Online resources

Molecular diagnosis and therapy

Ultra-Sensitive Detection of Circulating Cell-Free Human Papilloma Virus DNA Alvida Qvick^{1,*}, Elin Andersson², Anna Oldaeus Almerén³, Max Waenerlund^{4,5}, Bianca Stenmark¹, Christina Karlsson⁶, Mats G Karlsson¹, and Gisela Helenius⁷

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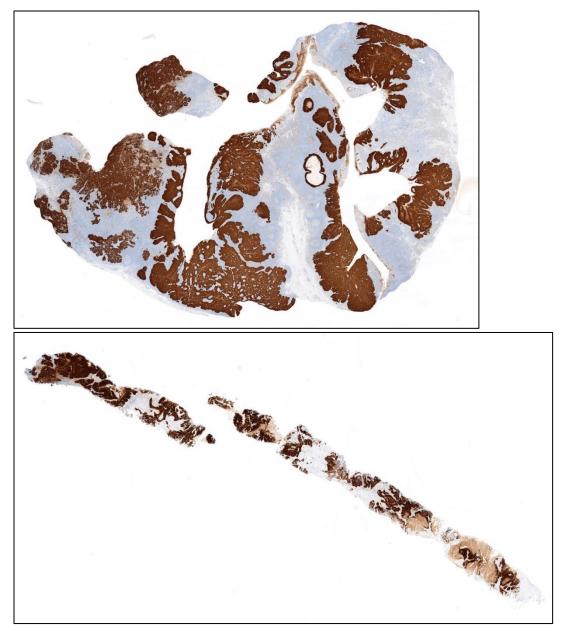
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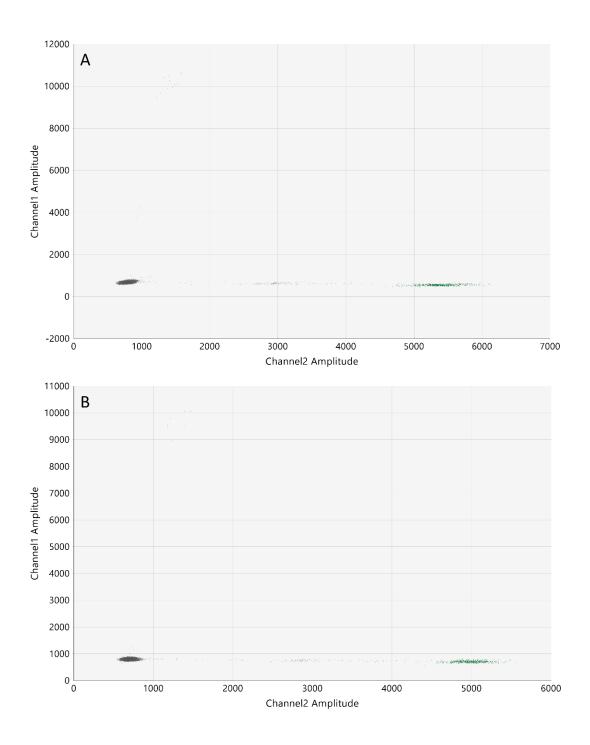
⁶ School of Health Sciences, Örebro University, Örebro, Sweden

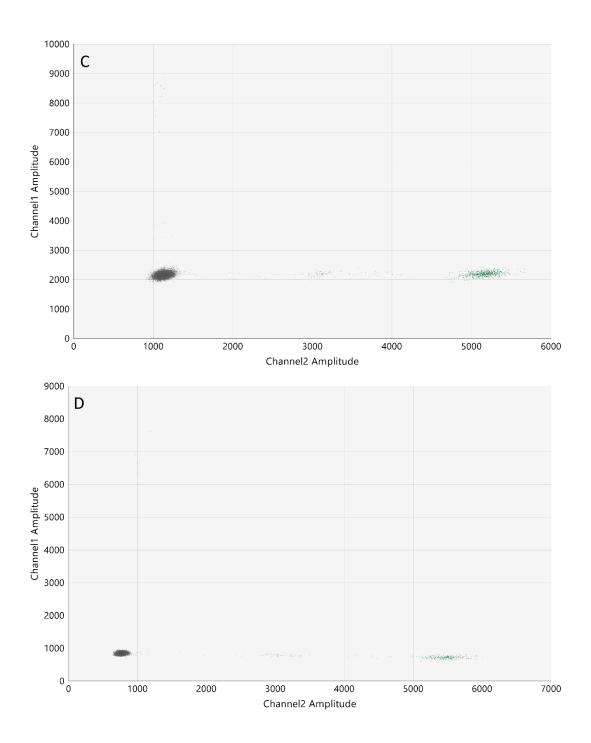
⁷ ATMP Center, Skåne University Hospital, Lund, Sweden

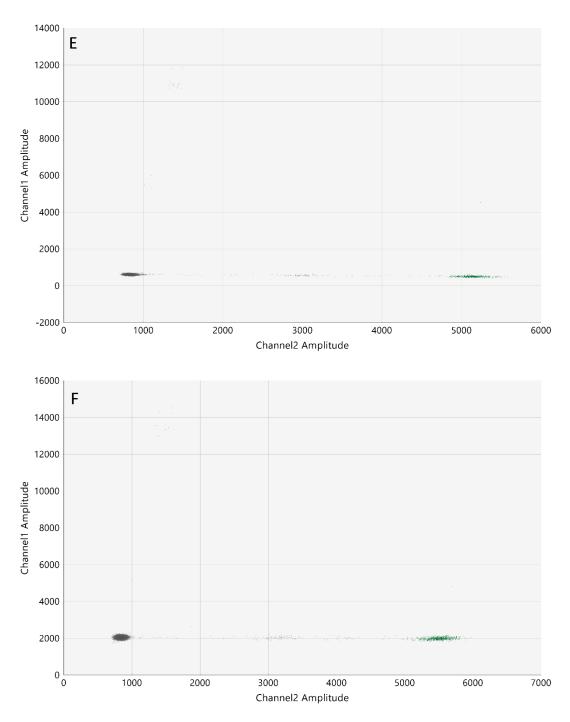
* Correspondence: <u>alvida.qvick@regionorebrolan.com</u>



Online resource 1 p16 Immunohistochemistry data. Two p16+ representative cases are shown.



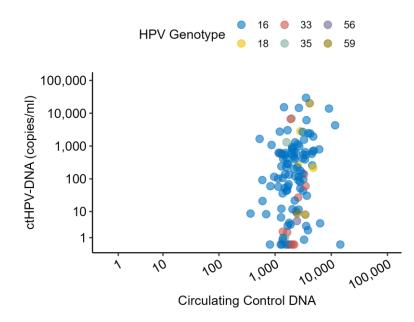




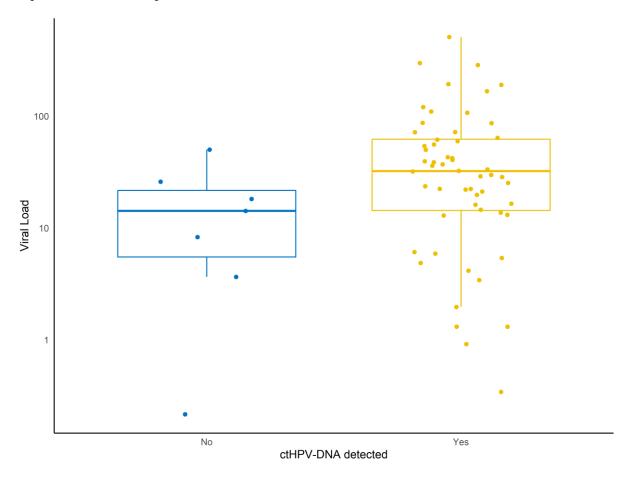
Online resource 2 Raw data from ctHPV-DNA assays. Representative experiments from A) HPV16, B) HPV18, C) HPV33, D) HPV35, E) HPV56, and F) HPV59 are shown. Channel 1 shows ctHPV-DNA+ droplets and Channel 2 shows droplets positive for the human control sequence on 12q24.32.

	HPV16		HPV18		HPV33		HPV35		HPV56		HPV59	
Time points	21		6		6		6		3		5	
(n)												
Duration	14		23		10		14		7		14	
(months)												
Target	HPV	12q										
HPV Low												
Mean	0.922	47.395	0.399	36.733	0.557	85.033	0.151	90.217	0.530	32.367	0.629	44.760
(copies/µl)												
SD	0.163	3.262	0.109	5.056	0.092	3.054	0.072	4.140	0.098	1.319	0.083	1.266
HPV High												
Mean	1.620	55.000	1.938	58.15	1.702	78.733	0.654	90.183	2.26	33.067	2.024	43.120
(copies/µl)												
SD	0.233	4.908	0.425	11.701	0.167	5.025	0.144	3.536	0.221	0.499	0.143	1.468

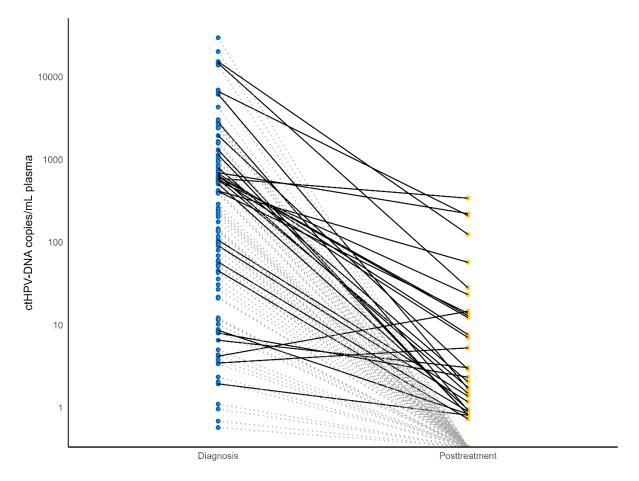
Online resource 3 Assay variation over time. Each assay was tested over time using one low HPV+ control and one higher HPV+ control. Each reaction included a human control sequence located on 12q24.32.



Online resource 5 ctHPV-DNA and the total cfDNA concentration as measured by the human control sequence located on 12q24.32.



Online resource 6 Viral load in ctHPV-DNA positive (n=56) and negative (n=7) cases.



Online resource 7 ctHPV-DNA levels at follow up (n=113). Diagnosis (blue) and posttreatment (yellow) are shown. Dotted grey lines represent samples with undetectable posttreatment ctHPV-DNA (n=82).