Abstract 7746

Human papilloma virus 16 viral load quantification using droplet digital PCR and correlation with cervical lesion

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Background: Human Papillomavirus (HPV) infection is known to be associated to cervical cancer development. Presently, many countries are changing their screening policy switching to HPV testing as primary screening as suggested by European guidelines. Even if cytology is the preferred triage method, HPV viral load measurement has been suggested to be a predictor of cervical lesion progression. The aim of this study was to evaluate the correlation between HPV16 viral load and cervical lesion grade using droplet digital PCR (ddPCR) among women referred to colposcopy.

Materials/methods: Cervical samples were collected from women attending the Colposcopy Clinic, San Gerardo Hospital (Monza, Italy) for abnormal cervical cytology. All samples were extracted using NucliSENS easyMAG and HPV detection was carried out using AnyplexII HPV28. All HPV16 positive samples were tested using ddPCR and read in a QX Droplet reader (Bio-Rad). PCR was set up in duplex, combining HPV16 with the CCR5 gene to obtain cell quantification. Viral copy number/cells were calculated as: HPV16 copies/(CCR5 copies/2).

Results: Currently, 47 HPV16 positive samples collected from women with different grade of lesion have been tested. A higher mean viral load/cells value was observed among women with a high-grade lesion (HSIL) compared to women with lower grade lesion (36.3 vs 21.9 copies/cells). The same result was obtained comparing viral load quantification with biopsy (22.1 vs 6.0 copies/cells, CIN2+ vs <CIN2). Moreover, 4 women showing HPV16 persistent cervical infection were followed-up at different time points to evaluate viral load changes with cervical cytology. Preliminary results show a possible correlation between HPV16 viral load and progression/regression of the cervical lesion (Figure 1).

Conclusions: Preliminary data suggest that ddPCR method represents a promising tool for HPV viral load estimation. Viral load measurement could represent a useful biomarker in the follow up of HPV-positive women although larger longitudinal clinical studies are required to assess its clinical value.
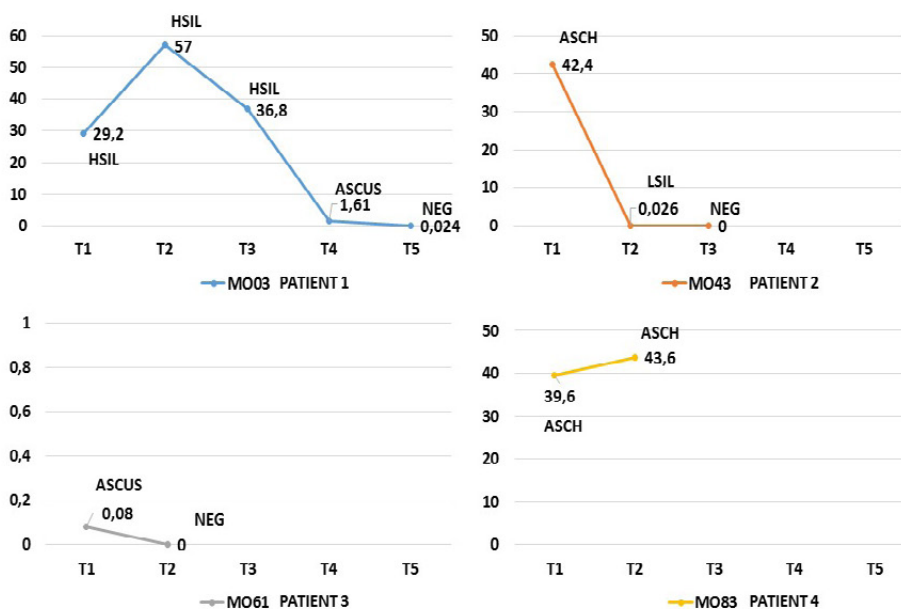
Figure 1. Correlation between HPV viral load and cytology of 4 women followed-up at different time points. HSIL: High grade squamous intraepithelial lesion; LSIL: Low-grade squamous intraepithelial lesion; ASCH: Atypical squamous cells cannot exclude HSIL; ASCUS: Atypical squamous cells of undetermined significance: NEG: Negative for cytological lesions

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