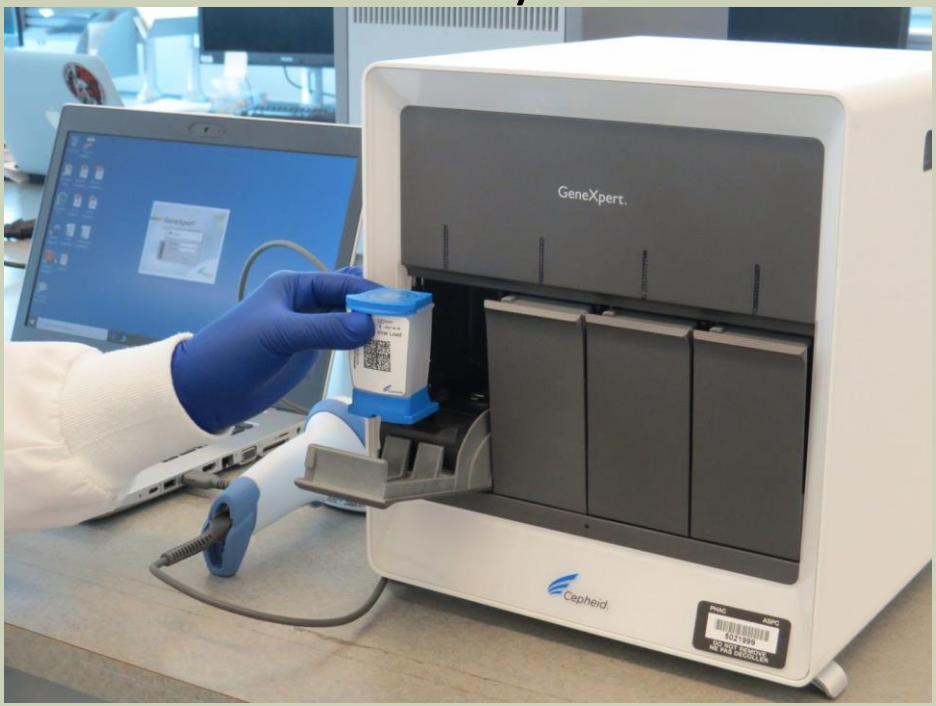
CEPHEID XPERT AND POINT OF CARE HPV TESTING

NCPTS National Training Day
5th of May 2022

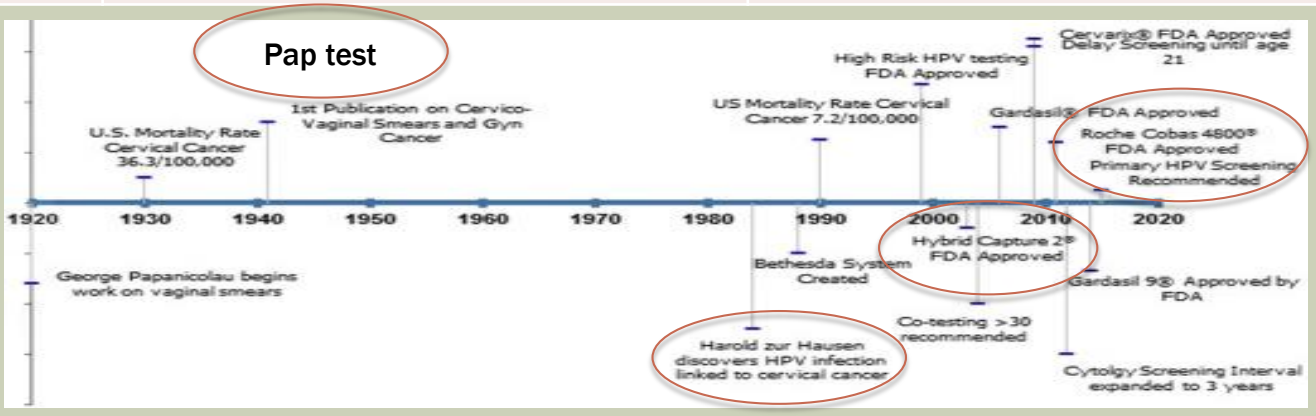


Meik Dilcher
Scientific Officer
Canterbury Health Laboratories, Christchurch

HUMAN PAPILLOMAVIRUS

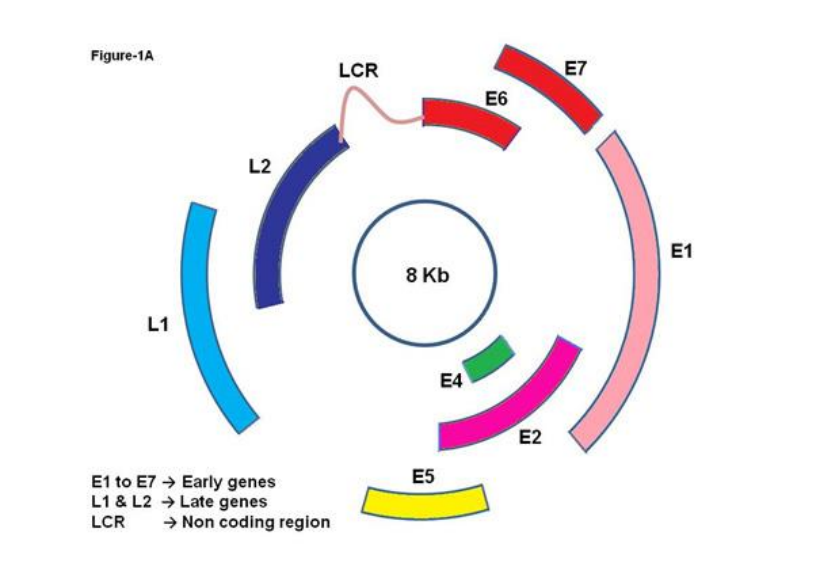
- HPV is one of the most common sexually transmitted viruses.
- Persistent HPV infection can lead to cervical cancer.

HPV Group	HPV Types	Clinical Association
Low Risk	6, 11, 42, 43, 44	Genital warts or benign lesions.
High Risk	16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, 66, 68	All types isolated from cancers of the cervix, but also vagina, vulva, penis, and anus.



- Strong evidence supports that screening using molecular assays that detect nucleic acids of oncogenic or high-risk human papillomavirus (hrHPV) types are more effective, in terms of reducing the incidence and mortality from this cancer, than cytology, and they offer better sensitivity and less frequent screening intervals.
- An increasing number of countries have switched from cytology to molecular HPV-based national screening programs or have decided to implement this change in the near future.

HPV NUCLEIC ACID DETECTION TESTS

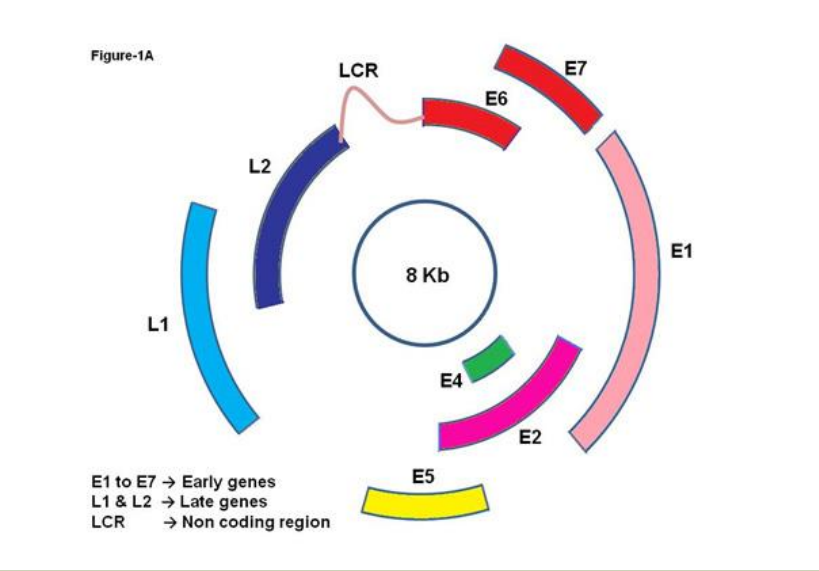


L1, L2, E2, E4, and E5 deletions may occur during integration

Company	Product	Target
Digene/Qiagen	Hybrid Capture 2	Whole genome probe DNA
Roche	Cobas HPV	L1 DNA
Abbott	RealTime HPV	L1 DNA
Abbott	Alinity m HR HPV	L1 DNA
Integrated Sciences	Seegene Anyplex II HPV	L1 DNA
Genera Biosystems	PapType	L1 DNA
Becton Dickinson	BD Onclarity	E6, E7 DNA
ESL Biosciences	EUROIMMUN EUROArray HPV	E6, E7 DNA
Hologic	Cervista HPV	L1, E6, E7 DNA
Hologic	Aptima HPV	E6, E7 mRNA

Most **HPV NAAT tests are complicated to use, have TAT's of several hours and batch testing can delay results** critical for scheduling patient consultations for follow-up testing or colposcopy.

HPV NUCLEIC ACID DETECTION TESTS



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Most **HPV NAAT tests are complicated to use, have TAT's of several hours and batch testing can delay results** critical for scheduling patient consultations for follow-up testing or colposcopy.

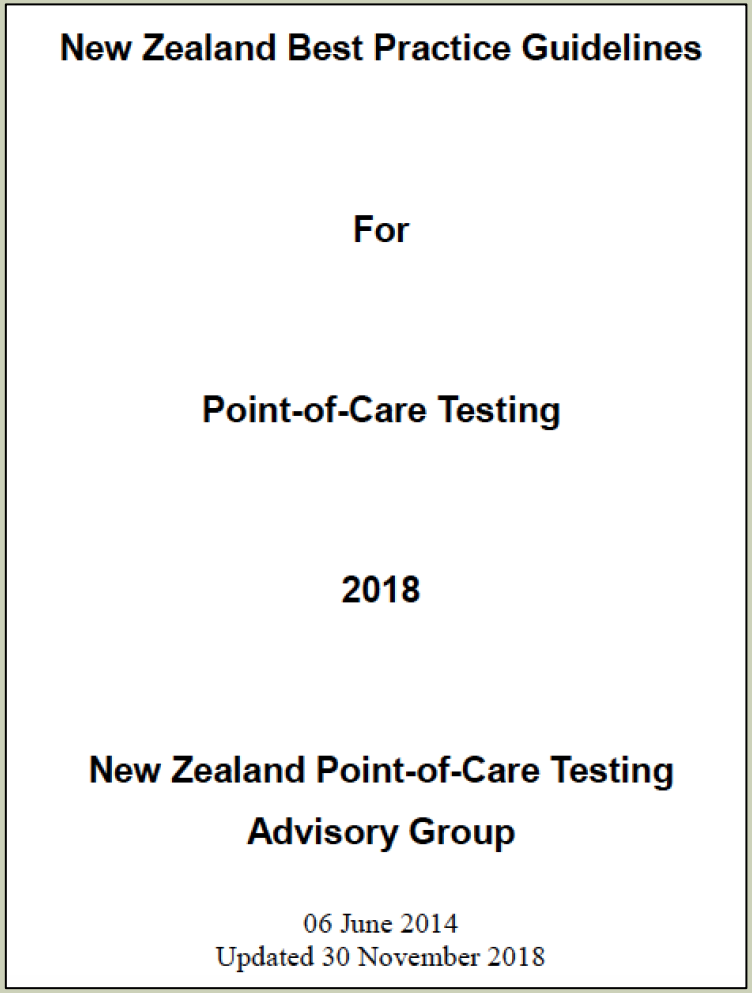
POINT-OF-CARE TESTING

„HIS URINE IS SWEET, HE HAS DIABETES MELLITUS“ SAID THE PHYSICIAN.

- POCT = analysis of clinical specimens outside the traditional laboratory, near to or at the site of patient care
  - e.g. hospital wards, operating theatres, ED, General Practice surgeries, health clinics, pharmacies, ambulance services or patient‘s homes
- Advantages
  - Reduced turnaround time
  - Easy to use
  - Ability to provide tests in remote locations
  - Improve convenience and access to health care service for patients
  - Facilitate opportunistic screening for early identification of certain conditions
- Challenges
  - Training and competency and potential increase of workload of clinical staff
  - Accuracy and reliability need to be ensured (quality management, eQA enrolment)
  - Potential for transcription errors of results if no interfacing with electronic patient records is established
  - Point-of-care devices are not subject to effective regulation and accreditation in NZ
    - There is a requirement for most medical devices to be notified to the WAND database, operated by Medsafe.

adapted from: Position Statement of the New Zealand Medical Association and 2018 New Zealand Best Practice POCT Guidelines

POCT POLICIES IN NZ


New Zealand Best Practice Guidelines

For

Point-of-Care Testing

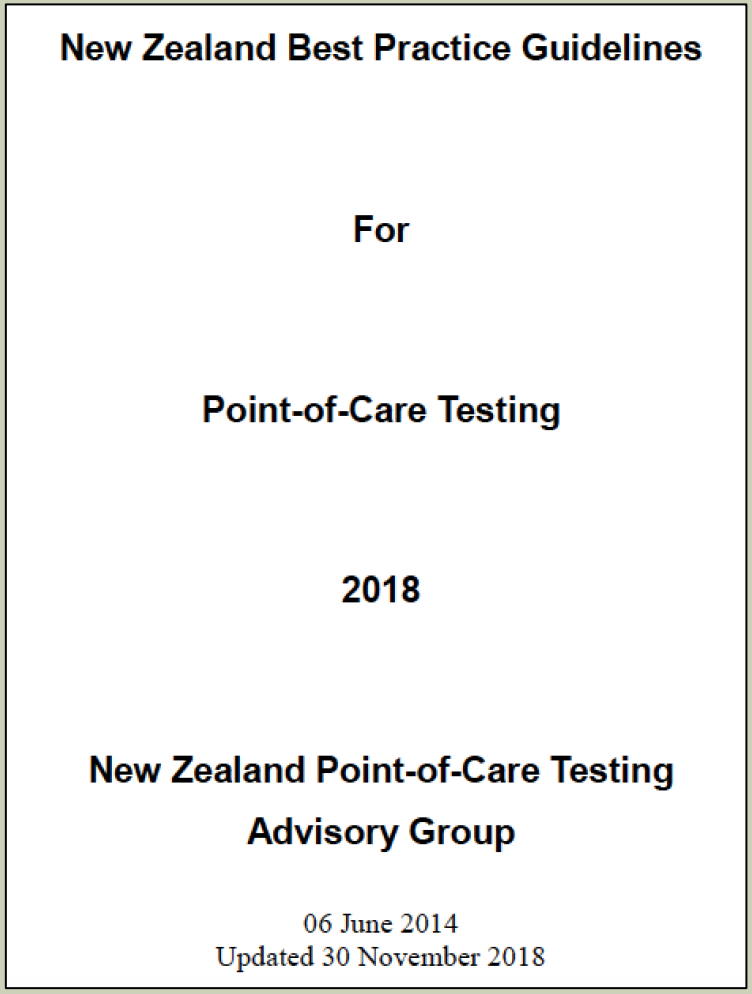
2018

New Zealand Point-of-Care Testing Advisory Group

06 June 2014
Updated 30 November 2018

- Governance for POCT
- Risk Management
- Assessment of clinical need
- Inclusion of Laboratory input
- Cost Benefit Analysis
- Validation technology
- Quality Mananagement System
- Sources of Errors
- Reporting of Results
- POCT device connectivity compliance
- Health and Safety

POCT POLICIES IN NZ



New Zealand Best Practice Guidelines

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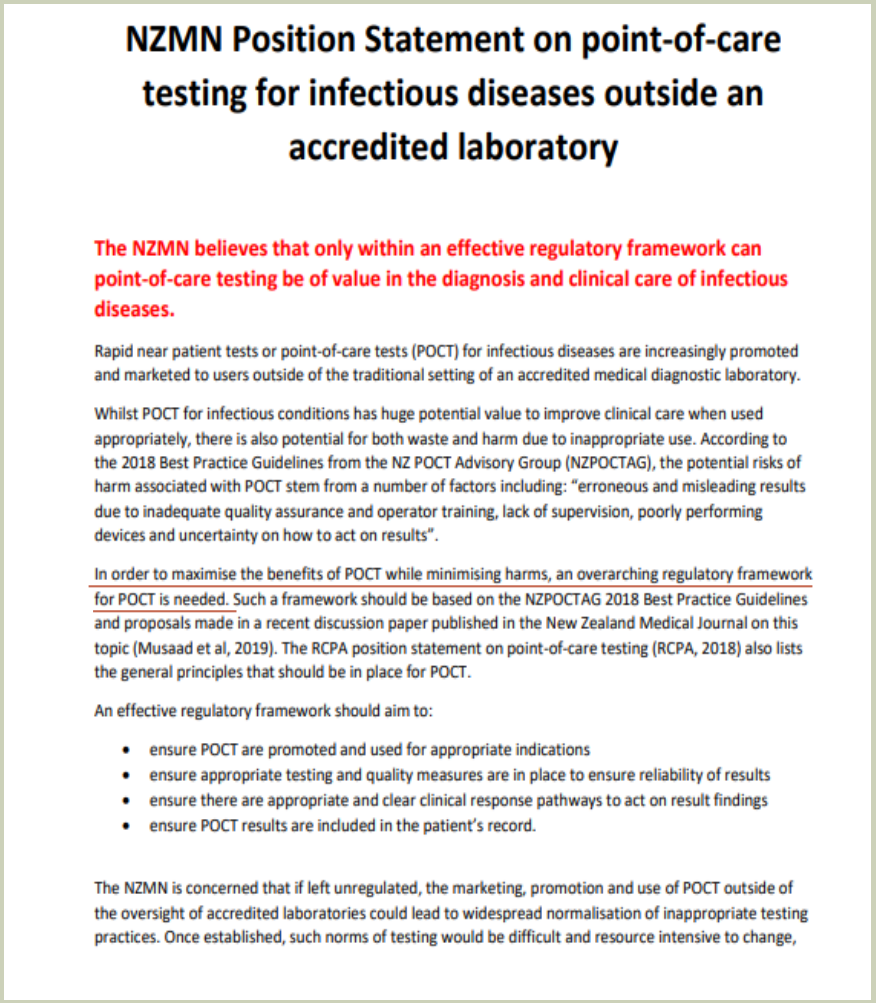


NZMN Position Statement on point-of-care testing for infectious diseases outside an accredited laboratory

The NZMN believes that only within an effective regulatory framework can point-of-care testing be of value in the diagnosis and clinical care of infectious diseases.

Rapid near patient tests or point-of-care tests (POCT) for infectious diseases are increasingly promoted and marketed to users outside of the traditional setting of an accredited medical diagnostic laboratory.

Whilst POCT for infectious conditions has huge potential value to improve clinical care when used appropriately, there is also potential for both waste and harm due to inappropriate use. According to the 2018 Best Practice Guidelines from the NZ POCT Advisory Group (NZPOCTAG), the potential risks of harm associated with POCT stem from a number of factors including: "erroneous and misleading results due to inadequate quality assurance and operator training, lack of supervision, poorly performing devices and uncertainty on how to act on results".

In order to maximise the benefits of POCT while minimising harms, an overarching regulatory framework for POCT is needed. Such a framework should be based on the NZPOCTAG 2018 Best Practice Guidelines and proposals made in a recent discussion paper published in the New Zealand Medical Journal on this topic (Musaad et al, 2019). The RCPA position statement on point-of-care testing (RCPA, 2018) also lists the general principles that should be in place for POCT.

An effective regulatory framework should aim to:

- ensure POCT are promoted and used for appropriate indications
- ensure appropriate testing and quality measures are in place to ensure reliability of results
- ensure there are appropriate and clear clinical response pathways to act on result findings
- ensure POCT results are included in the patient's record.

The NZMN is concerned that if left unregulated, the marketing, promotion and use of POCT outside of the oversight of accredited laboratories could lead to widespread normalisation of inappropriate testing practices. Once established, such norms of testing would be difficult and resource intensive to change,

LOW- AND MIDDLE-INCOME COUNTRIES (LMIC)

- Implementation of the traditional Pap smear or visual inspection after acetic acid application (VIA) in national screening programmes is not sustainable in under-resourced LMIC settings with a limited skilled cytologist workforce and where loss to follow-up and poor adherence to treatment are major obstacles.
- Where resources are available, the WHO recommends a "screen-and-treat" strategy for women aged 30 to 49 years with screening via DNA testing and treatment of HPV-positive women with timely cryotherapy.
- In LMIC there is a need for innovative non-batched point-of-care molecular diagnostic tools that are sensitive and specific and can be integrated into primary health care settings.
- There are multiple HPV point-of-care testing platforms on the market, e.g. **careHPV** test from Qiagen, but almost none of these have been fully validated in the clinical setting.
  - The **careHPV** laboratory processing time is still approximately 4 hours
  - Needs to be run in batched mode (90 samples)
- This hinders same-day results and treatment for HPV-positive women and poses a need for at least two visits (first for administration of the screening test and second for receiving results and treatment).

Shahin Sayed et al.,: Point-of-care HPV molecular diagnostics for a test-and-treat model in high-risk HIV populations, The Lancet, Vol 8, February 2020 and Campos et al,: Estimating the value of point-of-care HPV testing in three low- and middle-income countries: a modelling study BMC Cancer (2017) 17:791

CEPHEID XPERT HPV ASSAY

- The samples are processed as individual cartridges in individual modules
- The GeneXpert System is available in a 2, 4, 16, 48, or 80-module configuration

POCT

2 4 16 48-80

= tests/hour

1300 – 2300 test/24h
= 95 tests/hour

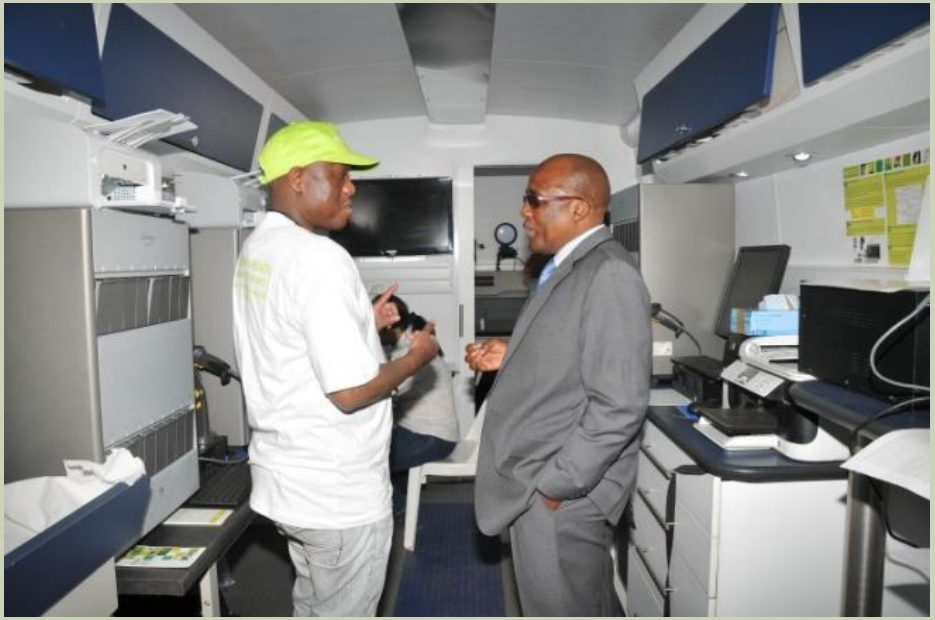
The GeneXpert is currently the only validated HPV point-of-care testing device.
It is CE marked, but is not yet FDA approved.

GENEXPERT SYSTEM: CE-IVD TEST MENU

Respiratory	Xpert® Xpress SARS-CoV-2/ Flu/RSV	Rapid detection and differentiation of SARS-CoV-2, Flu A, Flu B and RSV in approximately 36 minutes
	Xpert Xpress SARS-CoV-2	Rapid detection of SARS-CoV-2 in as soon as 30 minutes*
	Xpert Xpress Strep A	Rapid detection of Group A Streptococcus DNA in as soon as 18 minutes*
	Xpert Xpress Flu/RSV	Rapid detection and differentiation of Flu A, Flu B, and RSV in as soon as 20 minutes*
Healthcare-Associated Infections & Other Infectious Diseases	Xpert MRSA NxG	Active MRSA surveillance testing in around 45 minutes*
	Xpert SA Nasal Complete	Pre-surgical testing of *S. aureus* and MRSA in about an hour
	Xpert MRSA/SA BC	Detection of MRSA and *S. aureus* in positive blood cultures in about an hour
	Xpert MRSA/SA SSTI	Detection of MRSA and *S. aureus* skin and soft tissue infections in about an hour
	Xpert Carba-R	Detection and differentiation of KPC, NDM, VIM, IMP, and OXA-48 in 50 minutes
	Xpert Norovirus	Identification and differentiation of Norovirus GI and GII in less than 1 hour*
	Xpert EV	Detection of enteroviruses in CSF in 2.5 hours
	Xpert *C. difficile* BT	Detection of *Clostridium difficile* infection with an independent call-out of binary toxin and differentiation of the 027 strain in around 45 minutes
	Xpert *vanA/vanB*	Rapid VRE screening for active outbreak prevention and control in around 45 minutes
TB & Emerging Infectious Diseases	Xpert MTB/RIF	Detection of *Mycobacterium tuberculosis* complex and Rifampin-resistance associated mutations in less than two hours
	Xpert MTB/RIF Ultra	Detection of *Mycobacterium tuberculosis* complex and Rifampin-resistance associated mutations in less than 80 minutes
	Xpert MTB/XDR	Detection of *Mycobacterium tuberculosis* complex and mutations associated with drug resistance towards Isoniazid, Fluoroquinolones, Second-Line Injectable Drugs and Ethionamide in less than 90 minutes, leveraging 10-color GeneXpert technology
	Xpert Ebola	Detection of Ebola Zaire virus in around 90 minutes

Blood Virology, Women's Health, & Sexual Health	Xpert CT/NG	Detection of *Chlamydia trachomatis* and *Neisseria gonorrhoeae* infections in about 90 minutes
	Xpert HPV	Detection of high risk Human Papillomavirus (HPV) – identifies types HPV 16 and HPV 18/45; reports 11 other high risk types in pooled results in less than one hour
	Xpert GBS	Intrapartum detection for Group B Streptococcus (GBS) during labor/ delivery in less than one hour
	Xpert TV	Detection of *Trichomonas vaginalis* in male and female specimens in around one hour*
	*Resistance*Plus® MG FleXible#	Detection of *M. genitalium* and macrolide resistance in around two hours
	Xpert HBV Viral Load	Detection and quantitation of Hepatitis B virus (HBV) in less than one hour
	Xpert HCV Viral Load	Detection and quantitation of Hepatitis C virus (HCV) in 105 minutes
	Xpert HCV VL Fingerstick	Detection and quantitation of Hepatitis C virus (HCV) in about an hour
	Xpert HIV-1 Qual	Detection of Human Immunodeficiency Virus Type 1 (HIV-1) in around 90 minutes
	Xpert HIV-1 Viral Load	Detection and quantification of Human Immunodeficiency Virus type 1 (HIV-1) in around 90 minutes
Oncology & Human Genetics	Xpert Bladder Cancer Detection	Detection of the presence of bladder cancer in patients with hematuria in around 90 minutes
	Xpert Bladder Cancer Monitor	Qualitative monitoring for recurrence in patients previously diagnosed with bladder cancer in around 90 minutes
	Xpert Breast Cancer STRAT4	Semi-quantitative measurement of ESR1, PGR, ERBB2, and MKI67 from FFPE invasive breast cancer tissue in 70 minutes
	Xpert BCR-ABL Ultra	Standardized measurement of BCR-ABL p210 transcript levels for individuals with Chronic Myeloid Leukemia (CML) in under 2 hours
	Xpert FII & FV	Identification of genetic risk factors for thrombosis in around 30 minutes

MOBILE GENEXPERT LAB IN SA



EASY WORKFLOW, RAPID TAT

1

Obtain an endocervical specimen stored in the validated transport medium[#]

2

Transfer sample to cartridge

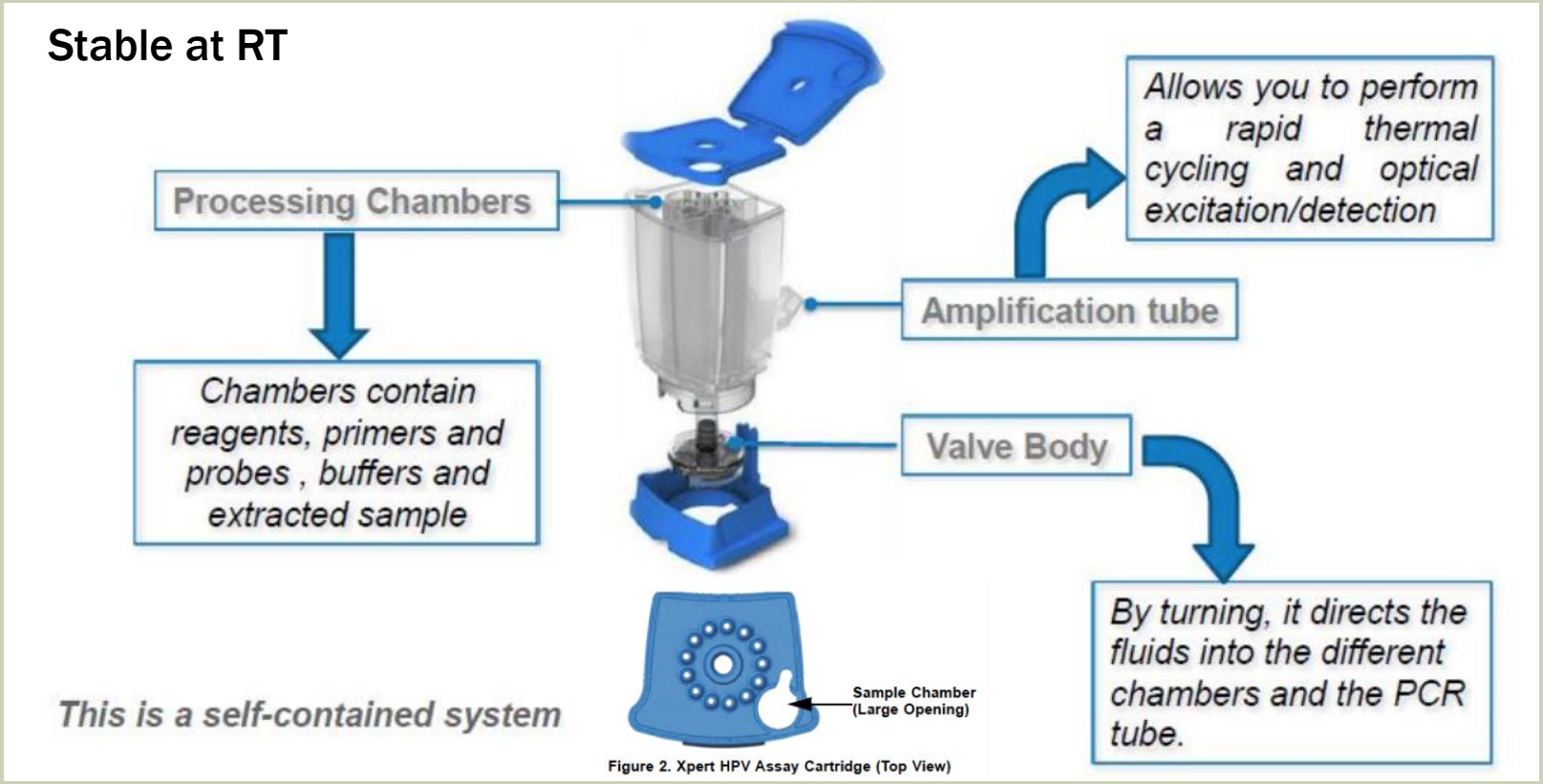
Sample volume: 1 ml

3

Insert cartridge and start test

TAT: 56 min

OVERVIEW OF THE GENEXPERT SINGLE-USE CARTRIDGE



https://www.youtube.com/watch?v=j-y3xi1K7JE&t=1s

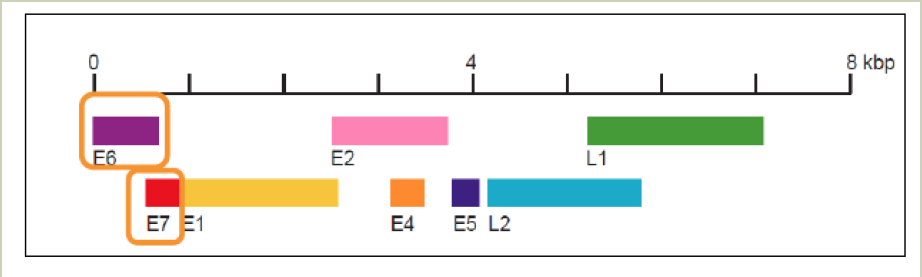
TARGETS AND PROBES

- Target
  - 14 High Risk HPV types

Result group	HPV types detected
HPV 16	HPV 16
HPV 18_45	HPV 18 and 45
P3	HPV 31, 33, 35, 52, 58
P4	HPV 51, 59
P5	HPV 39, 56, 66, 68

Cause more than 95% of cervical cancers

- Probes
  - One probe binds to the Sample Adequacy Control (SAC)
  - Remaining probes bind depending on the presence of hrHPV types detected in the patient sample



-> Targeting the E6/E7 oncogenes eliminates concerns in case of L1 gene deletion

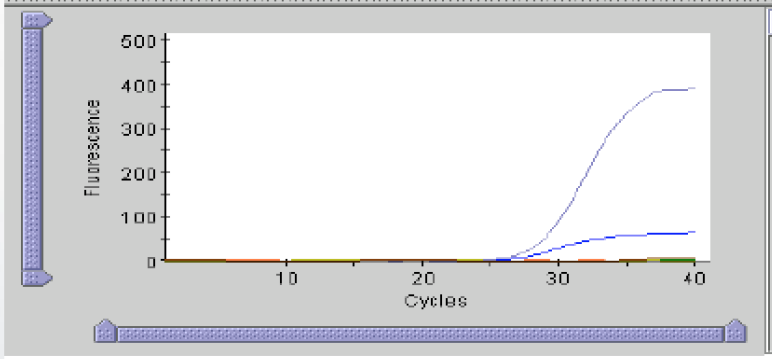
INTERNAL CONTROLS

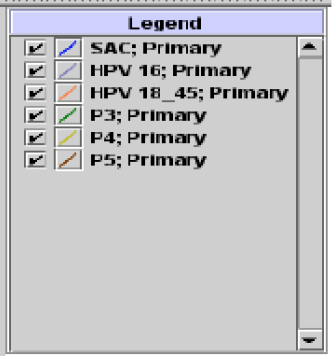
- Sample Adequacy Control (SAC)
  - Targets the human Hydroxymethylbilane Synthase gene
  - Ensures that human cells are present
  - Can indicate poor sampling if negative
  - Must be positive in HPV negative samples
  - Can be positive or negative in HPV positive samples
- Probe Check Controls (PCC)
  - Before the real-time PCR starts the fluorescence signal on all probes is measured and compared with pre-established factory settings to monitor for
    - Probe integrity
    - Dye stability
    - Reagent rehydration
    - PCR tube filling

HPV16 POS

Test Result | Analyte Result | Detail | Errors | History | Support

Analyte Name	Ct	EndPt	Analyte Result	Probe Check Result
SAC	28.7	63	NA	PASS
HPV 16	28.0	388	POS	PASS
HPV 18_45	0.0	4	NEG	PASS
P3	0.0	1	NEG	PASS
P4	0.0	-1	NEG	PASS
P5	0.0	5	NEG	PASS





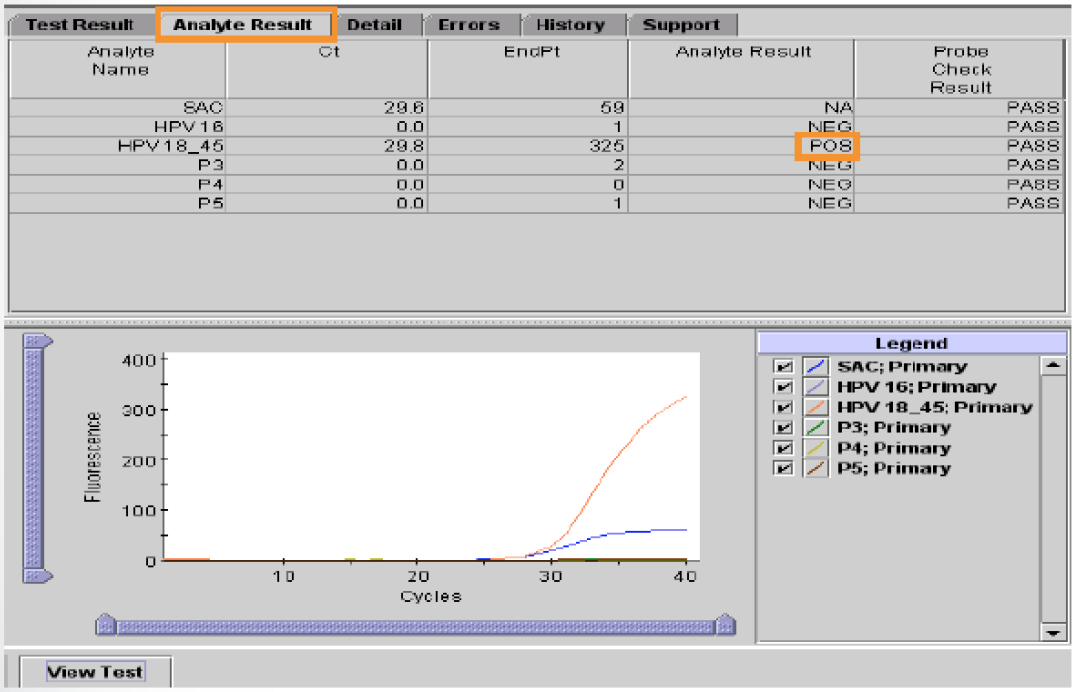
HPV 16 POS;
HPV 18_45 NEG;
OTHER HR HPV NEG

-HPV16 target DNA sequence has a Ct within the valid range and a fluorescence endpoint above the threshold setting.

-SAC: SAC is not applicable because the HPV target amplification can compete with this control.

-Probe Check: PASS

HPV18_45 POS

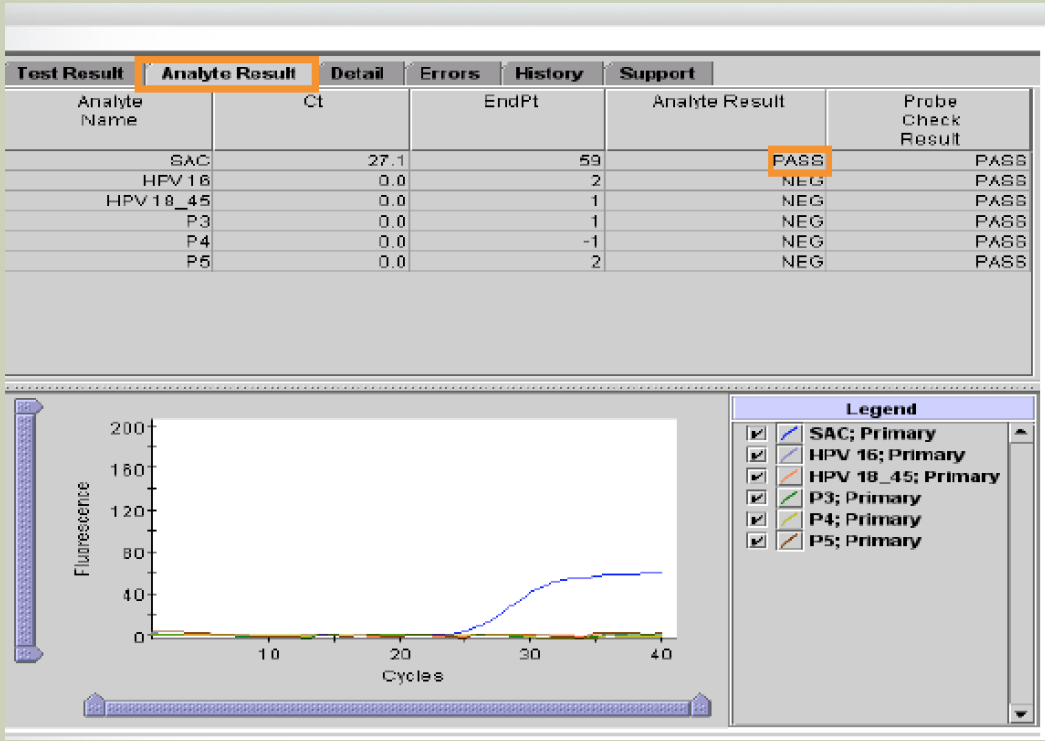


-HPV18_45 target DNA has a Ct within the valid range and a fluorescence endpoint above the threshold setting.

-SAC: SAC is not applicable because the HPV target amplification can compete with this control.

-Probe Check: PASS

NEG



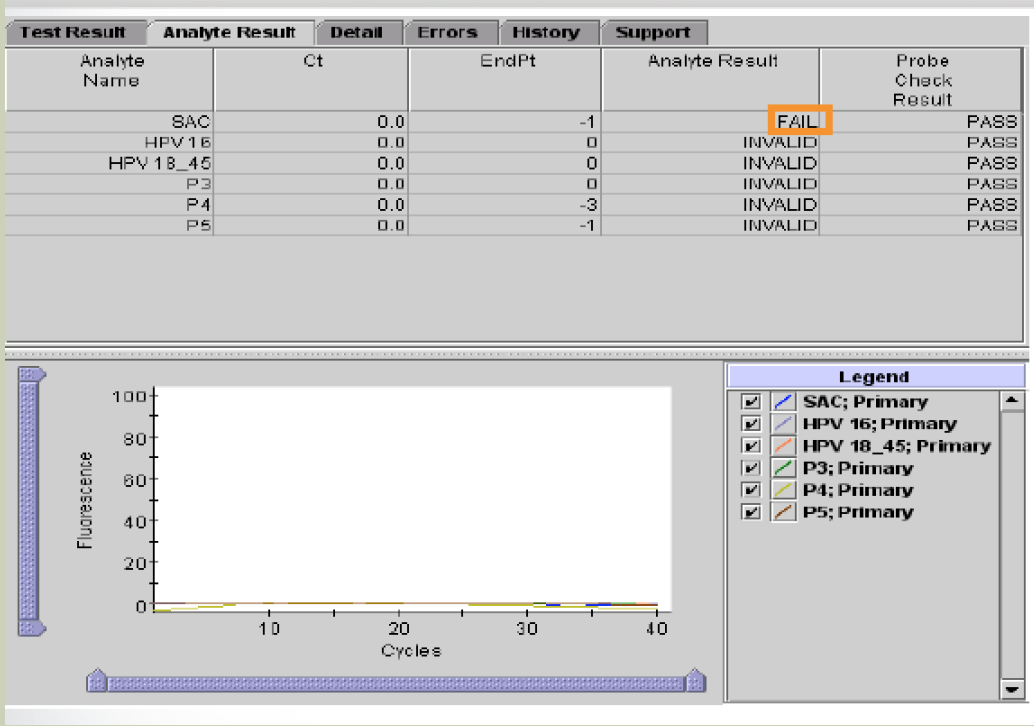
HPV 16 NEG;
HPV 18_45 NEG;
OTHER HR HPV NEG

-The targeted HPV DNA sequences have a Ct value that is not within the valid range and/or a fluorescence endpoint below the threshold setting.

-SAC: SAC has a Ct value within the valid range

- Probe Check: PASS

INVALID RESULT



Presence or absence of HPV target cannot be determined.

- SAC:FAIL The SAC does not meet the acceptance criteria
- Probe Check: PASS

Possible Causes

- Improper sample collection
- Incorrect sample preparation
- Improper storage of the cartridges
- Inefficient sample processing in cartridge
- Presence of interfering substances in the sample

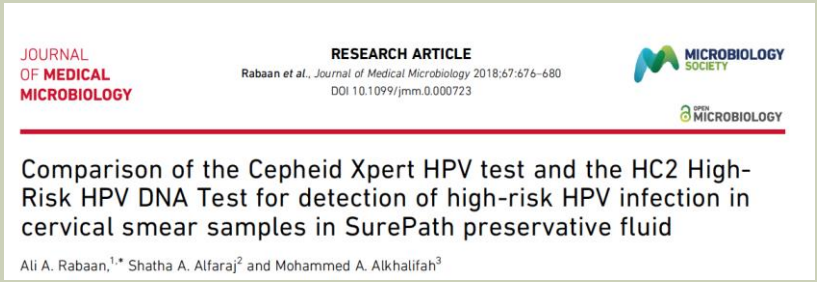
Solution

- Repeat the test with a new cartridge and new sample

SPECIMEN

Sample type:

	Prior to testing	Temperature (°C)	Storage Time
	Cervical specimens collected in PreservCyt Solutions	+30 +2 °C	Up to 6 months



JOURNAL OF MEDICAL MICROBIOLOGY

RESEARCH ARTICLE

Rabaan *et al.*, *Journal of Medical Microbiology* 2018;67:676–680
DOI 10.1099/jmm.0.000723

MICROBIOLOGY SOCIETY

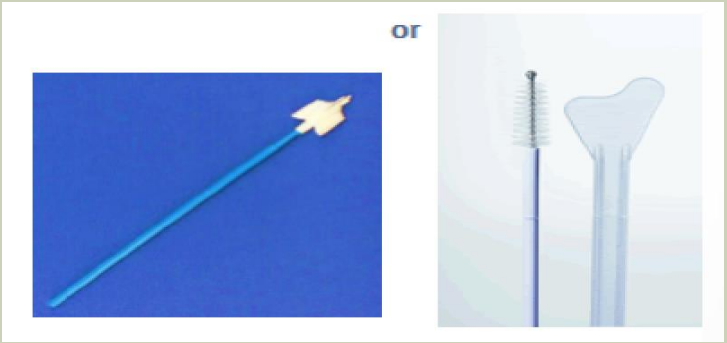
OPEN MICROBIOLOGY

Comparison of the Cepheid Xpert HPV test and the HC2 High-Risk HPV DNA Test for detection of high-risk HPV infection in cervical smear samples in SurePath preservative fluid

Ali A. Rabaan,[1,*] Shatha A. Alfaraj[2] and Mohammed A. Alkhalifah[3]

- Cervical cells collected in ThinPrep PreserveCyt® Solution (Hologic Corporation)
- Specimens pre-treated with Glacial Acetic Acid (GAA) have also been validated for use with the Xpert HPV assay

Sample collection:



Collected with either a broom-like device or an endocervical brush/spatula combination

SELF-COLLECTED SAMPLES

RESEARCH ARTICLE

Accuracy of self-collected vaginal dry swabs using the Xpert human papillomavirus assay

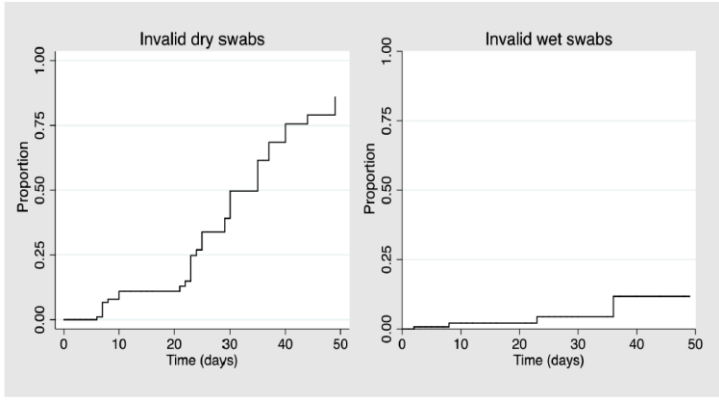
Rosa Catarino[1]*, Pierre Vassilakos[2], Aline Bilancioni[1], Stéphanie Bougel[3], Meriem Boukrid[1], Ulrike Meyer-Hamme[1], Patrick Petignat[1]

1 Division of Gynaecology, Department of Gynaecology and Obstetrics, Geneva University Hospitals, Geneva, Switzerland, 2 Geneva Foundation for Medical Education and Research, Geneva, Switzerland, 3 Biopath Lab SA, Lausanne, Switzerland

PlosOne, July 2017

- Cross-sectional study on 150 women
  - Each women first self-collected a vaginal sample using a dry swab
  - Then the physician collected a cervical specimen in ThinPrep
  - HPV analysis was performed with Xpert
  - Part of ThinPrep collected sample was also tested with the cobas HPV test
  - HPV test positivity and performance of the two collection methods was compared
- Results:
  - HPV positivity
    - 49.1% for dry swab on Xpert
    - 41.8% for wet swab (ThinPrep) on Xpert
    - 46.2% for wet swab (ThinPrep) on cobas
  - HPV16 detection and LSIL+
    - Excellent agreement between the two samples (dry/wet)
  - Sensitivity and specificity for CIN2+ detection
    - Dry swab: 84.2% and 47.7%
    - Wet swab: 73.1% and 58.7%
    - Cobas: 77.8% and 45.7%
- Conclusion:
  - Results suggest that dry swab performance is similar to the performance of clinically validated ThinPrep-collected samples.

36 (24%) **vs** **4 (2.7%)**



SELF-COLLECTED SAMPLES

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journal homepage: www.elsevier.com/locate/jcv

Analytical performance of HPV assays on vaginal self-collected vs practitioner-collected cervical samples: the SCoPE study

M Saville[a,b,c,d,1], D Hawkes[a,b,e,f,z,1], MHT Keung[a,b], ELO Ip[a,b], J Silvers[g], F Sultana[a,h], MJ Malloy[a,h], LS Velentzis[h,i], K Canfel I[i,j], CD Wrede[c,g], JML Brotherton[a,d,h]

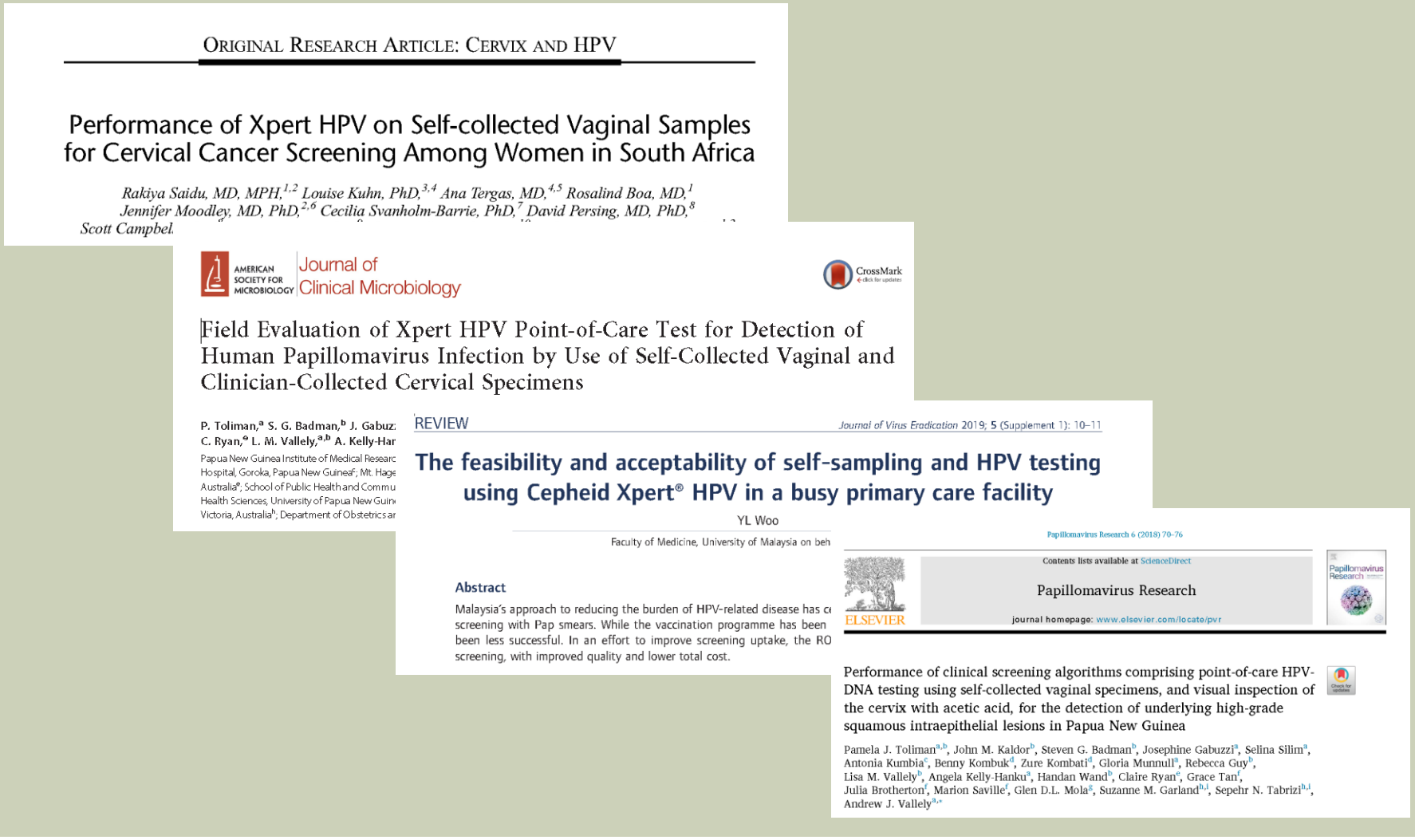
- Cross-sectional study on 303 women with either
  - Self-collected flocked swab
  - Practitioner-collected sample

HPV detection in self- and practitioner-collected samples using different HPV assays.

HPV assay type	Oncogenic HPV type	Self-collected			Practitioner-collected			P-value
		n/N	%	(95% CI)	n/N	%	(95% CI)	
cobas 4800	HPV 16	40/293	13.7	(9.9-18.1)	33/299	11.0	(7.7-15.1)	0.333
	HPV 18	9/293	3.1	(1.4-5.8)	5/299	1.7	(0.5-3.9)	0.263
	Other HPV (non-16/18)	180/295	61.0	(55.2-66.6)	148/299	49.5	(43.7-55.3)	0.005
	Any HPV[¥]	195/295	66.1	(60.4-71.5)	162/299	54.2	(48.3-59.9)	0.003
cobas	HPV 16	41/285	14.4	(10.5-19.0)	41/302	13.6	(9.9-18.0)	0.777
	HPV 18	15/280	5.4	(3.0-8.7)	10/302	3.3	(1.6-6.0)	0.224
	Other HPV (non-16/18)	173/292	59.2	(53.4-64.9)	151/302	50.0	(44.2-55.8)	0.024
	Any HPV[¥]	194/293	66.2	(60.5-71.6)	170/302	56.3	(50.5-62.0)	0.013
Onclarity	HPV 16	26/299	8.7	(5.8-12.5)	24/299	8.0	(5.2-11.7)	0.768
	HPV 18	6/299	2.0	(0.7-4.3)	4/299	1.3	(0.4-3.4)	0.524
	Other HPV (non-16/18)	149/300	49.7	(43.9-55.5)	129/299	43.1	(37.5-49.0)	0.110
	Any HPV[¥]	162/300	54.0	(48.2-59.7)	141/299	47.2	(41.4-53.0)	0.094
Xpert[βφ]	HPV 16	29/291	10.0	(6.8-14.0)	30/302	9.9	(6.8-13.9)	0.990
	HPV 18	21/291	7.2	(4.5-10.8)	18/302	6.0	(3.6-9.3)	0.537
	Other HPV (non-16/18)	148/291	50.9	(45.0-56.7)	125/302	41.4	(35.8-47.2)	0.021
	Any HPV[¥]	172/291	59.1	(53.2-64.8)	149/302	49.3	(43.6-55.1)	0.017
Anyplex II[β]	HPV 16	32/292	11.0	(7.6-15.1)	33/302	10.9	(7.6-15.0)	0.990
	HPV 18	9/292	3.1	(1.4-5.8)	7/302	2.3	(0.9-4.7)	0.565
	Other HPV (non-16/18)	171/296	57.8	(51.9-63.5)	163/302	54.0	(48.2-59.7)	0.350
	Any HPV[¥]	186/296	62.8	(57.1-68.4)	177/302	58.6	(52.8-64.2)	0.290
Abbott[μ]	HPV 16	26/295	8.8	(5.8-12.6)	26/299	8.7	(5.8-12.5)	0.960
	HPV 18	6/295	2.0	(0.7-4.4)	5/299	1.7	(0.5-3.9)	0.744
	Other HPV (non-16/18)	145/296	49.0	(43.2-54.8)	137/299	45.8	(40.1-51.7)	0.439
	Any HPV[¥]	162/296	54.7	(48.9-60.5)	151/299	50.5	(44.7-56.3)	0.302

Self-collection for HPV-based cervical screening shows good concordance and relative sensitivity when compared to practitioner collected samples across assays used in the Australian NCSP.

SELF-COLLECTED SAMPLES



ORIGINAL RESEARCH ARTICLE: CERVIX AND HPV

Performance of Xpert HPV on Self-collected Vaginal Samples for Cervical Cancer Screening Among Women in South Africa

Rakiya Saidu, MD, MPH,[1,2] Louise Kuhn, PhD,[3,4] Ana Tergas, MD,[4,5] Rosalind Boa, MD,[1] Jennifer Moodley, MD, PhD,[2,6] Cecilia Svanholm-Barrie, PhD,[7] David Persing, MD, PhD,[8] Scott Campbel

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Field Evaluation of Xpert HPV Point-of-Care Test for Detection of Human Papillomavirus Infection by Use of Self-Collected Vaginal and Clinician-Collected Cervical Specimens

P. Toliman,[a] S. G. Badman,[b] J. Gabuz:
C. Ryan,[a] L. M. Vallely,[a,b] A. Kelly-Har
Papua New Guinea Institute of Medical Researc
Hospital, Goroka, Papua New Guinea[a]; Mt. Hage
Australia[a]; School of Public Health and Commu
Health Sciences, University of Papua New Guin
Victoria, Australia[b]; Department of Obstetrics ar

REVIEW

Journal of Virus Eradication 2019; 5 (Supplement 1): 10–11

The feasibility and acceptability of self-sampling and HPV testing using Cepheid Xpert® HPV in a busy primary care facility

YL Woo

Faculty of Medicine, University of Malaysia on beh

Abstract

Malaysia's approach to reducing the burden of HPV-related disease has c
screening with Pap smears. While the vaccination programme has been
been less successful. In an effort to improve screening uptake, the RO
screening, with improved quality and lower total cost.

Papillomavirus Research 6 (2018) 70–76

Contents lists available at ScienceDirect

Papillomavirus Research

journal homepage: www.elsevier.com/locate/pvr

Performance of clinical screening algorithms comprising point-of-care HPV-DNA testing using self-collected vaginal specimens, and visual inspection of the cervix with acetic acid, for the detection of underlying high-grade squamous intraepithelial lesions in Papua New Guinea

Pamela J. Toliman[a,b], John M. Kaldor[b], Steven G. Badman[b], Josephine Gabuzzi[a], Selina Silim[a], Antonia Kumbia[c], Benny Kombuk[d], Zure Kombati[d], Gloria Munnull[a], Rebecca Guy[b], Lisa M. Vallely[b], Angela Kelly-Hanku[a], Handan Wand[b], Claire Ryan[e], Grace Tan[f], Julia Brotherton[f], Marion Saville[f], Glen D.L. Mola[g], Suzanne M. Garland[h,i], Sepehr N. Tabrizi[h,i], Andrew J. Vallely[a,*]

HAWKE'S BAY / TAIRAWHITI STUDY

(DAVID HAWKES, MELBOURNE AND JANE MACDONALD, WELLINGTON)

- Compares Xpert POC self-collection vs laboratory-based testing over a period of 2 years using different cohorts of women
- Aim is to explore the difference in pathway to colposcopy
  - Two pathways in rural settings in Wairoa (HB) and rural clinics in Tairawhiti
  - Time from positive result to colposcopy will be measured

ALL WĀHINE HAVE HPV SELF-TEST

Intervention	Control
POCResults in 1 hour 	Test swab to off-site lab
Immediate on-site result to patient with information and support HPV-negative information given with follow-up screening times	Results to GP/Nurse
Immediate referral date for colposcopy if HPV-positive	Patient notified by text or phone HPV-negative information given with follow-up screening times
Colposcopy	Letter/phone/text to patient if HPV-positive Letter of referral to gynaecologist Outpatient appointment generated and sent to patient
	Colposcopy

- Aim: to implement a community/iwi controlled cervical cancer prevention POC pathway to improve timely access to screening, diagnosis and treatment and to explore the acceptability and feasibility of this pathway to overcome the barriers for rural Māori communities.

HPV ASSAY VALIDATION CRITERIA

Int. J. Cancer: **124,** 516–520 (2009)

FAST TRACK

Guidelines for human papillomavirus DNA test requirements for primary cervical cancer screening in women 30 years and older

Chris J.L.M. Meijer[1]*, Johannes Berkhof[2], Philip E. Castle[3], Albertus T. Hesselink[1], Eduardo L. Franco[4], Guglielmo Ronco[5], Marc Arbyn[6,7], F. Xavier Bosch[8], Jack Cuzick[9], Joakim Dillner[10], Daniëlle A.M. Heideman[1] and Peter J.F. Snijders[1]

- A candidate test should have a clinical sensitivity for ≥CIN2 not less than 90% of the clinical sensitivity of a standard comparator test (HC2 or GP5+/6+ PCR EIA) in women of >30 years of age.
- A candidate test should have a clinical specificity for ≥CIN2 not less than 98% of the clinical specificity of a standard comparator test (HC2 or GP5+/6+ PCR EIA) in women of >30 years of age.

TABLE 4 Clinical performance of Xpert (Cepheid), cobas (Roche), and hc2 (Qiagen) for the detection of CIN2 or CIN2+ and CIN3 or CIN3+[a]

	Xpert		cobas		hc2	
Endpoint and parameter	Value	95% CI	Value	95% CI	Value	95% CI
CIN2+						
Sensitivity	90.8%	84.7–95.0%	90.8%	84.7–95.0%	81.6%	74.2–87.6%
Specificity	42.6%	38.5–46.9%	39.6%	35.5–43.8%	47.7%	43.4–51.9%
Positive predictive value	28.6%	24.5–33.1%	27.6%	23.6–31.9%	28.3%	24.0–33.0%
Negative predictive value	94.8%	91.3–97.2%	94.4%	90.6–97.0%	91.1%	87.2–94.1%
Odds ratio	7.32	4.07–13.2	6.45	3.58–11.6	4.03	2.56–6.34
Positive-likelihood ratio	1.58	1.45–1.73	1.50	1.38–1.64	1.56	1.39–1.74
Negative-likelihood ratio	0.216	0.128–0.216	0.233	0.137–0.395	0.387	0.270–0.553

Einstein et al., Journal of Clinical Microbiology 2014, 52:6, p.2089-2095.

HPV ASSAY VALIDATION CRITERIA

Evaluated index HPV assay	Study	Index assay		Comparator assay			Index/comparator assay		Non-inferiority test[a]		Validation level[c]
		Absolute		Comparator HPV assay	Absolute		Relative				
		Sensitivity	Specificity		Sensitivity	Specificity	Sensitivity	Specificity	p_{sens}	p_{spec}	
Standard comparator HPV tests											
GP5+/6+ EIA	Meijer, 2009 [13]	98.7%	96.0%	HC2	98.7%	94.1%	1.00	1.02	0.0037	<0.0001	⊕⊕⊕
Evaluated index HPV tests											
PapilloCheck	Hesselink, 2010 [49]	95.8%	96.7%	GP5+/6+ EIA	96.4%	97.7%	0.99	0.99	<0.0001	0.0072	⊕⊕⊕
	Heard, 2016 [50]	96.1%	89.7%	GP5+/6+ EIA	94.1%	90.4%	1.02	0.99	0.0002	0.0970	
Abbott RT	Carozzi, 2011 [33]	96.4%	92.3%	HC2	97.6%	92.6%	0.99	1.00	0.0040	0.0087	
hrHPV test	Poljak, 2011 [34]	100.0%	93.3%	HC2	97.4%	91.8%	1.03	1.02	0.0112	0.0000	⊕⊕⊕
	Hesselink, 2013 [35]	95.6%	92.0%	GP5+/6+ EIA	98.5%	91.8%	0.97	1.00	0.0278	0.0003	
Cobas 4800	Heideman, 2011 [42]	90.0%	94.6%	HC2	91.7%	94.4%	0.98	1.00	0.0216	0.0009	⊕⊕⊕
	Lloveras, 2013 [43]	98.3%	86.2%	HC2	98.3%	85.3%	1.00	1.01	0.0093	0.0012	
	Ejegod, 2020 [20]	92.6%	91.2%	GP5+/6+ EIA	92.6%	89.2%	1.00	1.02	0.0006	<0.0001	
RIATOL	Depuydt, 2012 [60]	93.5%	95.6%	HC2	83.9%	94.4%	1.11	1.01	0.0001	<0.0001	⊕⊕
qPCR	Benoy, 2019 [61]	96.0%	89.5%	GP5+/6+ EIA	96.0%	89.7%	1.00	1.00	0.0006	0.0069	
APTIMA	Heideman, 2013 [53]	95.5%	94.5%	GP5+/6+ EIA	100.0%	93.6%	0.96	1.01	0.0394	0.0002	X
Cervista	Boers, 2014 [58]	89.0%	91.2%	HC2	93.4%	88.8%	0.95	1.03	0.0043	<0.0001	⊕
	Alameda, 2015 [59]	98.4%	85.2%	HC2	100.0%	86.4%	0.98	0.99	0.0122	0.3170[b]	
BD Onclarity	Ejegod, 2014 [38]	92.9%	87.7%	HC2	94.2%	88.8%	0.99	0.99	0.0009	0.0216	
	Cuschieri, 2015 [39]	96.7%	89.6%	HC2	98.4%	89.9%	0.98	1.00	0.0245	0.0155	⊕⊕⊕
	Ejegod, 2016 [40]	96.1%	89.7%	GP5+/6+ PCR	94.1%	90.4%	1.02	0.99	0.0002	0.0970	
	Bonde, 2019 [41]	92.6%	92.6%	GP5+/6+ EIA	92.6%	89.6%	1.00	1.04	<0.0001	<0.0001	
HPV-Risk assay	Hesselink, 2014 [46]	97.1%	94.3%	GP5+/6+ EIA	97.1%	94.1%	1.00	1.00	0.0056	0.0003	
	Polman, 2017 [47]	93.7%	91.8%	HC2	96.1%	89.9%	0.98	1.02	<0.001	<0.001	⊕⊕⊕
	Heideman, 2019 [48]	93.4%	92.6%	GP5+/6+ EIA	92.6%	89.%	1.01	0.99	0.0006	<0.0001	
Anyplex II HPV HR	Hesselink, 2016 [36]	98.3%	93.6%	GP5+/6+ PCR	98.3%	94.1%	1.00	0.99	0.0052	0.0232	
HPV HR	Jung, 2016 [37]	92.5%	81.7%	HC2	87.5%	81.8%	1.06	1.00	0.0067	0.0354	⊕⊕⊕
	Ostrbenk, 2018 [19]	96.9%	94.1%	HC2	95.9%	92.7%	1.01	1.01	0.001	<0.0001	
Xpert HPV	Cuschieri, 2016 [102]	94.1%	90.3%	GP5+/6+ PCR	94.1%	90.3%	1.00	1.00	0.0171	0.0269	⊕⊕
INNO-LIPA	Xu, 2018 [64]	96.9%	87.9%	HC2	95.9%	97.0%	1.01	0.95	0.0002	0.9998	⊖
Linear Array[e]	Xu, 2018 [62]	98.0%	94.3%	HC2	95.9%	92.7%	1.02	1.02	0.0076	<0.0001	⊕⊕
EUROArray	Viti, 2018 [57]	93.7%	89.9%	HC2	96.1%	90.1%	0.98	1.00	0.0076	0.0070	⊕[d]
Cobas 6800	Saville, 2019 [54]	98.3%	88.4%	Cobas 4800	100.0%	89.4%	0.98	0.98	0.0157	.0442	⊕⊕⊕
	Frayle, 2019 [45]	98.3%	92.1%	Cobas 4800	100.0%	92.8%	0.98	0.99	0.0157	0.0056	
Alinity	Ostrbenk, 2020 [52]	100%	92.4%	HC2	95.6%	91.9%	1.05	1.01	0.0006	<0.0001	⊕⊕
HBRT-H14	Xu, 2020 [63]	93.9%	93.7%	GP5+/6+ PCR	95.9%	92.9%	0.98	1.01	0.0159	<0.0001	⊕[d]
CLART	Ejegod, 2020 [56]	92.6%	88.8%	mod GP5+/6+	90.1%	89.1%	1.03	1.00	0.0021	0.0083	⊕
		97.5%	93.8%	PCR LMNX	100%	86.7%	0.98	1.08	0.0127	<0.0001	

Arbyn et al., Clin Microbiol Infect. 2021 Aug;27(8):1083-1095

HPV ASSAY VALIDATION CRITERIA

Currently, there are 254 distinct commercial HPV tests and 425 assay variants available on the global market. The large majority of them lack any analytical or clinical evaluation published in the peer-reviewed literature and more than 90% have not undergone regulatory evaluation or have not been evaluated following a stringent clinical validation protocol.

ELSEVIER

Journal of Clinical Virology
Volume 76, Supplement 1, March 2016, Pages S14-S21

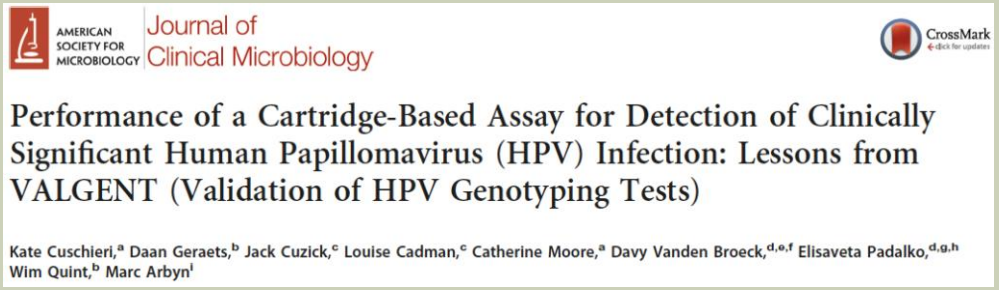
Review

VALGENT: A protocol for clinical validation of human papillomavirus assays

Marc Arbyn [a], Christophe Depuydt [b], Ina Benoy [b], Johannes Bogers [b], Kate Cuschieri [c], Markus Schmitt [d], Michael Pawlita [d], Daan Geraets [e], Isabelle Heard [f], Tarik Gheit [g], Massimo Tommasino [g], Mario Poljak [h], Jesper Bonde [i], Wim Quint [e]

- A candidate test should display intra-laboratory reproducibility and inter-laboratory agreement with a lower confidence bound not less than 87%.

HPV ASSAY VALIDATION CRITERIA



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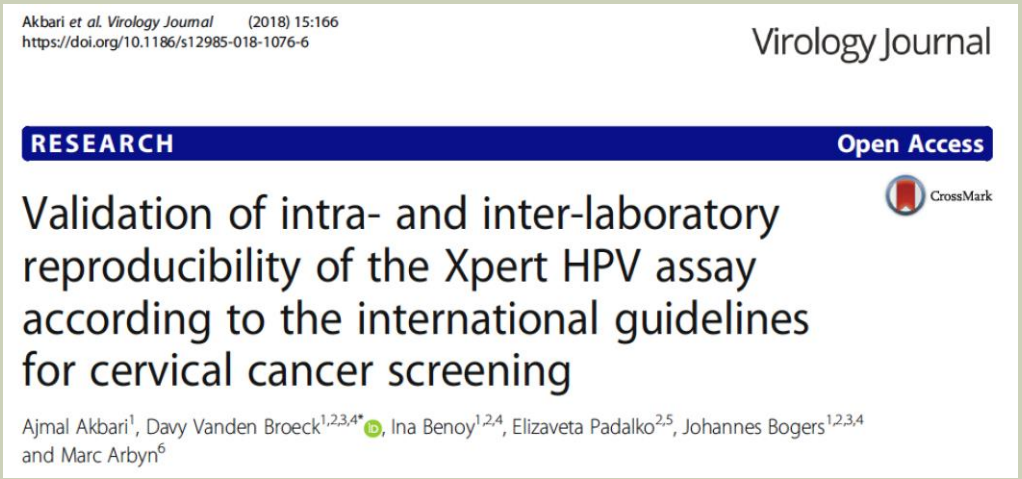
CrossMark

Performance of a Cartridge-Based Assay for Detection of Clinically Significant Human Papillomavirus (HPV) Infection: Lessons from VALGENT (Validation of HPV Genotyping Tests)

Kate Cuschieri,[a] Daan Geraets,[b] Jack Cuzick,[c] Louise Cadman,[c] Catherine Moore,[a] Davy Vanden Broeck,[d,e,f] Elisaveta Padalko,[d,g,h] Wim Quint,[b] Marc Arbyn[i]

September 2016 Volume 54 Number 9 Journal of Clinical Microbiology jcm.asm.org 2337

-> Conclusion: The clinical performance and reproducibility of the Xpert HPV are comparable to those of well-established HPV assays and fulfil the criteria for use in primary cervical cancer screening.



Akbari *et al. Virology Journal* (2018) 15:166
https://doi.org/10.1186/s12985-018-1076-6

Virology Journal

RESEARCH Open Access

CrossMark

Validation of intra- and inter-laboratory reproducibility of the Xpert HPV assay according to the international guidelines for cervical cancer screening

Ajmal Akbari[1], Davy Vanden Broeck[1,2,3,4*], Ina Benoy[1,2,4], Elizaveta Padalko[2,5], Johannes Bogers[1,2,3,4] and Marc Arbyn[6]

-> Conclusion: The Xpert HPV assay fulfils the HPV test reproducibility criterion requirement for use in cervical cancer screening.

VALIDATED HPV ASSAYS

Currently, there are 11 commercial hrHPV DNA (*) assays that are completely validated to be used for cervical cancer diagnostics based on primary HPV testing:

1) Qiagen HC2
2) GP5+/6+ PCR-EIA
3) Abbott RealTime
4) Abbott Alinity
5) Seegene Anyplex HR
6) Roche Cobas 4800
7) {Roche Cobas 6800}
8) Greiner PapilloCheck
9) BD Onclarity
10) Self-Screen BV HPV-Risk
11) Cepheid Xpert HPV

Systematic Review

2020 list of human papillomavirus assays suitable for primary cervical cancer screening

Marc Arbyn [1, 2, *], Marie Simon [3], Eliana Peeters [1], Lan Xu [1, 4], Chris J.L.M. Meijer [5], Johannes Berkhof [6], Kate Cuschieri [7], Jesper Bonde [8], Anja Ostrbenk Vanlencak [9], Fang-Hui Zhao [10], Remila Rezhake [1, 10, 11], Murat Gultekin [12], Joakim Dillner [13], Silvia de Sanjosé [14], Karen Canfell [15, 16], Peter Hillemanns [17], Maribel Almonte [18], Nicolas Wentzensen [19, †], Mario Poljak [9, †]

Clin Microbiol Infect. 2021 Aug; 27(8).

(*): The APTIMA HPV Assay targeting E6/E7 mRNA of hrHPV was fully validated in one formal validation study and showed slightly lower pooled sensitivity but higher specificity than the standard comparator tests in seven screening studies. However, the current international validation criteria relate to DNA assays. The additional requirement for longitudinal performance data required for non-DNA based HPV assays was not assessed in this review.

SUMMARY

- GeneXpert HPV test is an easy to use POCT with fast TAT, and comparable sensitivity and specificity to established testing platforms
- It can be used in remote settings and requires minimal training, however test kits are expensive and the cost is far beyond the budget available for screening in many countries
- It's the only fully validated POCT for HPV testing
- It shows good concordance for self-collected specimens
- It fulfils all criteria for use in primary cervical cancer screening
- It is a test for low-throughput fast TAT situations, but may not be that useful in a molecular screening lab with high sample numbers

ACKNOWLEDGEMENTS

- Dr Margaret Sage, NCPTS
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