

Frickenhausen, June 21st 2012**STATEMENT Greiner Bio-One****Usage of the CYT-ALL collection medium by Alphapath in combination with the PapilloCheck® HPV genotyping Test**

The CYT-ALL collection medium by Alphapath was tested with the PapilloCheck® system comprising of the Greiner Bio-One oCheck® DNA Extraction Kit (formerly PapilloCheck® DNA Extraction Kit), the PapilloCheck® Kit and further approved equipment according the respective instructions for use. Additionally the chemagic Prepito (PerkinElmer chemagen Technologie GmbH, formerly chemagen Biopolymer-Technologie AG) was tested as an alternative DNA extraction system.

The main objectives were to investigate (I) the stability of fixed cells in the CYT-ALL collection medium, (II) PapilloCheck® analyses using fixed reference cells at different concentrations and (III) the analysis of clinical cervical samples collected in CYT-ALL with the PapilloCheck® system. For comparison, all tests for (I) an (II) were performed in parallel using the validated transport medium of the PapilloCheck® Collection Kit and the CYT-ALL medium, tests for (II) an (III) were performed in parallel using the oCheck® DNA Extraction Kit (Greiner Bio-One) and the chemagic Prepito (PerkinElmer chemagen Technologie GmbH).

(I) Stability: Testing replicates of samples with different amounts of fixed HPV-positive reference cells (500 and 10^4 per extraction) at different time points (up to 30 days) and in combination with a transport simulation, did not result in relevant differences between the two media for HPV-specific, sample and PCR control signals.

(II) Results for analytical samples: Replicates of samples with fixed HPV-positive reference cells at concentrations of 0.5×10^2 to 10^5 cells per extraction were analyzed. All tests were valid and no relevant differences between the two media were detected for HPV-, sample and PCR control signals.

(III) Results for clinical samples: Applying the oCheck® DNA Extraction Kit, analyzing altogether 155 cervical samples collected in the CYT-ALL transport medium lead to 62 HPV-positive and 93 HPV-negative results. The on-chip quality controls (Sample- and PCR-Control) performed adequately, with no invalid results.

Using the chemagic Prepito as an alternative DNA extraction method together with a newly established protocol based on the combination of the Prepito Pathogen NA Kit (art. No. 2022, PerkinElmer chemagen Technologie GmbH) and the external lysis steps of the dedicated Prepito Pathogen NA Extension Kit (art. No. 2023, PerkinElmer chemagen Technologie GmbH), resulted in a positive and negative concordance of both DNA extraction methods of 96,8 % and 94,6 %, respectively.

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Greiner Bio-One GmbH, Maybachstr. 2, D-72636 Frickenhausen

Conclusion: The CYT-ALL collection medium by Alphapath, effective June 2012, is comparable with the validated transport medium of the PapilloCheck® Collection Kit (Greiner Bio-One) regarding stability and the analysis of defined analytical samples. Clinical samples collected in CYT-ALL medium are compatible with the PapilloCheck® system.

In comparison to the validated manual DNA extraction method, automated DNA isolation using the chemagic Prepito has proved to be a reliable and efficient option for cervical specimens stored in CYT-ALL medium that will subsequently be analyzed using PapilloCheck® when using the above described protocol.

Sincerely,
Greiner Bio-One



Dr. Jörg Stappert
Head of Biochip Group