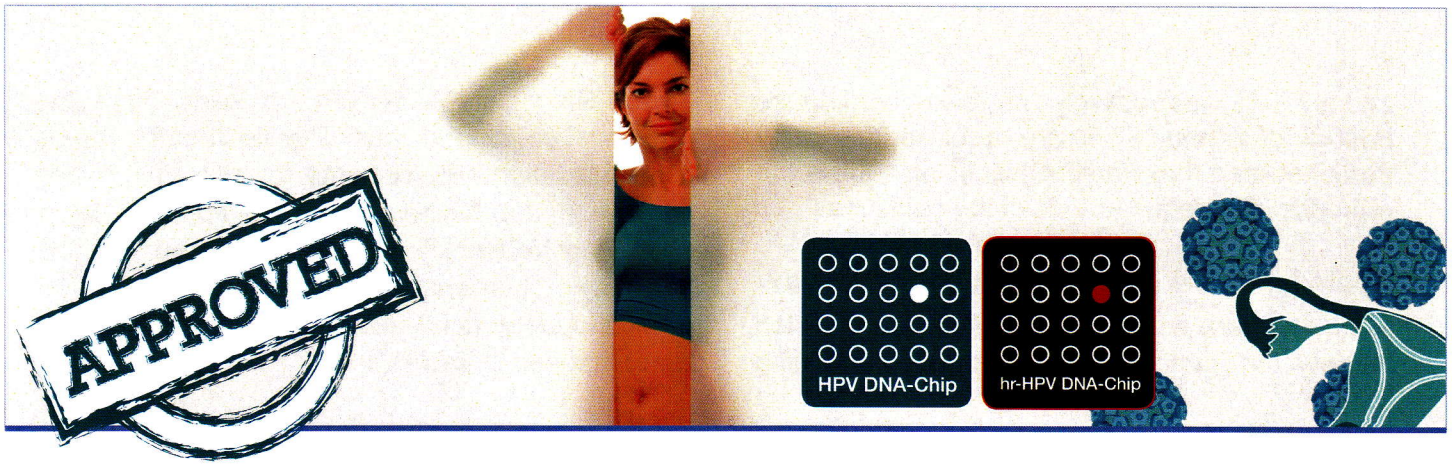

PapillоCheck® - a clinically validated HPV genotyping assay

Greiner Bio-One HPV assay fulfils international guidelines for HPV DNA test requirements

Testing for high-risk types of human papillomaviruses (HPV) has become an important part of cervical cancer screening, management and treatment. Only HPV tests with a documented clinical performance as well as a proved reliability, reproducibility and accuracy should be used in clinical management. A group of international experts published guidelines defining the requirements for DNA tests for primary cervical cancer screening in women 30 years and older[1]. These requirements are widely accepted and often used to rate the clinical performance of an HPV DNA test.

PapillоCheck® - an IVD for the detection and genotyping of 24 different high-risk and low-risk HPV types by DNA chip technology - proved its performance in several clinical studies. Two of them were carried out according to the international guidelines mentioned above.

Clinical Sensitivity and Specificity

In 2010 Hesselink and colleagues published a study which evaluated the clinical sensitivity and specificity of PapillоCheck® in comparison to the clinically validated GP5+/6+-PCR-enzyme immunoassay[2]. Cervical scrapings collected during the Dutch population based randomised-controlled implementation trial POBASCAM were used as samples. PapillоCheck® showed a clinical sensitivity for the detection of ≥CIN3 of 95.8 % and a clinical specificity for the detection of ≥CIN2 of 96.7 %. For calculations the 14 hrHPV types detectable by both, PapillоCheck® and the clinically validated reference test were considered. According to the guidelines, a new HPV DNA test should show at least 90 % of the clinical sensitivity and 98 % of the clinical specificity or the clinically validated reference test. Applying these critera, the non-inferiority of PapillоCheck® to the used reference test was demonstrated for both clinical key parameters. Consequently, main requirements of the guidelines were fulfiled.

Reproducibility

To complete the validation data set the reproducibility of the PapillоCheck® assay was assessed in a separate study carried out in 2012 in the Netherlands and Germany. In the study cervical specimens obtained from a cohort of 10,000 women who participated in regular cervical cytology screening in the Utrecht province of the Netherlands were used. Using a defined set of 550 pretested samples PapillоCheck® showed an intra-laboratory reproducibility and inter-laboratory agreement, with lower confidence bounds of 96.3 %

[1] Meijer et al., Int J Cancer. 2009 Feb 1;124(3):516-20. [2] Hesselink et al., J Clin Microbiol. 2010 Mar;48(3):797-801.

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and 92.1 %, respectively. As lower confidence bounds of at least 87 % are required, this study demonstrated that Papill**oCheck**® also fulfils the reproducibility requirements of the guidelines.

Approval by the Dutch Society for Pathology

Adequate data sets were submitted to the Dutch Society for Pathology (Nederlandse Vereniging voor Pathologie – NVVP). In early 2013 the society concluded that Papill**oCheck**® meets the validation requirements for use in cytology triage in the Netherlands. The approval letter for Papill**oCheck**® can be accessed via the society's homepage (http://www.pathology.nl/vakinhoudelijk/richtlijnen). The submitted data is summarised in Tables 1-4.

Table 1: Relative Clinical Sensitivity for ≥CIN2

	GP5+/6+-PCR pos	GP5+/6+-PCR neg
Papill**oCheck**® pos	108	1
Papill**oCheck**® neg	4	4

Requirements: minimum n = 60, p-value < 0,05
Results: Non-inferiority scoring test-statistic = 2,455, p-value = 0,007
Conclusion: APPROVED

Table 2: Relative Clinical Specificity for <CIN2

	GP5+/6+-PCR pos	GP5+/6+-PCR neg
Papill**oCheck**® pos	29	18
Papill**oCheck**® neg	4	1386

Requirements: minimum n = 800, p-value = 0,05
Results: Non-inferiority scoring test-statistic = 2,450, p-value = 0,007
Conclusion: APPROVED

Table 3: Intra-Laboratory Reproducibility

	Papill**oCheck**® time 1 pos	Papill**oCheck**® time 1 neg
Papill**oCheck**® time 2 pos	147	6
Papill**oCheck**® time 2 neg	7	390

Requirements: minimum n = 500, Lower confidence bound [%] > 87, Kappa > 0
Results: Lower confidence bound [%] = 96.3; Kappa = 0.941
Conclusion: APPROVED

Table 4: Inter-Laboratory Agreement

	Papill**oCheck**® Lab 1 pos	Papill**oCheck**® Lab 1 neg
Papill**oCheck**® Lab 2 pos	123	2
Papill**oCheck**® Lab 2 neg	31	394

Requirements: minimum n = 500, Lower confidence bound [%] > 87, Kappa > 0.5
Results: Lower confidence bound [%] = 92.1; Kappa = 0.842
Conclusion: APPROVED

The validation according to the international guidelines and the approval by the NVVP based on the validation results is a milestone in the performance evaluation of Papill**oCheck**®. Its clinical performance together with an extensive set of on-chip controls, e.g. internal PCR control, sample control and hybridisation control render the Papill**oCheck**® HPV genotyping test a highly reliable tool in HPV detection.

Sensitivity + **Specificity** + **Reproducibility** = **PapilloCheck®**

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