American Journal of Human Pathology

American Journal of Human Pathology
http://www.ivyunion.org/index.php/pathology
Vol.1, Article ID 201400351, 10 pages

Research Article

Establishing an Anal Neoplasia Screening Service for a Unique HIV Positive Patient Population in a Small City: Lessons Learned

Charles D. Sturgis,[1,2] Lacey L. Siekas,[3] Juan C. de la Ossa,[4] Stephanie Hamilton[1], Sheryl A. Dreyer[5]

1. CellNetix Pathology and Laboratories, PLLC, Everett, WA, United States
2. Providence Regional Medical Center, Everett, WA, United States
3. Virginia Mason Medical Center, Seattle, WA, United States
4. Polyclinic, Seattle, WA, United States
5. The Everett Clinic, Harbour Pointe, Mukilteo, WA, United States

Abstract

Mortality from human papillomavirus (HPV) mediated squamous carcinoma of the uterine cervix has dramatically decreased in recent decades as a result of broad scale population screening for early detection of cervical cancer precursors. Concomitantly, deaths associated with HPV related carcinomas of the anus have oppositely trended upward. At this time, there are no national guidelines for anal screening. We herein report our experience with establishing an anal cytology screening program for HIV infected patients in a small city. The HIV positive population studied is unique in that 75% of patients had undetectable viral loads by PCR with average CD4+ cell/uL counts of 550. In addition 40% are adult females. 45% of patients in this relatively healthy HIV+ population were discovered to have atypical squamous cells or worse on entry into screening, and 20% of patients were ultimately shown on high resolution anoscopic biopsy histology to harbor high grade squamous intraepithelial lesions (AIN 2/3). Meaningful small scale anal cytology screening programs are possible with clinical and anoscopic collaborations. It seems possible that this simple and inexpensive test may prevent morbidity and mortality from HPV mediated anal carcinoma.

Keywords: Anus; Screening; Dysplasia; Cytology; HIV

Reviewers: Houda Alatassi, MD, Department of Pathology and Laboratory Medicine, University of Louisville, United States; Francesca Sanguedolce, MD, PhD, Department of Pathology, University of Foggia, Italy

Academic Editor: Sihua, PhD, Shanghai Ocean University, China

Received: March 23, 2014 **Accepted**: May 14, 2014 **Published:** June 11, 2014

Competing Interests: The authors have declared that no competing interests exist.

*Correspondence to: Charles D. Sturgis, CellNetix Pathology and Laboratories, PLLC, United States

Email: csturgis@cellnetix.com

Introduction

Studies of screening for anal dysplasia and carcinoma in human immunodeficiency virus (HIV) infected patients are increasingly appearing in the pathology literature. As an historical comparison, exfoliative cytology of the female genital tract (cervix and vagina) has been employed as a routine cancer screening test in North America for decades. This testing is now standard of care and has resulted in a significant diminution in the number of invasive cervical squamous carcinomas through early detection and subsequent management of precursor lesions. Prior to implementation of broad scale screening, cervical cancer was the number one cause of cancer mortality for women in the United States. Today, invasive cervical cancers are less commonly encountered, and this disease now falls outside of the "top ten" most frequently diagnosed and most commonly fatal malignancies for women [1]. It is well known that nearly all cervical cancers are HPV mediated [2, 3]. As the incidence and frequency of death from invasive cervical cancer has decreased, the incidence and frequency of death from anal cancer (another HPV mediated condition) has increased for both men and women [4, 5]. Anal cancer precursors (like cervicovaginal dysplasias) are often asymptomatic, and because population based screening for anal neoplasia is not practiced, it is not uncommon for persons with HPV mediated anal disease to be diagnosed with mass lesions by digital anal exam or to present with symptoms such as bleeding. Certain specific populations may be predisposed to developing anal warts, dysplasias and carcinomas. Perhaps the most widely studied populations include HIV infected persons and men who have sex with men (whether HIV positive or not) [6-11]. Availability of anal cytology screening is currently variable, and as anal dysplasia clinics open, some are reporting their findings [12]. Most anal dysplasia screening programs are currently located in urban and highly populated geographies. Herein, we report our experiences with establishing an anal cytology screening program for HIV infected persons in Everett, Washington (a small Pacific Northwest community with a population of approximately 100,000 people). It is our hope that targeted screening of patient cohorts who are at highest risk of developing HPV mediated anal neoplasia may stem the tide of increasing disease incidence, ideally preventing both morbidity and mortality.

Materials and Methods

An electronic medical record retrieval for first time anal cytology screening tests (CellNetix Pathology and Laboratories, Everett, Washington, U.S.A.) was performed for the two year period 03/2011 through 03/2013. All patients in this cohort were HIV+. All anal cytology samples were collected by one physician (S.A.D., Internal Medicine, HIV Specialist, American Academy of HIV Medicine) working in an outpatient clinic (The Everett Clinic, Harbour Pointe, U.S.A.). All samples were collected using Dacron swabs. The employed collection technique was based upon the instructional video Anal Pap Smear: A Simple, Fast & Easy Procedure HRSA Grant #6 H4AHA006002, 2004, Johns Hopkins University School of Medicine, Infectious Disease Division. During the first seven months of the twenty-four month period, the samples were submitted in SurePath vials (Becton Dickinson Company). During the last seventeen months, the samples were submitted in ThinPrep vials (Hologic). Samples were processed per product guidelines. Cytomorphologic interpretation was based upon The Bethesda System for Reporting Cervical Cytology, Second Edition [13]. Cytologic interpretations were performed by five board certified anatomic pathologist, some but not all of whom were board certified in cytopathology ("non-specialty" sign out). HPV DNA testing was not performed. Results of the retrieval were entered into a spread sheet for data organization and were correlated with a review of the electronic clinical / laboratory medical records of each of the identified patients. Data points gathered for spread sheet entry for each patient included age at time of cytology sample collection,

gender, HIV viral load (measured within preceding six months), CD4+ cell count (measured within preceding six months), documentation of current use of highly active antiretroviral therapy (HAART), and cytologic interpretation. All patients interpreted to have squamous intraepithelial lesions on cytology were referred to high resolution anoscopy. Most patients with atypical squamous cells were also referred to anoscopy, including all of those with atypical squamous cells cannot exclude high grade squamous intraepithelial lesion. Follow up information (12/2013, with time frames ranging from a minimum of 9 months to a maximum of 33 months) was also recorded in spread sheet columns. Follow up data points included all subsequently available cytology or histology interpretations and corresponding chronologic intervals from the time of initial entry into the screening program. Institutional review board (IRB) approval for this retrospective project was sought and granted by Western IRB, Olympia, WA, letter dated 08/30/2013.

Results

The anatomic pathology laboratory information system data retrieval identified 138 patients who underwent first-time anal cytology testing in the initial two years of the screening program at The Everett Clinic. The majority of patients were male (60%), and all patients were adults. Patient ages ranged from 20 to 72 years with a mean age of 44 years. (See Table 1 for demographic data). The majority (88%) of patients was receiving combination antiretroviral therapy at the time of anal cytology screening, and the majority of patients (75%) had viral loads that were undetectable by polymerase chain reaction testing. The mean CD4+ cell count was 595 cells/uL.

Table 1 Demographics

Number of patients for first time anal cytology screening	**138**
Male	83 (60%)
Female	55 (40%)
Youngest age (years)	20
Oldest age (years)	72
Mean age (years)	44
Lowest absolute CD4+ cell count (cells/uL)	86
Highest absolute CD4+ cell count (cells/uL)	1253
Mean CD4+ cell count (cells/uL)	595
Lowest viral load (copies / ml)	0 (75%)
Highest viral load (copies / ml)	148,000
Mean viral load (copies / ml)	3,105
Currently on HAART	121 (88%)
Currently not on HAART	17 (12%)
Number of patients with lesional (ASC & above) anal cytology	**62 (45%)**
Number of lesional patients with no follow up	21 (34%)
Number of lesional patients with cytology follow up	21 (34%)
Number of lesional patients with biopsy follow up	20 (32%)

HAART = highly active antiretroviral therapy； ASC = atypical squamous cells

Of the 138 patients who underwent anal cytology screening, 62 (45%) were identified with abnormal squamous epithelial findings. (See Table 2 for cytology interpretations and subsequent biopsy follow up results). Thirty patients (22%) were interpreted to have atypical squamous cells of undetermined significance (ASC-US). Three patients (2%) were interpreted to have atypical squamous cells, cannot exclude high grade squamous intraepithelial lesion (ASC-H). Twenty-six patients (19%) were interpreted to harbor a low grade squamous intraepithelial lesion (LSIL). Three patients (2%) were interpreted to have high grade squamous intraepithelial lesions (HSIL). No invasive carcinomas were identified. Of the patients with lesional (initial cytology of ASC and above) interpretations, 41 (66%) had either additional cytology or histology follow up. Histologic follow up was made possible through routine processing of high resolution anoscopic biopsies (performed by L.L.S and J.C.d.l.O). The majority of patients without follow up were those in the ASC-US category. The percentages of patients with HSIL [anal intraepithelial neoplasia (AIN) 2/3] on biopsy follow up were 100% for those patients with HSIL cytology, 67% for those patients with ASC-H cytology, 42% for those patients with LSIL cytology, and 3% for those with ASC-US cytology.

Table 2 Initial Cytology and Biopsy Follow Up

Initial Cytology Interpretation	# (%)	HSIL (AIN2/3) on Biopsy Follow Up, # (%)
Unsatisfactory	1 (1%)	
NILM	75 (54%)	
ASC-US	30 (22%)	1 (3% of patients with ASC-US)
ASC-H	3 (2%)	2 (67% of patients with ASC-H)
LSIL	26 (19%)	11 (42% of patients with LSIL)
HSIL	3 (2%)	3 (100% of patients with HSIL)
Totals:	138 (100%)	27 (20% of all patients in study)

NILM = negative for intraepithelial lesion / malignancy, ASC-US = atypical squamous cells of undetermined significance, ASC-H = atypical squamous cells cannot exclude high grade dysplasia, LSIL = low grade squamous intraepithelial lesion, HSIL = high grade squamous intraepithelial lesion, AIN = anal intraepithelial neoplasia.

Discussion

Contrary to the noted decreasing trends of preinvasive and invasive squamous tumors of the cervix and vagina, age-adjusted incidence rates for preinvasive and invasive squamous anal malignancies have significantly increased over the last several decades [4]. The incidence of anal cancer is known to be elevated in HIV infected men who have sex with men compared to the general population, and anal HSIL has clear potential to progress to invasive disease in this group [6, 11]. The goal of clinical management of patients with HPV mediated lesions of the lower anogenital tract should be to identify and treat patients with high grade precursor lesions (HSIL) to decrease the risk of developing invasive cancers [14]. Some investigators have suggested that risk factor assessment and stratification in HIV infected patients may help to disentangle the influences of anal exposure to HPV, immunodeficiency, tobacco smoking, and combined antiretroviral therapy [15, 16]. Such stratification might allow for development of algorithms as to which subpopulations are in greatest need of anal cytology screening. No clear correlations between degree of dysplasia and CD4+ cell counts or measurable viral loads could be made in our study; however, our population is relatively well and our cohort number is low. The small sample size is a limiting factor in our study. Some authors suggest annual screening for all patients with HIV, and this is our preferred approach [7].

One unique characteristic of the population in our current retrospective review is the percentage of female patients presenting for first time screening and follow up (40%). The female patients in this cohort acquired their HIV infections through heterosexual practices and/or through intravenous drug use. It is uncertain what percentage of these women engaged in heterosexual anal intercourse. More than 20% of U.S. women ages 20-39 reported having anal sex in the past year in a recent nationally representative probability sample study [17]. While women represented 40% of the patients initially screened, they represented only 13% of the patients who were ultimately found to harbor high grade squamous intraepithelial lesions (AIN 2/3) on high resolution anoscopy with biopsy over the average of 21 months of follow up time. (See Table 3 for HSIL follows up by gender). The comparatively large percentage of female patients with HIV in this study may be one explanation for the somewhat lower fractions of patients with abnormal cytology in comparison to other reports.

Table 3 Follow Up HSIL by Gender

	Undergoing Primary Anal Cytology Screening	**Follow Up (9 to 33 Months) of HSIL (AIN 2/3) by HRA Biopsy**
Men	83/138 (60%)	14/16 (87%)
Women	55/138 (40%)	2/16 (13%)

HSIL = high grade squamous intraepithelial lesion, AIN = anal intraepithelial neoplasia, HRA = high resolution anoscopy.

Another finding from this retrospective review is that cytology appears to underestimate the grade of dysplasia when compared to results of corresponding anoscopically guided biopsy histology. The sensitivity and specificity of a single anal cytology specimen have been reported to be comparable with those for a single cervical cytology test; however, lesional severity appears more likely to be underestimated on anal collections. High resolution anoscopy with biopsy and histologic interpretation is most often viewed as the gold standard [6, 18-19]. In the current review, 42% of patients (mostly men) interpreted to have LSIL on screening cytology were subsequently found to have HSIL on biopsy follow up. This figure is more than four times higher than the 10% we see in our geographically identical general cervical screening program (for all women of all ages and all immune statuses).

In our local pathology professional group cervicovaginal and anal cytology cases are interpreted by all of the anatomic pathologists (non-specialty sign out). In revisiting the cytology and histology slides from all of the patients initially classified as LSIL on cytology, at least three of these patient's could have been originally cytologically interpreted as HSIL. This would have increased the number of HSIL patients from 3 (2%) to 6 (4%). (See Figures 1, 2 and 3 for a case example of a patient interpreted with LSIL whose follow up revealed HSIL and retrospective cytology review confirmed rare overlooked high grade cells in the original screening cytology). While it cannot be said with absolute certainty, it may be that assignment of anal cytology cases to a focused group of pathologists with cytopathology expertise might result in greater accuracy and reproducibility. The literature on this topic suggests moderate to good agreement between cytopathohlogists evaluating anal cytology specimens from HIV positive men who have sex with men [20]. A recent College of American Pathologists Interlaboratory Comparison

Program in Nongynecologic Cytology showed poor performance on anal cytology, especially in regard to the correct identification of HSIL, and indicated a need for continued education about anal cytology [21].

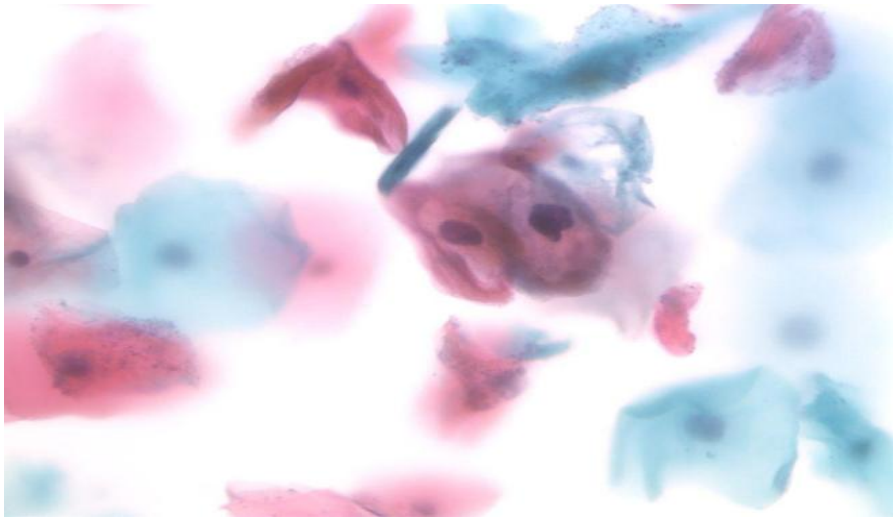

Figure 1 Koilocytes identified in initial anal screening cytology from a 57 year-old, HIV+, male patient. Anal cytology interpreted as LSIL (low grade squamous intraepithelial lesion). Papanicolaou stain, 630X.

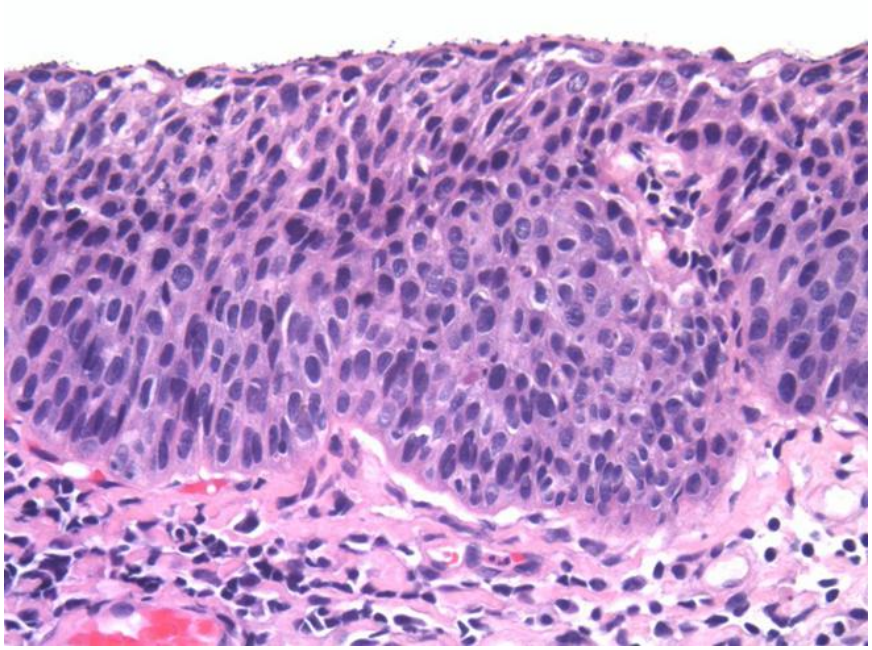

Figure 2 Moderate to severe squamous dysplasia (anal intraepithelial lesion [AIN 2/3], high grade squamous intraepithelial lesion [HSIL]) in a histologically processed high resolution anoscopic biopsy from a 57 year-old, HIV+, male (same patient as Figure 1) whose screening cytology was originally interpreted as low grade squamous intraepithelial lesion (LSIL). Hematoxylin and eosin, 400X.

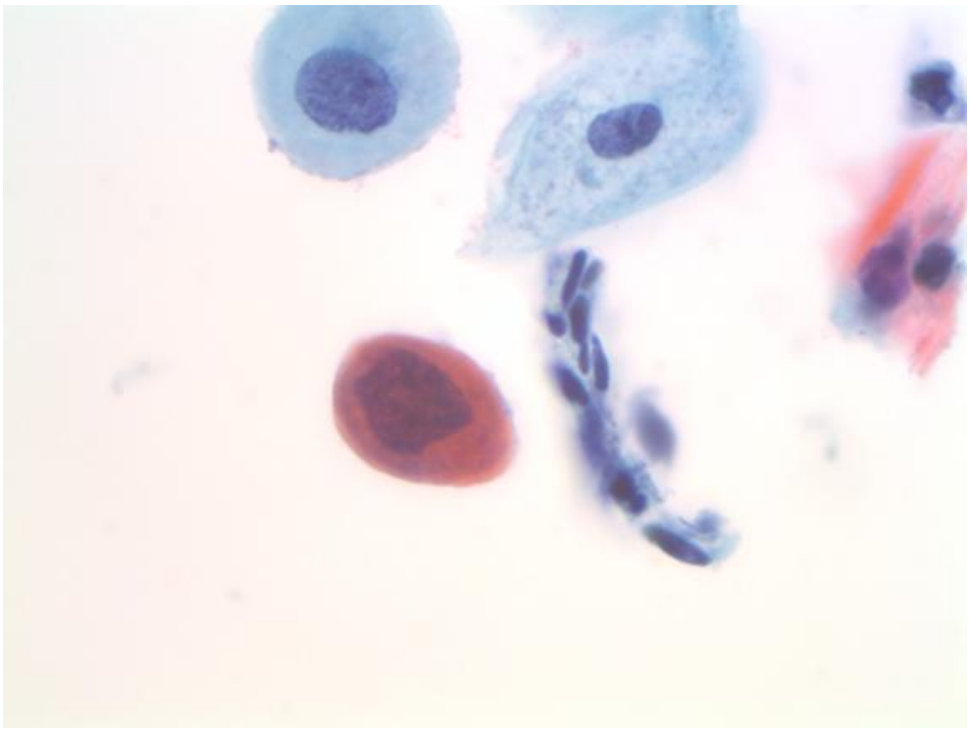

Figure 3 Rare cells of high grade squamous intraepithelial lesion (HSIL) in a 57 year-old, HIV+, male patient (same patient as in Figures 1 and 2) were identified on retrospective review for cyto-histologic correlation. Papanicolaou stain, 630X.

Currently, there are no national guidelines or organizationally approved recommendations for anal cytology screening of the general public. This lack of published guidelines for the general public results in many "unanswered" questions about which patients should be screened and at what screening intervals testing should be performed. Recent primary care guidelines from the Infectious Diseases Society of America (IDSA) for the specific management of persons infected with HIV provide a "weak recommendation based upon moderately quality evidence" that men who have sex with men, women with a history of receptive anal intercourse or abnormal cervical Pap tests, and all HIV infected individuals with genital warts should have an anal Pap test [22]. With these new HIV related guidelines, there seems likely to be little argument that persons living with HIV and men who have sex with men may benefit from anal screening, and some authors suggest that patients in these categories who are screened and are interpreted to have atypical squamous cells (ASC) or above should be referred to high resolution endoscopy [18]. In our practice in Everett, Washington, following this paradigm would result in a 45% anoscopy referral rate. It is important to remember that screening for screening's sake alone is of no value, and patients who are discovered to have ASC or worse on cytology need to be examined by a well trained anoscopist. In establishing our program in Everett, we were limited in that no such person exists in our community. We do have the good fortune of being located within 40 miles of two high quality clinician anoscopists, and patients with abnormal findings are triaged to these providers in the Seattle metropolitan area for anoscopy with biopsy and appropriate follow up. In communities without high resolution anoscopy services, it is of paramount importance to establish clinical connections with a referral anoscopist prior to starting a screening service.

Because the prevalence of anal HPV infection in HIV infected men is high (more than half of patients

in many studies), we chose not to reflex atypical cytology results to HPV DNA testing in designing our triage approach [23-27]. There is literature to suggest that commercially available DNA tests are valid for use in liquid based anal cytology samples [28]. It is possible that HPV DNA testing might have utility in centers where anal cytology screening is performed on patient populations other than men who have sex with men and those with HIV. Other populations in which anal cytology screening might be considered are patients who are solid organ transplant recipients and well (non-immunocompromised women) who are known to have high grade squamous intraepithelial neoplasia (HSIL) of the cervix [29,30]. In those populations, reflex HPV DNA testing may be relevant. It seems possible that if patients were HPV DNA tested and found to be negative for high risk infections, they might be candidates for vaccination. There is some literature to suggest that combined high resolution anoscopy and anal cytology may be cost effective surveillance strategies after treatment for HSIL in HIV infected men [31]. Broad scale outcomes studies that systematically assess the efficacy of anal cancer screening programs in reducing the incidence and morbidity and mortality of invasive anal cancers are needed [32].

References

1. Siegel R, Naishadham D, Jemal A. Cancer statistics, 2012. ***CA Cancer J Clin.*** 2012, 62:10-29
2. Stoler MH. Human papillomavirus and cervical neoplasia: a model for carcinogenesis. ***Int J Gynecol Pathol.*** 2000, 19:16-28
3. Thomison J, Thomas LK, Shroyer KR. Human papillomavirus: molecular and cytologic/histologic aspects related to cervical intraepithelial neoplasia and carcinoma. ***Hum Pathol.*** 2008, 39:154-166
4. Kurdgelashvili G, Dores GM, Srour SA, Chaturvedi AK, Huycke MM, Devesa SS. Incidence of potentially human papillomavirus-related neoplasms in the United States, 1978 to 2007. ***Cancer.*** 2013, 119:2291-2299
5. Jemal A, Simard EP, Dorell C, Noone AM, Markowitz LE, Kohler B, Eheman C, Saraiya M, Bandi P, Saslow D, Cronin KA, Watson M, Schiffman M, Henley SJ, Schymura MJ, Anderson RN, Yankey D, Edwards BK. Annual report to the nation on the status of cancer, 1975-2009, features the burden and trends in human papillomavirus (HPV) - associated cancers and HPV vaccination coverage levels. ***J Natl Cancer Inst.*** 2013, 105:175-201
6. Betancourt EM, Wahbah MM, Been LC, Chiao EY, Citron DR, Laucirica R. Anal cytology as a predictor of anal intraepithelial neoplasia in HIV-positive men and women. ***Diagn Cytopathol.*** 2013, 41:697-702
7. Mallari AO, Schwartz TM, Luque AE, Polashenski PS, Rauh SM, Corales RB. Anal cancer screening in HIV infected patients: is it time to screen them all? ***Dis Colon Rectum.*** 2012, 55:1244-1250
8. Reed SF, Britt RC, Novosel TJ, Collins JN, Weireter LJ, Britt LD. Screening human immunodeficiency virus positive men for anal intraepithelial neoplasia. ***Am Surg.*** 2012, 78:901-903
9. Amirian ES, Fickey PA, Sheurer ME, Chiao EY. Anal cancer incidence and survival: comparing the greater San Francisco bay area to other SEER cancer registries. ***PLOS One.*** 2013, 8:e58919
10. Phanuphak N, Teeratakulpisarn N, Triratanachak S, Keelawat S, Pankam T, Kerr SJ, Deesua A, Tantbirojn P, Numto S, Phanuphak P, Palefsky JM, Ananworanich J. High prevalence and incidence of high grade anal intraepithelial neoplasia among young Thai men who have sex with men with and without HIV. ***AIDS.*** 2013, 27:1753-1762
11. Berry JM, Jay N, Cranston RD, Darragh TM, Holly EA, Welton ML, Palefsky JM. Progression of anal high grade squamous intraepithelial lesions to invasive anal cancer among HIV-infected men who have sex with men. ***Int J Cancer.*** 2014, 134:1147-1155
12. Siekas LL, Aboulafia DM. Establishing an anal dysplasia clinic for HIV infected men: initial experience. ***AIDS Read.*** 2009, 19:178-186
13. Darragh TM, Birdsong GG, Luff RD, Davey DD. Anal Rectal Cytology. In: Solomon D, Nayar R, eds. The Bethesda System for Reporting Cervical Cytology Definitions, Criteria, and Explanatory Notes, Second

Edition. New York, U.S.A.: ***Springer-Verlag.*** 2004, 169-175

14. Darragh TM, Colgan TJ, Cox JT, Heller DS, Henry MR, Luff RD, McCalmont T, Nayar R, Palefsky JM, Stoler MH, Wilkinson EJ, Zaino RJ, Wilbur DC. The lower anogenital squamous terminology standardization project for HPV associated lesions: background and consensus recommendations from the College of American Pathologists and the American Society for Colposcopy and Cervical Pathology. ***Arch Pathol Lab Med.*** 2012, 136:1266-1297
15. Bertisch B, Franceschi S, Lise M, Vernazza P, Keiser O, Schoni-Affolter F, Bouchardy C, Dehler S, Levi F, Jundt G, Ess S, Pawlita M, Kovari H, Wandeler G, Calmy A, Cavassini M, Stockle M, Clifford G. Risk factors for anal cancer in persons infected with HIV:a nested case control study in the Swiss HIV cohort study. ***Am J Epidemiol.*** 2013, 178:877-884
16. Chiao EY, Hartman CM, El Serag HB, Giordano TP. The impact of HIV viral control on the incidence of HIV associated anal cancer. ***J Acquir Immune Defic Syndr.*** 2013, 63:631-638
17. Herbenick D, Reece M, Schick V, Sanders SA, Dodge B, Fortenberry JD. Sexual behavior in the United States:results from a national probability sample of men and women ages 14-94. ***J Sex Med.*** 2010, 7:255-265
18. Bean SM, Chhieng DC. Anal rectal cytology:a review. ***Diagn Cytopathol.*** 2010, 38:538-546
19. Richel O, Hallensleben ND, Kreuter A, van Noesel CJ, Prins JM, de Vries HJ. High resolution anoscopy:clinical features of anal intraepithelial neoplasia in HIV positive men. ***Dis Colon Rectum.*** 2013, 56:1237-1242
20. Darragh TM, Tokugawa D, Castle PE, Follansbee S, Borgonovo S, LaMere BJ, Schwartz L, Gage JC, Fetterman B, Lorey T, Wentzensen N. Interrater agreement of anal cytology. ***Cancer Cytopathol.*** 2013, 121:72-78
21. Darragh TM, Winkler B, Souers RJ, Laucirica R, Zhao C, Moriarty AT. Room for improvement: initial experience with anal cytology oberservations from the College of American Pathologists interlaboratory comparison program in nongynecologic cytology. ***Arch Pathol Lab Med.*** 2013, 137:1550-1554
22. Aberg JA, Gallant JE, Ghanem KG, Emmanuel P, Zingman BS, Horberg MA, Primary care guidelines for the management of persons infected with HIV:2013 update by the HIV Medicine Association of the Infectious Diseases Society of America. ***Clin Infectious Diseases.*** 2014, 58:1-34
23. Colon-Lopez V, Ortiz AP, Del Toro-Mejias L, Clatts MC, Palefsky JM. Prevalence, genotype diversity and determinant of anal HPV infection in high risk men attending a sexually transmitted infection clinic in Puerto Rico. ***Sex Health.*** 2013, 10:572-573
24. Darwich L, Canadas MP, Videla S, Coll J, Molina-Lopez RA, Sirera G, Clotet B. Prevalence, clearance, and incidence of human papillomavirus type specific infection at the anal and penile site of HIV infected men. ***Sex Transm Dis.*** 2013, 40:611-618
25. Hu Y, Qian HZ, Sun J, Gao L, Yin L, Li X, Xiao D, Li D, Sun X, Ruan Y, Milam DF, Pan SW, Shao Y, Vermund SH. Anal human papillomavirus infection among HIV-infected and uninfected men who have sex with men in Beijing, China. ***J Acquir Immune Defic Syndr.*** 2013, 64:103-114
26. Beachler DC, D'Souza G, Sugar EA, Xiao W, Gillison ML. Natural history of anal vs oral HPV infection in HIV infected men and women. ***J Infect Dis.*** 2013, 208:330-339
27. Machalek DA, Grulich AE, Jin F, Templeton DJ, Poynten IM. The epidemiology and natural history of anal human papillomarivurs infection in men who have sex with men. ***Sex Health.*** 2012, 9:527-537
28. Lowe B, Goldstone SE, Rus S, Salim H, Chen G, Rothmann T, Nazarenko I. Detection of human papillomavirus in anal specimens using the hybrid capture 2 assay. ***Diagn Mol Pathol.*** 2012, 21:150-156
29. Madeleine MM, Finch JL, Lynch CF, Goodman MT, Engels EA. HPV related cancers after solid organ transplantation in the United States. ***Am J Transplant.*** 2013, 13:3202-3209
30. Sehnal B, Dusek L, Cibula D, Zima T, Halaska M, Driak D, Slama J. The relationship between the cervical and anal HPV infection in women with

cervical intrapeithleial neoplasia. ***J Clin Virol.*** 2013, 1386-6532

31. Assoumou SA, Mayer KH, Panther L, Linas BP, Kim J. Cost effectiveness of surveillance strategies after treatment for high grade anal dysplasia in high risk patients. ***Sex Transm Dis.*** 2013, 40:298-303
32. Cachay ER, Mathews WC. Human papillomavirus, anal cancer, and screening considerations among HIV infected individuals. ***AIDS Rev.*** 2013, 15:122-133