Screening for Anal and Cervical Dysplasia in HIV-Infected Patients

Joel Palefsky, MD, FRCP(C)
Professor of Laboratory Medicine, University of California San Francisco
San Francisco, California

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SUMMARY BY TIM HORN
EDITED BY KATHRYN ANASTOS, MD, AND STEPHEN GOLDSTONE, MD

One need not be a cynic, or even the slightest bit jaded, to acknowledge that HAART is truly a double-edged sword. While it continues to have a positive impact on both the incidence and prevalence of "classic" AIDS-related diseases, it has also paved the way for infections with long latency periods to fulfill their pathogenic potential and fulminate into life-threatening complications. A prime example of this therapeutic catch-22 is human papillomavirus (HPV) and its sinister sequalae: squamous intraepithelial lesions and invasive cervical or anal carcinoma.

HPV Primer

Human Papillomavirus (HPV)

"One of the most striking and underappreciated things about HPV," began Dr. Joel Palefsky, "is that it is the most common sexually transmitted agent. It is more common than herpes; more common than chlamydia. It has been estimated that more than 75% of sexually active adolescents and adults, between the ages of 15 and 49, acquire at least one type of HPV infection during their lifetime. "The problem with HPV," Dr. Palefsky added, "is that only a small percentage of people actually know they have HPV. They have genital warts; they itch and they burn and they can be felt. But these are only the tip of the iceberg. There are a lot of people out there who don't have any noticeable symptoms but may still be walking around with precancerous lesions of the anus or cervix. These are the patients we need to remain conscious of and beef up our screening practices."

More than 100 different types of HPV exist, approximately 30 of which infect genital mucosal tissues. These include nononcogenic varieties—primarily types 6 and 11, as well as types 42, 43, and 44—that are associated with the development of genital condylomas or mild dysplasia and generally do not progress to either higher-grade lesions or cancer. Oncogenic varieties—types 16, 18, 31, and 35—are associated with more dysplastic lesions and account for the majority of invasive cervical or anal carcinoma cases. HPV type 16 accounts for approximately 50% of all cervical cancers; types 18, 31, and 35 account for an additional 20%; types 39, 45, 51, 52, 56, 58, 70, and others likely account for the rest.

"If our patients were infected with only one HPV type, such as a nononcogenic type, and somehow developed immunity against all the rest, we wouldn't have too much to worry about," Dr. Palefsky said. "But the fact is, a number of people are infected with more than just one type." In one San Francisco-based study conducted by Dr. Palefsky and his colleagues, 196/269 (73%) HIV-positive men who have sex with men (MSM) were infected with multiple HPV types, with type 16 being the most common viral type (Palefsky, 1998b). These findings also extend to a cohort of HIV-positive women with cervical HPV infection (see: Prevalence of HPV and Cervical Dysplasia in HIV-Positive Women, below). "The bottom line is that you can't rule out the possibility of high-grade dysplasia being present in someone who presents with genital warts. Where there is one type of HPV infection, there may be another lurking nearby."

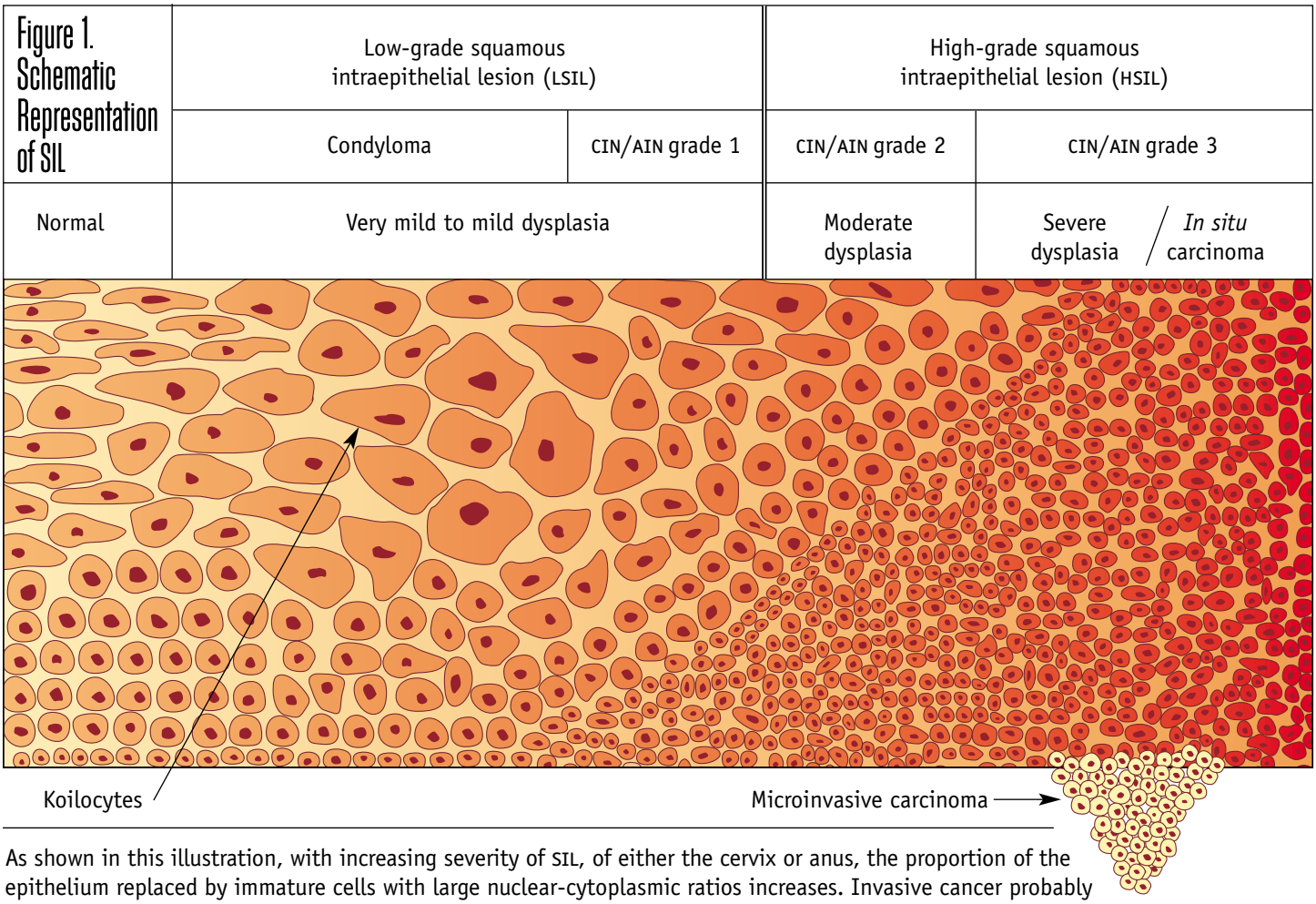
Infection of the Cervix and Anus

HPV infection initially takes place at the basal cell layer of the anogenital epithelium. In the cervix, this usually occurs at the level of the transformation zone or squamocolumnar junction, where the columnar epithelium of the endocervix meets the squamous epithelium of the exocervix. In the anal canal, HPV infection also occurs in the transformation zone, located at the junction of the stratified squamous epithelium of the anus with the columnar epithelium of the rectum. Cervical and anal epithelia are histologically very similar and are typically the place where most HPV-associated lesions, including invasive carcinoma, develop.

Classifying HPV Lesions and Carcinoma

Several classification systems have been used for the purpose of grading the cellular atypia seen in HPV lesions, varying from relatively benign lesions to invasive malignant neoplasms. The Bethesda system—the system employed by Dr. Palefsky—is the accepted standard for classifying and staging both cervical and anal precancerous lesions.

At one end of the HPV disease spectrum are anogenital warts (condyloma acuminatum) and mild dysplasia: cervical intraepithelial neoplasia grade 1 (CIN I) and anal intraepithelial neoplasia grade 1 (AIN I). The Bethesda system combines CIN I and AIN I into one single category: low-grade squamous intraepithelial lesions (LSIL). At the other end of the spectrum, are CIN/AIN grades 2 and 3, also known as moderate and severe dysplasia, respectively, and carcinoma *in situ*. The Bethesda system groups all these changes under



As shown in this illustration, with increasing severity of SIL, of either the cervix or anus, the proportion of the epithelium replaced by immature cells with large nuclear-cytoplasmic ratios increases. Invasive cancer probably arises from one or more foci of high-grade SIL (HSIL), as depicted in the drawing by epithelial cells crossing the basement membrane below the region of HSIL.

Source: Joel Palefsky, MD, FRCP(C)

high-grade squamous intraepithelial lesion (HSIL). Another cytological classification is that of atypical squamous cells of undetermined significance (ASCUS), which refers to cells that cannot be classified as either completely normal or dysplastic.

Similarly, different degrees of cervical or anal cancer may be staged using classification systems set up by the International Federation of Gynecologists and Obstetricians (for cervical cancer) and the National Cancer Institute (for anal cancer). The IFGO staging system for cervical cancer and the NCI staging system for anal cancer can both be accessed through the NCI web site: **http://cancernet.nci.nih.gov**.

Cervical Dysplasia and Carcinoma

General Screening Issues

SINCE ITS CLINICAL DEBUT IN THE 1940S, Papanicolaou (Pap) smear testing has been the gold standard for cytologic screening of the cervix. Although the false-negative rate of Pap smears in the general population has been reported to be as high as 20% to 45%—one-half of the false negatives are likely because of inadequate specimen sampling and the other half are attributed to failure to identify the abnormal cells or to interpret them accurately—its role in reducing the rates of cervical cancer over the past 50 to 60 years is undisputed. Prior to the introduction of Pap smears, the incidence of cervical cancer was 40–50/100,000 women. More recent data show that the incidence in the United States is approximately 8/100,000 women—a marked improvement—indicating that cervical cancer is, by and large, completely avoidable.

Yet, despite the recognized benefits of Pap smear screening, substantial subgroups of American women have not been screened or are not screened at regular intervals. According to a 1996 consensus statement issued by the National Institutes of Health (NIH), one-half of the women with newly diagnosed invasive cervical carcinoma have never had a Pap smear, and another 10% have not had a smear in the past five years (NIH Consensus Statement Online, 1996).

The unscreened populations include older women, the uninsured, ethnic minorities, especially Latinas and elderly African Americans, and poor women—not unlike the populations of women at the greatest risk of HIV infection. One-fourth of the cases of cervical cancer and 41% of the deaths occur in women age 65 and older. Data from the 1992 National Health Interview Survey, conducted by the U.S. Centers for Disease Control (CDC), indicated that one-half of all women age 60 and older have not had a Pap smear in the past three years. Although older women are screened less frequently, they have the same number of recent physician visits as younger women, which indicates the need to educate older women and their healthcare providers about the importance of Pap smear screening. For patients who are not involved in routine screening pro-

grams, any health-care encounter should be an opportunity to obtain a Pap smear and offer other screening modalities.

Pap smears that suggest abnormal cells require further evaluation by colposcopy, colposcopic-directed biopsy, and endocervical curettage. Colposcopy offers direct visualization of the cervix with an opportunity to biopsy sites of abnormality, and the results can be clinically correlated—by assessing characteristic color changes, vascular patterns, and margins—with the results of the Pap smear. Colposcopy-directed biopsy usually provides enough clinical evidence for an accurate diagnosis. If colposcopic evaluation is unsatisfactory or inconclusive, a cervical conization biopsy is required, performed by a loop electrical excision procedure (LEEP), laser, or cold knife.

Prevalence of HPV and Cervical Dysplasia in HIV-Positive Women

SINCE HIV AND HPV ARE TRANSMITTED IN somewhat similar fashions, the prevalence of HPV and cervical dysplasia among HIV-infected women would be expected to be relatively high. Data from the Women's Interagency HIV Study (WIHS) suggest that this may be true.

With respect to cervical HPV infection, a team headed by Dr. Palefsky employed PCR to check for HPV in cervicovaginal lavage fluid collected from 2015 HIV-infected women and 577 HIV-negative controls (matched for age, drug use, and number of sexual partners) (Palefsky, 1999). Evidence of HPV infection was found in 58% of the HIV-positive women, compared to 26% of the controls.

HIV-positive women participating in the WIHS were also more likely to be infected with multiple HPV types. Approximately 42% of the HIV-positive women, compared with 16% of the HIV-negative controls, had evidence of more than one type of HPV in lavage samples. Almost one quarter of the HIV-positive women were infected with three or more types of HPV.

Also of interest was an association between HPV infection, CD4+ cell counts, and HIV-RNA levels in the HIV-positive women. As the CD4+ cell count declined, a greater percentage of HIV-positive women were found to have HPV: approximately 45% of women with CD4+ counts >500 cells/mm^3, 55% of women with CD4+ counts between 200 and 500 cells/mm^3, and 70% of women with CD4+ counts <200 cells/mm^3 had PCR evidence of HPV infection. As for viral load, 71% of HIV-infected women with CD4+ counts above 500 cells/mm^3 and plasma HIV-RNA levels in excess of 100,000 copies/mL were found to be positive for HPV, compared to 44% of HIV-positive women with similarly high CD4+ cell counts and less than 44,000 HIV-RNA copies/mL. These data were recently corroborated by a report presented at the 13th International AIDS Conference in Durban (Sewell, 2000).

The WIHS has also demonstrated that HIV-infected women are more likely to have abnormal cervical cytology results (Massad, 1999). An analysis of Pap smears collected from 2054 HIV-positive women and 568 HIV-negative controls yielded a 40% overall prevalence of any type of abnormal Pap result (including ASCUS) in HIV-positive women, compared to an abnormal cytology rate of 17% among otherwise healthy controls. As with rates of HPV infection, rates of abnormal cytology corresponded with CD4+ cell counts in the HIV-positive women: approximately 26% of women with CD4+ counts >500 cells/mm^3, 35% of women with CD4+ counts between 200 and 500 cells/mm^3, and 53% of women with CD4+ counts <200 cells/mm^3 had abnormal Pap results.

While the CDC expanded its case definition of AIDS to include invasive cervical cancer, there hasn't been much in the way of hard evidence to conclude that HIV-positive women do, in fact, have a higher incidence of cervical cancer. While it is likely that HIV-positive women are more likely to develop cervical cancer than their HIV-negative counterparts—given the higher rates of HSIL among HIV-positive women—it's not at all clear if HIV-positive women progress faster to cervical cancer. "This is really a tough question to answer, given that we can't very well follow HIV-positive women with high-grade lesions to see how fast they progress to cancer," Dr. Palefsky pointed out. "This would be unethical. If we discover a high-grade lesion, in anyone, we need to treat it, not watch it progress."

A review of New York City AIDS surveillance data collected between 1990 and 1995 found that the observed cervical cancer cases were two to three times higher than the expected number of cases (Chiasson, 1997). Another study reported a higher prevalence of invasive cervical cancer in HIV-positive patients compared with HIV-negative hospitalized patients, particularly among those aged 20 to 34 and among African-American women and Latinas (Weber, 1998). And in a recent analysis of women in the HER study, HIV-positive women had an invasive cervical cancer rate of 144 per 1,000 person-years as compared with 0 per 1,000 person-years in HIV-negative women (Phelps, 2000).

There are also data to suggest that HIV-positive women may see their cervical cancer metastasize to unusual locations (e.g., psoas muscle, the clitoris, and the central nervous system), have poorer responses to standard therapy, and have higher recurrences and death rates, as well as shorter intervals to recurrence or death, compared with HIV-negative women of similar stage (Klevens, 1996; Maiman, 1990).

Guidelines for the Assessment of Cervical SIL in HIV-Positive Women

BOTH THE CDC AND THE AGENCY FOR HEALTH Care Policy and Research recommend that HIV-infected women have a complete gynecologic evaluation, including a Pap smear and pelvic exam, as part of their initial evaluation (see Figure 2). As reiterated by Dr. Palefsky, a Pap smear should be obtained twice in the first year following an HIV diagnosis. If these results are normal, annual examinations are then indicated. However, more frequent Pap smears should be obtained from HIV-positive women with previous abnormal Pap smears (including ASCUS or low-grade lesions) and after treatment for cervical dysplasia. The American College of Obstetricians and Gynecologists recommends Pap smears every three to four months for the first year after treatment of precancerous cervical lesions, followed by Pap smears every six months.

As for colposcopy, the indications include any cytologic abnormality (including ASCUS and atypical glandular cells of undetermined significance [AGCUS]); after treatment of cervical dysplasia; and, perhaps, as an initial screening tool in HIV-positive women with less than 200 CD4+ cells/mm^3. Biopsies should be obtained at the time of colposcopy to confirm cytologic abnormalities.

Treatment Considerations in HIV-Positive Women with Cervical SIL

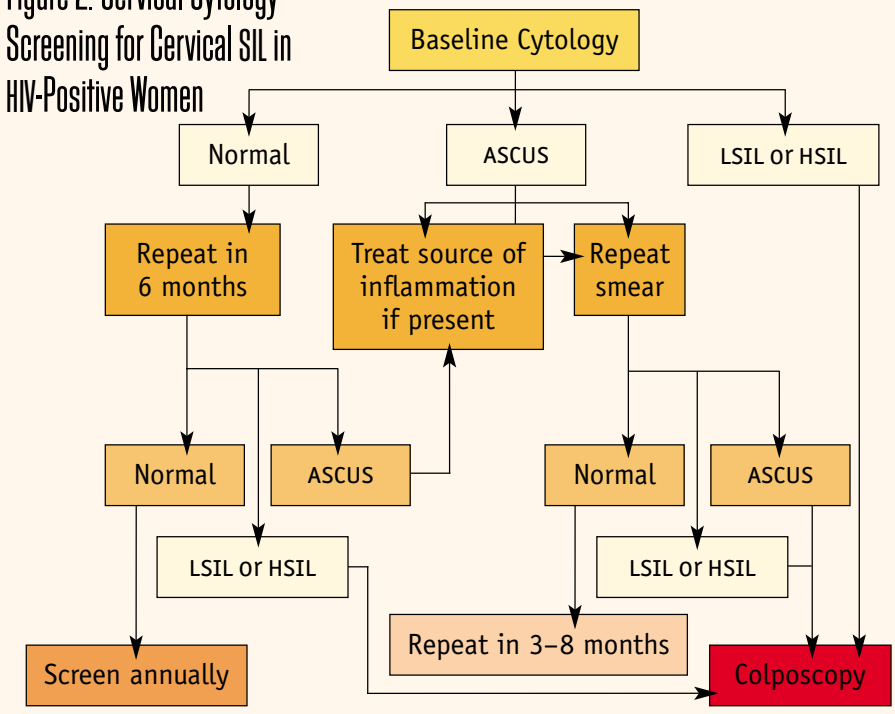
WHILE LOW-GRADE DYSPLASIA GENERALLY does not require treatment, given that it typically does not progress to high-grade dysplasia or cervical cancer, women with

HSIL require therapy. Loop electric excision, laser ablation, and cryotherapy are highly effective when the entire lesion and transformation zone can be seen by colposcopy and when there is an absence of endocervical involvement. For women who do not meet these criteria, cervical conization remains the gold standard.

Compared to HIV-negative women, in whom the risk of HSIL recurrence two years after specific therapy is between 5% and 10%, the recurrence rate in HIV-positive women is in the ballpark of 50% within 12 months after completion of therapy (Maiman, 1993). Thus, there is an urgent need for new treatments—and new treatment strategies—to reduce the recurrence of medium- and high-grade cervical dysplasia in HIV-infected women.

According to a study conducted by the AIDS Clinical Trials Group (ACTG 200), topical vaginal 5-fluorouracil (5-FU) cream (2 g biweekly for 6 months) was shown to reduce recurrence rates after standard treatment for high-grade cervical dysplasia in HIV-positive women (Maiman, 1999). Fifty women were randomized to receive 5% 5-FU; 51 women served as a control group and were simply monitored for CIN recurrence. After 18 months of follow-up, approximately 31% of the women in the control group experienced a recurrence of CIN, compared to 8% of women who received 5-FU. What's more, there were no grade III or IV toxicities reported in any women receiving 5-FU.

Other treatments in various stages of development include HPV peptide vaccines (e.g., Stressgen's HspE7) and difluoromethyl ornithine (DFMO), a chemotherapeutic agent being studied for its prophylactic potential.



Source: Joel Palefsky, MD, FRCP(C). Adapted from 1993 sexually transmitted diseases treatment guidelines. Centers for Disease Control and Prevention. *Morb Mortal Wkly Rep* 42(RR14):89-91, 1993.

The U.S. Centers for Disease Control and the Agency for Health Care Policy and Research recommend that HIV-infected women have a complete gynecologic evaluation, including a Pap smear and pelvic exam, as part of their initial evaluation. A Pap smear should be obtained twice in the first year following an HIV diagnosis. If these results are normal, annual examinations are then indicated. However, more frequent Pap smears should be obtained from HIV-positive women with previous abnormal Pap smears (including ASCUS or low-grade lesions) and after treatment for cervical dysplasia. The American College of Obstetricians and Gynecologists recommends Pap smears every three to four months for the first year after treatment of precancerous cervical lesions, followed by Pap smears every six months. As for colposcopy, the indications include any cytologic abnormality (including ASCUS and atypical glandular cells of undetermined significance [AGCUS]); after treatment of cervical dysplasia; and, perhaps, as an initial screening tool in HIV-positive women with less than 200 CD4+ cells/mm^3. Biopsies should be obtained at the time of colposcopy to confirm cytologic abnormalities. ASCUS: atypical squamous cells of undetermined significance; LSIL: low-grade squamous intraepithelial lesions; HSIL: high-grade squamous intraepithelial lesions.

Anal Dysplasia and Carcinoma

Prevalence of HPV, Anal Dysplasia, and Anal Cancer

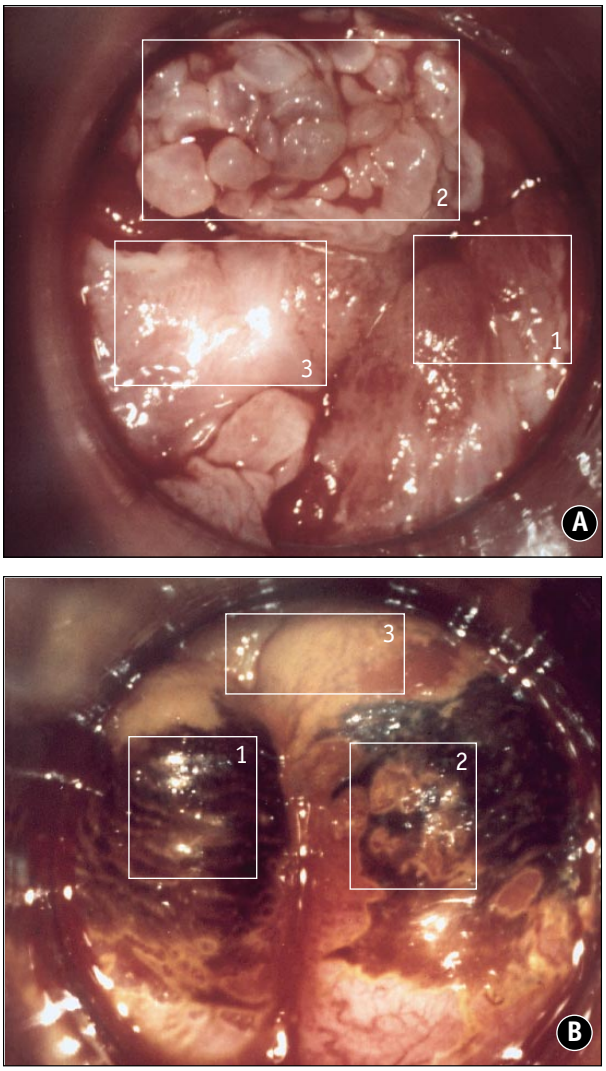
ANAL CANCER IS FAIRLY RARE. ITS INCIDENCE in the general population is less than one per 100,000 people and is one-tenth the current rate of cervical cancer in the United States. However, when evaluating the incidence of anal cancer among specific populations, more startling numbers come into play. In a 1987 paper published in the *New England Journal of Medicine,* the incidence of anal cancer among HIV-negative men who engage in receptive anal intercourse with other men was estimated to be 35/100,000—a rate on a par with the incidence of cervical cancer before routine Pap smears were initiated in the 1940s (Daling, 1987).

Even more troubling is the incidence rate among HIV-positive men who have sex with men. During the late 1980s, the incidence of anal cancer among gay men with AIDS was reported to be twice that of men of the same age, race, and sexual orientation in the years before AIDS (1975 to 1979) (Goedert, 1998). "In other words," commented Dr. Palefsky, "the incidence of anal cancer may be more than 70 of every 100,000 HIV-infected men who have a history of receptive anal intercourse with other men. This is really a shocking statistic."

No data are available regarding the incidence of anal cancer among HIV-positive or HIV-negative women, although it has been said that anal cancer is about twice as common in women as it is among men (Holmes, 1989; Holly, 1989).

Research into the natural history of anal cancer has been limited—no study has actually been conducted to prove that anal HSIL leads to anal cancer. However, it requires little more than common sense to conclude that HSIL represents the true pre-

Figure 3. Images of Anal SIL



Images taken during high-resolution anoscopy (HRA) after applying 3% acetic acid. Areas of acetowhiteness correlate with higher nuclear density; a mild acetowhite epithelium correlates with less severe disease, whereas intensely acetowhite epithelium correlates with more advanced forms of SIL.

A) healthy epithelial tissue appears pink and shiny [area 1]; a cluster of condylomas [area 2]; a thick acetowhite flat HSIL plague within the squamocolumnar junction [area 3]. While the condylomas would likely have been felt upon digital examination, the HSIL lesion would have been missed without HRA and application of acetic acid.

B) Epithelium coated with acetic acid and Lugol's iodine solution. Healthy epithelial tissue is dubbed Lugol's positive and appears mahogany/black in color [area 1]; Lugol's iodine solution is not fully absorbed by areas of low-grade dysplasia (Lugol's positive LSIL) and appears dark yellow/mustard in color [area 2]; high-grade dysplastic lesions of the epithelium absorb very little Lugol's iodine solution (Lugol's negative HSIL) and appear white or light yellow [area 3].

Source: Stephen E. Goldstone, MD

cursor lesion to anal cancer, based on what is already known about cervical HSIL and its association with cervical cancer. This assumption is based on a number of factors, including the histologic similarities between anal and cervical cancer and the observation that, like cervical cancer, anal cancer is often found with overlying HSIL.

As with cervical cancer, it is also not clear if HIV-positive men or women with anal SIL progress to anal cancer faster than their HIV-negative counterparts. "We know that HIV-positive women are more likely to have cervical dysplasia and that they progress faster to HSIL than HIV-negative women, but we don't know if they have an accelerated rate of progression to cervical cancer," Dr. Palefsky explained. "Similarly, we see a higher prevalence of anal dysplasia in HIV-positive women and gay and bisexual men, but it's not at all clear if ongoing immune suppression increases their risk of anal cancer. I'm inclined to say that there is a higher risk of anal cancer in these patients, but we really don't have the data to support this."

While HIV-positive men and women might not necessarily progress to anal cancer faster than their HIV-negative peers, they are certainly at greater risk for anal cancer, given the highly disproportionate rate of HSIL among HIV-positive patients. In one San Francisco cohort consisting of more than 600 MSM, LSIL and HSIL were present in 124/346 (36%) of HIV-positive men and 19/262 (7%) HIV-negative men (Palefsky, 1998). The relative risk of SIL among HIV-positive men was inversely correlated with CD4+ cell count, when compared with HIV-negative men. For those with a CD4+ count greater than 500 cells/mm^3, the relative risk was 3.8; for those with a CD4+ count between 200 and 500 cells/mm^3, the relative risk was 5.6; and for those with a CD4+ count less than 200 cells/mm^3, the relative risk was 7.3.

As for the natural history of SIL in gay and bisexual men, prospective studies conducted in Seattle and San Francisco have yielded interesting results. In the Seattle cohort, 158 HIV-positive and 147 HIV-negative MSM without initial evidence of anal SIL were monitored for an average of 21 months (Critchlow, 1995). In less than two years, HSIL developed in 24/158 (15%) of the HIV-positive men and 8/147 (5%) HIV-negative men.

In a study evaluating the natural history of HSIL in another of Dr. Palefsky's San Francsico cohorts, 277 HIV-positive MSM and 221 HIV-negative MSM—all of whom entered the study with either normal anal Pap test results, ASCUS, or LSIL—were followed prospectively for approximately four years (Palefsky, 1998a). During this period, 49% of the HIV-infected men developed HSIL, compared with 17% of the HIV-negative men. What's more, HIV-positive men with either low-grade dysplasia or ASCUS at baseline were more likely to develop HSIL during the four years of follow-up, compared with HIV-positive men with normal anal Paps upon entering the study (57% vs. 38%, respectively). As for HIV-negative men with ASCUS or LSIL at baseline, 33% developed HSIL during the four years of follow-up, compared with 14% of HIV-negative men who entered the study with normal anal cytologies.

As for anal HPV infection in both HIV-positive and HIV-negative men and women, several studies have provided a startling glimpse at the prevalence of HPV infection, indicating that current SIL and anal cancer rates may really only be the tip of the iceberg. Results from cross-sectional analyses have shown that anal HPV infection is found in nearly all HIV-positive men, as well as in a substantial proportion of HIV-negative men. In one of Dr. Palefsky's San Francisco cohorts, 93% of the HIV-positive men and 61% of the HIV-negative men had anal HPV infection detected by PCR in single samplings (Palefsky, 1998b). According to Dr. Palefsky, HPV type 16 was the most common viral type in both groups of men and, as discussed above, the ma-

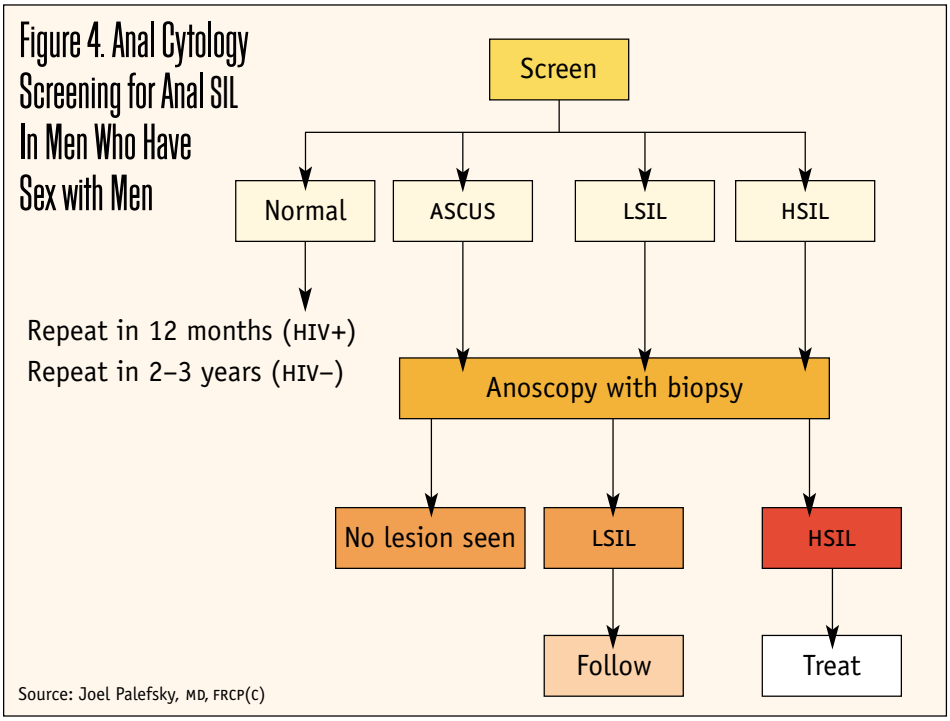
jority of HIV-infected men had evidence of multiple HPV types in their anuses (73% vs. 23% of HIV-negative men).

Data from cross-sectional analyses involving women also paint a grim picture. In one recent WIHS analysis published by Dr. Palefsky and his colleagues, anal HPV infection was more frequent than cervical infection in both HIV-positive and high-risk HIV-negative women (Palefsky, 2001). One hundred seventy of 223 (76%) HIV-positive women and 24/57 (42%) HIV-negative women had anal HPV infection detected by PCR. Among 200 women for whom there were concurrent anal and cervical HPV data, anal HPV was more common than cervical HPV in both HIV-positive (79% vs. 53%) and HIV-negative women (43% vs. 24%). Dr. Palefsky also reported that detection of HPV was inversely associated with CD4+ cell counts, but was not associated with plasma HIV-RNA levels.

Screening Issues

THERE IS NO DENYING THAT CERVICAL PAP smear and colposcope screenings have had a profound effect on the incidence of cervical cancer, among both HIV-positive and HIV-negative women. If we are to assume that anal dysplasia is similar to cervical dysplasia in its natural history and pathogenesis, compounded by the seemingly high prevalence and incidence of HSIL in certain populations, then isn't it possible that anal cytology screenings might play an invaluable role in detecting (and treating) high-grade dysplastic lesions, before they progress to anal cancer? According to Dr. Palefsky, the answer is most likely "yes," but not without potential caveats to consider.

Discussing results from one of his cohorts of MSM in San Francsico, Dr. Palefsky indicated that the sensitivity of anal Paps—"tush Paps" as he refers to them—to detect abnormal cytology was approximately 80% in HIV-positive men and 51% in HIV-negative men, roughly similar to the accuracy of cervical Pap smears in HIV-positive and HIV-negative women. However, anal Pap smears often yielded incorrect results regarding the grade of anal dysplasia present—a number of lesions that were said to be of a low-grade variety upon conducting a Pap smear were actually high grade upon conducting high-resolution anoscopy and biopsy. "Anal Pap smears are definitely appropriate as initial screening strategies," commented Dr. Palefsky.



While routine anal cytology screening has not been standardized by the United States Public Health Service (including the U.S. Centers for Disease Control), there is growing evidence to suggest that an anal screening program should be incorporated into the care of all at-risk individuals (e.g., men with a history of anal intercourse with other men [regardless of HIV serostatus], all HIV-positive men and women, and all women with high-grade cervical or vulvar lesions or cancer). At the time of the initial screening, if the cytology is normal, it is recommended that an anal Pap smear be repeated annually for HIV-positive men, and every two to three years for HIV-negative men. In the event of abnormal Pap findings—whether it be ASCUS, LSIL, or HSIL—high-resolution anoscopy with biopsy should be performed. ASCUS: atypical squamous cells of undetermined significance; LSIL: low-grade squamous intraepithelial lesions; HSIL: high-grade squamous intraepithelial lesions.

"But it's important that any abnormality be followed up with high-resolution anoscopy and that lesions be biopsied to confirm the grade of dysplasia."

To conduct an anal Pap smear, a Dacron swab is inserted approximately 1.5 to 2 inches (approximately 3 to 5 cm) into the anal canal. "It's important to use a Dacron swab, not a cotton swab, as cotton clings to the cells and won't give them up easily for cytology," recalled Dr. Palefsky. "And be sure to moisten the swab with water, not lubricant. You'll want the Pap smears to be comfortable for patients without botching the sample."

Once inserted deep enough into the anus—this is necessary, in order to collect both rectal columnar and anal squamous cells—the swab should be pulled out slowly, applying some pressure to the wall of the anus, rotating the swab in a spiral motion along the way. From there, the cells collected with the swab can be placed in the "ThinPrep" methanol fixative medium or spread onto a glass slide and dipped in fixative solution and, finally, shipped off to a cytology lab for analysis.

In the event of abnormal Pap findings—whether it be ASCUS, LSIL, or HSIL—high-resolution anoscopy should be performed. To visualize the anal wall, a disposable anoscope is used. As explained by Dr. Palefsky, it is inserted through the anal canal using a water-based lubricant. A swab, wrapped in gauze and soaked in 3% acetic acid, is then passed through the anoscope. The anoscope is then removed, leaving the acetic acid-soaked swab in the anus, pressed up against the anal epithelia, for approximately one minute. The swab is then removed and the anoscope reinserted. A colposcope is then inserted, through the anoscope, to conduct the visual inspection.

Whereas healthy epithelial tissue inside the anus is pink and shiny, the application of acetic acid will turn dysplastic lesions dull and white. These should be inspected and a biopsy should be taken. Another dye

that can be used is Lugol's solution. When taken up by healthy epithelial tissue, it will render the anal wall a deep mahogany color. Conversely, dysplastic lesions do not fully absorb this iodine-rich solution and will likely turn a mustard or light yellow color. These, too, should be examined and biopsied for staging purposes.

As for anal warts found during visual inspection of the anus and anal wall, Dr. Palefsky pointed out that, in some cases, condylomas may contain high-grade lesions and should be biopsied and/or removed. "Even though these warts rarely, if ever, progress to high-grade dysplasia or anal cancer, there may be high-grade disease mixed in with them and, as a result, require treatment," he said.

With that said, Dr. Palefsky discussed the elements of an anal screening program that should be incorporated into the care of all gay and bisexual men (see Figure 4). At the time of the initial screening, if the cytology is normal, it is recommended that an anal Pap smear be repeated annually for HIV-positive men, and every two to three years for HIV-negative men. If the patient is found to have LSIL, routine follow-up should occur every six to 12 months without necessitating therapy. As with cervical lesions, only those patients with HSIL should routinely receive therapy.

During the question-and-answer period that followed Dr. Palefsky's PRN lecture, Dr. Stephen Goldstone, a colleague of Dr. Palefsky's and a surgeon specializing in anorectal care in New York, suggested that clinicians, at a minimum, should be conducting visual and digital rectal examinations in their patients at least once a year. In a *Diseases of the Colon and Rectum* paper published in May 2001, Dr. Goldstone and his colleagues reported that a sizeable percentage (39%) of 39 gay and bisexual men with noncondylomatous benign anal disease—e.g., hemorrhoids, fissures, etc.—referred for surgical treatment had anal HSIL on biopsy (Goldstone, 2001). "What this tells us," Dr. Goldstone said, "is that men who have sex with men who have any signs of anal disease should be evaluated for anal SIL. It's also important for primary care providers to keep lines of communication open in order to determine which patients are at risk for anal SIL. Patients might not volunteer information about their sexual history or behaviors. It's up to providers to bring the topic up and to make their patients feel comfortable discussing sexual issues."

As for women, screening strategies for anal dysplasia have not yet been elucidated, but are definitely warranted—especially for women with HIV or a history of cervical HPV infection and HSIL. Dr. Palefsky and Dr. Goldstone have both hinted that anal cytology guidelines for HIV-positive and HIV-negative women would look very much like the screening recommendations for HIV-positive and HIV-negative MSM.

Treating Dysplasia and the Effects of HAART

Treatment Considerations

WHILE NOT A WHOLE LOT IS KNOWN ABOUT the most effective ways to treat anal dysplasia, a number of lessons can be learned from the experience of cervical dysplasia and its various treatment modalities. For starters, Dr. Palefsky does not routinely recommend that low-grade cervical or anal dysplasia be treated, given the likelihood of pain associated with treatment, the high recurrence rate of low-grade lesions, and the low risk of progression to cancer. "Still," he added, "many of our patients opt for therapy to relieve symptoms, especially when warts are involved. They can itch and they can burn and they can be rather unsightly. What's more, treatment may help reduce the spread of HPV from one person to another."

Therapies for condylomas that can be applied at home include: imiquimod (Aldara), a cream that is applied three times weekly; podophyllotoxin (Condylox), a gel applied in cycles of three days on/four days off; and 5-FU cream. Condyloma treatments that can be administered by a clinician are similar to those used for the treatment of HSIL. These include: liquid nitrogen, 80% trichloroacetic acid, surgical excision, laser ablation, thermocoagulation/infrared coagulation, LEEP, and intralesional interferon. "Again, these aren't routinely recommended for patients with condylomas or low-grade lesions," reiterated Dr. Palefsky. "However, when we're dealing with high-grade dysplasia, treatment becomes necessary."

As with cervical lesions, a number of patients with HSIL experience a recurrence or persistence of high-grade lesions. In turn, the need to retreat HSIL is commonplace in both HIV-negative and HIV-positive men who have sex with men.

The Effects of HAART

MUCH OF WHAT IS KNOWN ABOUT THE PREVALence and natural history of cervical and anal HPV infection, dysplasia, and the risk of carcinoma—in the setting of HIV—comes from studies conducted prior to the widespread use of HAART. In fact, many of the cohort studies described above involved patients who were not receiving antiretroviral therapy and, as a result, paint a slightly skewed picture of what to expect in patients who are now able to achieve durable suppression of HIV-RNA levels and substantial increases in CD4+ cells. In this way, there are two possible scenarios to consider: 1) that the virologic and immunologic benefits of HAART will ultimately heighten the immune response to HPV infection and lead to the regression of high-grade lesions and halt the development of new lesions, or 2) that HAART will have little impact on the natural history of HPV infection and possibly increase the risk of cervical or anal cancer, given that patients with HSIL will be living longer and, as a result, see their HPV disease progress to more advanced stages.

It is not yet clear which of these two scenarios is correct, given that follow-up data are still limited among HIV-positive men and women with HSIL who have initiated HAART. And the results that have been reported thus far are decidedly mixed.

In one study evaluating 49 women with advanced HIV disease screened before starting HAART and then five months after initiating therapy, no changes in the rate of HPV infection or in the level of HPV-DNA in cervical tissue was found (Heard, 1998). However, some improvement in clinical disease was demonstrated, with the prevalence of cervical SIL falling from 66% before HAART to 49% five months after therapy was started. Regression from high-grade to low-grade cervical SIL or normal cytology occurred in 23%, while regression from low-grade SIL to normal was documented in 43%. It is important to note, however, that some women who had normal cytologies or low-grade dysplasia prior to intiating HAART went on to develop high-grade lesions within five months after starting therapy.

As for the impact of HAART on the prevalence and natural history of anal dysplasia, Dr. Palefsky does not anticipate much of a change for the better (Palefsky, 2000). Among 28 HIV-positive MSM who had high-grade dysplasia at the time of initiating HAART, cytology and biopsy tests

conducted six months later found that 16 (57%) had no change in their dysplasia, six (21%) regressed to low-grade dysplasia, five (18%) to ASCUS, and one (4%) had normal cytology. However, 4/30 (13%) men with low-grade dysplasia at baseline went on to develop high-grade disease within six months after initiating HAART.

"Basically, we have a lot more to learn about the effects of HAART on cervical and anal dysplasia," summarized Dr. Palefsky. "With other viral diseases, such as HHV-8 and CMV, we've seen some really encouraging improvements with the introduction and continued use of HAART. We did see some encouraging results in our patients who initiated HAART with relatively high baseline CD4+ counts—they were much more likely to see a regression from high-grade disease to either low-grade dysplasia or ASCUS than patients with lower CD4+ cell counts. So maybe there is something going on with immune reconstitution. But we need to study this further to figure out what we can expect with the use of HAART."

Conclusion

IN CONCLUDING HIS LECTURE, DR. PALEFSKY was careful to illustrate some of the most pressing issues regarding the diagnosis and care of both HIV-positive and HIV-negative men and women with cervical and/or anal dysplasia. First, there is a dire need to develop and implement anal SIL screening programs in high-risk populations, given the success of similar programs focusing on cervical HPV disease. A handful of studies, including cost-benefit models, show that anal cytology screenings are both clinically useful and cost-effective to prevent anal cancer in both HIV-positive and HIV-negative MSM (Goldie, 1999; 2000). While there are insufficient data to make official recommendations for anal screening in women, early data suggest that those at highest risk include HIV-positive women and women with high-grade cervical or vulvar SIL or cancer. Second, it is important to continue research into the natural history and pathogenesis of anal and cervical HPV infection and dysplasia, particularly as they relate to each other. Third, there is an urgent need for the development of therapies that are HPV-specific and that do not require a completely intact immune response to be efficacious.

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