 | 0.6 | 5.85 | 9.7 |
| Total | 139 | 15 | 6 | 13 | 173 |
| %   | 80.4 | 8.7 | 3.5 | 7.5 | 100.0 |

Asymptotic symmetry test: $\chi^2 = 46.4$, d.f. = 6, $p < 0.00001$

Table 4. Contrast of AIN and PAIN
6. References

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The past few decades have seen the escalation of HIV-infections and the 'frantic' search for new drugs to treat the millions of people that live with HIV-AIDS. However because HIV-AIDS cannot be cured, but only controlled with drugs, and the Antiretroviral (ARV) treatment itself results in some undesirable conditions, it is important to generate wider awareness of the plight of people living with this condition. This book attempts to provide information of the initiatives that have been used, successfully or unsuccessfully, to both prevent and combat this 'pandemic' taking into consideration the social, economic, cultural and educational aspects that involve individuals, communities and the countries affected.

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