

Detection of anal dysplasia is enhanced by narrow band imaging and acetic acid

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Abstract

Aim Anal intraepithelial neoplasia precedes the development of anal squamous cell carcinoma. Detection of the lesion is essential to management. This paper describes a prospective study to detect and ablate anal squamous intraepithelial lesions (SILs) using white light narrow band imaging (NBI) and NBI with acetic acid (NBIA).

Method Sixty patients with abnormal anal cytology and risk factors for anal dysplasia underwent examination of the anoderm with a high definition gastroscope and NBIA. Targeted biopsies were taken and the lesions were ablated and characterized histopathologically. Visualization of the anal transitional zone was facilitated by retroflexion and examination through a disposable anoscope.

Results Targeted biopsies were taken from lesions in 58 patients. No lesion was seen in two patients. Histopathology showed SIL in 48 (80.0%) of 60 biopsies. One biopsy showed lymphoid aggregates. Biopsies in nine (15%) of the 60 patients showed normal mucosa. Lesions were seen in white light in 27 (45%) of

the 60 cases, NBI in 39 (65%) and NBIA in 57 (95%). There was no major morbidity. Sensitivity analysis showed that all methods were significantly different from each other.

Conclusion Anal SIL in the anal transitional zone and anal canal can be identified by NBIA. Patient selection influences findings. Limitations include small sample size and non-randomization.

Keywords Anal dysplasia, anal transitional zone, human immunodeficiency virus, human papillomavirus, narrow band imaging, squamous intraepithelial lesion

What does this paper add to the literature?

Individual facets of narrow band imaging with acetic acid are not new. What is new is the application of this technique to identify and destroy anal intraepithelial lesions. The procedure can be done by any practitioner familiar with endoscopy in a single setting with high yield and good patient tolerance.

Introduction

Anal cancer and precursor lesions develop within the anal transitional zone (ATZ) defined by Fenger as the 'zone interposed between uninterrupted colorectal type mucosa above and uninterrupted squamous epithelium below' [1]. In 1989, Scholefield *et al.* [2] prospectively studied the use of an 'endoscope' (colposcope) and acetic acid to examine the anal canal for the detection of premalignant lesions. In 1997, Jay *et al.* modified Scholefield *et al.*'s method by adding anoscopy [3]. In 2009, Chou *et al.* [4] described a novel diagnostic method for early-stage squamous cell carcinoma of the

anal canal by colonoscopy with retroflexion and narrow band imaging (NBI) plus autofluorescence imaging. In 2010, Oono *et al.* [5] described NBI imaging colonoscopy with transparent hood for diagnosis of squamous cell carcinoma *in situ* in the anal canal. In 2012, Morisaki *et al.* [6] reported the use of magnifying endoscopy with NBI for diagnosing a patient with squamous cell carcinoma of the anal canal. In the same year, Horimatsu *et al.* [7] described the diagnosis of early-stage squamous cell carcinoma of the anal canal by colonoscopy with NBI. Also in 2012, Tanaka *et al.* [8] demonstrated that flexible endoscopy with retroflexion and rectal air insufflation provides a clear view of the anorectal line and the dentate line, the boundaries of the ATZ. In this report, we apply flexible endoscopy using white light (WL), NBI and NBI/acetic acid

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(NBIA) to detect and ablate anal dysplasia. We used this method because examination of the anoderm is a part of lower endoscopy and this study is an extension of our normal daily practice as endoscopists.

Method

In 2011, we obtained approval from the Cleveland Clinic Institutional Review Board to identify and evaluate all patients within our health system who had received the diagnosis of anal intraepithelial neoplasia.

This was a non-randomized prospective study. History and physical examination, anal brushing, anal human papillomavirus (HPV) serotyping (digene HC2 High-risk HPV DNA test, Qiagen™, Valencia, California, USA) and human immunodeficiency virus (HIV) testing were done. Those who were tested further had an abnormal anal brushing. The anal canal and perianal skin were examined by inspection, palpation, digital rectal examination and anoscopy. Informed consent for endoscopic examination of the anorectum was obtained. The procedure was performed in our hospital endoscopy units using a high definition gastroscope (Olympus GIF 180, Olympus, Center Valley, Pennsylvania, USA). A bowel preparation was given as for colonoscopy. The patient was positioned in the left lateral decubitus position. Intravenous moderate sedation with fentanyl or meperidine plus midazolam was administered. Blood pressure, pulse oximetry and electrocardiogram/heart rate were monitored before, during and after the procedure. Topical intra-anal 2% lidocaine gel was applied for local anaesthesia. The gastroscope was introduced through the anus into the rectum. The rectum was inspected up to the rectosigmoid junction. Biopsies were taken of the rectum if appropriate. The ATZ and anal canal were inspected both with WL and NBI using *en face* and retroflexed views. The anoderm was treated with 3% acetic acid by dripping the solution onto the surface using an endoscopic injection needle inserted through the biopsy channel. Changes in the appearance of the anoderm occurred after about 30 s. Evaluation of the anal canal was facilitated by passing the endoscope through a self-lighted beveled Anospec™ (Lawrence, Massachusetts, USA) anoscope. Lesions that were flat or elevated, showed acetowhite changes and/or abnormal vascular features, including punctuation or mosaicism [9], were biopsied and ablated. Hot biopsy forceps were preferred for small lesions. A Gold Probe™ Electrohemostasis catheter (Boston Scientific, Marlborough, Massachusetts, USA) set at 20 W was used to ablate larger and irregular lesions. Photodocumentation was preserved as part of the permanent electronic medical record (Provation®, Provation Medical, Minneapolis,

Minnesota, USA). Patients were recovered and discharged home. When surgery was required, endoscopy was done in the operating room. Histopathology was reported according to the lower anogenital squamous terminology standardization (LAST) project for HPV-associated lesions [10]. Cytology was reported according to the Bethesda system [11].

Results

Patient demographics are shown in Table 1. The majority of patients were male and were positive for HPV and HIV. The remainder were at risk for anal dysplasia because of a prior history of cervical dysplasia or solid organ transplant.

Figure 1(a, b) illustrate the anatomical relationship of the ATZ and its borders, the anorectal line and the dentate line as seen with WL and NBI during retroflexion in a normal patient. Circumferential assessment of the ATZ was achieved by rotating the endoscope. Spatial position was determined by reference to water located in the dependent portion of the rectum.

Figure 2 depicts the view of a high grade squamous intraepithelial lesion (HSIL) seen with retroflexion. This is a raised lesion demonstrating abnormal vasculature and mosaicism and was seen in WL, NBI and NBIA.

Figure 3 demonstrates a different lesion that is flat and only identified with the use of NIBA. It was not seen with WL or NBI. Pathology showed HSIL.

Pathology results

Abnormal cytology diagnoses were categorized as either low grade squamous intraepithelial lesion (LSIL), HSIL or atypical cells of uncertain significance. Biopsy speci-

Table 1 Demographics of 60 patients evaluated by NBIA.

	N
Sex	
Male	54
Female	6
Age, years	
Median	45
Range	21–74
MSM	43
HIV positive (all male)	39
HPV positive	46
History of anal dysplasia	23
History of genital dysplasia	5
Solid organ transplant recipients	2

MSM, men who have sex with men.

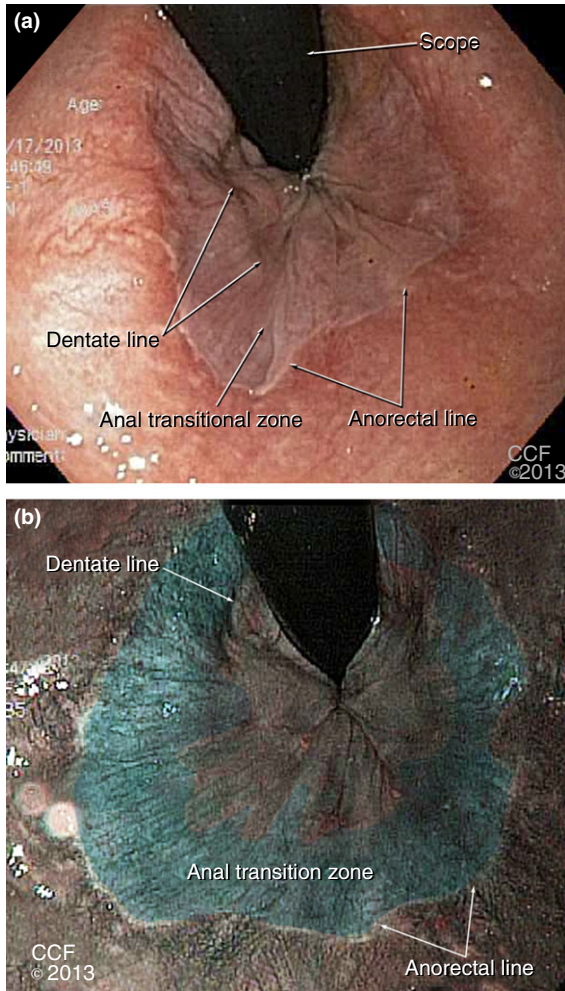


Figure 1 (a) Cross-sectional depiction of the anal canal (after Tanaka *et al.* [8]). The anal transitional zone is located between the dentate line and anorectal line. (b) Retroflexion with white light shows the dentate line, the anal transitional zone and the anorectal line.

mens were classified as negative for dysplasia, LSIL or HSIL. Results for 60 patients with abnormal anal cytology are shown in Table 2. SIL was found in 41/60 (68.3%) patients (28 LSIL, 13 HSIL). Non-SIL lesions associated with HPV infection were seen in 7/60 (11.7%). Overall, SIL and non-SIL HPV-associated lesions were found in 48/60 (80.0%) patients examined. A biopsy from one patient showed lymphoid aggregates. Normal mucosa was seen in the remaining 9/60 (15%) biopsied patients. The distribution of anal pathology compared with major risk factors for SIL is shown in Table 3. HPV-related pathological findings were highest for the HPV-positive group of patients.

Specificity could not be evaluated as all patients had an abnormal anal Pap smear and no comparison was made with another procedure. Sensitivity analysis is

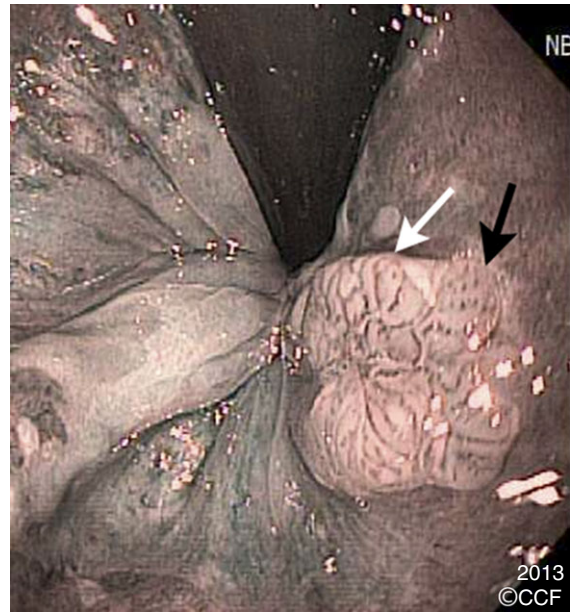


Figure 2 ATZ lesion in the left anterior position illuminated with NBI. Note the enhanced punctuation (black arrow) and mosaicism (white arrow). Figure shows a left anterior raised lesion with punctuation and mosaicism arising from the ATZ; pathology showed HSIL.

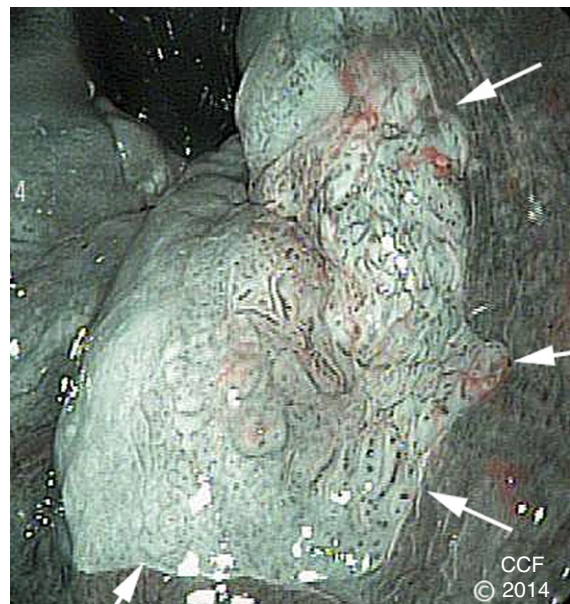


Figure 3 Examination with NBIA revealed a complex collection of slightly raised lesions with enhanced vascularity. Multiple lesions (see arrows) not visible with white light are seen on retroflexion in the ATZ. All lesions were ablated with hot biopsy forceps. Pathology showed HSIL.

shown in Table 3. Lesion visibility was compared under examination in WL, NBI and NBIA. McNemar's test was used to examine the paired sensitivities of the three

Table 2 Anal pathology in 60 patients compared with abnormal cytology.

Cytology	Histopathology					
	Dysplasia		Negative for dysplasia			
	HSIL	LSIL	Non-SIL HPV-related*	Lymphoid aggregates	Normal mucosa [†]	No biopsy [‡]
ASCUS	4	14	5	1	6	1
LSIL	7	12	2		3	1
HSIL	2	2				

ASCUS, atypical cells of uncertain significance.

*Hyperkeratosis/parakeratosis, squamous papilloma.

[†]Random biopsies were taken in one case because a lesion was not seen; pathology showed normal mucosa.

[‡]Biopsy was not done because the mucosa looked normal.

Table 3 Lesion visibility by high definition endoscopy with WL, NBI and NBIA in 60 patients with abnormal anal cytology.

Lesion visibility	N	Sensitivity
White light		
Lesion seen	27	0.45
Lesion not seen	33	
NBI		
Lesion seen	39	0.65
Lesion not seen	21	
Lesion visibility better than white light alone	29	
NBI plus acetic acid		
Lesion seen	57	0.95
Lesion visibility not better than NBI alone	12	
Lesion visibility better than NBI alone	45	

All sensitivities are significantly different from each other.

test methods. All methods are significantly different from all others: WL *vs* NBI, $P = 0.0015$; WL *vs* NBIA, $P < 0.0001$; and NBI *vs* NBIA, $P < 0.0001$. Bonferroni corrections were applied for multiple comparisons.

Discussion

Although anal dysplasia is thought to progress to anal cancer, investigation in the area is limited. The technique presented in this paper allows detection and ablative treatment of SILs with techniques familiar to all endoscopists with minimal additional training. A full bowel preparation was used to guarantee a clean field. We inspected the rectum and biopsied any abnormalities. Assessment of the anal margin was done at the initial office consultation. NBIA was done to assess abnormal anal Pap smears. We used sedation to ensure tolerance of both hot biopsy and cauterly ablation in the one proce-

dure. This obviates the need for multiple procedures to accomplish both biopsy and lesion removal. Hot biopsy of lesions for pathology leads to less bleeding and with small lesions can obliterate the area at the same time. The gold probe is used to ablate larger lesions.

Retroflexion with air insufflation provides a circumferential view of the effaced ATZ

NBIA enhances SIL/HPV-related lesions that can then be biopsied and ablated endoscopically. The ATZ can be visualized with NBI because, although the major type of epithelium is stratified columnar, mucin-producing cells can be present on the surface and the cells can be cuboidal or more flattened. These differences are highlighted to a varying degree in each patient [8]. Because of the smaller diameter and increased flexibility, it is easier to rotate a gastroscope in this area to obtain a 360° view. The left lateral view can be noted by looking for the site of the water when first entering the rectum as it will be in the dependent position. We take digital photographs of each aspect of the anal canal, with each type of light. Documentation is stored automatically in the Provation™ program which is then automatically loaded into the patient's electronic medical record. Photographs are available to anyone who wishes to look at them. An anoscope facilitates visualization of lesions distal to the ATZ such as those that extend into the anal canal. The anoscope dilates the internal anal sphincter and keeps it open. At this time we have not had any adverse events that would lead to either an emergency visit or hospitalization.

NBIA is able to detect lesions not visible in either WL or NBI alone; NBIA can also enhance the appearance of lesions that are visible in WL or NBI. Targeted biopsies of lesions had a yield of SILs that was 80.0% (Table 1). Random biopsies were not done except in

one case. Correlation between lesion size and pathology findings was not done as this was beyond the scope of this report. The study is limited by a small patient number.

Practices differ in different countries. We have no comparative data of our own that would allow us to make any statements about the relative benefit of this procedure to others in the literature. Long-term follow-up will be important to assess the ultimate impact of this technique in the detection of dysplastic lesions, the progression of LSIL and HSIL after ablation and prevention of anal cancer.

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Author contributions

Conception and design: Michelle Inkster. Acquisition of data: Michelle Inkster and James Wu. Analysis and interpretation of data: Michelle Inkster, Homer Wiland, James Wu. Drafting of the article: Michelle Inkster, Homer Wiland, James Wu. Final approval of the version to be published: Michelle Inkster, Homer Wiland, James Wu.

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