AML

La charge virale et le génotypage ont-ils un intérêt dans le suivi d'une femme HPV positive?

Depuydt Christophe

Pas de conflits d'intérêts

Pathologie cervicale:

La charge virale et le génotypage ont-ils un intérêt dans le suivi d'une femme HPV positive?

- Introduction charge virale HPV type spécifique
- L'experience AML (2000-2023): changements de la charge virale prédisent la progression vers le cancer du col (4 types: infectieux, sériel transitoire, régression, progression)
- Etude Medipath NICE-CIMIEZ (2014-2023) Suivit avec typage HPV et charge virale sur cytologie en milieu liquide
- Atelier et discussion autours de cas cliniques

AML

Méthodes

- Frottis liquide (BD-SurePath 2000-2014; ThinPrep 2014-2023)
- Dépistage du cancer du col en Flandre > 2 250 000 frottis depuis 2000
- PCR à temps réel quantitative spécifique pour chaque type d'HPV:
  6, 11, 16, 18, 31, 33, 35, 39, 45, 51, 52, 53, 56, 58, 59, 66, 67 et 68
- Mesure en série de frottis consécutif permet de calculer le statut (infectieux et ou clonal) des types d'HPV présent (Verhelst et al 2016)
- Lecture automatique guidé avec connaissance d'HPV (18 types)
- Les résultats cytologiques ont été classifié en utilisant le système Bethesda 2001 (NILM, ASC-US, ASC-H, L-SIL, H-SIL) et avec connaissance des résultats HPV
- Au cours du suivi, les femmes ont eu une colposcopie avec biopsie

Real time quantitative HPV PCR on liquid based cytology

- TaqMan™ based assays

  Typing and quantification of HPV types:

  6E6,11E6,16E7,18E7,31E6,33E6/L1,35E4/E6,39E7,45E7,51E7,
  52E7/L1,53E6,56E7,58E7/L1,59E7,66E6,67L1 and 68E7

  β-Globin control PCR:

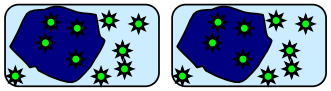
  For each sample a control PCR's is performed

- Viral load was expressed as HPV copies / cell

=> 20 real time PCR's / sample

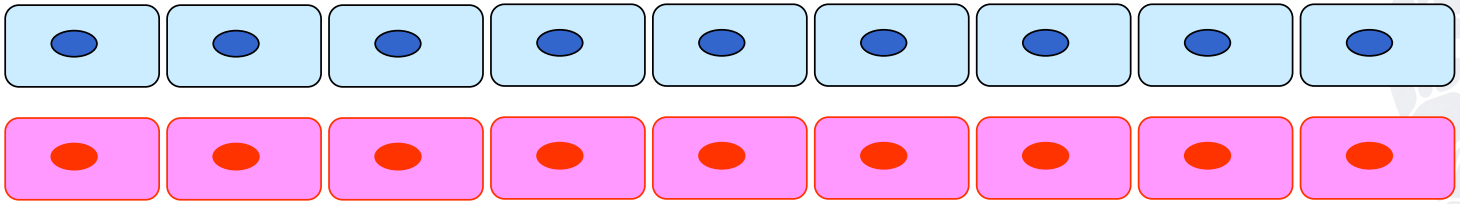
Moberg et al 2003; J Clin Microbiol: 41(7):3221-3228
Flores-Mungia et al 2004; J Mol Diagn; 6(2): 115-124
Depuydt et al 2006; Cytopathology 17: 1-8
Micalessi et al 2011; Clin Chem Lab Med; 20;50(4):655-61
Depuydt et al 2012; J Clin Microbiol;50(12):4073-7

Charge virale HPV
copies HPV / cellule



40 copies HPV (HPV qPCR)

Σ = 20 cellules (β-globine qPCR)



Charge virale = 40 copies HPV / 20 cellules
= 2 copies HPV / cellule

Test qPCR HPV cliniquement validé

- Validation du test qPCR sur la base des critères de Meijer (CIN2+):

Clinical Validation of a Type-Specific Real-Time Quantitative Human Papillomavirus PCR against the Performance of Hybrid Capture 2 for the Purpose of Cervical Cancer Screening.

Depuydt CE, Benoy IH, Beert JF, Criel AM, Bogers JJ, Arbyn M. J Clin Microbiol. 2012 Dec;50(12):4073-7.

	hc2 (RLU)	qPCR (log hrHPV copies/cell)
For sensitivity of 95%		
Specificity	69.2 %	95.2 % [a]
95% CI	55.7 – 81.0	94.2 – 96.6
Threshold value	> 0.2015	> -4
For specificity of 95%		
Sensitivity	78.31 %	93.55 % [a]
95% CI	63.86 – 85.54	87.1 – 96.77
Threshold value	> 1.477	> -4

- 20 qPCR / échantillion
- 14 hr HPV – 4 lr HPV
- Génotypage quantitative
- Région E6/E7
- Entièrement automatisé
- Haute sensibilité
- Certifié ISO

BELAC ISO 15189 352 - MED

Les tests HPV détectent à la fois le virus et les particules infectieuses (virions)

L'ADN HPV E6/E7 est mesurable dans:

1. Virions HPV libres (en dehors des cellules) (INFECTIEUX)
2. Dans les cellules qui desquament (virions)

   (cellules qui ne se divisent pas; INFECTIEUX)
3. Dans les cellules en division (transformation, épisomique et intégrée)

   (cellules clonales; NON-INFECTIEUX)

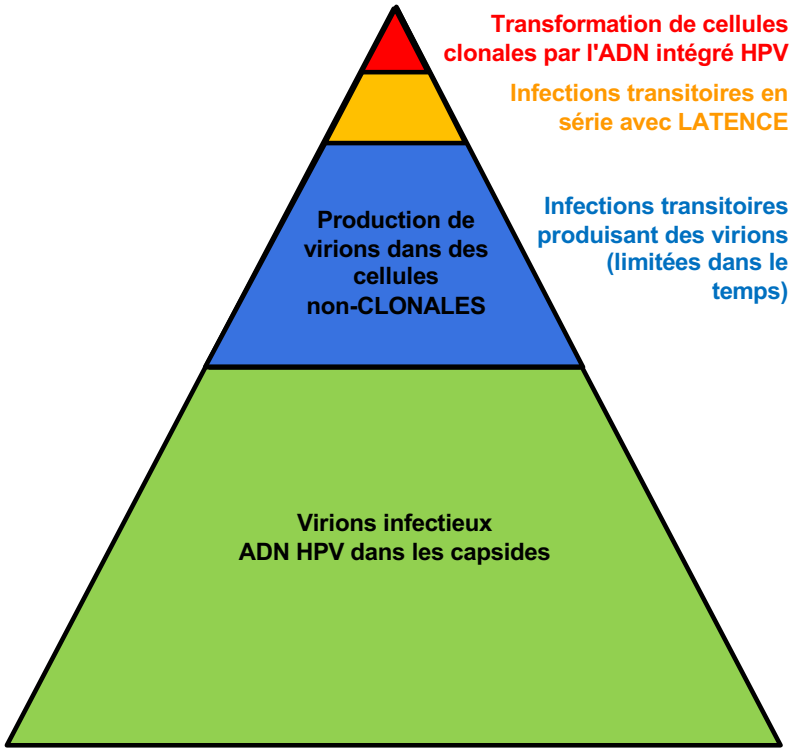
Charge virale HPV E6/E7 = Σ 1 + 2 + 3 / nombre de cellules

L'iceberg ADN HPV

♀ NON-INFECTIEUX
CLONALE

INFECTIEUX
♀♂

♀ Prélèvement cervicale:
82% INFECTIEUX; 18% CLONALE



CELLULES CLONALES ♀

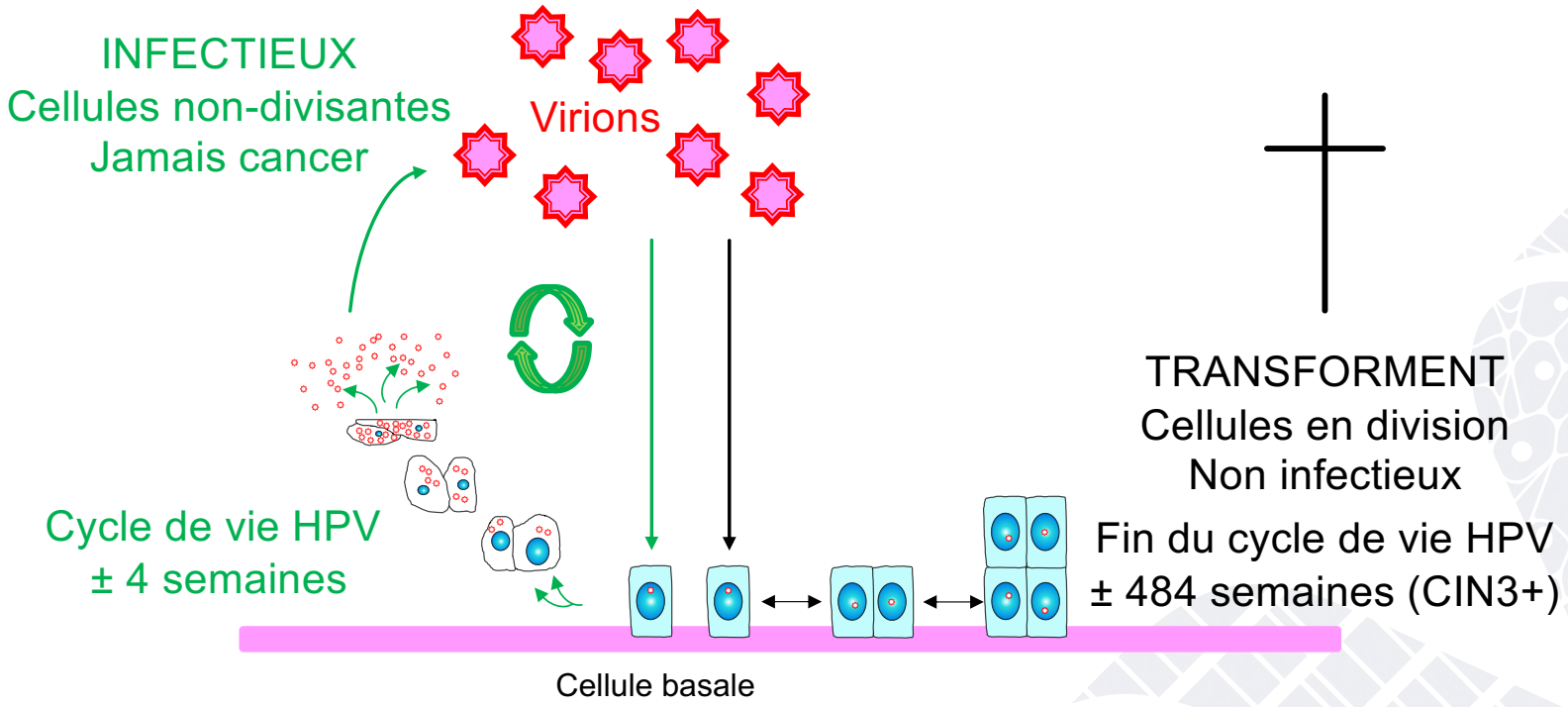
CELLULES QUI NE SE DIVISENT PAS ♀♂

HORS DES CELLULES
♀♂

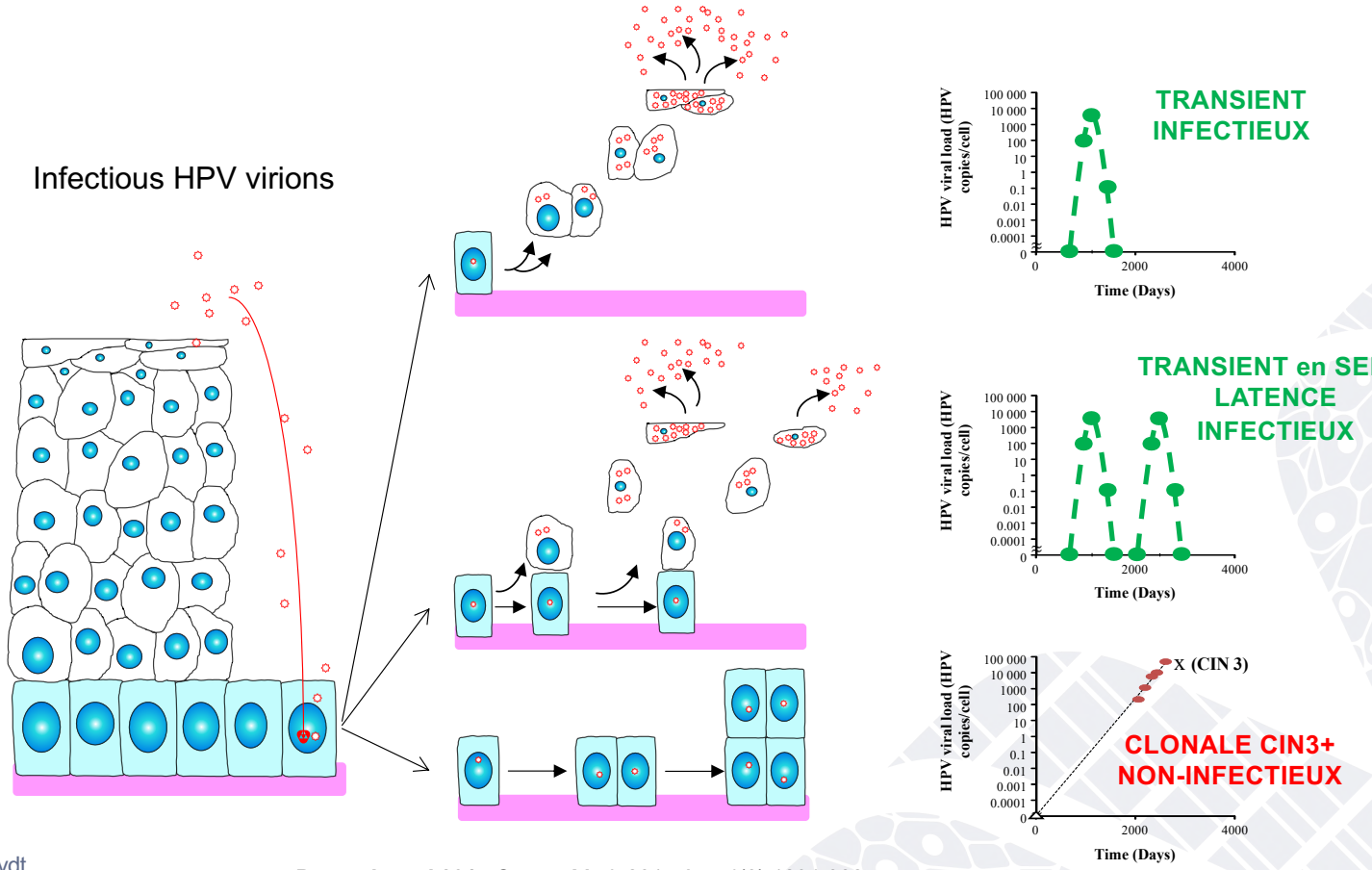
♂ Sperme:
absence de cellules en division
100% INFECTIEUX

Depuydt et al 2016 Human Papillomavirus (HPV) virion induced cancer and subfertility, two sides of the same coin. Facts Views Vis Obgyn, 2016, (4): 211-222,

Le Papillomavirus humain est un organisme avec 2 processus causés par des virions



Catégorisation de l'infection selon la division cellulaire basale



Depuydt et al 2015 Cancer Med. 2015 Aug;4(8):1294-302.

Les changements de la charge virale de l'HPV (type spécifique) prédisent la progression vers le cancer du col de l'utérus

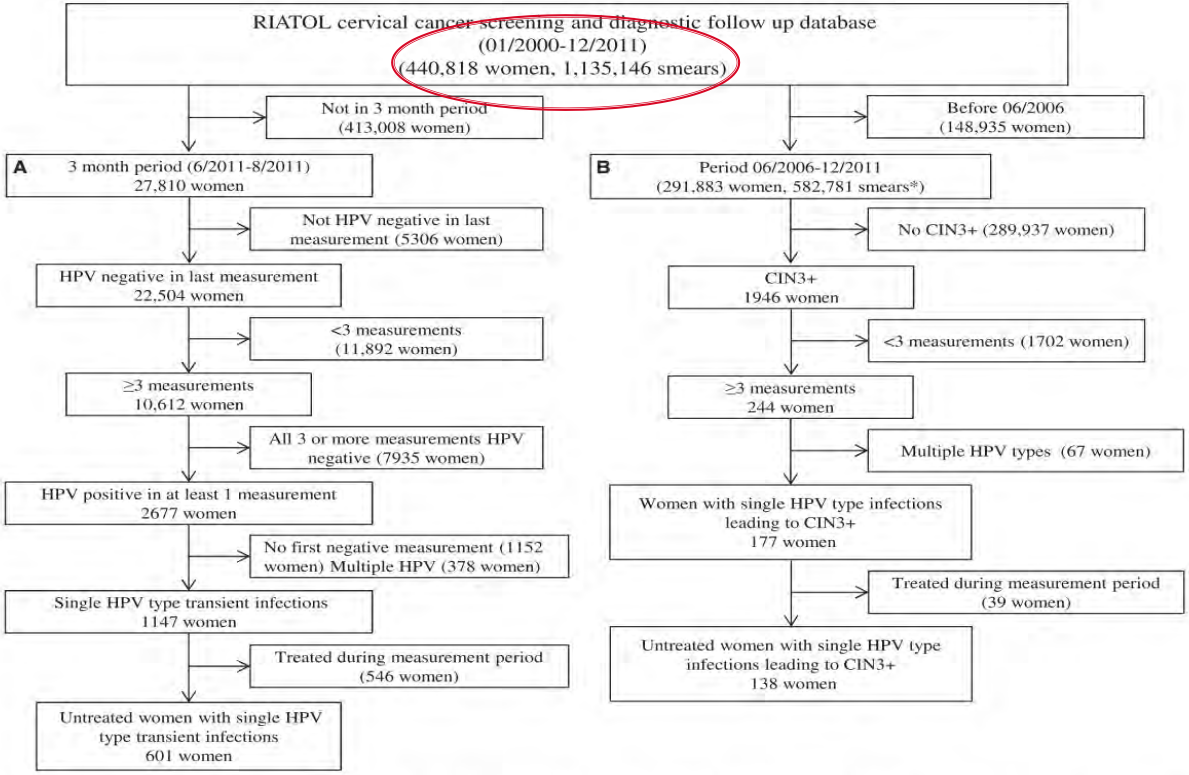
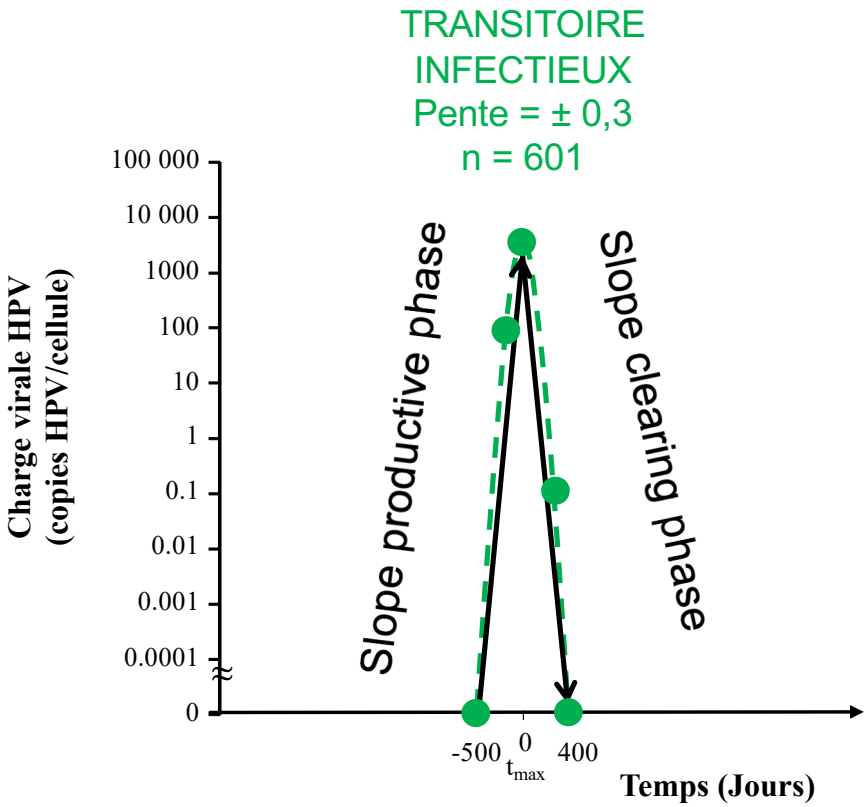
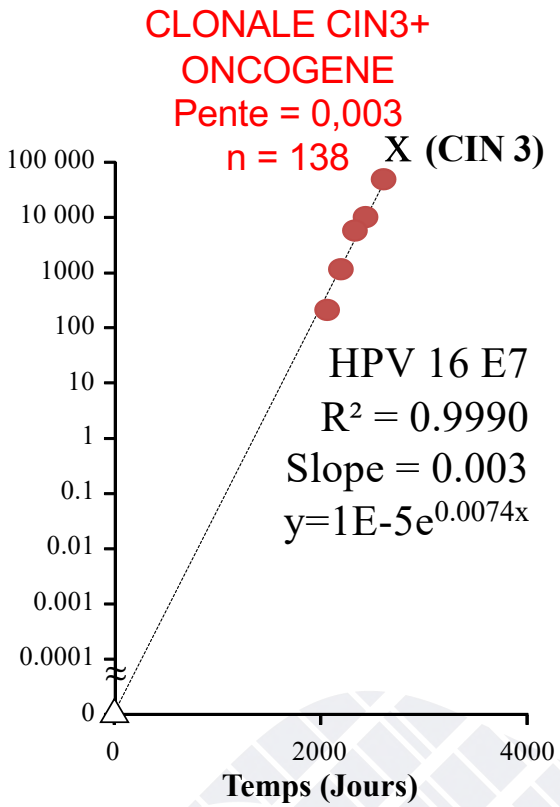


Fig. 1 Study Profile. (**A**) Selection of women with transient single-type HPV infections. (**B**) Selection of women with single-type HPV infections leading to CIN3+. * = Since June 2006 HPV genotyping was always performed before cytology reading.

Depuydt et al 2012. Changes in Type-specific Human Papillomavirus Load Predict Progression to Cervical Cancer. J Cell Mol Med. 2012;16(12):3096-104

Les changements de la charge virale spécifique au type d'HPV prédisent la progression vers le cancer du col de l'utérus





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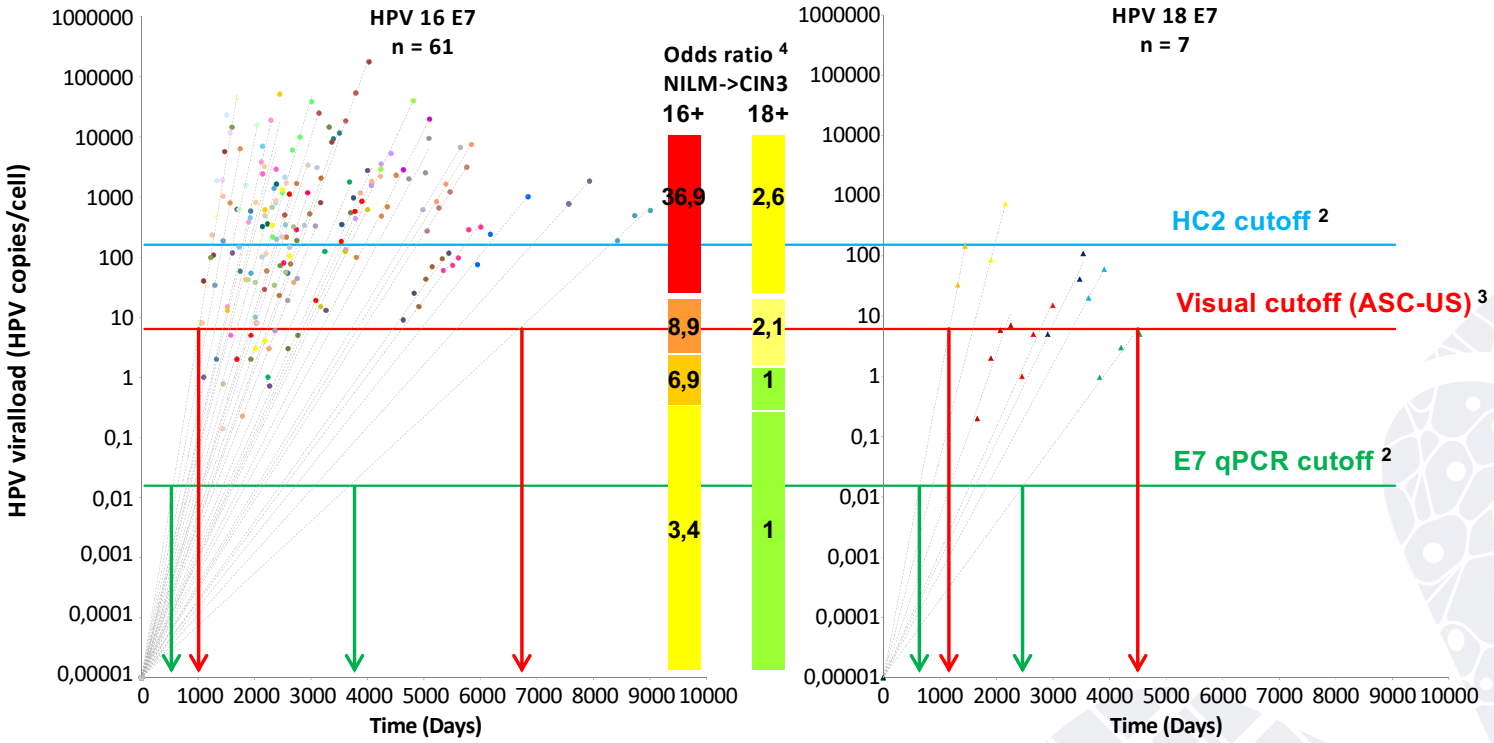
Depuydt et al 2016. Linear viral load increase of a single HPV-type in women with multiple HPV infections predicts progression to cervical cancer. Int J Cancer. 2016;139(9):2021-32.

Augmentation linéaire des cas HPV 16 et HPV 18 CIN3+[1]



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4. Moberg et al 2004. Type-specific associations of Human Papillomavirus load with risk of developing cervical carcinoma in situ. Int J Cancer 2004;112:854-859,

HPV 45 in cervical cancer cell lines

Cervical carcinoma cell line MS751

Geisbill 1997; Journal of General Virology, 78,655-658.

HPV-45 in MS751

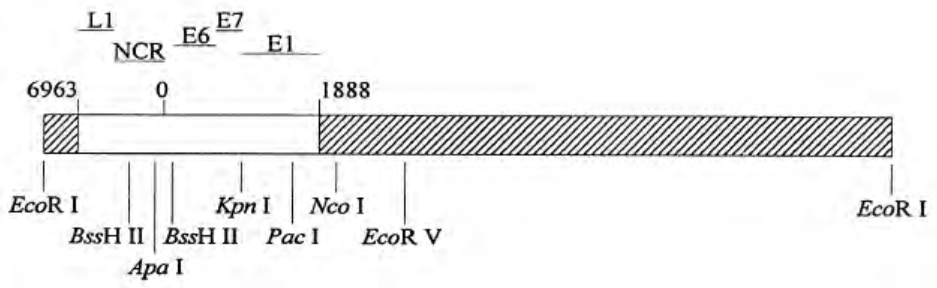
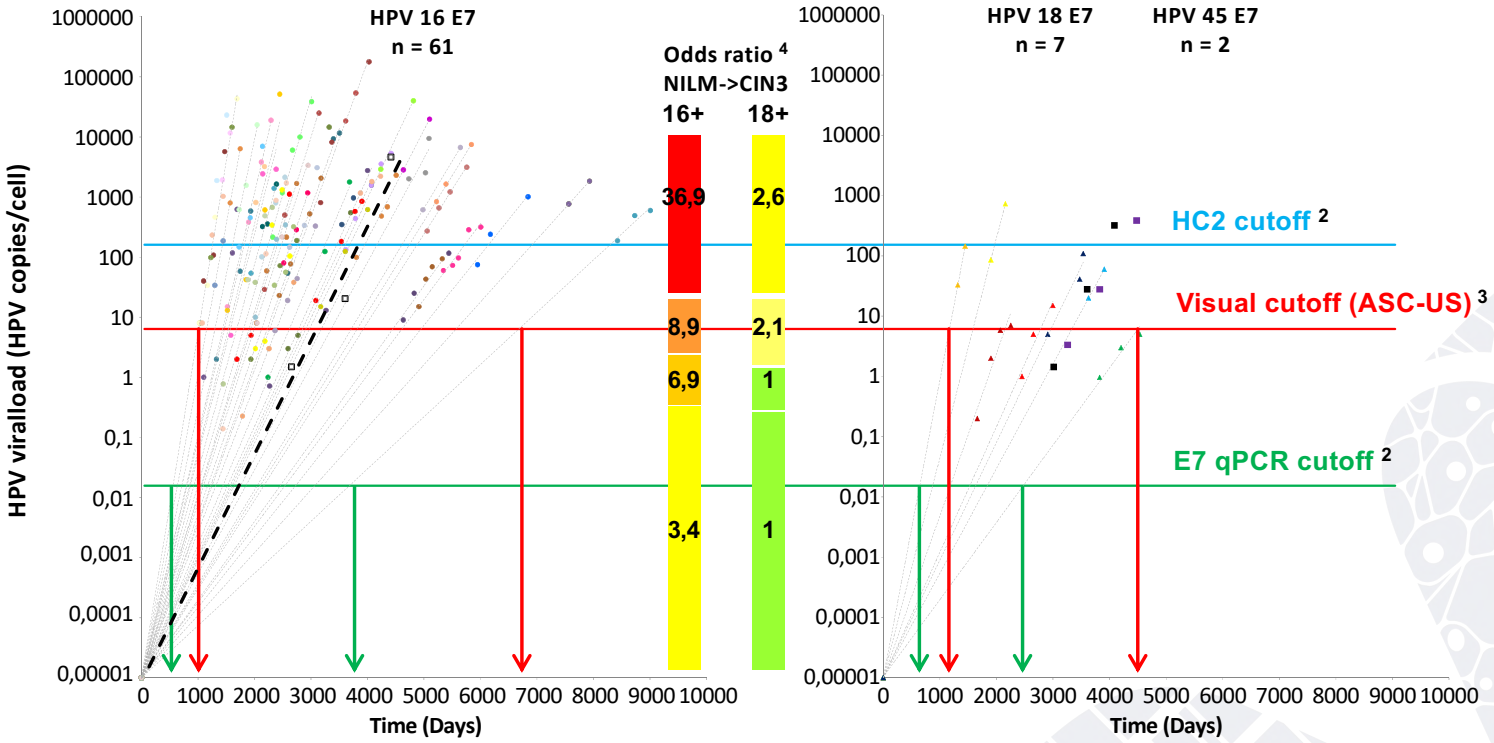


Fig. 2. Physical map of the 9·5 kb *EcoRI* restriction fragment cloned from MS751 genomic DNA and location of viral and cellular sequences. The positions of cleavage sites for restriction enzymes are shown. Cellular sequences are represented by regions with diagonal hatching. HPV sequences are given as an open bar and the open reading frames are indicated on top.

HPV 45 copies in tumor cell lines	
MS751	1 copy
CC-8	2 copies
CC-10A	2 copies
CC-10B	2 copies

=> Low viral load in HPV 45 cxca

Augmentation linéaire des cas HPV 16, 18 et HPV 45 CIN3+[1]



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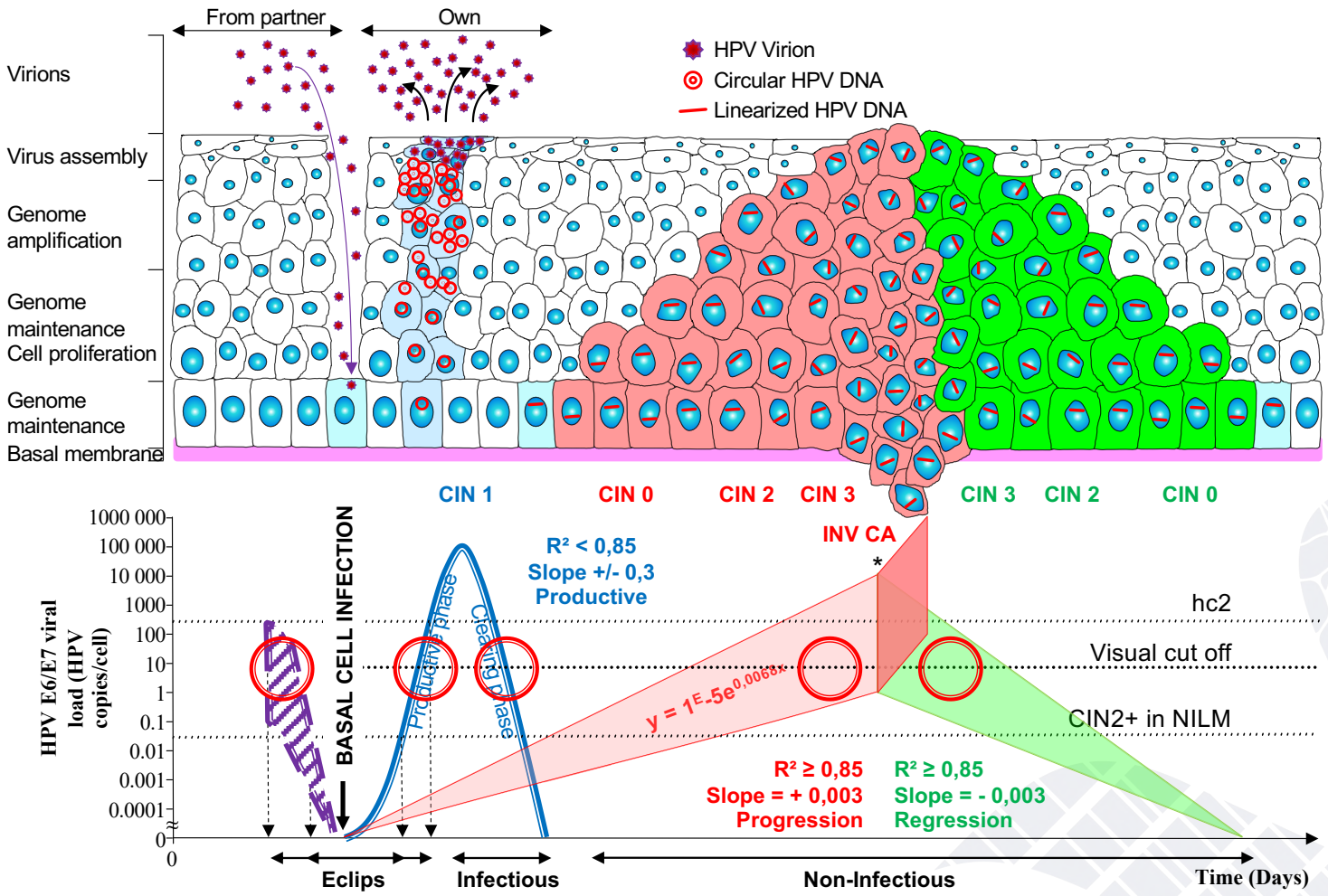
Type-specific Median HPV Slope in Infections with single or multiple HPV types Leading to CIN3+

	Single HPV		Multiple HPV		All HPV	
HPV type	n	Slope	n	Slope	n	Slope
6	1	0.0026	ND		1	0.0026
11	ND		ND		ND	
16	61	0.0030	21	0.0031	82	0.0031
18	4	0.0019	4	0.0038	8	0.0034
31	19	0.0026	9	0.0018	28	0.0023
33	15	0.0031	6	0.0035	21	0.0032
35	3	0.0025	1	0.0039	4	0.0029
39	4	0.0027	2	0.0024	6	0.0026
45	1	0.0018	1	0.0017	2	0.0018
51	8	0.0032	3	0.0040	11	0.0034
52	10	0.0035	3	0.0034	13	0.0035
53			ND		ND	
56	2	0.0038	ND		2	0.0038
58	8	0.0034	4	0.0026	12	0.0031
59	1	0.0029	ND		1	0.0029
66	1	0.0032	2	0.0022	3	0.0025
68			1	0.0054	1	0.0054
All	138	0.0028	57	0.0029	185	0.0029

48,6%

49,7%



5 fois la même charge virale !!!

Verhelst et al 2017. Serial measurement of type-specific Human Papillomavirus (HPV) load allows classification of CIN lesions (n=307) according to occurring HPV induced pathway. Eur Jour Cancer Prev 2017 Mar;26(2):156-164. doi: 10.1097/CEJ.0000000000000241.

- Ce n'est pas la charge virale mais l'augmentation HPV type spécifique qui prédit le cancer
- CLONALE 0,003 vs TRANSITOIRE 0,3 copies HPV/cellule/jour
- Mesurer la charge virale 3 fois !

Conclusions mesures en série charge virale HPV type spécifique

Seuls 4 modèles différents de charge virale spécifique de type séquentiel plus long ont été identifiés, représentant les 2 voies différentes induites par l'HPV

Voie productive (infectieuse)

1. produisant des virions (infection transitoire)
   (+/- 0,3 copies HPV/cellule/jour)
2. latence, production de virions en série
   (entre 0,0010 and –0,0010 copies HPV/cellule/jour)

Voie de transformation (CLONALE; non infectieuse)

3. Lésions progressives (R^2>0,9 and 0,003 copies HPV/cellule/jour)
4. Lésions régressives (R^2>0,9 and -0,003 copies HPV/cellule/jour)

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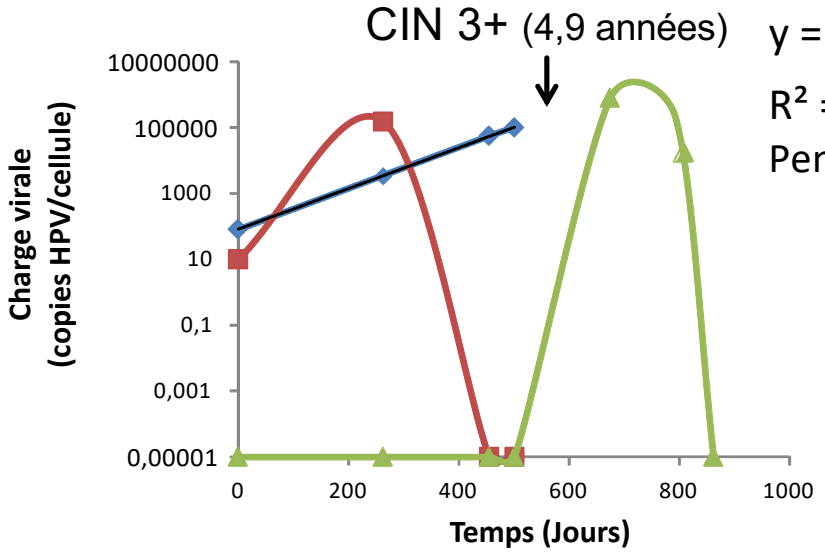
Calculation of infectious and clonal HPV infections

- Measure the quantitative type specific HPV viral load on 3 different smears during follow-up (3x; for all HPV types)
- Calculate R^2 (for each HPV type)
  - $R^2 \geq 0{,}85$
    - calculated slope is positive (clonal; progression)
    - calculated slope is negative (clonal; regression)
  - $R^2 < 0{,}85$
    - Calculate individual slopes delta 1-2 and delta 2-3
    - Look up ongoing processes in table 1 (Verhelst et al 2016)

Verhelst et al 2017. Serial measurement of type-specific Human Papillomavirus (HPV) load allows classification of CIN lesions (n=307) according to occurring HPV induced pathway. Eur Jour Cancer Prev 2017 Mar;26(2):156-164. doi: 10.1097/CEJ.0000000000000241.

Infections transitoires et persistantes avec plusieurs types d'HPV (CIN3+)



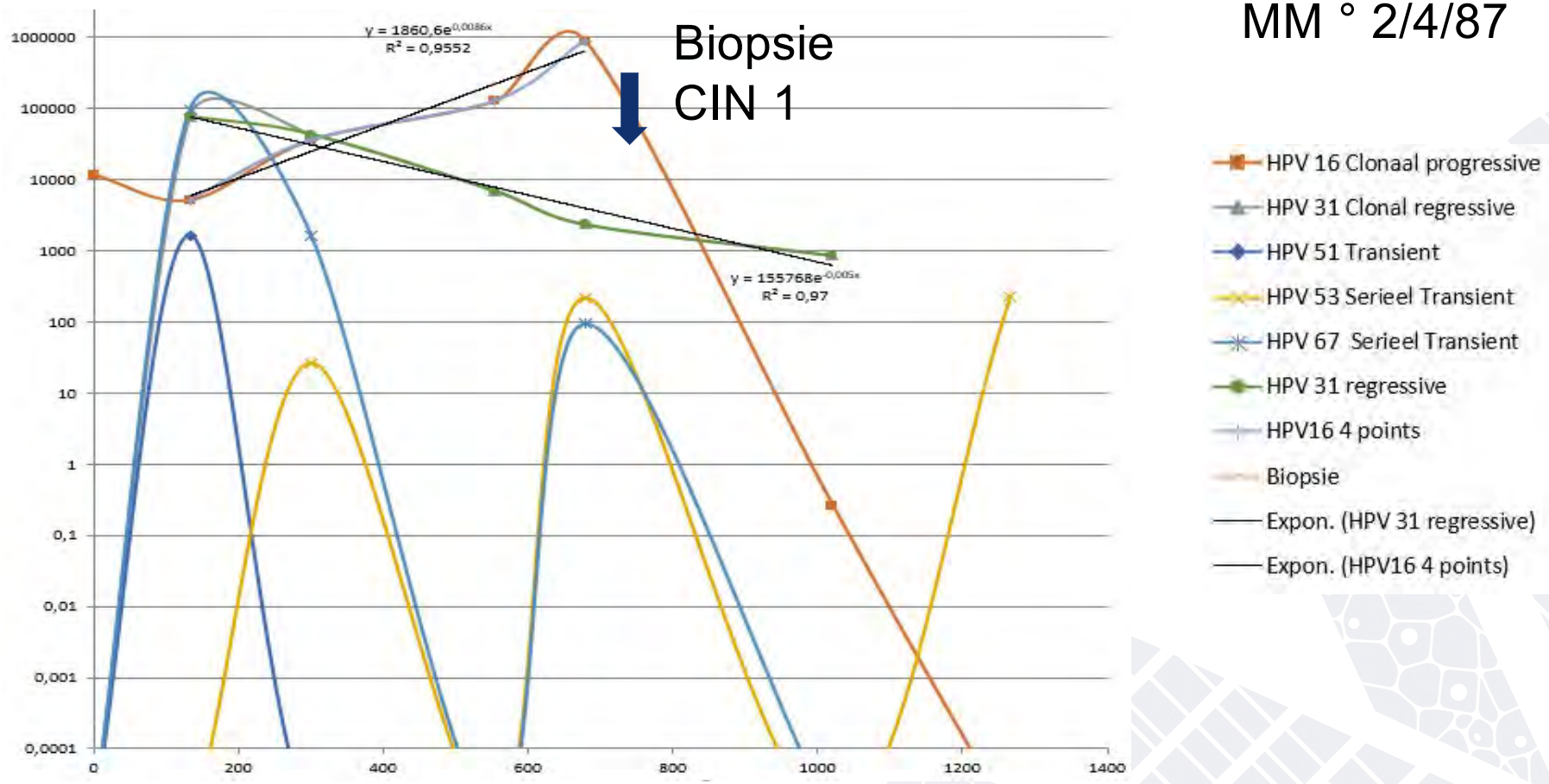
$y = 81{,}263e^{0{,}0143x}$

$R^2 = 0{,}9999$

Pente = 0,0061 copies HPV/cellule/jour

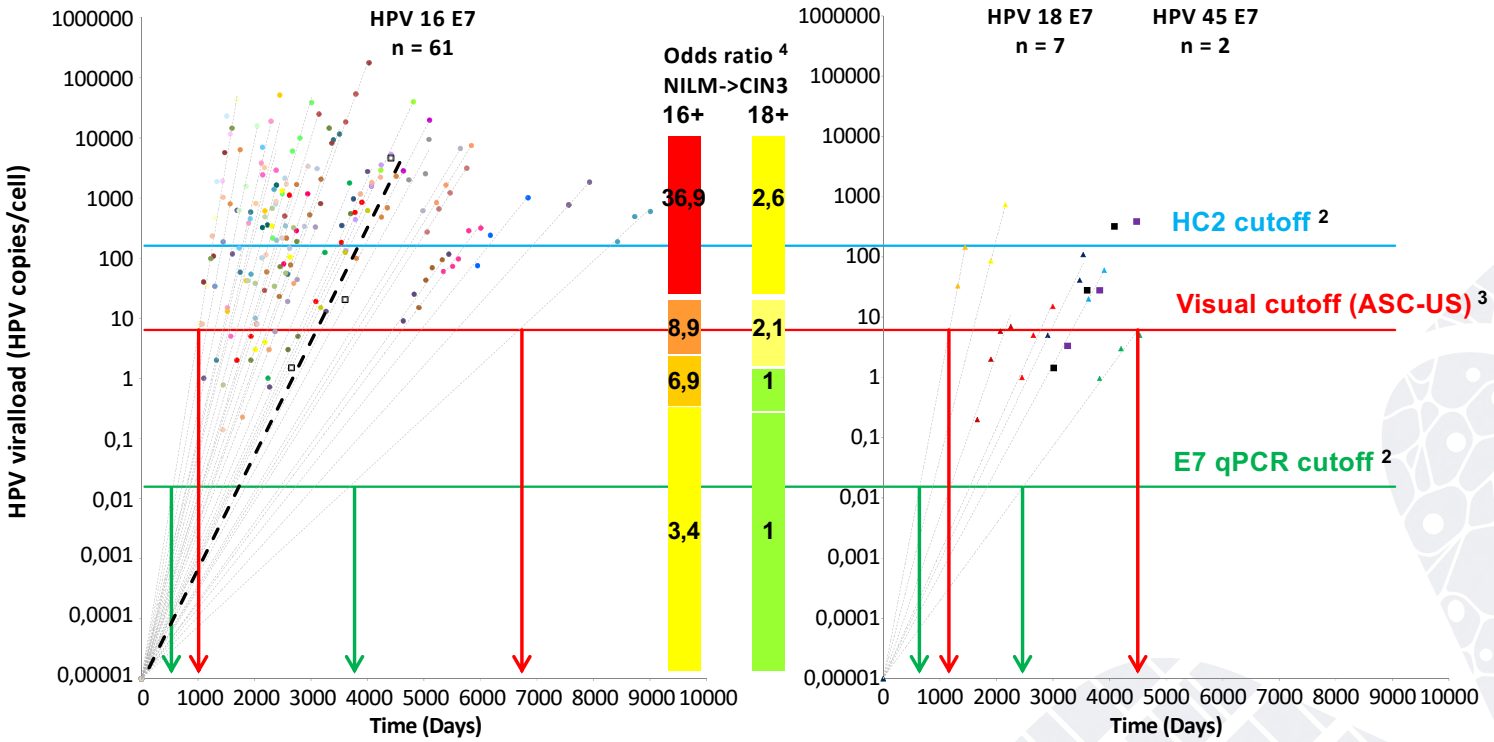
Patiente a des charges virales HPV type spécifique qui montes et qui descend ?

MM ° 2/4/87



Patiente a un HPV 18 avec une charge virale qui monte régulièrement et une colpo-biopsie normale ?

Augmentation linéaire des cas HPV 16, 18 et HPV 45 CIN3+[1]



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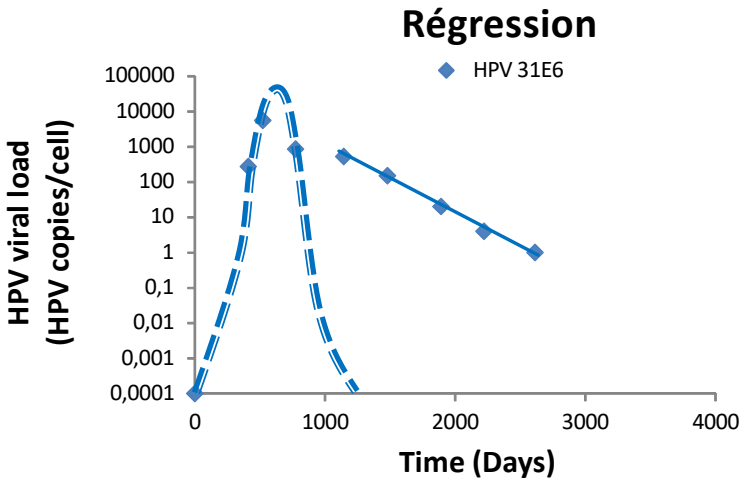
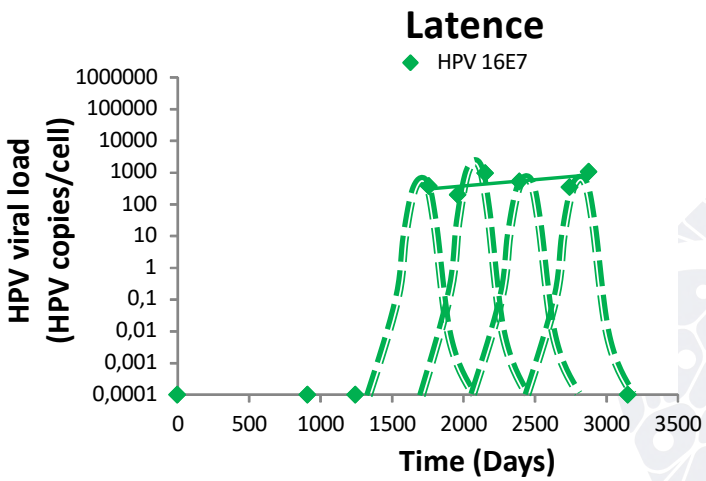
- L'âge moyenne d’une lésion CLONALE CIN3+ est de 9,3 ans
- La transformation des cellules clonales prend du temps
- Apres colposcopie avec biopsie normale, contrôler si la charge virale a diminuée ou a été négativée
- Si la charge remonte a nouveau la biopsie a été prise a cote de la lésion
- Proposer une conisation diagnostique

Patiente a une charge virale qui monte et qui descend ?

Régression vs latence



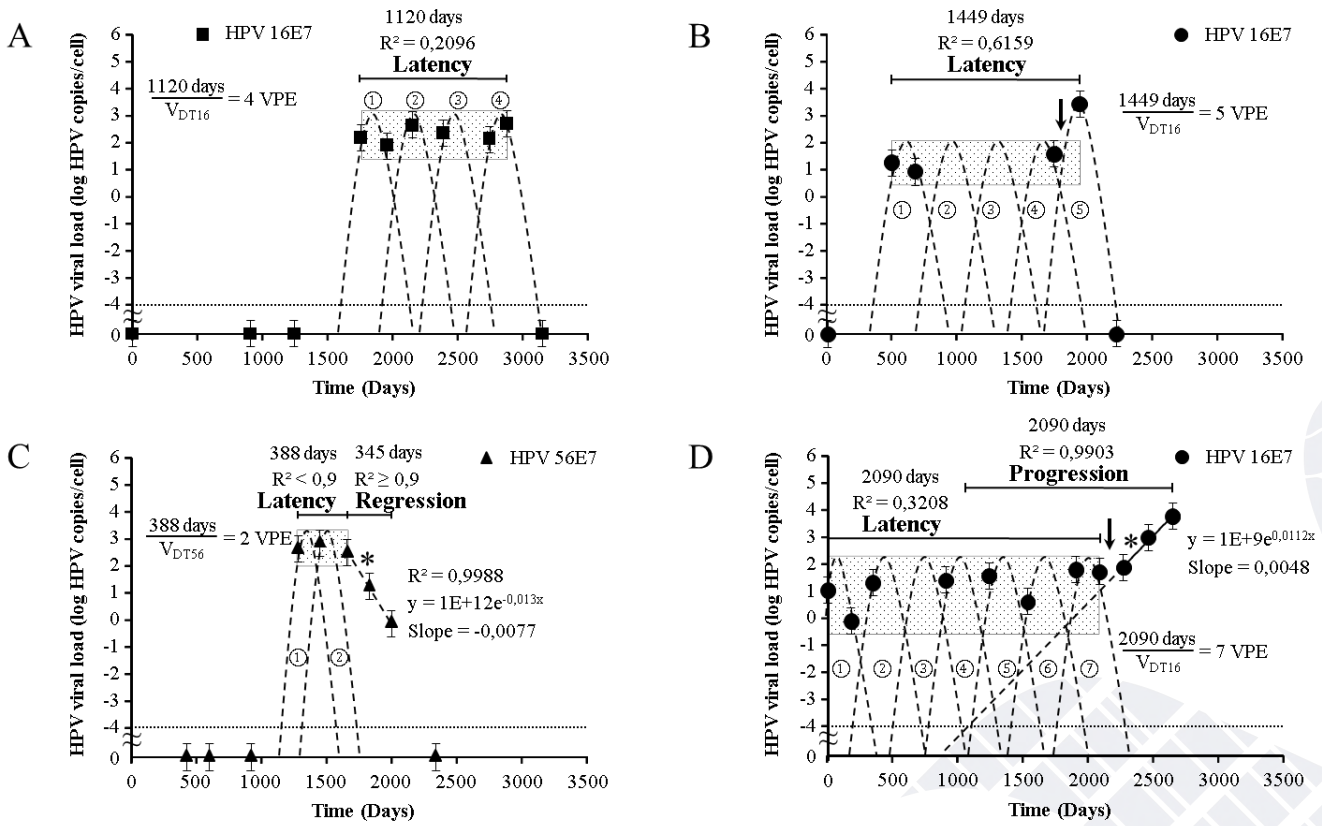
R^2 = 0,9966
Slope = -0,002



R^2 = 0,2096
Slope = 0,0003

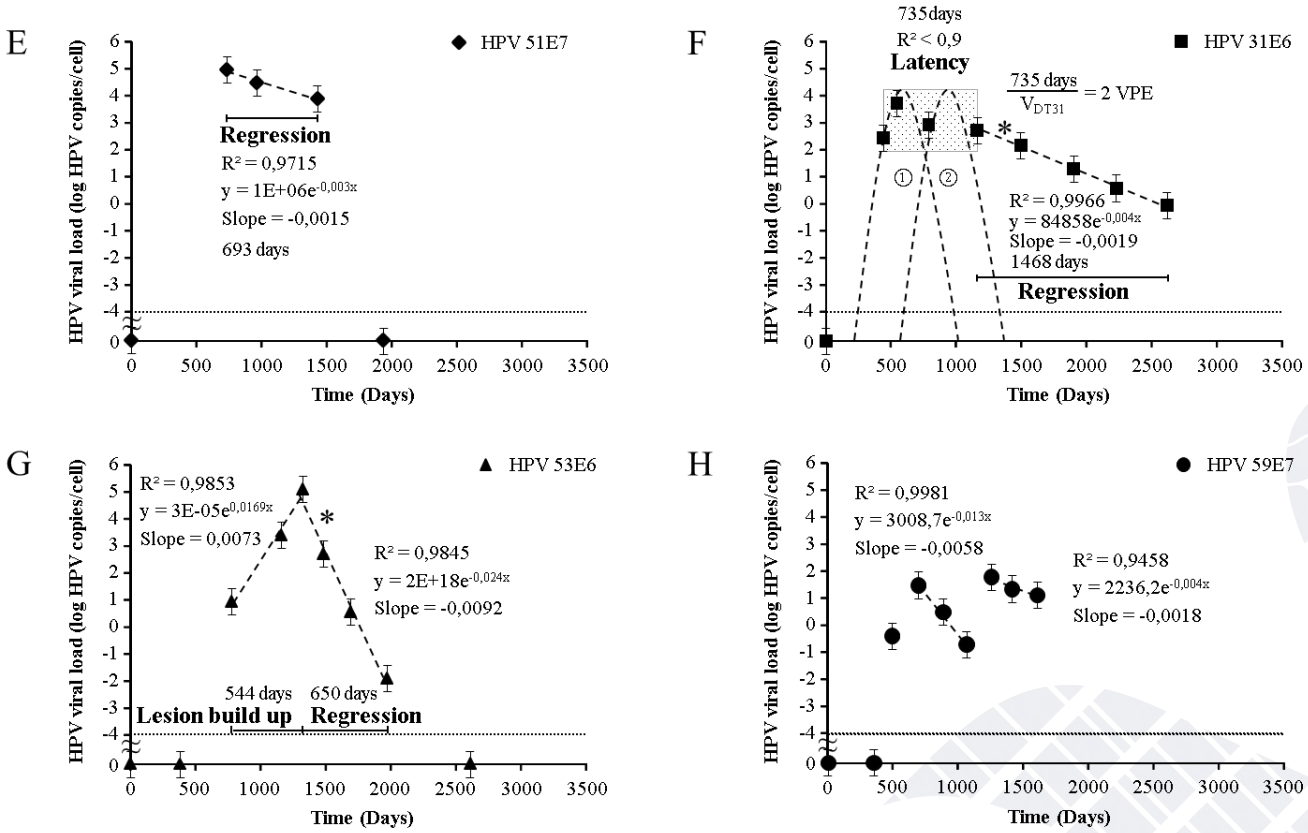
Depuydt et al 2015 Cancer Med. 2015 Aug;4(8):1294-302.

Cas représentatifs montrant les différents modèles de charge virale en latence



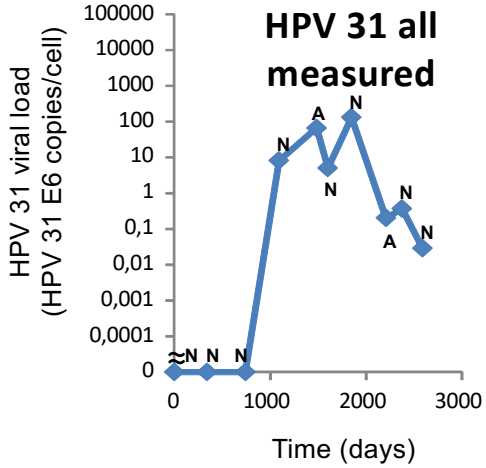
Depuydt et al 2015 Cancer Med. 2015 Aug;4(8):1294-302.

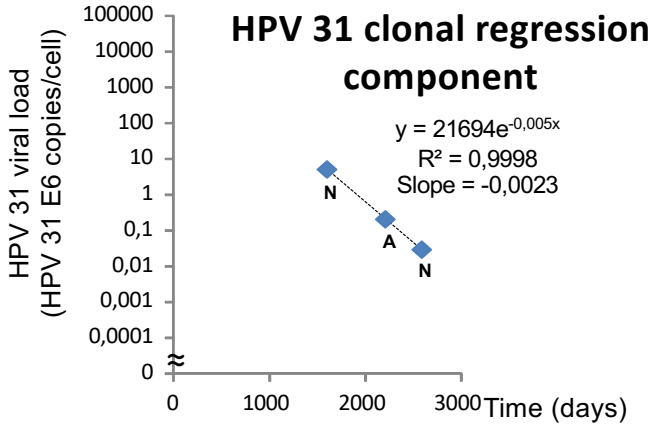
Cas représentatifs montrant les différents modèles de charge virale en régression

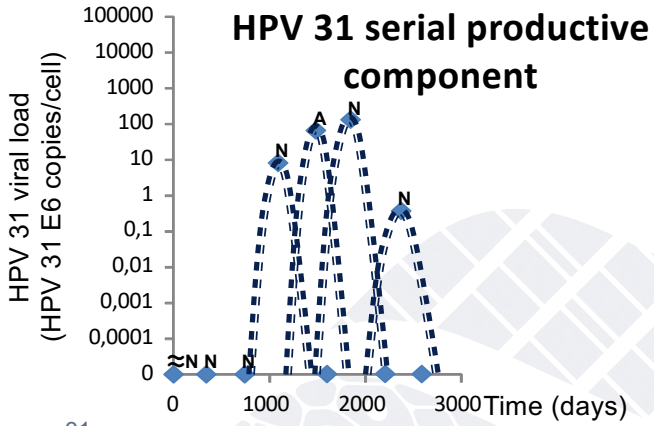


Depuydt et al 2015 Cancer Med. 2015 Aug;4(8):1294-302.

Clonal HPV:
Breakdown in productive and regressing components



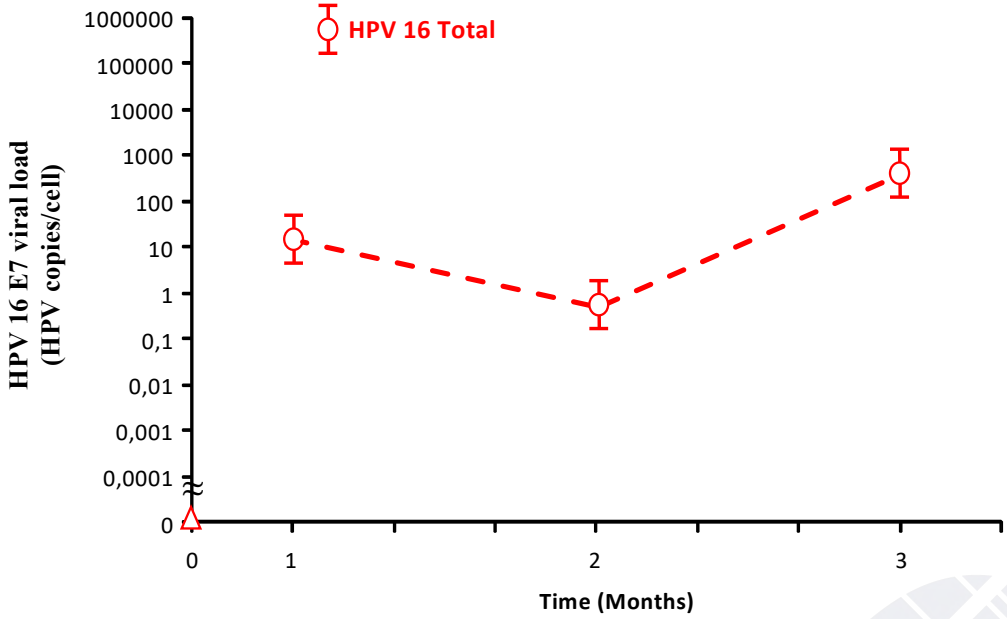




N = Negative for intraepithelial neoplasia or malignancy;
A = Atypical cells of unknown significance

Not pregnant and HPV DNA positive

(3 x IUI; 36 years)



Not pregnant and HPV DNA positive
(3 x IUI; 36 years)

With 3 measurements we can calculate which HPV induced processes are present (table 1 Verhelst et al 2017):

- 1e IUI on 18 January 2017 = 447 HPV 16 copies/cell
- 2e IUI on 14 February 2017 = 191 HPV 16 copies/cell (after 27 days)
- 3e IUI on 13 March 2017 = 1067 HPV 16 copies/cell (after 32 days)

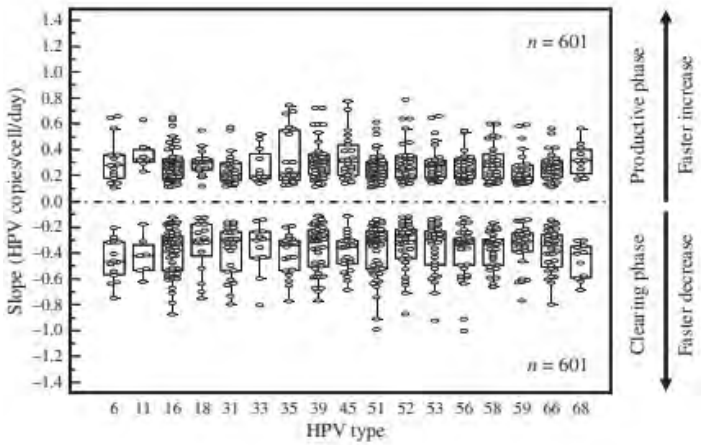
R^2 for the 3 measurements (delta 1-2-3) was < 0.85 namely 0,2306

First calculate delta1-2 (27 days), slope = -0.0136 HPV 16 copies/cell/day
Secondly calculate delta 2-3 (32 days), slope = 0.0233 HPV 16 copies/cell/day

Calculation of HPV type specific slopes in transient infections

$$\text{Slope}_{\text{productive phase}} = \frac{(\text{Log load}_{t0-1}) - (\text{Log load}_{tmax})}{(D_{t0-1}) - (D_{tmax})}$$

$$\text{Slope}_{\text{clearing phase}} = \frac{(\text{Log load}_{tmax}) - (\text{Log load}_{t0+1})}{(D_{tmax}) - (D_{t0+1})}$$



Slope in Productive and Clearing Phase of Transient Infections by HPV type.

Depuydt et al 2012 Changes in type-specific human papillomavirus load predict progression to cervical cancer. J Cell Mol Med 2012: 16 (12): 3096-3104

Depuydt et al 2016 Linear viral load increase of a single HPV-type in women with multiple HPV infections predicts progression to cervical cancer. Int J Cancer 2016: 139: 2021-2032

Not pregnant and HPV DNA positive

(3 x IUI; 36 years)

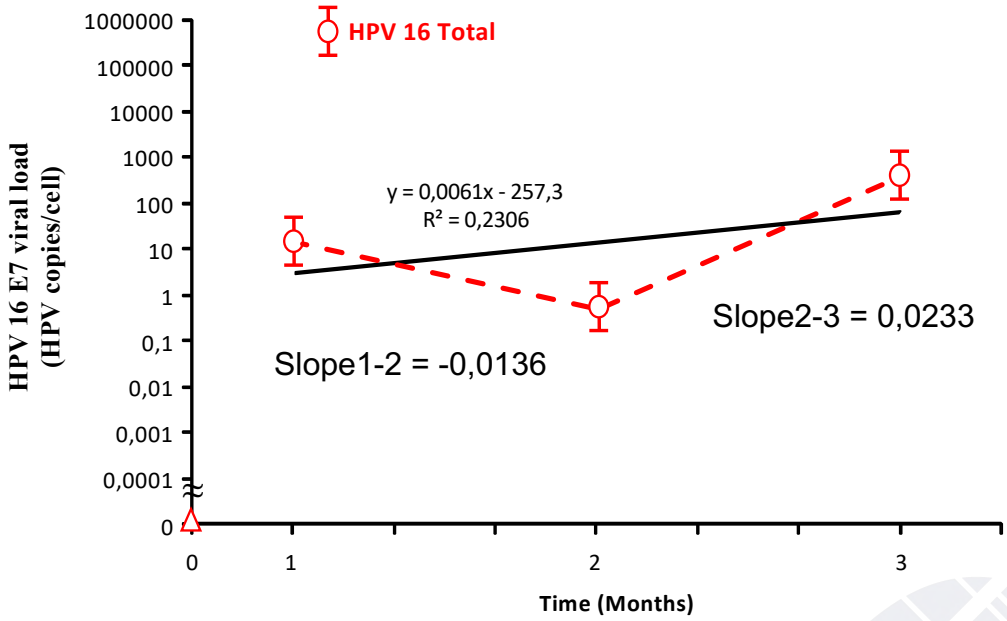


Table 1 Calculated different slope combinations per woman for each cervical intraepithelial neoplasia category with three consecutive measurements

$R^2 \geq 0.85$			CIN0		CIN1		CIN2		CIN3		1 process	2 processes
Δ1–2–3			S	M	S	M	S	M	S	M		
Pos.			5	5	2	10	3	5	4	11	Progressive	
Neg.			3	2		3	1	4	1	1	Regressive	
	Δ1–2	Δ2–3										
$R^2 < 0.85$		> 0.0099	–	–	–	1	–	–	–	–	Transient	–
		0.0010 to 0.0099	1	–	–	–	–	–	–	–	Transient	–
	> 0.0099	– 0.0010 to 0.0010	–	–	–	–	–	–	–	–	Transient	–
		– 0.0099 to – 0.0010	–	–	–	1	–	–	–	–	Transient	–
		> – 0.0099	–	1	–	1	–	–	–	–	Transient	–
		> 0.0099	1	–	–	–	–	–	–	–	Transient	–
		0.0010 to 0.0099	–	–	1	–	–	–	–	–	Transient	Transient
	0.0099 to 0.0010	– 0.0010 to 0.0010	3	1	2	–	2	–	–	–	Transient	Transient
		– 0.0099 to – 0.0010	3	4	1	2	2	–	1	–	Transient	Prog.+Tr.
		> – 0.0099	–	2	–	–	–	–	–	–	Transient	–
		> 0.0099	–	–	–	–	–	–	–	–	Transient	–
		0.0010 to 0.0099	–	2	1	1	1	–	1	–	Transient	Prog.+Tr.
	0.0010 to – 0.0010	– 0.0010 to 0.0010	–	–	–	2	–	1	–	–	Transient	Ser. Tr.
		– 0.0099 to – 0.0010	1	–	1	1	–	4	–	–	Transient	Transient
		> – 0.0099	–	–	–	–	–	–	–	–	Transient	–
		> 0.0099	–	–	1	–	–	1	–	–	Transient	Transient
		0.0010 to 0.0099	–	5	1	2	2	–	2	5	Transient	Prog.+Tr.
	– 0.0010 to – 0.0099	– 0.0010 to 0.0010	–	–	2	–	–	1	–	–	Transient	Transient
		– 0.0099 to – 0.0010	–	–	–	–	–	–	–	–	Transient	–
		[illegible]	–	–	–	–	–	1	–	–	Transient	
		> 0.0099	1	–	–	1	–	–	–	–		Ser. Tr.
		[illegible]	–	–	–	–	–	–	–	–	Transient	
	> – 0.0099	– 0.0010 to 0.0010	–	–	–	–	–	–	–	–	Transient	–
		– 0.0099 to – 0.0010	–	–	–	–	–	1	–	–	Transient	–
		> – 0.0099	–	–	–	–	–	–	–	–	Transient	–
$R^2 < 0.85^{(1)}$			–	–	–	–	–	–	–	–	–	–
		> 0.0099	–	2	–	1	3	2	–	–	Transient	–
		0.0010 to 0.0099	3	3	2	2	–	1	–	–	Transient	–
	> 0.0099[(1)]	– 0.0010 to 0.0010	–	2	–	–	4	–	–	–	Transient	–
		– 0.0099 to – 0.0010	1	–	1	1	1	3	–	–	Transient	–
		> – 0.0099	3	2	–	1	–	–	–	–	Transient	–
	0.0099 to 0.0010	> – 0.0099[(2)]	–	–	–	–	–	–	–	–	Transient	–
		> 0.0099[(2)]	–	–	–	–	–	–	–	–	Transient	–
		> 0.0099[(2)]	3	9	10	6	2	10	–	–	Transient	–
		0.0010 to 0.0099	–	1	–	–	–	–	–	–	Transient	–
	0.0010 to – 0.0010	– 0.0010 to 0.0010	–	–	–	–	–	–	–	–	Transient	–
		– 0.0099 to – 0.0010	–	–	–	–	–	–	–	–	Transient	–
		> – 0.0099[(2)]	–	–	1	1	–	–	–	–	Transient	–
			–	–	–	–	–	–	–	–	Transient	–
			–	–	–	–	–	–	–	–	Transient	–
	– 0.0010 to – 0.0099		–	–	–	–	–	–	–	–	Transient	–
			–	–	–	–	–	–	–	–	Transient	–
		> – 0.0099[(2)]	1	–	–	–	–	–	–	–	Transient	–
		> 0.0099[(2)]	–	–	1	2	–	2	–	–	–	Ser. Tr.
	> – 0.0099	0 (2.3)	1	3	–	–	–	–	–	–	Transient	–
Total			30	44	27	39	21	36	9	17	–	–

Verhelst et al 2017. Serial measurement of type-specific Human Papillomavirus (HPV) load allows classification of CIN lesions (n=307) according to occurring HPV induced pathway. Eur Jour Cancer Prev 2017 Mar;26(2):156-164. doi: 10.1097/CEJ.0000000000000241.

Not pregnant and HPV DNA positive
(3 x IUI; 36 years)

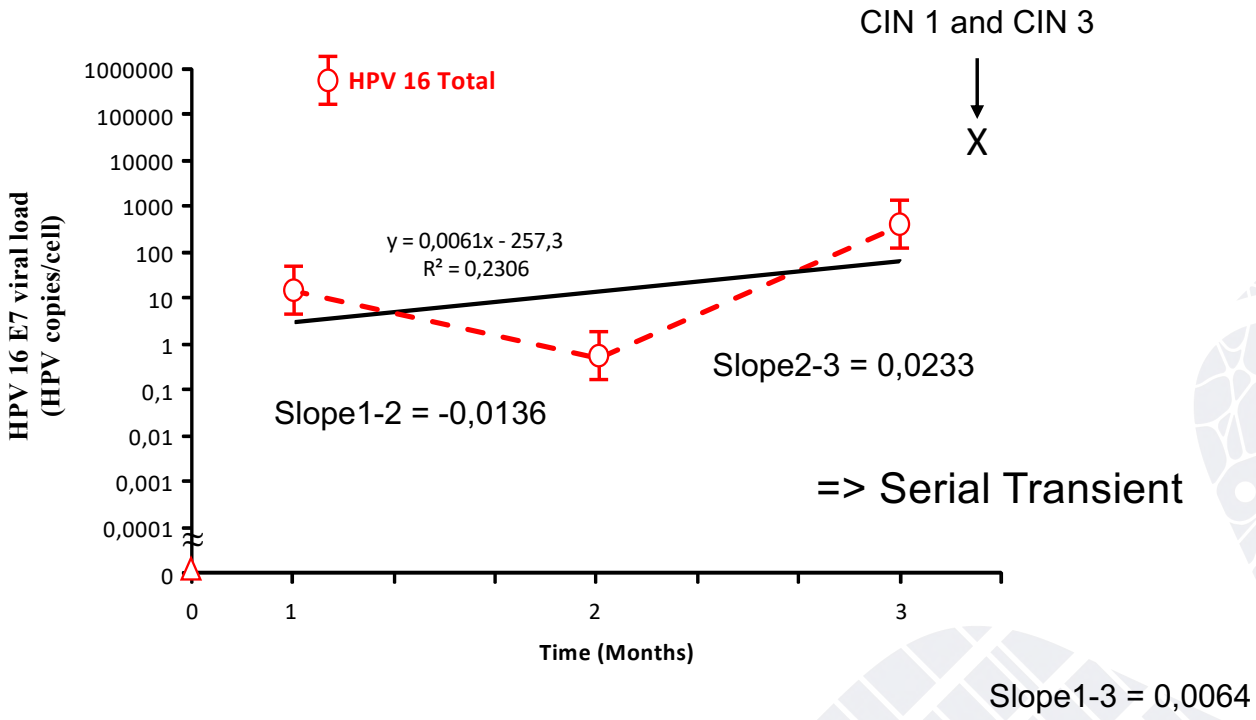
Table 1 (Verhelst et al 2017): HPV 16 DNA originates from a serial transient infection (R^2<0.85 en delta 1-2 >-0.0099 en delta 2-3 >0.0099).

- **2 HPV 16 processes present !**
  - **Serial transient virion producing HPV 16 infection**
  - **Clonal, HPV 16 transformed process**

- **Colposcopy with biopsies to exclude CIN3+**

Not pregnant and HPV DNA positive

(3 x IUI; 36 years)

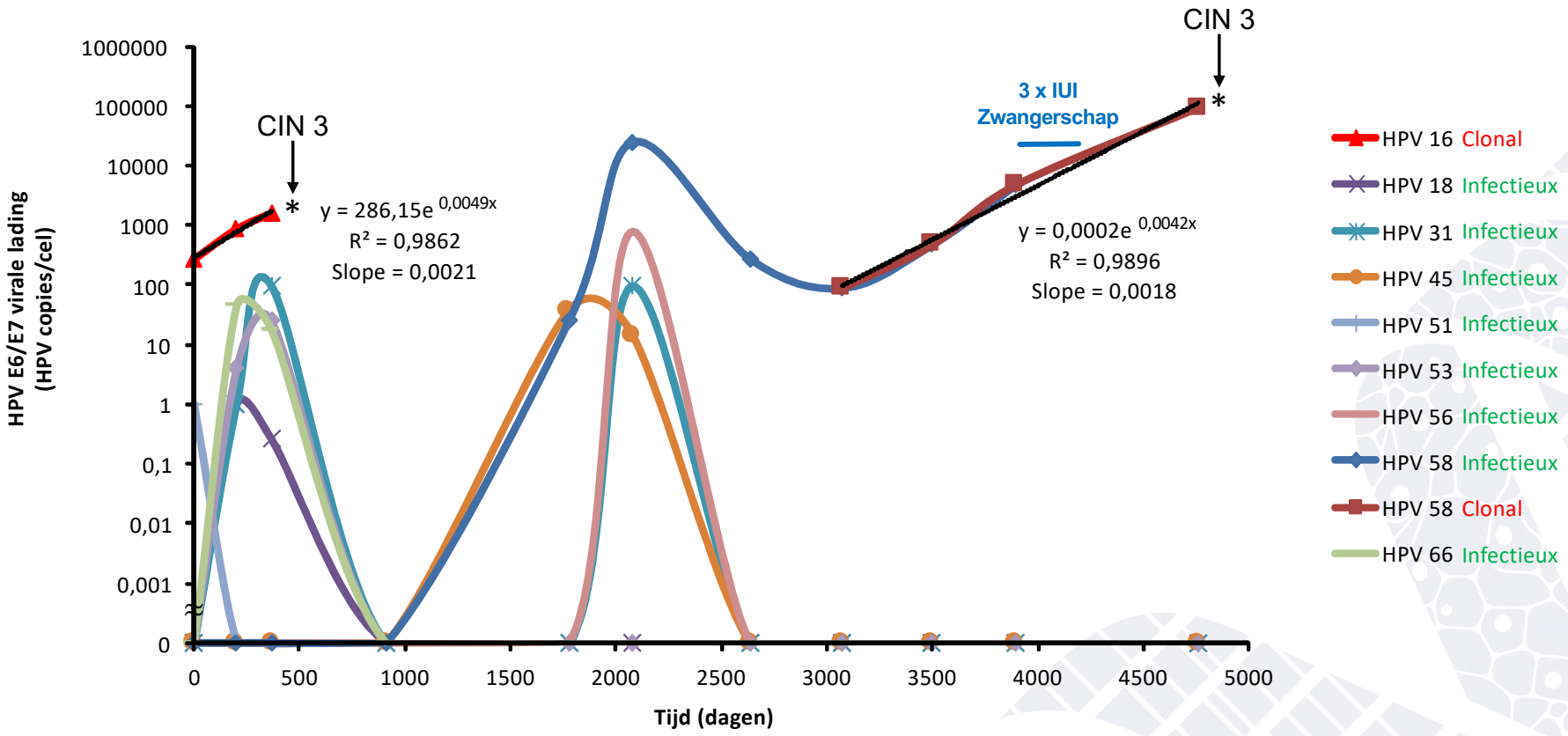


- Quand la charge virale monte et descend il est possible qu'il y ait 2 types de lésions (avec le même type d'HPV), une lésion INFECTIEUSE et / ou une lésion CLONALE
- Colposcopie avec biopsie pour exclure CIN3+

Patiente avait un HPV il y a 3 ans, puis qui a disparu, puis a à nouveau un HPV : le même, une autre contamination ?

HPV est il infectieux ou clonal ?

(35 ans, follow-up de 2006 à 2019)



- Chaque fois qu’on détecte une infection CLONALE
- Colposcopie + biopsie

Patiente pose la question de l’éventuelle incidence de la présence d’un HPV dans ses Frottis et qui fait des fausses couches à répétition ?

Prospective multi-centric study: influence of HPV infections on the outcome of Intra Uterine Insemination

Genk Institute for Fertility Technology, ZOL Hospitals, Genk, Belgium and AML intermediary structure for human body material, Sonic Healthcare, Antwerp Belgium

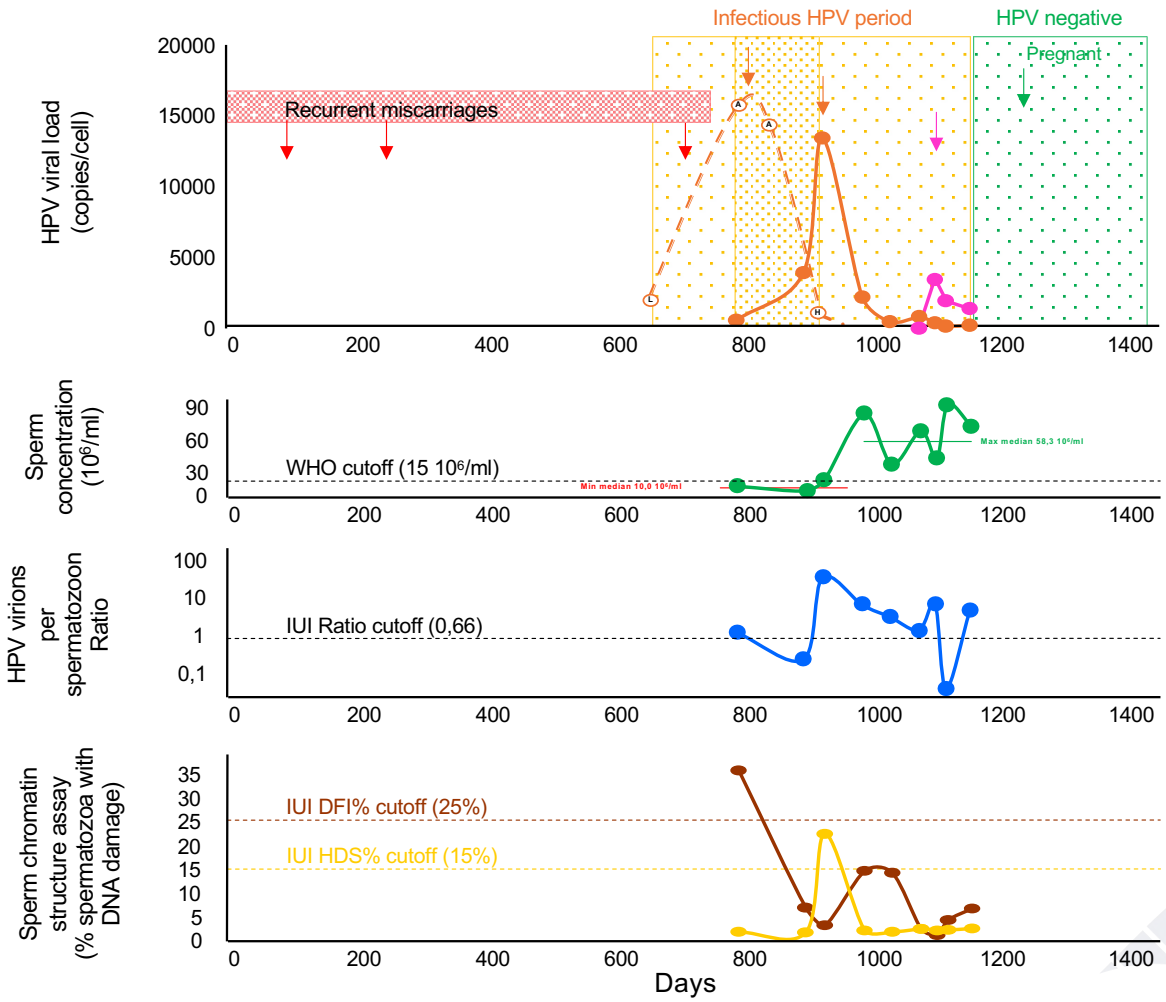
HPV status		Pregnancy			Total # cycles
Man	Woman	Clinical	Misc/bioch	Not pregnant	
-	-	141 (13,3)	38 (3,6)	883 (83,1)	1062
+	-	5 (4,3)	3 (2,6)	107 (93,0)	115
-	+	8 (3,9) *	11 (5,4)	184 (90,6)	203
+	+	0 (0,0)	3 (4,7)	61 (95,3)	64
Total		154 (10,7)	55 (3,8)	1235 (85,5)	1444

X^2 for trend $p < 0,0001$

Diminished number of clinical pregnancies in HPV+ couples

* Total of infectious and clonal HPV DNA

HPV prevalence per IUI cycle = 26,5% (382 HPV+/1444)



Repeated miscarriages

Female partner (40 yrs)

- HPV 31

7 PAP smears with HPV detection:
A:ASC-US; L: L-Sil; H: H-Sil
Last 3 PAP smears are Normal and HPV negative

Male partner (34 yrs)

- HPV 16
- HPV 31

9 Semen samples with HPV and DNA fragmentation detection
Last 5 semen samples were normal (WHO)

- Sperm concentration
- HPV virions per sperm ratio
- DFI%, DNA Fragmentation Index; % spermatozoa with measurable DNA damage
- HDS%; High DNA stainability; % spermatozoa with immature chromatin and abnormal proteins

HPV virions can bind the head of spermatozoa

- HPV virions bind the syndecan-1 receptor on sperm head

  Garolla et al., 2012, Hum Reprod;27:967-73.

- HPV virion binding causes sperm DNA damage

  Boeri et al., 2019. Hum Reprod;34:209-217.

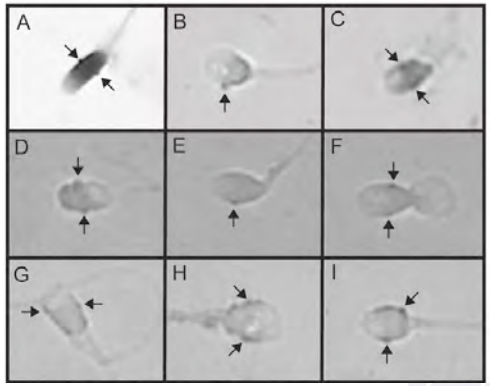
  Depuydt et al., 2021. J Clin Med;10:717.

- Spermatozoa can transfer HPV virions to the oocyte

  Foresta et al., 2011. PLoS One. 2011;6:e15036.

- The transferred HPV virions also induce stage-specific maturation arrest in infected embryos

  Henneberg et al., 2006. et al. J Assist Reprod Genet 2006;23:255-9.

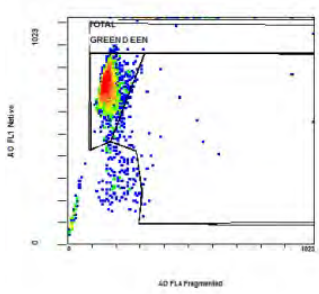


Kaspersen MD, Larsen PB, Ingerslev HJ, Fedder J, Petersen GB, Bonde J, Hollsberg P: Identification of multiple HPV types on spermatozoa from human sperm donors. PLoS One 2011;6:e18095.

Sperm chromatin structure assay (SCSA) measures sperm DNA damage

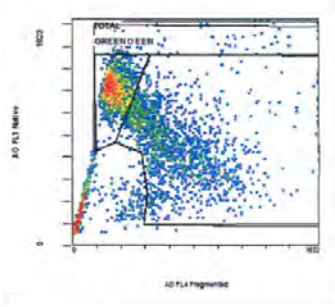
(%DFI; DNA Fragmentation Index; % spermatozoa with measurable DNA damage)
(%HDS; High DNA stainability; % spermatozoa with immature chromatin and abnormal proteins)

Donor (HPV-)



DFI% = 2,5%
HDS% = 0,5%

Subfertile patient (HPV+ in semen)



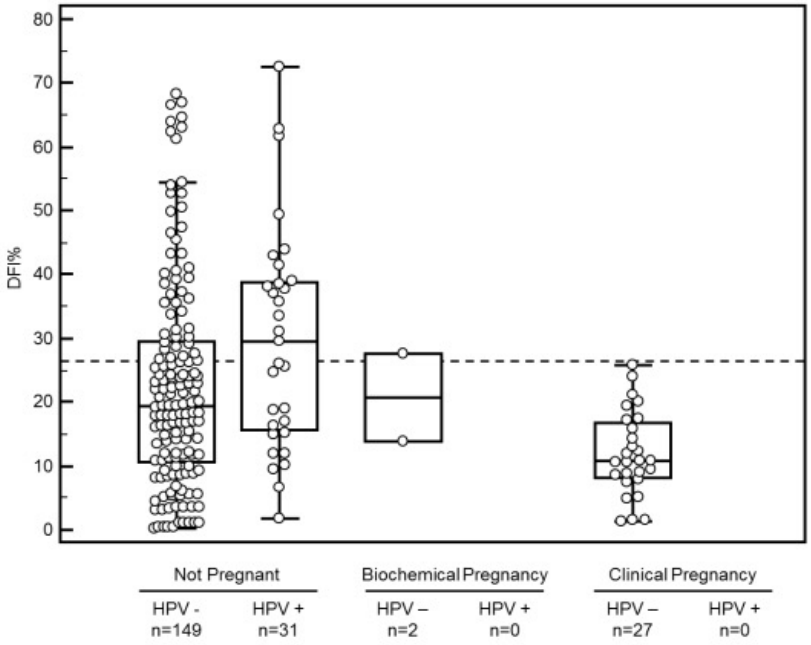
DFI% = 44,7%
HDS% = 2,0%

Cut-off clinical pregnancy for IUI: DFI% <25%; DFI% ≥25% → IVF/ICSI

Depuydt 2021 Negative Impact of Elevated DNA Fragmentation and Human Papillomavirus (HPV) Presence in Sperm on the Outcome of Intra-Uterine Insemination (IUI). J Clin Med. 2021; 10(4):717. doi: 10.3390/jcm10040717.

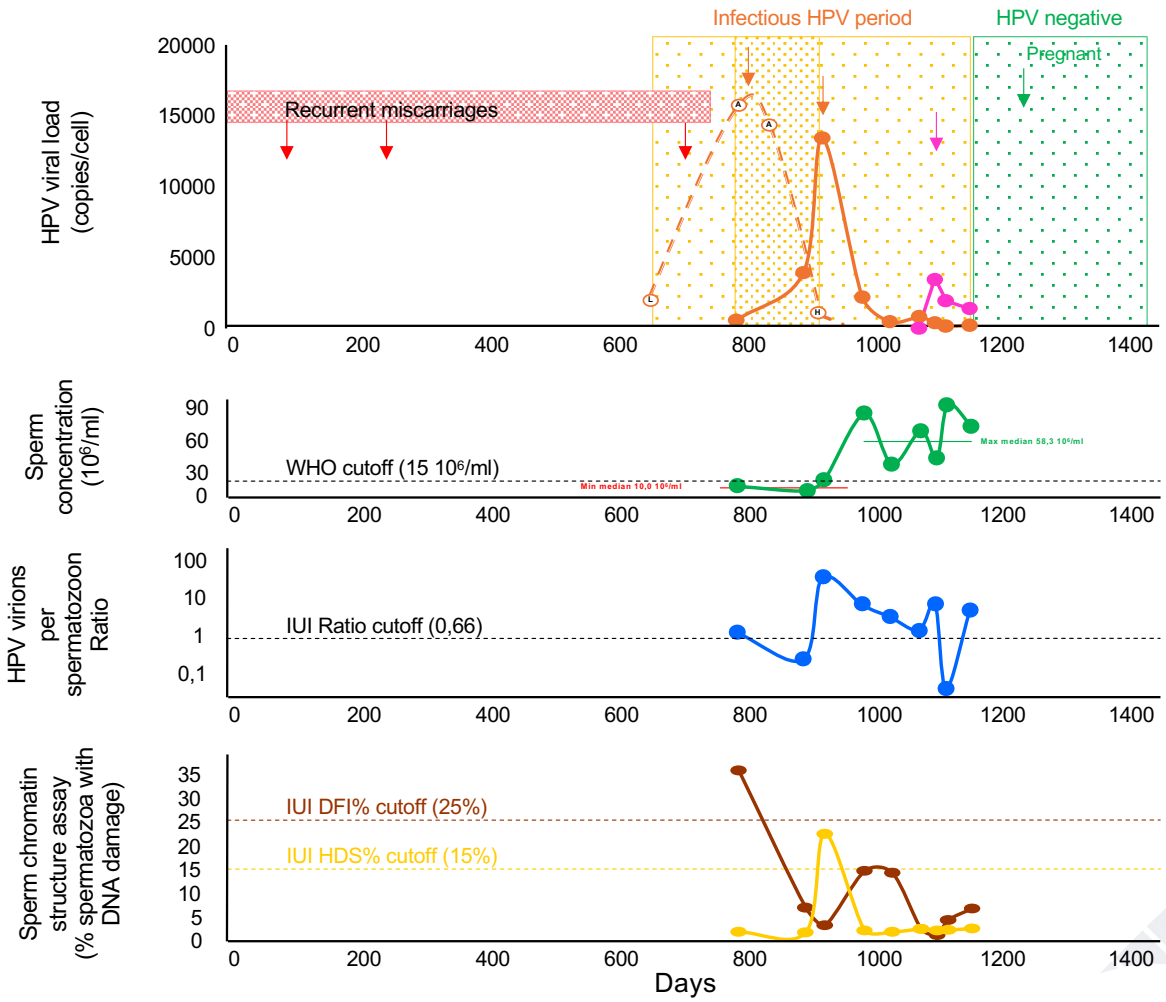
Depuydt 2018 Time has come to include Human Papillomavirus (HPV) testing in sperm donor banks. Facts Views Vis Obgyn, 2018, 10(4): 201-205.

DFI% versus pregnancy outcome and HPV status (DFI% cut-off)



No clinical pregnancies when DFI% is above 26% !

Depuydt et al 2021 Negative Impact of Elevated DNA Fragmentation and Human Papillomavirus (HPV) Presence in Sperm on the Outcome of Intra-Uterine Insemination (IUI). J. Clin. Med. 2021, 10, 717. https://doi.org/10.3390/jcm10040717



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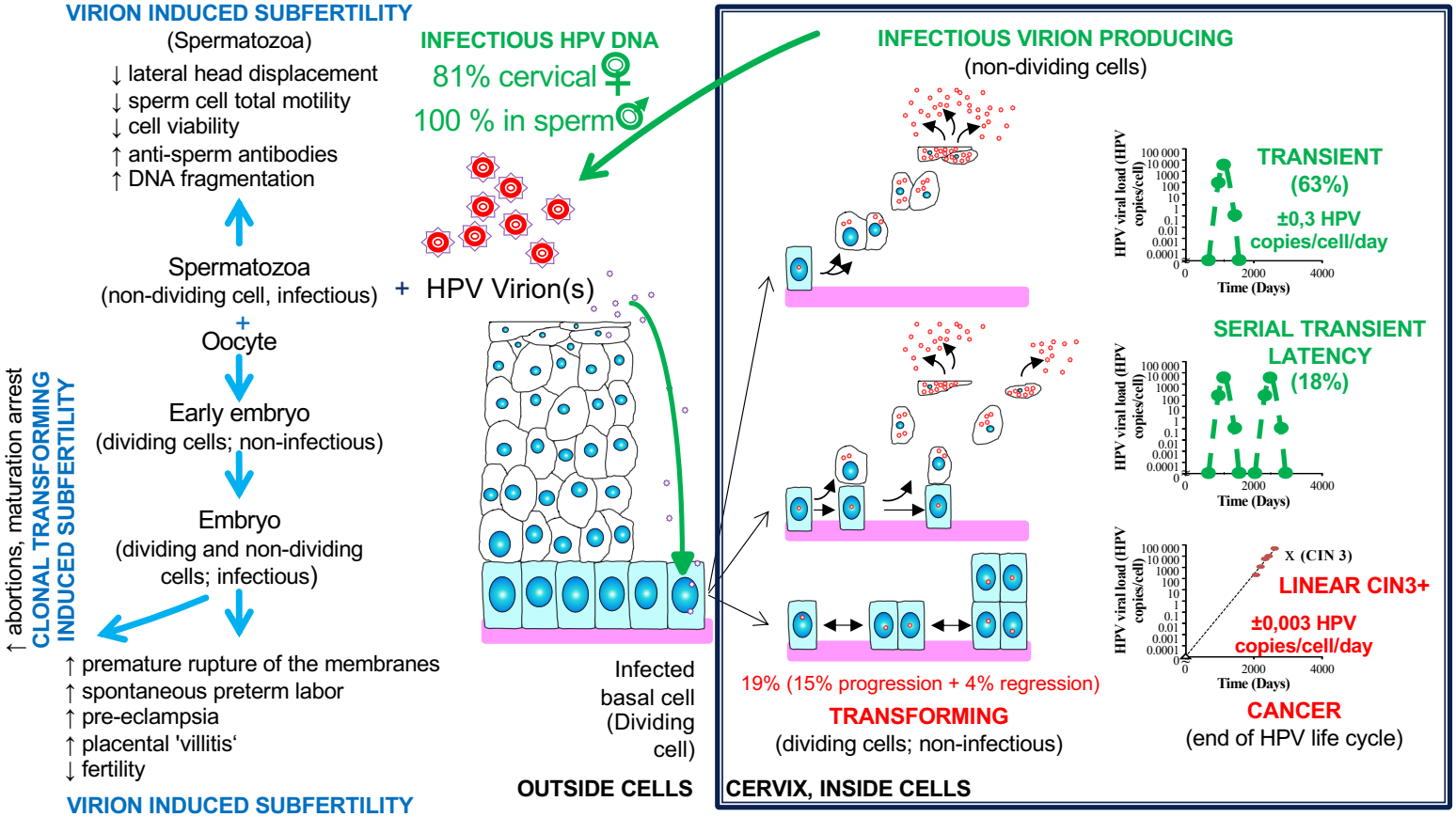
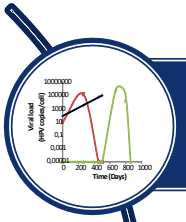
HPV induced processes: infectious virion producing pathway in non-dividing HPV DNA harboring cells and the clonal transforming pathway in dividing HPV DNA harboring cells

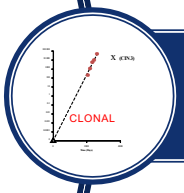


Depuydt et al 2016 Human Papillomavirus (HPV) virion induced cancer and subfertility, two sides of the same coin. Facts Views Vis Obgyn, 2016, (4): 211-222,

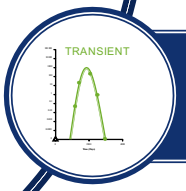
Conclusions



Le genotypage HPV en milieu liquide permet le tri chez les patientes entre infection et évolution clonale



Pendant le suivi la détection de CIN 3+ ne peut survenir que chez les femmes porteuses d'une infection clonale



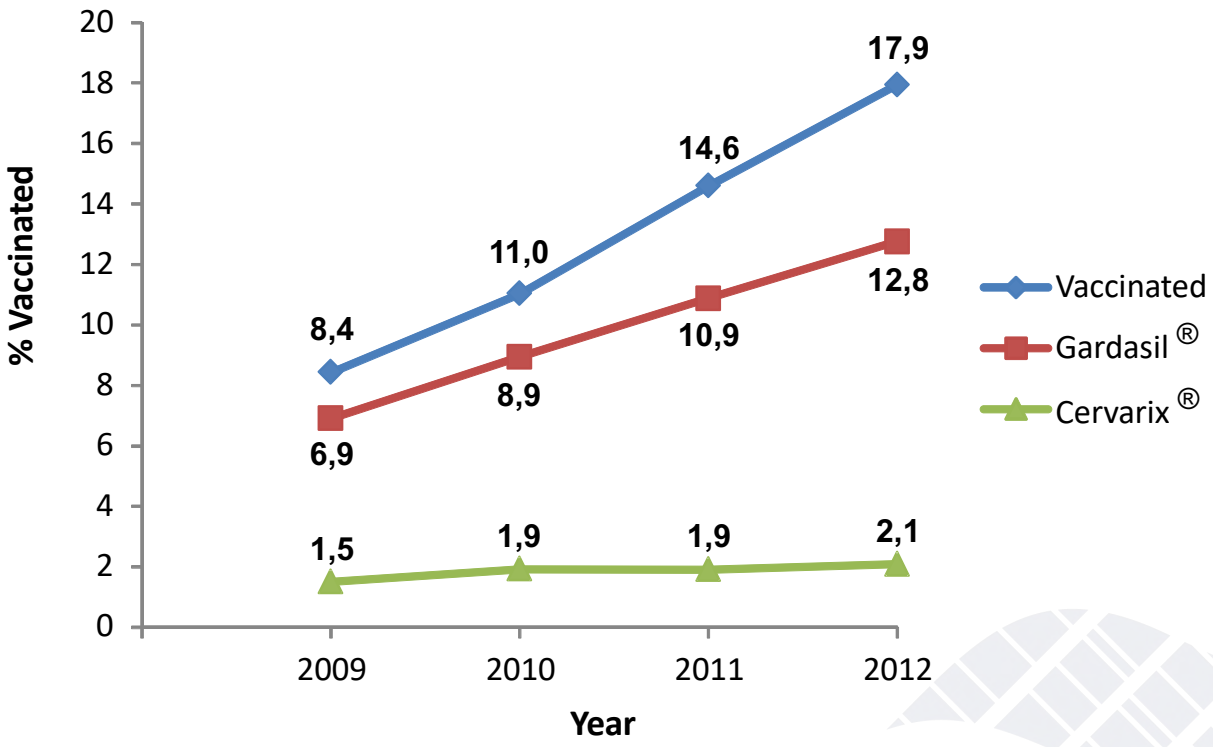
Les femmes avec une infection HPV transitoire se débarrasseront de leur infection et n'auront jamais de CIN 3

A été vaccinée il y a 15 ans et qui est HPV + ?

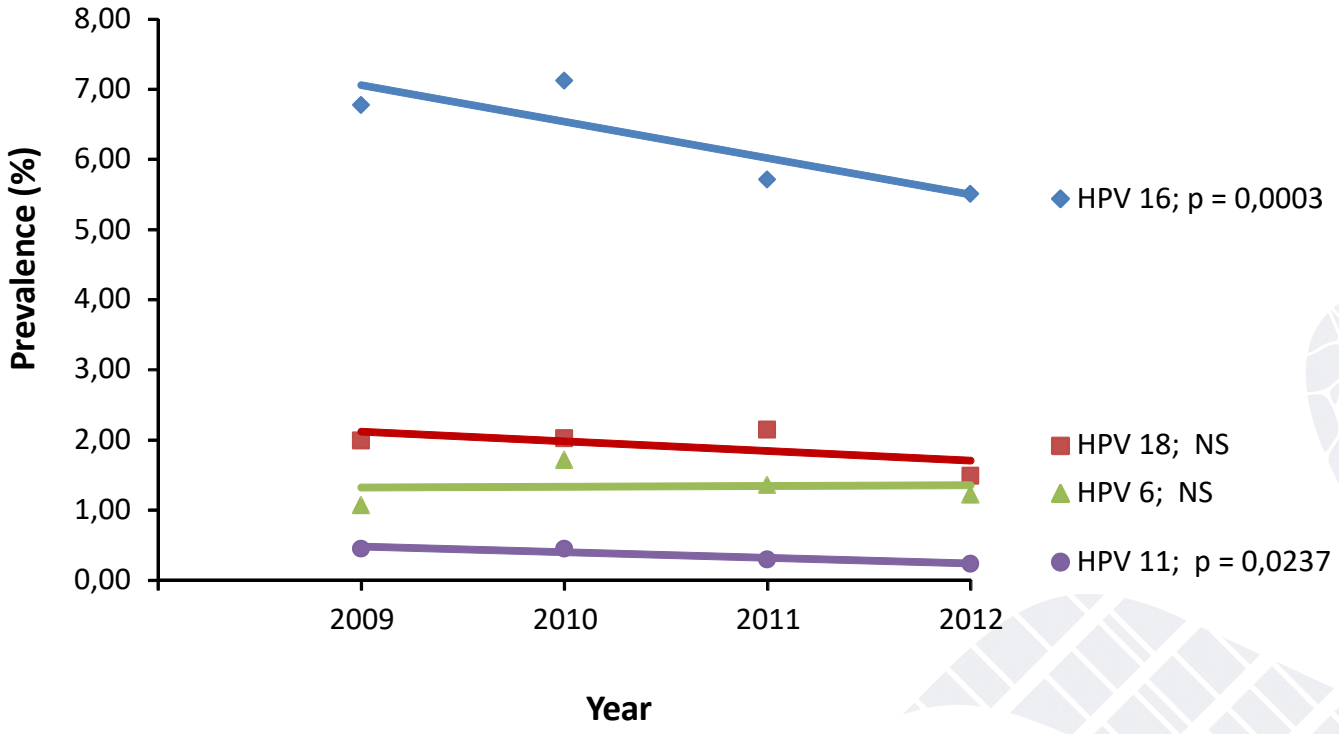
Vaccinated and unvaccinated women with screening smears between 07/2009 and 10/2012

Age	<25 yrs	40 - 45 yrs
	Vaccinated	Unvaccinated
2009	3573	3222
2010	7417	7826
2011	8490	10273
2012	5557	6534
Total	25037	27855

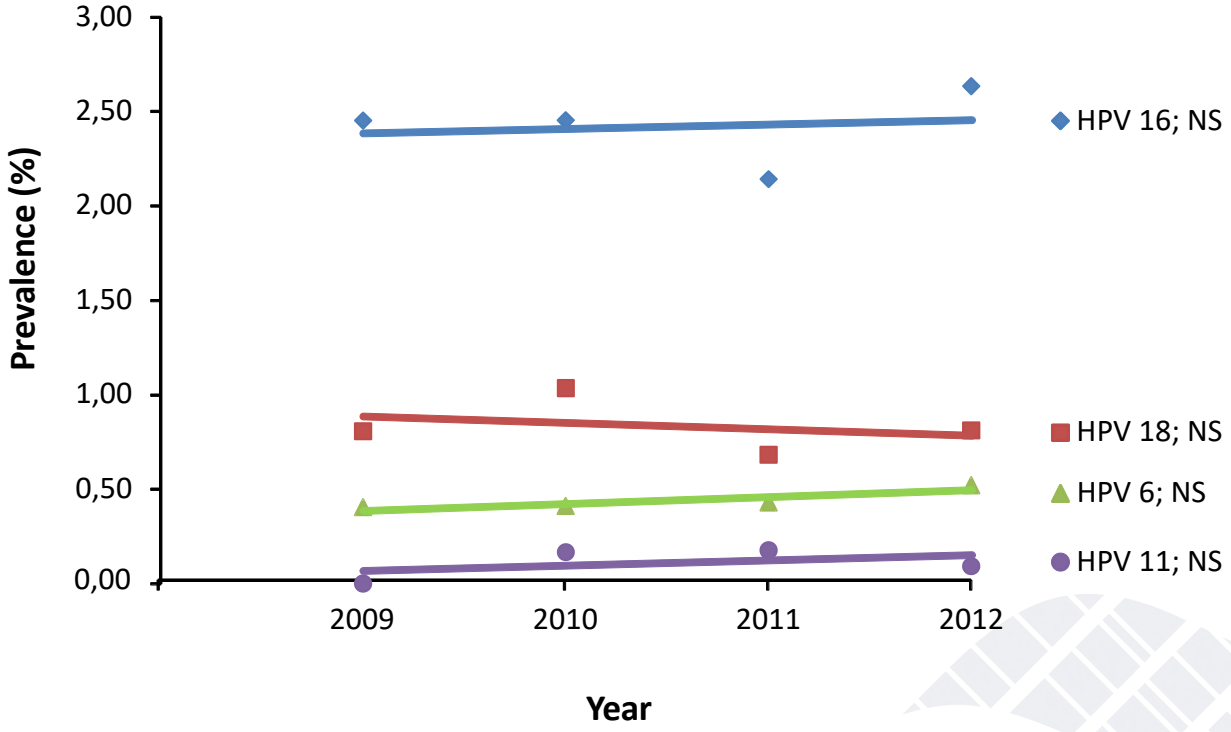
Vaccinated women under 25 years with screening smear (n = 25037)



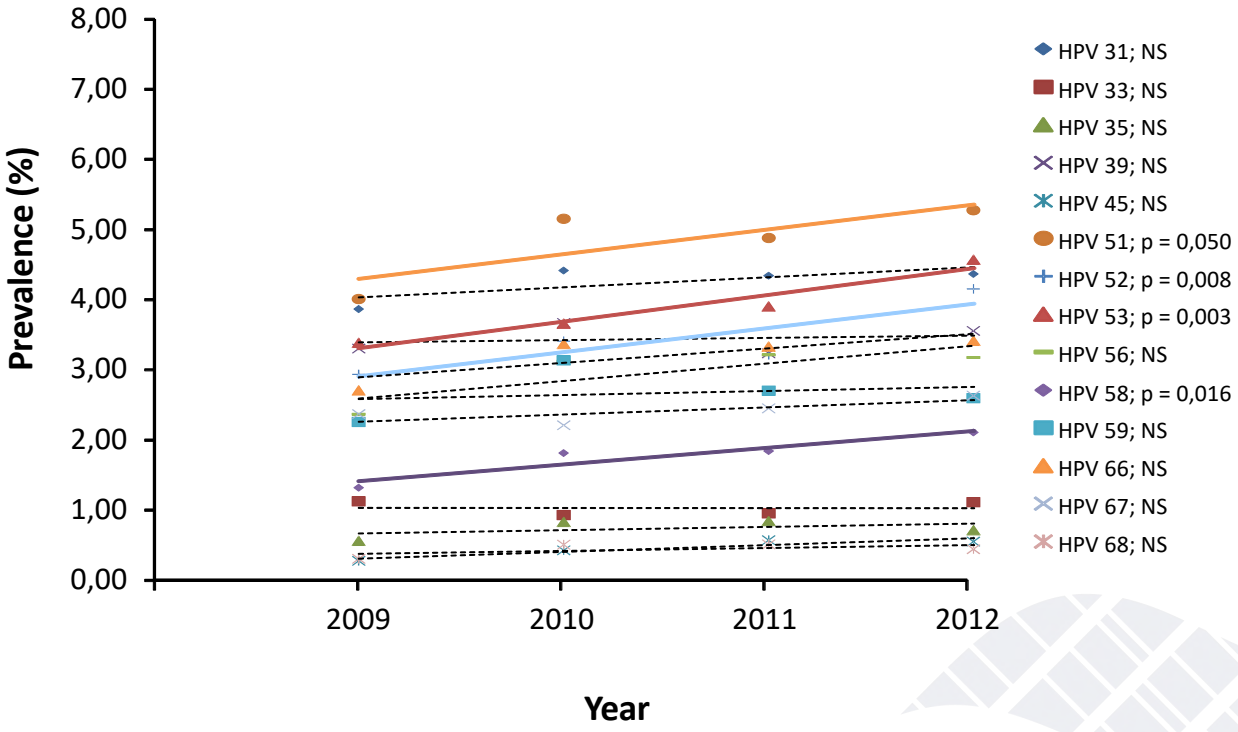
Prevalence of HPV-vaccine types in screening smears from women <25 years (VACCINATED)



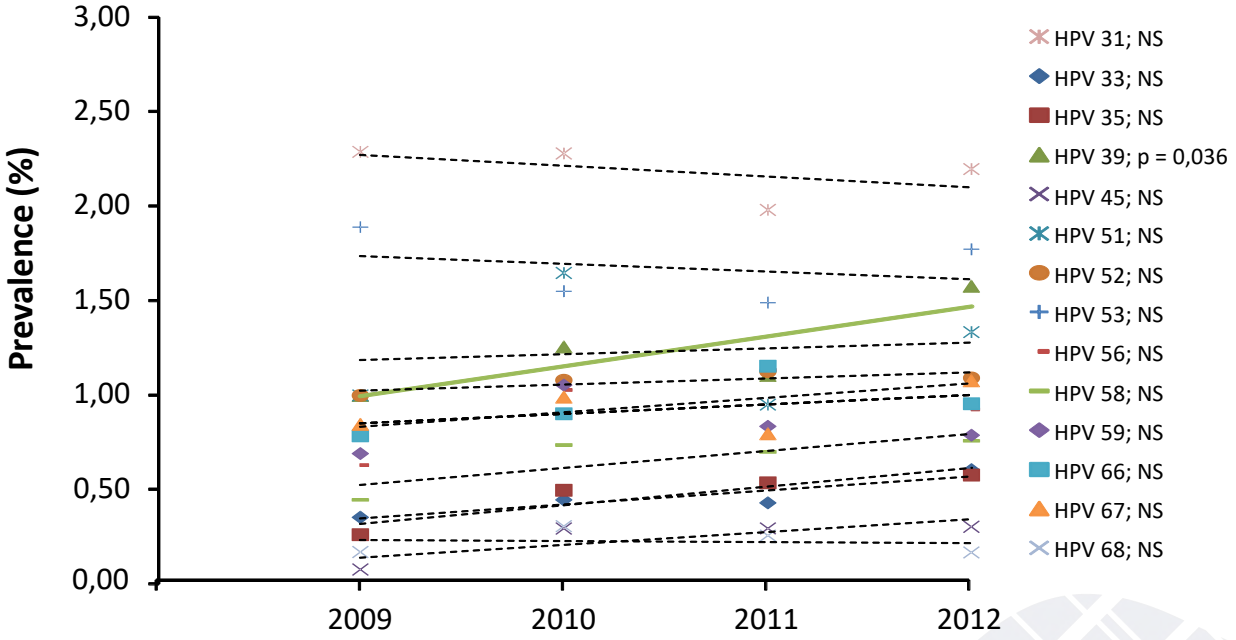
Prevalence of HPV-vaccine types in screening smears from women 40 - 45 years (UNVACCINATED)



Prevalence of non HPV-vaccine types in screening smears from women <25 years (VACCINATED)



Prevalence of non HPV-vaccine types in screening smears from women 40 - 45 years (UNVACCINATED)



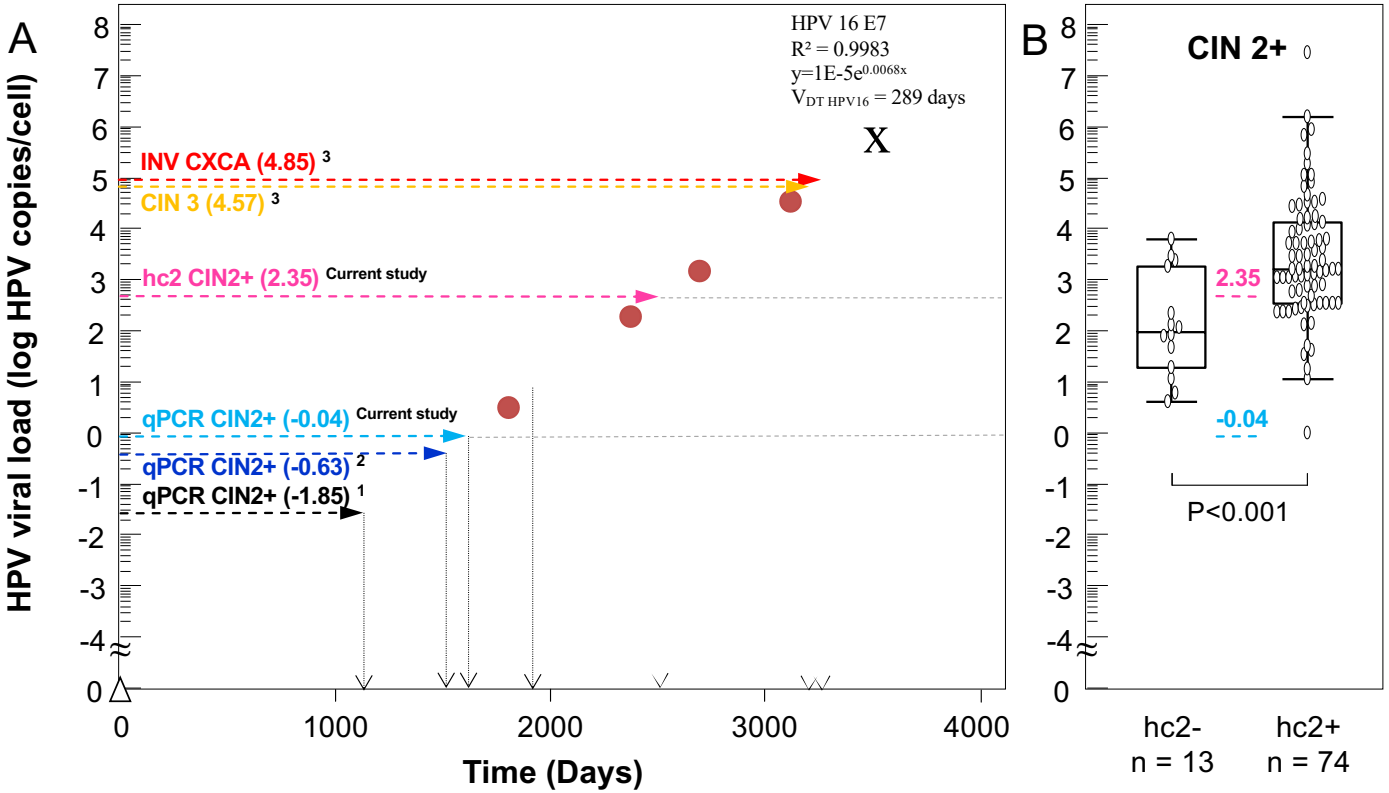
Conclusion

- Reduction in HPV-vaccine types only in vaccinated women (<25 yrs)
- Vaccination significantly reduced the prevalence of HPV 11 (48,9%) and 16 (18,6%)
- No significant reduction of HPV 6 and 18 prevalence
- No cross protection observed for any non HPV-vaccine types
- Type replacement in vaccinated group, significant increase in HPV 51, 52, 53 and 58 prevalence

Se pose la question d’un cancer ORL ou du canal anal ?

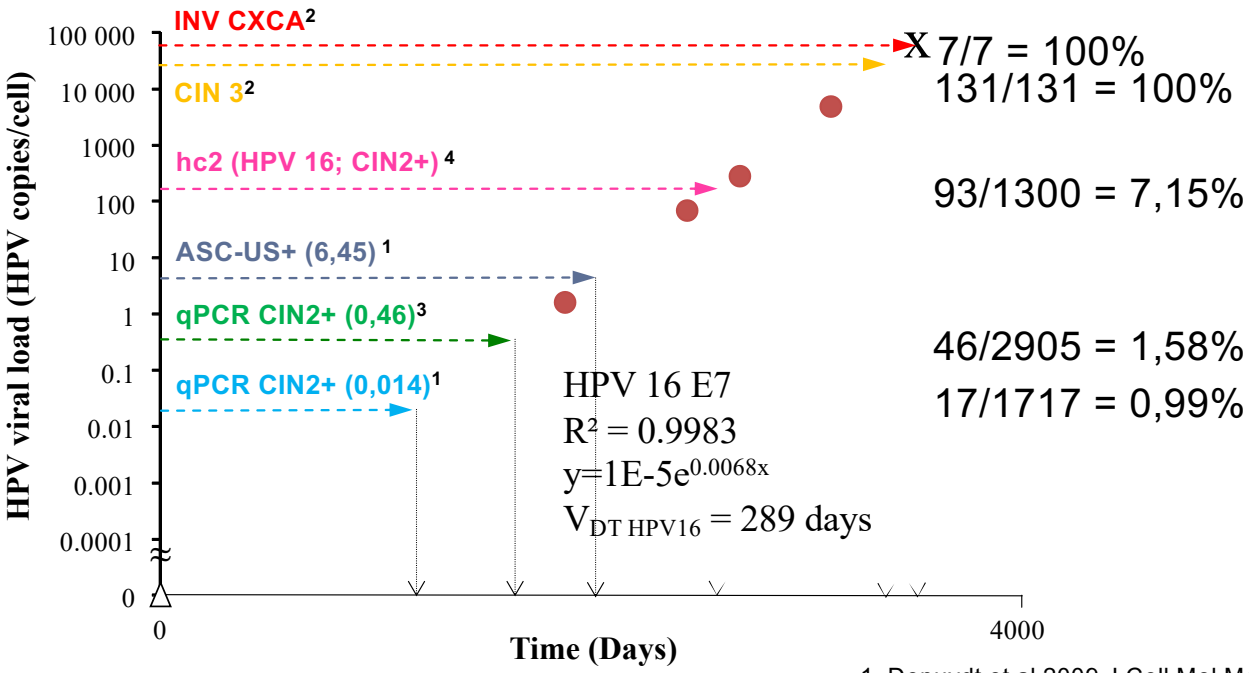
Demande combien coûte cet examen ?

Viral load cuttoff hc2 vs qPCR



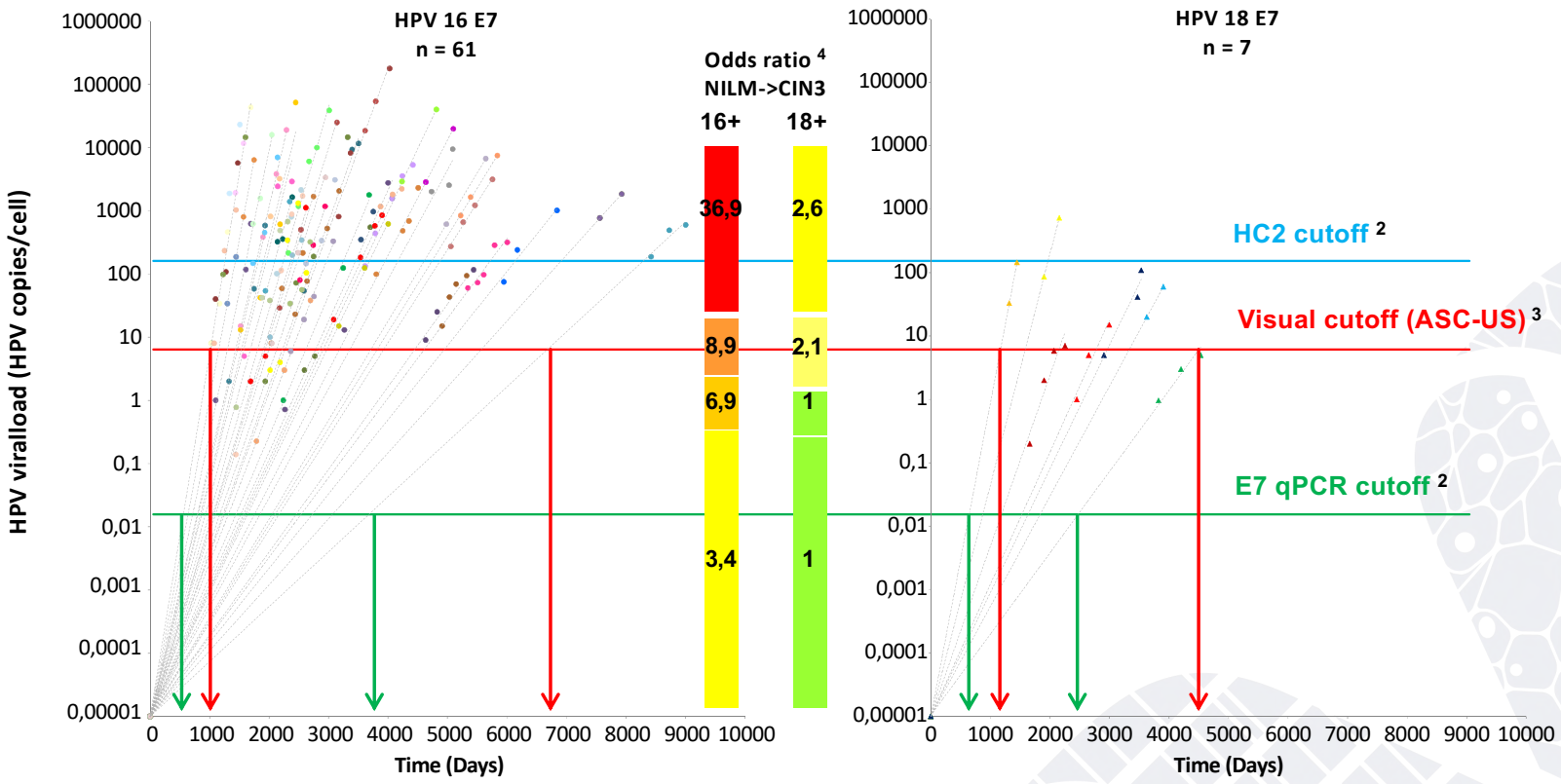
A linear increase of viral load in time according to y = 1E-5e$^{0.0068x}$, in function of different cutoffs. Circles represent viral load measurements in the linear phase with $R^2 > 0.9$, prior to detection of CIN3+. x = detection of CIN3+. Triangle represents the calculated starting point of the linear increase leading to CIN3+ with viral load of 0.00001 HPV copies/cell. 1) Depuydt et al 2009, J Cell Mol Med; 13: 4051-60. 2) Depuydt et al 2011, CEBP; 3) Depuydt et al 2012 J, Cell Mol Med **B** log hr viral load of CIN2+ cases for hc2 negative (n = 13) and hc2 positive cases (n = 74). For hc2 ROC curve analysis showed a cut-off for CIN2+ at 2.35 log hrHPV copies/cell with a sensitivity of 90.5 and specificity of 69.2 with an area under the ROC curve of 0.791 and a 95%CI between 0.691 to 0.871.

Viral load cut off for detection of CIN2+ is dependent on the ratio of CIN2+ vs non-CIN2+ cases



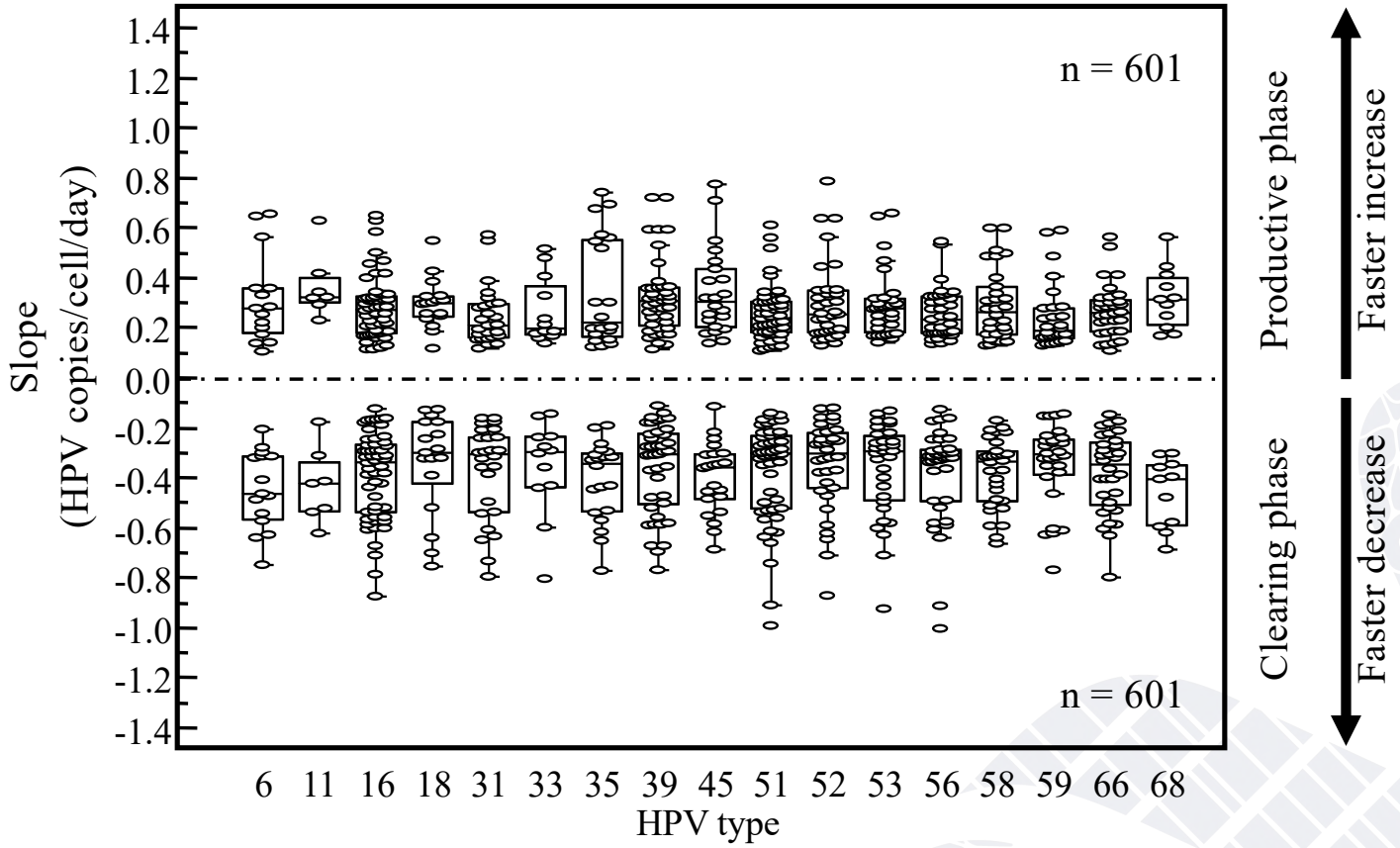
1. Depuydt et al 2009 J Cell Mol Med; 13: 4051-60.
2. Depuydt et al 2012 J Cell Mol Med; 16: 3096-3104.
3. Depuydt et al 2011 CEBP; 20:628-637.
4. Depuydt et al 2012 JCM; 50(12):4073.

Augmentation linéaire des cas HPV 16 et HPV 18 CIN3+[1]



1. Depuydt et al 2012. Changes in Type-specific Human Papillomavirus Load Predict Progression to Cervical Cancer. J Cell Mol Med. 2012;16(12):3096-104
2. Depuydt et al 2012. Clinical Validation of a Type-Specific Real-Time Quantitative Human Papillomavirus PCR against the Performance of Hybrid Capture 2 for the Purpose of Cervical Cancer Screening. J Clin Microbiol. 2012;50(12):4073-7.
3. Depuydt et al 2009. Quality control for normal liquid-based cytology: rescreening, high-risk HPV targeted reviewing and/or high-risk HPV detection? J Cell Mol Med. 2009;13(9B):4051-60.
4. Moberg et al 2004. Type-specific associations of Human Papillomavirus load with risk of developing cervical carcinoma in situ. Int J Cancer 2004;112:854-859,

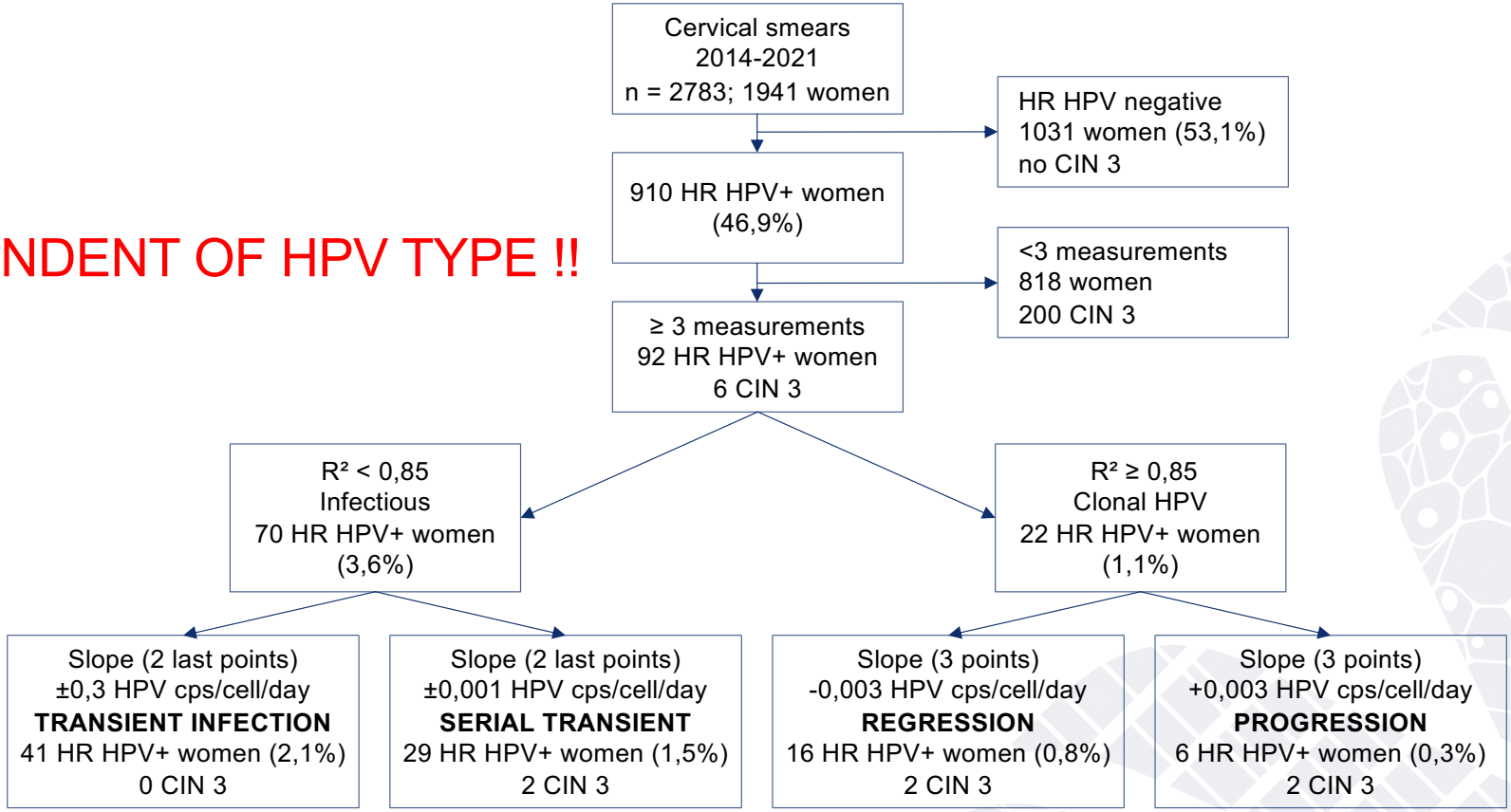
Slope in Productive and Clearing Phase of Transient Infections by HPV type



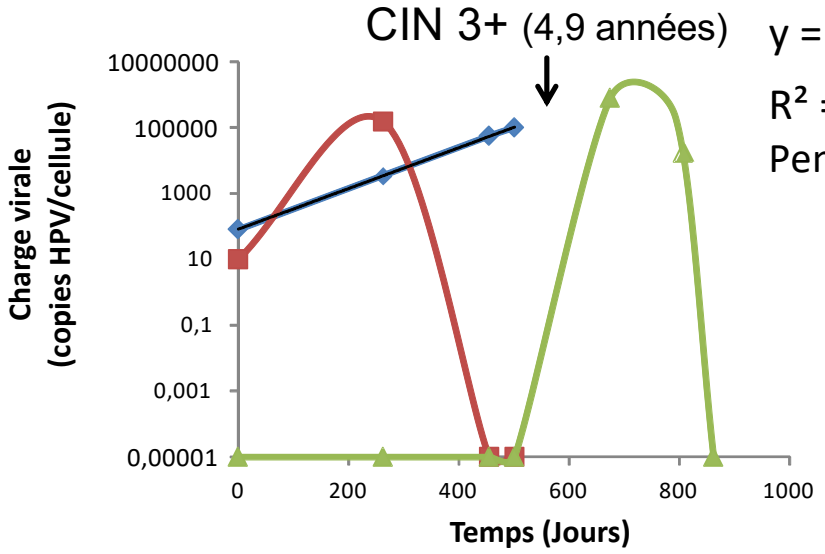
Depuydt et al 2012. Changes in Type-specific Human Papillomavirus Load Predict Progression to Cervical Cancer. J Cell Mol Med. 2012;16(12):3096-104
Depuydt et al 2016. Linear viral load increase of a single HPV-type in women with multiple HPV infections predicts progression to cervical cancer. Int J Cancer. 2016;139(9):2021-32.

Prevalence of the 4 different serial type specific viral load measurement patterns in cervical smears

!! INDEPENDENT OF HPV TYPE !!



Infections transitoires et persistantes avec plusieurs types d'HPV (CIN3+)



$y = 81{,}263e^{0{,}0143x}$

$R^2 = 0{,}9999$

Pente = 0,0061 copies HPV/cellule/jour

HPV 16 E7

HPV 51 E7

HPV 56 E7