MAYO CLINIC LABORATORIES

Test Definition: AHPV

Anal Pap Human Papillomavirus (HPV) with Genotyping, High-Risk Types, PCR, Varies

Overview

Useful For

Detection of high-risk (HR) genotypes associated with the development of anal cancer

Individual genotyping of human papillomavirus (HPV)-16 and HPV-18 if present

May aid in triaging men and women with positive HR HPV but negative anal Papanicolaou (Pap) smear test results

This test is **not intended for use** in medical-legal applications.

Method Name

Real-Time Polymerase Chain Reaction (PCR)

NY State Available

Yes

Specimen

Specimen Type

Varies

Necessary Information

Specimen source collection date, and patient identifiers are required.

Specimen Required

Submit one of the following specimens:

Preferred

Specimen Type: Anus or rectum

Supplies: ThinPrep Media with Spatula and Brush Kit (T434)

Collection Container/Tube: Brush

Submission Container/Tube: ThinPrep

Specimen Volume: A minimum of 20 mL or entire collection

Collection Instructions:

1. Visualize the anal opening by retracting the buttocks.
2. Insert brush past internal anal sphincter until it abuts the distal rectal wall (4-5cm).
3. Rotate the brush 360 degrees and maintain lateral pressure on brush against the walls of the anus. The brush should bow slightly due to the pressure.
4. While rotating, slowly withdraw the brush.

5. Rotate at least 10 times while withdrawing the brush. This should take 20 to 30 seconds.
6. Immediately place brush in the ThinPrep solution and swish at least 20 to 30 seconds.
7. Dispose of the brush.
8. Tighten the cap so that the torque line on the cap passes the torque line on the vial.
9. Bag ThinPrep specimens individually as they tend to leak during transport.
10. Place labels on the vial and on the biohazard bag.

Acceptable
Specimen Type: Anus or rectum
Supplies: PreservCyt Vial (T536)
Collection Container/Tube: Dacron/polyester swab
Submission Container/Tube: PreservCyt solution vial
Specimen Volume: A minimum of 20 mL or entire collection
Collection Instructions:
1. Visualize the anal opening by retracting the buttocks.
2. Insert Dacron or polyester swab past internal anal sphincter until it abuts the distal rectal wall (4-5cm).
3. Rotate the swab 360 degrees and maintain lateral pressure on swab against the walls of the anus. The swab should bow slightly due to the pressure.
4. While rotating, slowly withdraw the swab.
5. Rotate at least 10 times while withdrawing the swab. This should take 20 to 30 seconds.
6. Immediately place swab in the ThinPrep solution. Swish at least 20 to 30 seconds in the ThinPrep solution.
7. Dispose of the swab.
8. Tighten the cap so that the torque line on the cap passes the torque line on the vial.
9. Bag ThinPrep specimens individually as they tend to leak during transport.
10. Place labels on the vial and on the biohazard bag.

Specimen Minimum Volume

1 mL

Reject Due To

Specimen containing CytoRich Red preservative fluid	Reject
Perirectal or perianal	Reject

Specimen Stability Information

Specimen Type	Temperature	Time	Special Container
Varies	Ambient	21 days	

Clinical & Interpretive

Clinical Information

Persistent infection with human papillomavirus (HPV) can cause anal cancer, with approximately 90% of all anal cancers being associated with HPV infection. HPV is a small, nonenveloped, double-stranded DNA virus, with a genome of approximately 8000 nucleotides. There are more than 118 different types of HPV and approximately 40 different HPVs that can infect human anogenital mucosa. However, data suggests that 14 of these types (HPV types 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68) are high risk (HR) for the development of cervical and anal cancer and precursor lesions. Furthermore, HPV types 16 and 18 have been regarded as the genotypes most closely associated with progression to cancer. HPV-16 is the most carcinogenic and is associated with approximately 60% of all HPV-related cancers, while HPV-18 accounts for approximately 10% to 15% of HPV-related cancers.(1-3)

Sexual transmission of HPV is extremely common, with estimates of up to 75% of all women being exposed to HPV at some point. However, almost all infected patients will mount an effective immune response and clear the infection within 2 years without long-term health consequences.

Testing for HPV DNA by real-time polymerase chain reaction (PCR) is a noninvasive method for determining the presence of HPV infection. Proper implementation of DNA testing for anal HPV may increase the sensitivity of anal cancer detection and reduce the need for unnecessary biopsy and treatment.

Data suggests that individual genotyping for HPV types 16 and 18 can assist in determining appropriate follow-up testing and triaging of patients who are at risk for possible anal cancer. Detection of HR HPV DNA, especially genotypes 16 and 18, may assist in triaging patients and determining appropriate management strategies.

Reference Values

Negative for human papillomavirus (HPV) genotypes 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68

Interpretation

A positive result indicates the presence of human papillomavirus (HPV) DNA from one or more of the following genotypes: 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68. Management of abnormal results requires consideration of baseline risk of anal cancer and prior anal cytology and HPV results.

For patients with an anal Papanicolaou (Pap) smear test result showing atypical squamous cells of undetermined significance (ASC-US) and who are positive for high-risk (HR) HPV, consider referral for anoscopy if clinically indicated.

A negative result indicates the absence of HPV DNA from the targeted genotypes.

For men and women with a negative anal Pap smear test result but who are positive for HPV-16 or HPV-18, consider referral for anoscopy if clinically indicated.

For men and women with a negative anal Pap smear test and positive-HR HPV test result but are negative for HPV-16 and HPV-18, consider repeat testing by both cytology and HR HPV RT-PCR in 12 months.

Cautions

The cobas human papillomavirus (HPV) test is US Food and Drug Administration (FDA)-approved for cervical and endocervical samples collected in PreservCyt (ThinPrep) media. Other specimen types (eg, anal) are not considered FDA-approved sources; however, verification studies have been completed by Mayo Clinic Laboratories and Mayo Clinic in compliance with CLIA regulations.

The cobas HPV test detects DNA from high-risk genotypes 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68.

This test does not detect DNA of HPV low-risk types (eg, 6, 11, 42, 43, 44) since these are not associated with cervical cancer and its precursor lesions.

Prevalence of HPV infection in a population may affect performance. Positive-predictive values decrease when testing populations with low prevalence or individuals with no risk of infection.

Infection with HPV is not an indicator of cytologic high-grade squamous intraepithelial lesion (HSIL) or underlying high-grade cervical intraepithelial neoplasia (CIN), nor does it imply that CIN2-3 or cancer will develop. Most patients infected with 1 or more high-risk (HR) HPV types do not develop CIN2-3 or cancer.

A negative HR HPV result does not exclude the possibility of future cytologic HSIL or underlying CIN2-3 or cancer.

Anal specimens may show visibly detectable levels of whole blood or stool as a pink or light brown coloration. These specimens are processed normally on the cobas system. If concentrations of whole blood or stool are high, results may be impacted.

The cobas HPV test performance has been validated with PreservCyt specimens that have been treated with up to 5% glacial acetic acid for removal of red blood cells. Addition of glacial acetic acid over 5% in PreservCyt specimens prior to HPV testing would invalidate the cobas HPV test results.

The cobas HPV test performance has not been validated with PreservCyt specimens that have been filled past the maximum fill line of the primary vial. ThinPrep vials that have had any additional PreservCyt fluid volume added or any dissimilar fluid volume added to the initial specimen should not be submitted for testing.

Human beta-globin amplification and detection is included in the cobas HPV test to differentiate HPV-negative specimens from those that do not exhibit HPV signal due to insufficient cell mass in the specimen. All HPV-negative specimens must have a valid beta-globin signal within a predefined range to be identified as valid negative results.

Human papillomavirus-negative cancers of the cervix or anus do occur in rare circumstances. Also, no cancer screening test is 100% sensitive. Use of this device for primary anal cancer screening should be undertaken after carefully considering the performance characteristics put forth in the cobas HPV test label as well as recommendations of professional guidelines.

The presence of real-time polymerase chain reaction inhibitors may cause false-negative or invalid results.

Anal specimens that are grossly contaminated with stool may yield invalid or false-negative results.

Clinical Reference

1. Swanson AA, Hartley C, Long ME, et al. Evaluation of high-risk human papillomavirus testing and anal cytology to detect high-grade anal intraepithelial neoplasia. J Am Soc Cytopathol. 2021;10(4):406-413. doi:10.1016/j.jasc.2021.03.007
2. Silva Dalla Libera L, Almeida de Carvalho KP, Enocencio Porto Ramos J, et al. Human papillomavirus and anal cancer: Prevalence, genotype distribution, and prognosis aspects from Midwestern region of Brazil. J Oncol. 2019;2019:6018269. Published 2019 Sep 18. doi:10.1155/2019/6018269
3. Wieland U, Kreuter A. Anal cancer risk: HPV-based cervical screening programmes. Lancet Infect Dis. 2019;19(8):799-800

Performance

Method Description

The cobas HPV (human papillomavirus) test is a qualitative real-time polymerase chain reaction (PCR) test that detects 14 high-risk HPV genotypes. The test uses primers to define a sequence of approximately 200 nucleotides within the polymorphic L1 region of the HPV genome. A pool of HPV primers present in the Master Mix is designed to amplify HPV DNA from 14 high-risk types (16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, and 68). An additional primer pair targets the human beta-globin gene (330 base pair amplicon) as an internal control to monitor the entire sample preparation and PCR amplification process. Fluorescent oligonucleotide probes bind to polymorphic regions within the sequence defined by these primers. The test utilizes a low titer positive and a negative control.(Package insert: cobas HPV: Qualitative nucleic acid test for the cobas 5800/6800/8800 Systems. Roche Diagnostics; Rev. 2.0, 09/2024)

PDF Report

No

Day(s) Performed

Monday through Saturday

Report Available

3 to 6 days

Specimen Retention Time

2 weeks

Performing Laboratory Location

Mayo Clinic Laboratories - Rochester Superior Drive

Fees & Codes

Fees

- Authorized users can sign in to Test Prices for detailed fee information.
- Clients without access to Test Prices can contact Customer Service 24 hours a day, seven days a week.
- Prospective clients should contact their account representative. For assistance, contact Customer Service.

Test Classification

This test has been modified from the manufacturer's instructions. Its performance characteristics were determined by Mayo Clinic in a manner consistent with CLIA requirements. This test has not been cleared or approved by the US Food and Drug Administration.

CPT Code Information

87626

LOINC® Information

Test ID	Test Order Name	Order LOINC® Value
AHPV	HPV Anal Detect / Genotyping PCR	77376-2

Result ID	Test Result Name	Result LOINC® Value
AHPVS	Specimen Source	31208-2
607831	HPV High Risk type 16, PCR	77396-0
607832	HPV High Risk type 18, PCR	77395-2
607833	HPV other High Risk types, PCR	77394-5