

DNA versus RNA based methods for HPV testing in Norway. Evaluation of Hybrid Capture II and PreTect HPV-Proofer, a validation study.



Lie AK¹, Risberg B¹, Sandstad B², Delabie J¹, Rimala R³, Hagen B⁴, Onsrud M⁵, Thoresen S⁶

Department of Pathology (1) and Clinical Research (2), The Norwegian Radium Hospital, Oslo, Norway (1), Laboratory of Pathology, Oslo (3), Gynecologic Department, St. Olavs University Hospital, Trondheim (4), Gynecologic Department, Ullevål University Hospital, Oslo (5), Institute of Population-based Cancer Research, Oslo, Norway

Background

The specificity as well as the positive predictive value of HPV DNA testing in screening is low, because most high-risk HPV infections are transient. Expression of E6/E7 oncogenes are required for the development and maintenance of a malignant phenotype. HPV RNA testing can be used for risk evaluation and may have a superior positive predictive value in screening.

Aims

To validate two commercially available assays for HPV testing in order to investigate the prevalence of high-risk HPV infections in women with negative and positive cytology. To evaluate the outcome of DNA-based and RNA-based testing compared to cytology and histology.

Material and Methods

The study population was selected from outpatient departments and gynecologists in private practice. Included in this study were 628 women with median age 40 years (range, 19-85). A conventional Pap smear was taken first, and the remaining material was transferred to a PreservCyt™ vial (Cytoc Corporation). Testing for high-risk HPV DNA (type 16, 18, 31, 33, 35, 39, 45, 51, 52, 56, 58, 59, and 68) was performed with the Hybrid Capture II assay (Digene Corporation) and individual identification of E6/E7 mRNA transcripts from HPV 16, 18, 31, 33, and 45 with the Pre Tect HPV-Proofer assay (NorChip AS), a real-time NASBA technique. Biopsies were taken when HPV test was positive or cytology revealed HSIL. Histology was regarded as the “gold standard”.

Results

Concordance between cytology and histology were found in 53% of cases. High-grade histology (CIN 2+) was detected in 61% of the women with benign or low-grade cytology. Kappa value was 0.31. Different outcomes of the two tests were present in 17% (109/628) of cases (table 1). The HPV results related to cytology and histology diagnoses are shown in figure 1 and 2. Both HPV tests showed significant association with grade of the lesions ($p < 0.001$). The DNA test was more often positive in benign and low-grade lesions. The DNA test revealed higher sensitivity but lower specificity compared to the RNA test (Table 2).

Conclusion

The RNA test revealed a higher prognostic value and higher specificity than the DNA test. Larger scale studies are necessary to evaluate the predictive values of these tests in the Norwegian screening program. National monitoring of HPV testing should be obligate and incorporate cytology/histology results.

Table 1: HPV testing related to histology in cases with different outcome of the two HPV tests.

HPV test	Histology		
	< CIN 2	≥ CIN 2	Total
DNA +/ RNA-	40	59	99
RNA +/ DNA-	1	9	10
Total	41	68	109

Table 2: The performance of the HPV testing for detection of histological confirmed CIN 2+

	Sensitivity (%)		Specificity (%)	
	DNA	RNA	DNA	RNA
Age				
< 30 years (n=102)	98	82	20	70
≥ 30 years (n=281)	93	76	40	81
Cytology*				
normal (n=105)	89	62	79	87
low-grade (n=73)	96	72	22	72
high-grade (182)	96	83	67	50

* Twenty-three PAP smears with an unsatisfactory diagnosis were excluded from this analysis.

Fig 1: Cytological diagnoses related to HPV test.

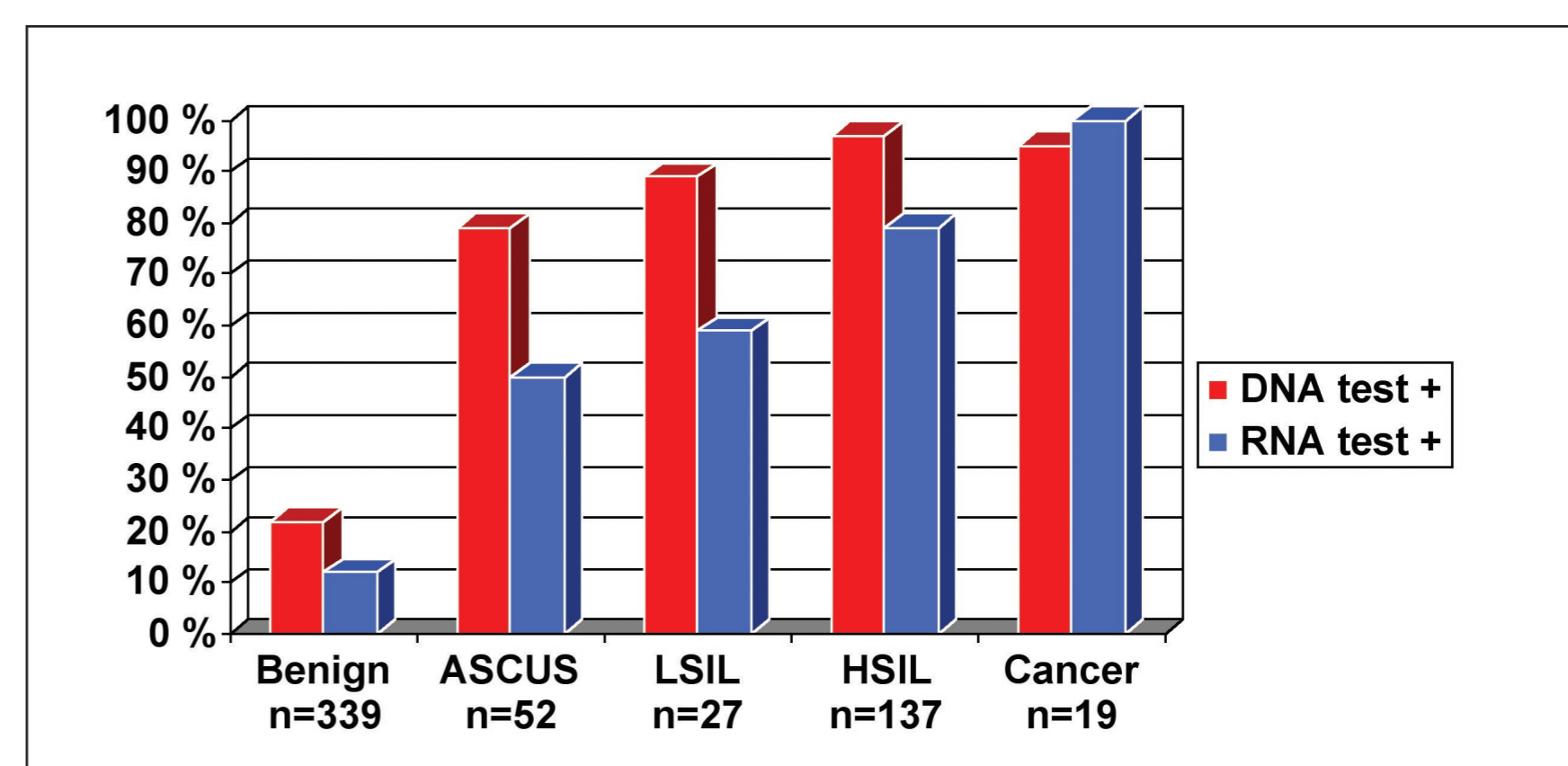


Fig 2: Histological diagnoses of squamous lesions related to HPV test.

