
P-16; FLOW CYTOMETRY IN RAPID SCREENING FOR E6/E7 mRNA DETECTION

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Background: For the development of cervical cancer, apart from the simple presence of HPV, the viral oncoproteins E6/E7 have to be over-produced.

Objectives: The aim of this study was to evaluate two different techniques for E6/E7 HPV mRNA detection.

Materials and Methods: 44 cervical samples in PreserveCyt vials were typed for HPV DNA with ClinicalArrays HPV (Genomica, Spain). Flow cytometric evaluation of E6/E7 mRNA of high risk HPV types was performed with HPV OncoTect (Invirion Diagnostics, USA). Endocervical cells were gated according to their FSC-SSC parameters and the cut-off was set at 1,5%. NucliSENS EasyQ® HPV (BioMerieux, France) was used to amplify E6/E7 mRNA from HPV types 16, 18, 31, 33 and 45.

Results: 7 samples were negative with both FC and NASBA. 2 samples were negative with FC, but positive with NASBA. 11 samples (5 HgSIL, 3 SCC, 3 AdenoCa) were positive for both methods only if the gating for FC was set according to CAM5.2 and CD16 expression. Another 17 samples (1 WNL, 4 LgSIL, 11 HgSIL, 1 SCC) were positive with both methods. 7 more samples (6 LgSIL, 1 SCC) were only positive in FC, out of which 5 harbored coinfection of HPV-16 with at least another high risk type. The remaining 2 were single HPV-16 positive samples.

Conclusions: Total agreement of the two methods was at 93,18%. Flow cytometry proved to be more efficient a screening method, since all high risk types can be detected. On the other hand, due to the morphological changes in size and granularity of cells in HgSILs and carcinomas, a three color staining procedure is needed. To conclude with, the two methods seem to be complementary since FC can be easily used as a screening method, while NASBA as a typing one.