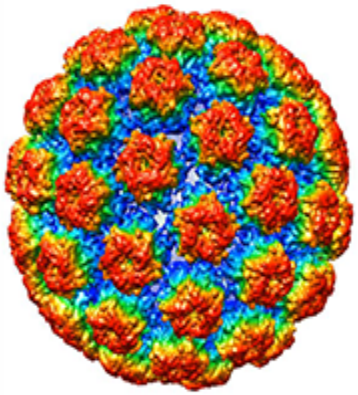
Cervical Cancer

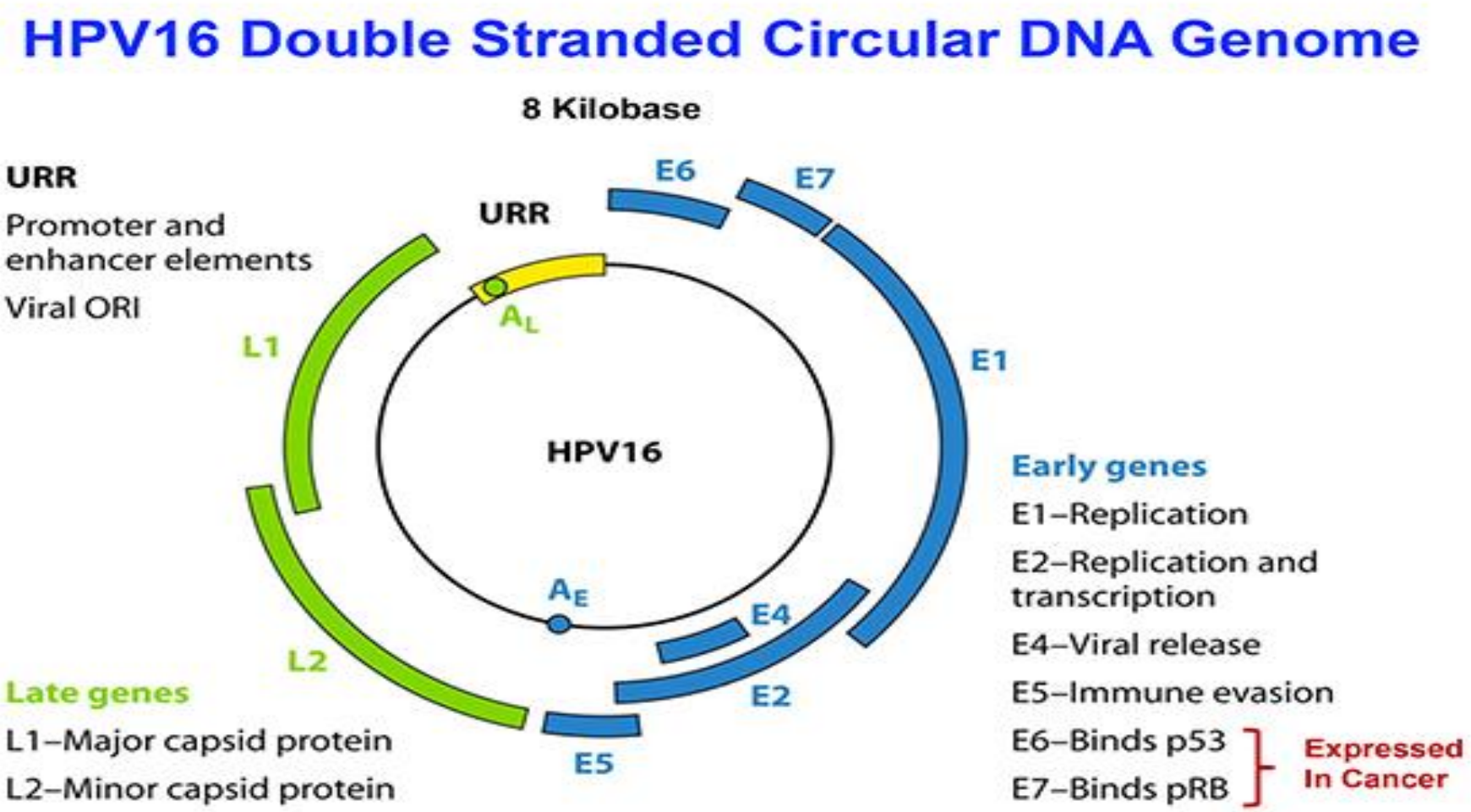
HPV Vaccines to Prevent Cervical Cancer and other HPV-associated Diseases

John Schiller, Center for Cancer Research, NCI



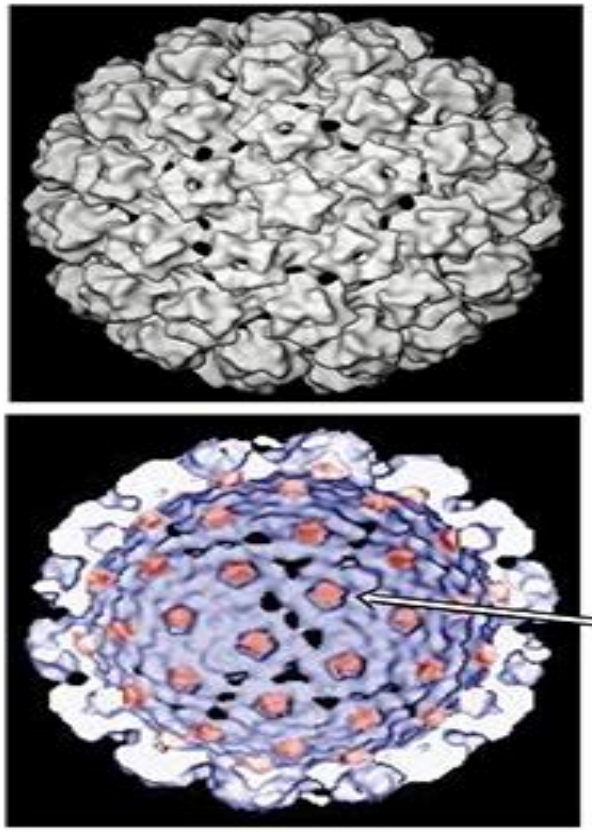
- HPV and Cancer
- Vaccine Efficacy/Effectiveness
- Key Implementation Issues
- Why they work so well

HIV genome


HPV16 Double Stranded Circular DNA Genome
8 Kilobase
URR
Promoter and enhancer elements
Viral ORI
URR
E6
E7
E1
L1
A_L
HPV16
A_E
E4
E2
L2
E5
Late genes
L1–Major capsid protein
L2–Minor capsid protein
Early genes
E1–Replication
E2–Replication and transcription
E4–Viral release
E5–Immune evasion
E6–Binds p53
E7–Binds pRB
Expressed In Cancer

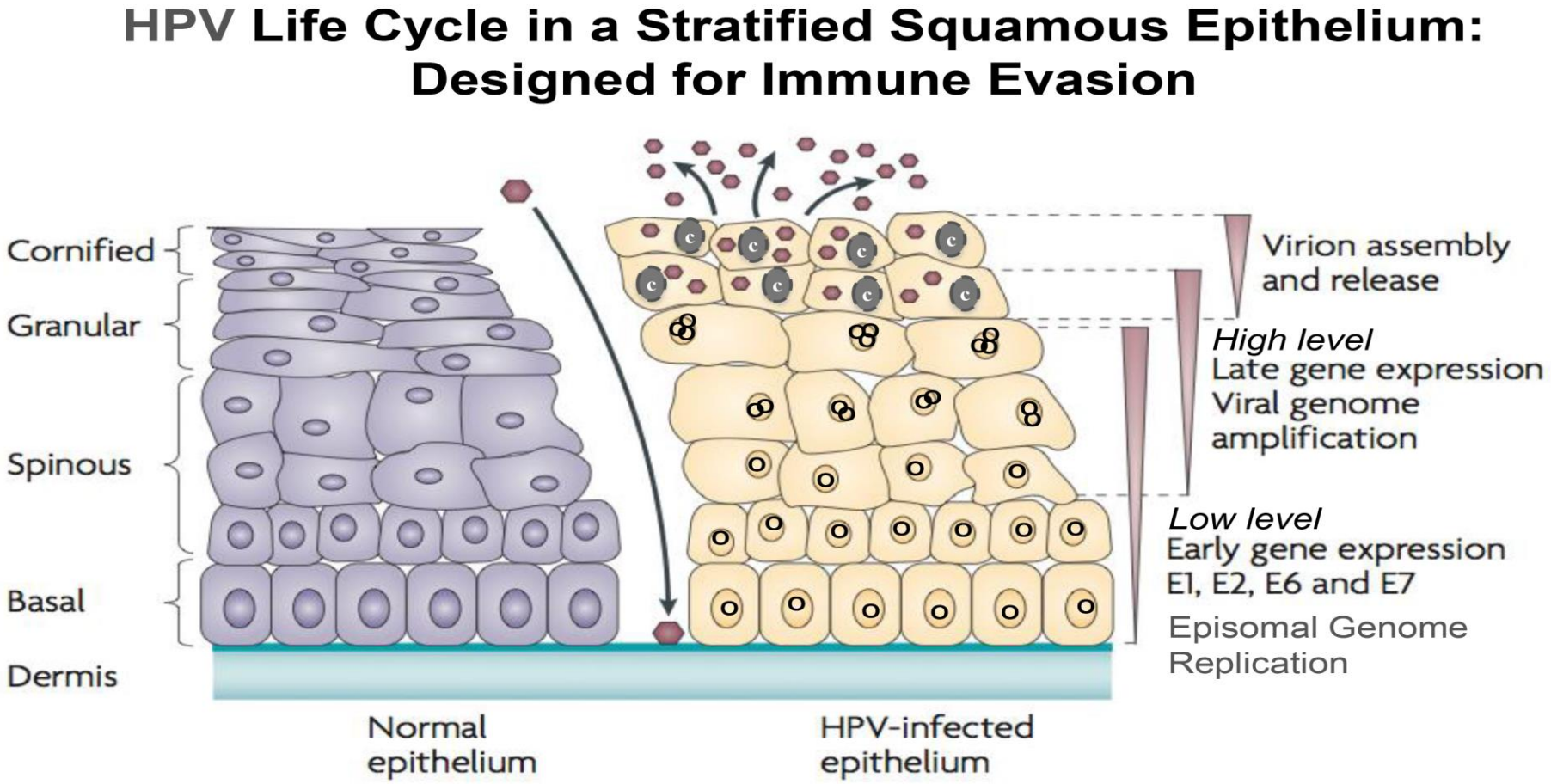
Virion

Papillomavirus Virion



- Non-enveloped icosahedral shell formed by 72 pentamers of L1
- 60 nanometer diameter
- A second capsid protein L2 is present at up to 72 copies
- 8kb circular dsDNA genome (chromatinized)

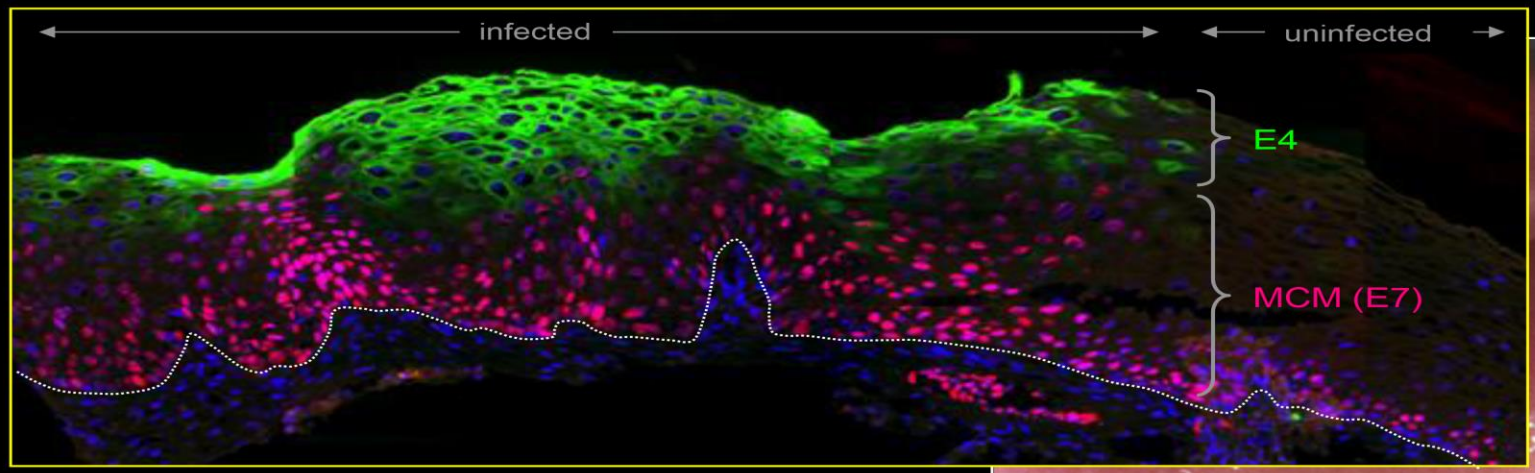
HPV life cycle

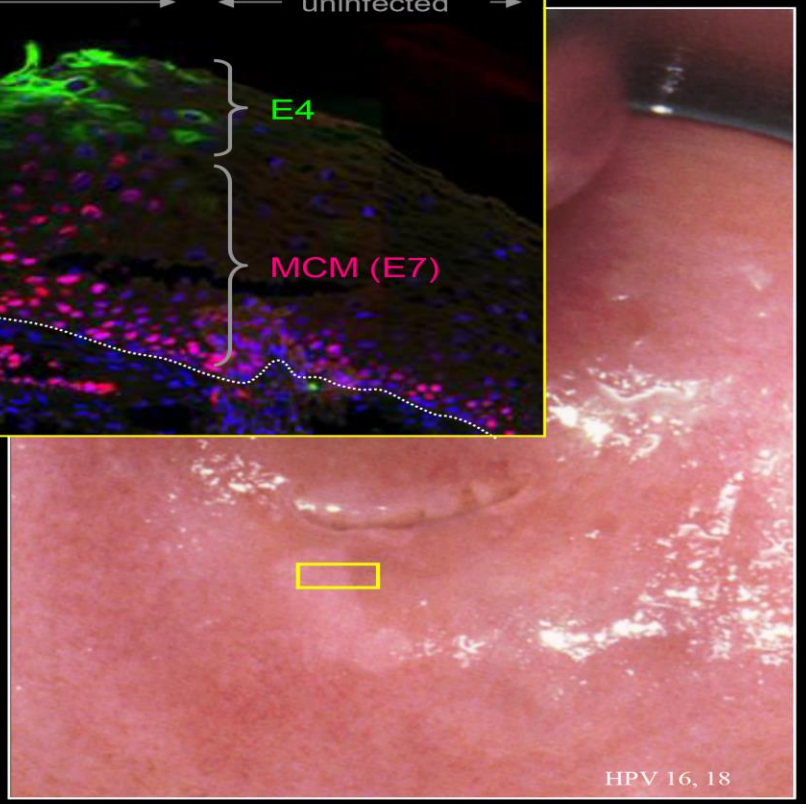


Moody and Laimins Nat Rev Micro 2010

HPV infection

Productive HPV Infection: Hiding in Plain Site



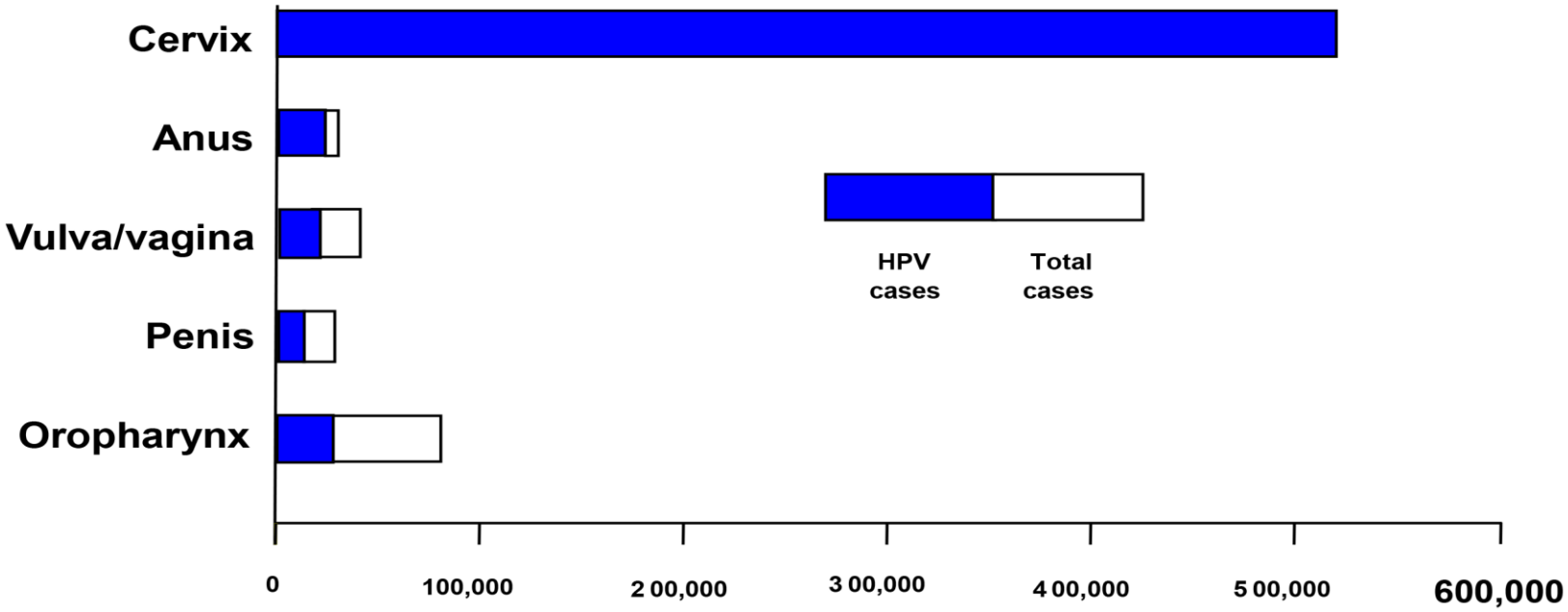


HPVs have evolved to exploit the limited immuno-surveillance of the upper layers of skin and mucosal membranes.

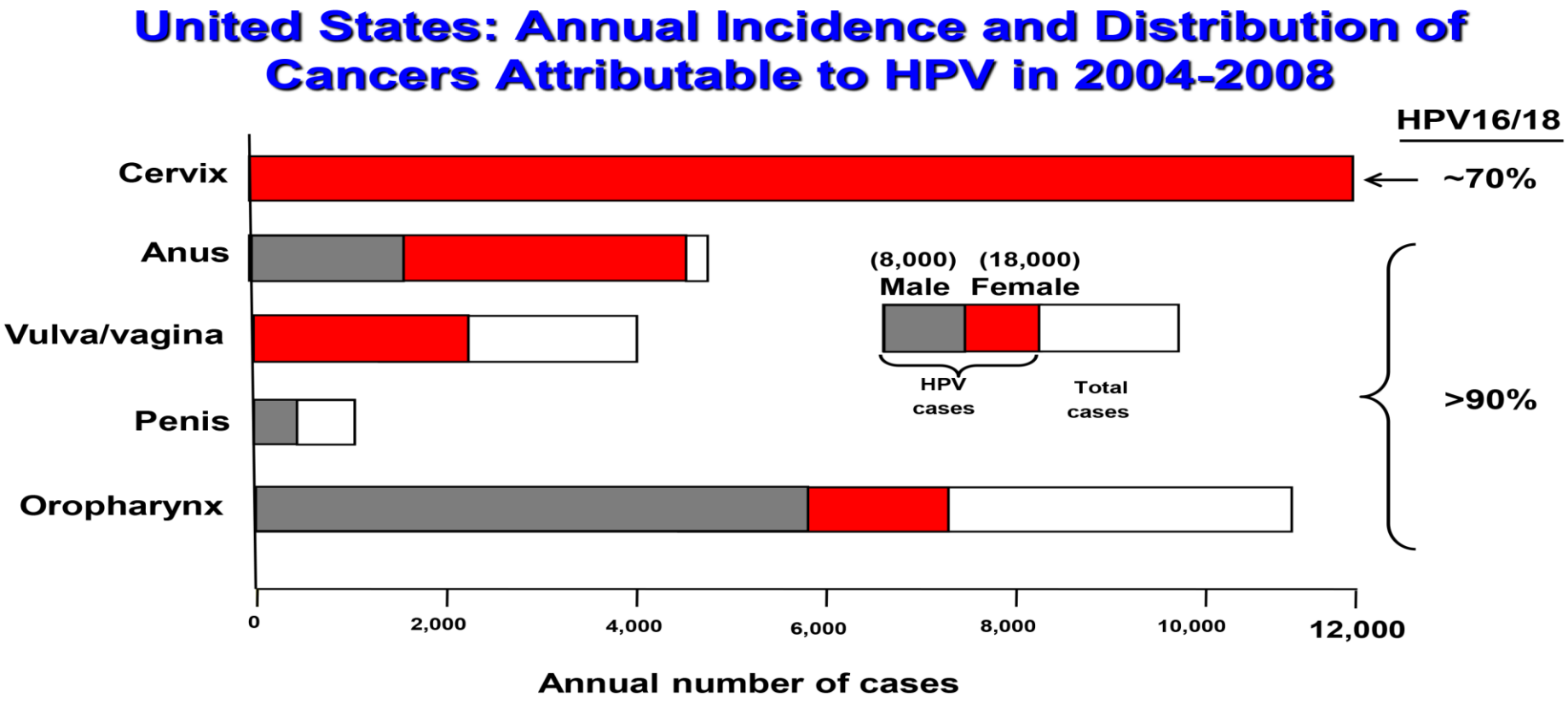
John Doorbar, Heather Griffin, NIMR, London

Cancers and HPV

Worldwide Incidence and Distribution of Cancers Attributable to HPV



HPV cancers

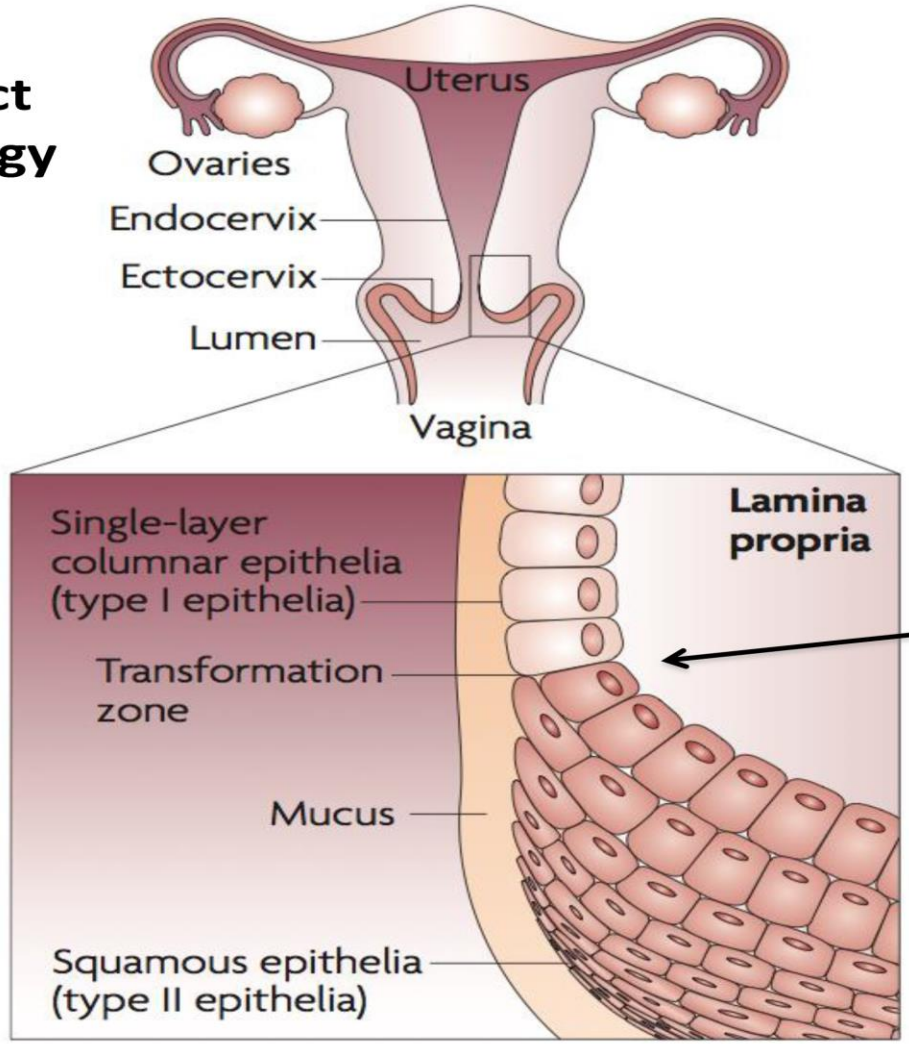


- Pap screening has reduced the incidence of cervical cancer by ~ 80%
- Incidence of HPV-positive oropharynx cancer 1988-2004 increased 225%

MMWR Weekly 61:253-80, 2012

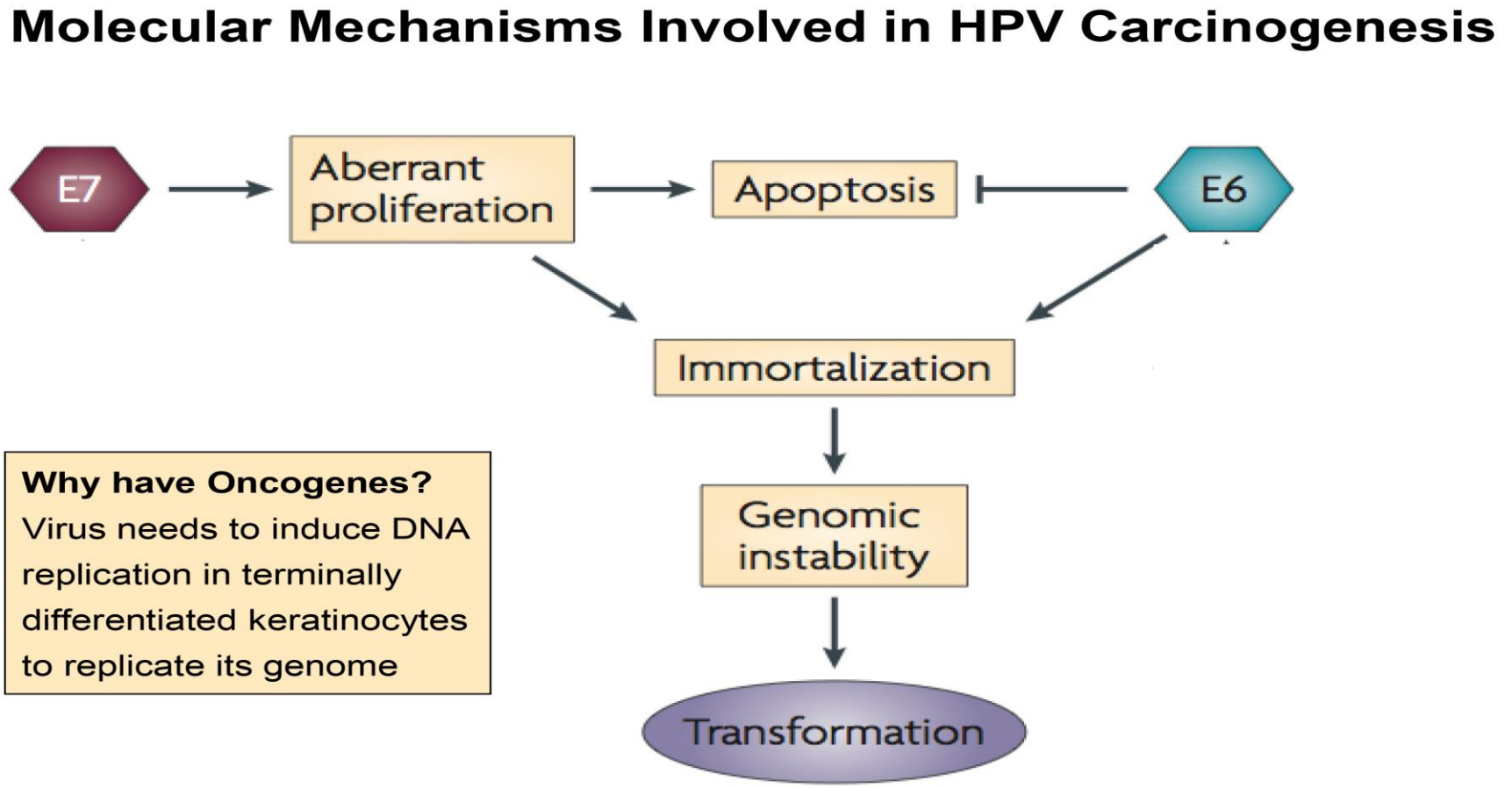
Cervical cancer

Female Reproductive Tract Anatomy & Histology



Iwasaki A Nat. Rev. Imm. 2010; 10:699.

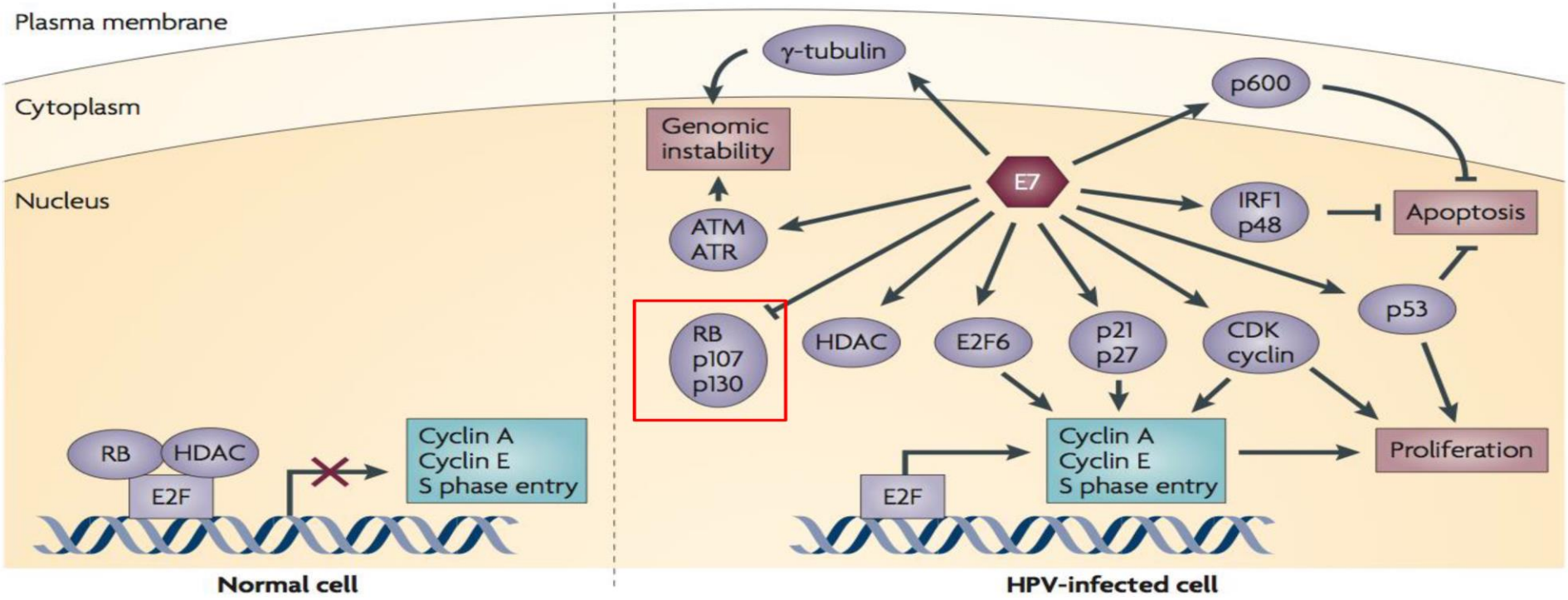
HPV carcinogenesis



Moody and Laimins Nat Rev Micro 2010

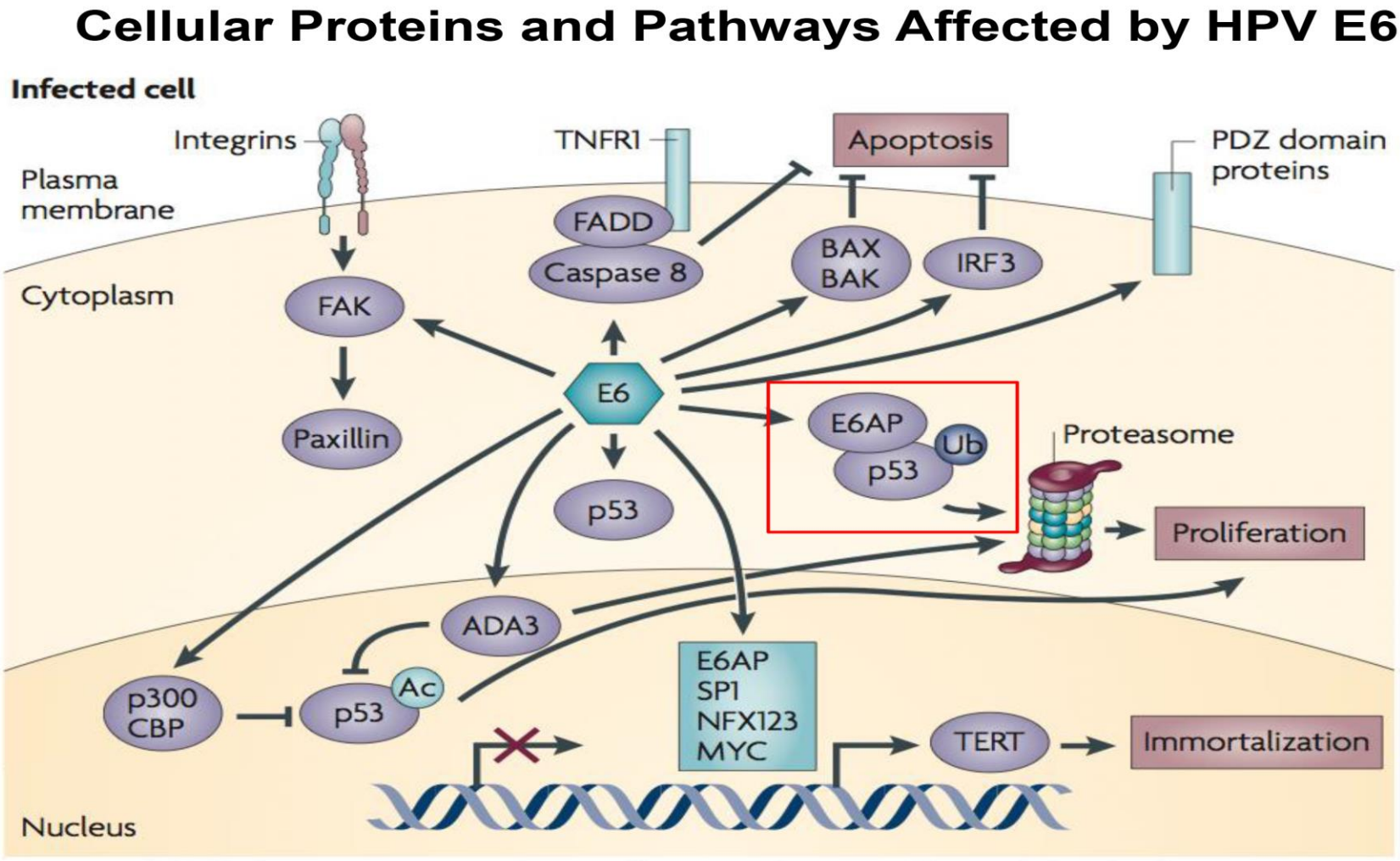
Cellular proteins

Cellular Proteins and Pathways Affected by HPV E7



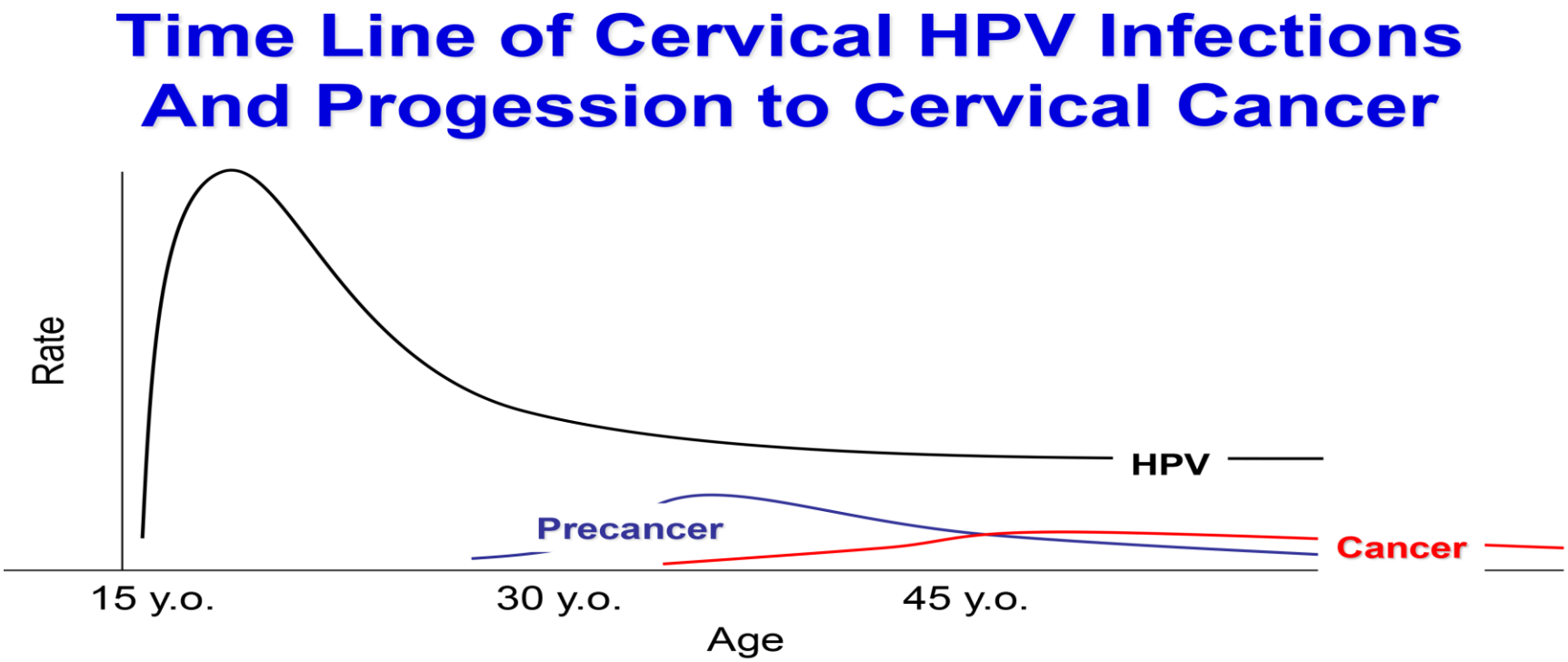
Moody and Laimins Nat Rev Micro 2010

HPV pathways



Moody and Laimins Nat Rev Micro 2010

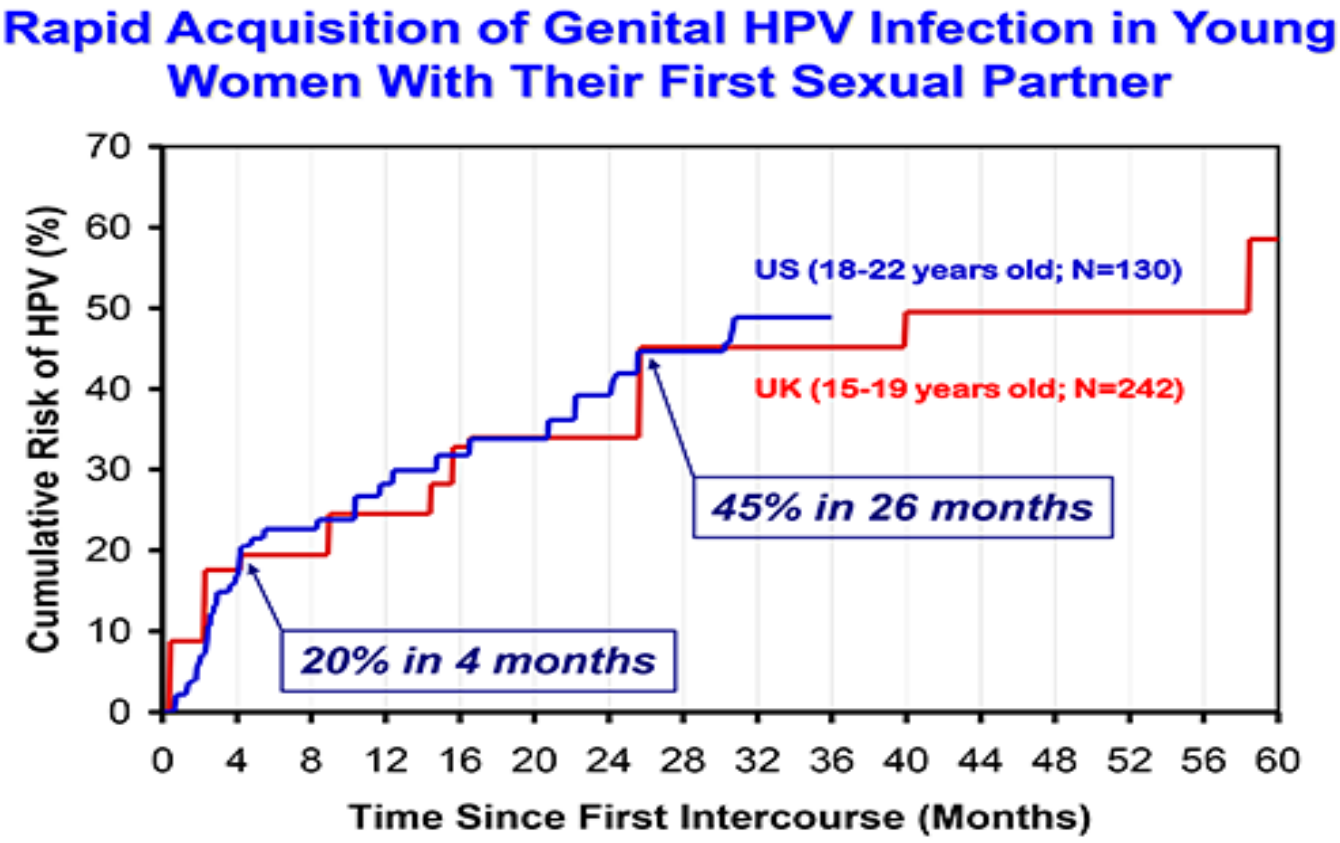
HPV infection time line



- **Lifetime incidence of genital HPV infection >80% in U.S.**
- **Most infections clear spontaneously, eliminating cancer risk for that infection.**
- **Persistent infection with a high-risk HPV, especially HPV16 or 18, is the single most important risk factor for progression to precancer and cancer.**

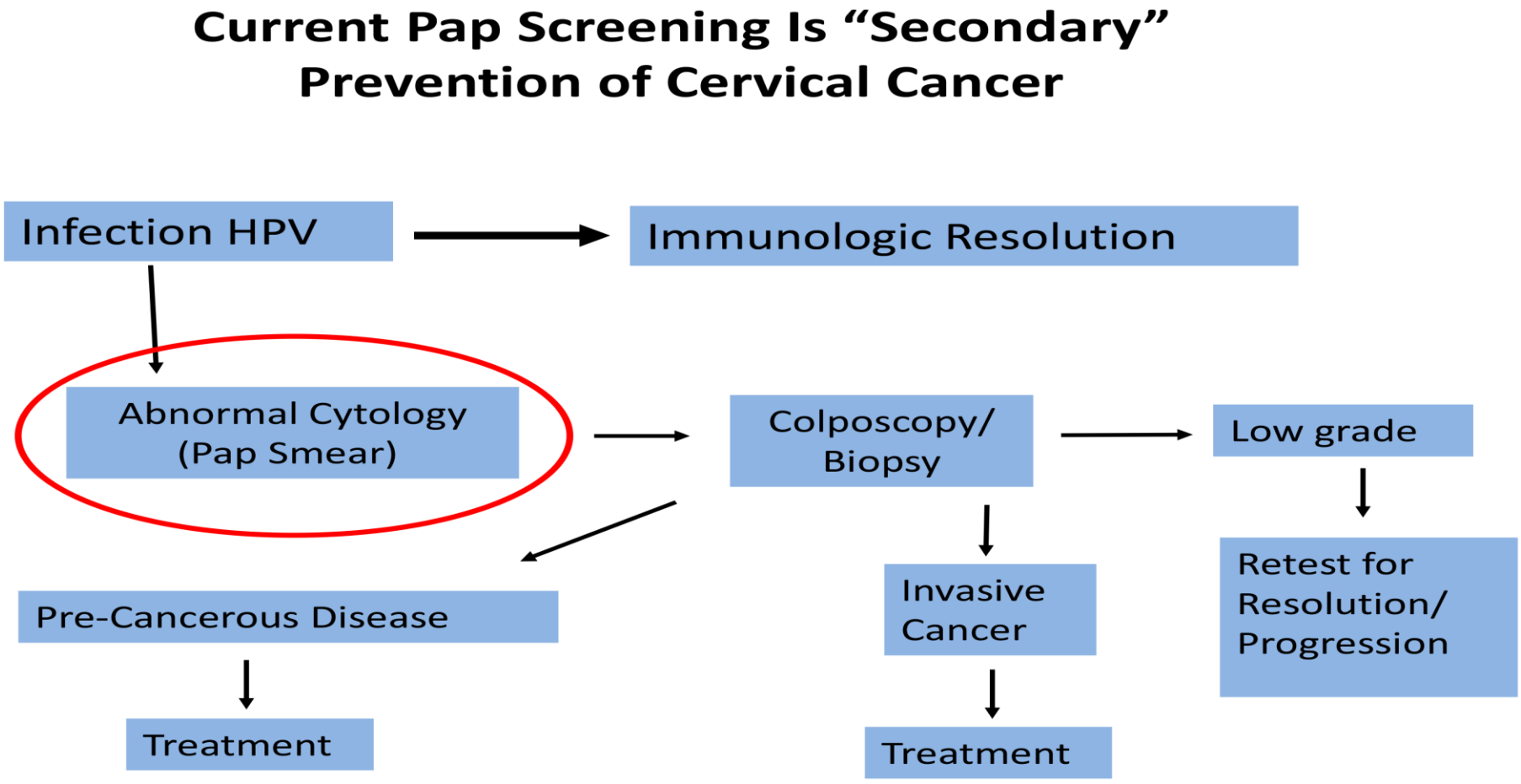
Adapted from Schiffman & Castle, New Eng J Med 353:2101-4, 2005

HPV infection

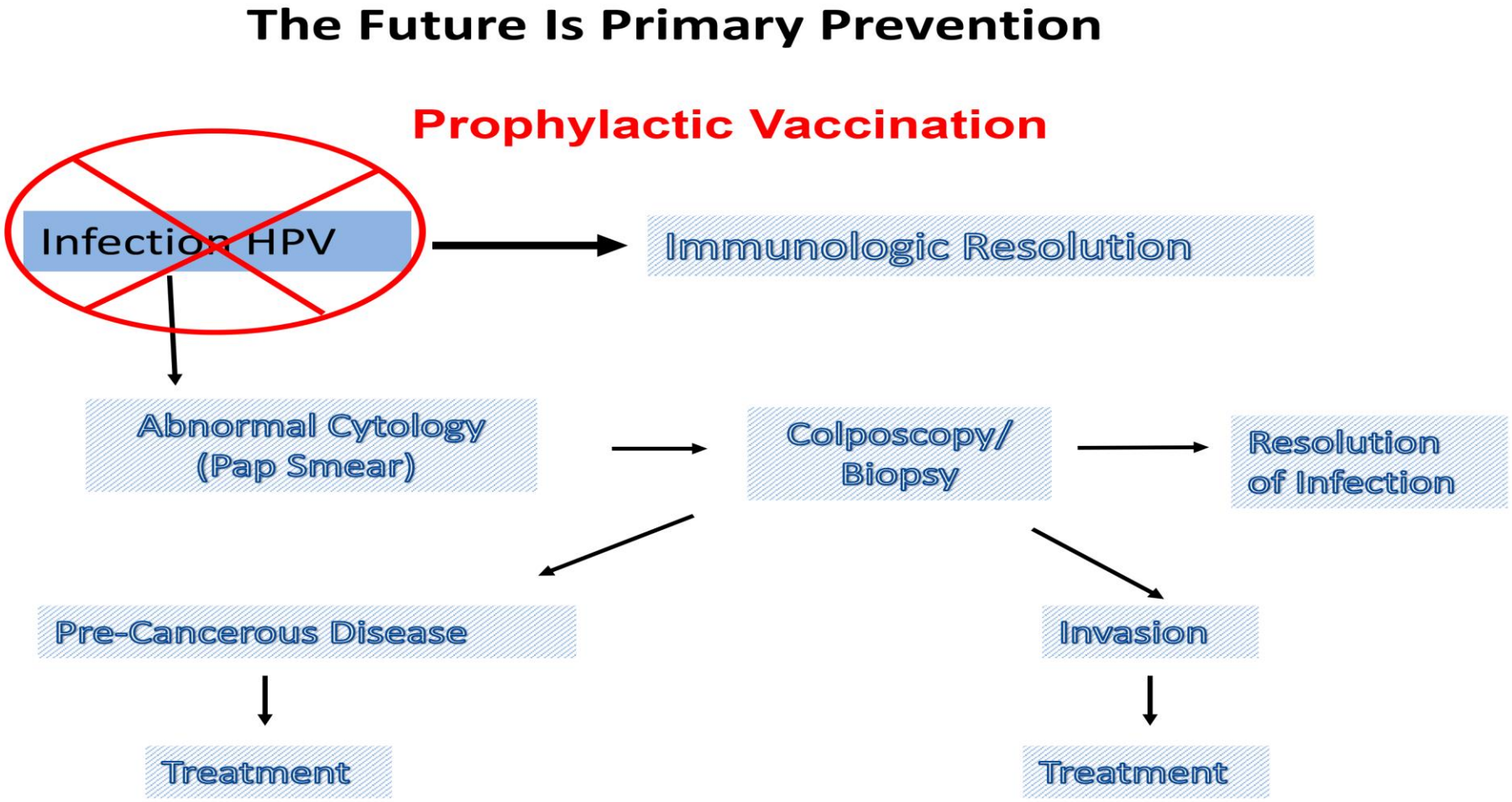


UK data adapted from *Collins et al, BJOG 109: 96-98, 2002* US data adapted from *Winer et al, J Inf Dis 197:279-282, 2008*

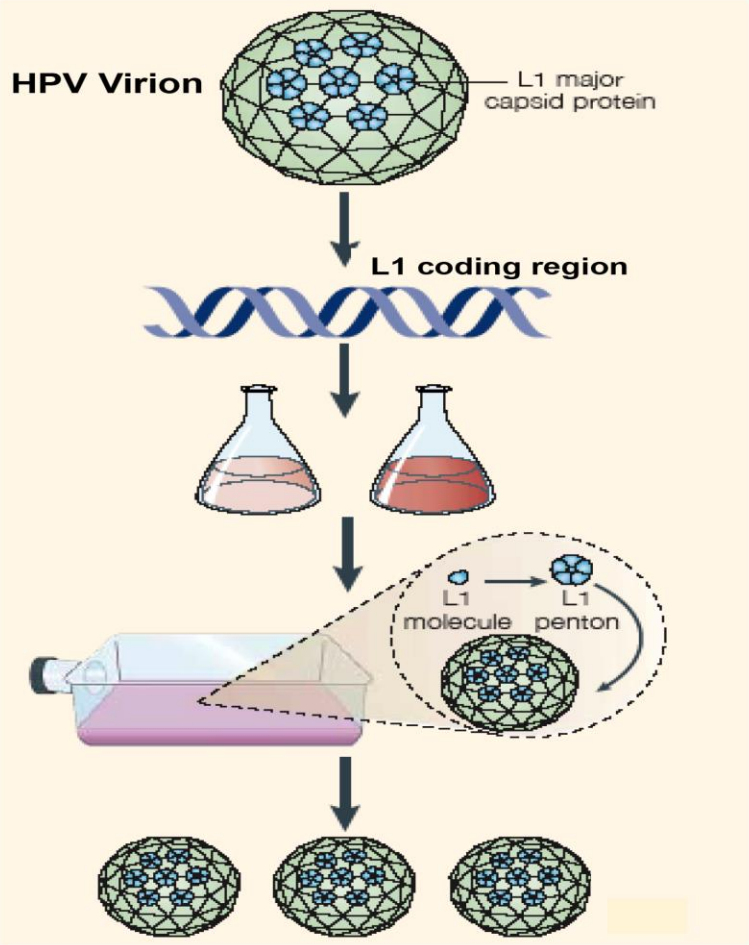
Pap screening



Primary prevention



Virus like particles



Prophylactic HPV Vaccines Are L1 Virus Like Particles (VLPs)

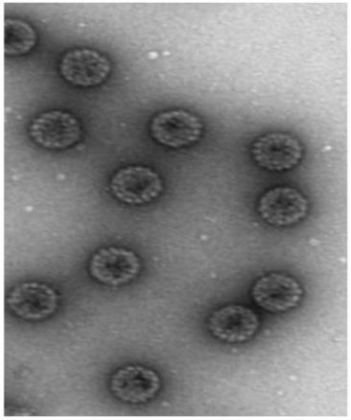
L1 Insertion into a Baculovirus Expression Vector

Production in Insect Cells

Spontaneous assembly of L1 into VLPs

Induce high titers of virion neutralizing antibodies

HPV16 L1 VLPs



Non-infectious, Non-oncogenic

Reinhard Kirnbauer et al. PNAS 1992

Three vaccines

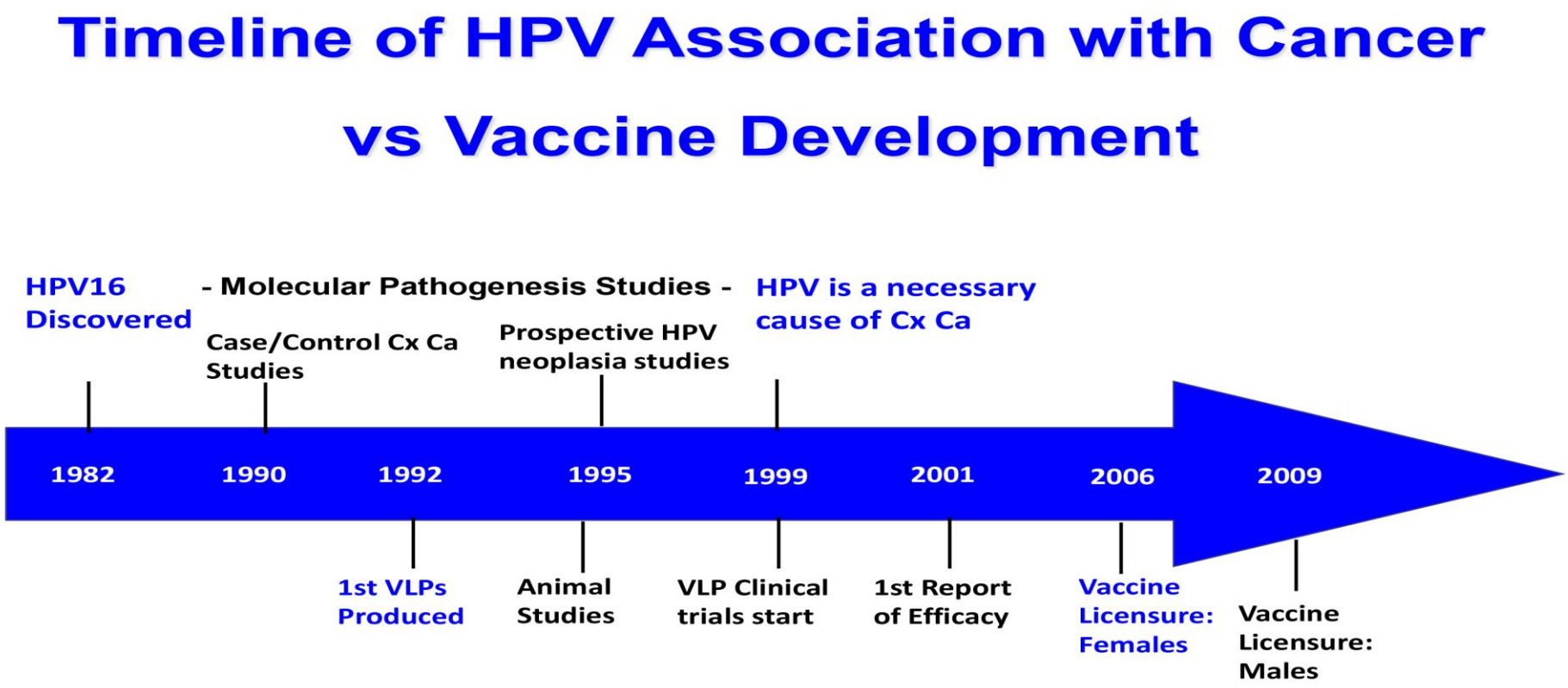
Three Distinct HPV L1 VLP Vaccines Have Been Commercialized

Name	Producer	VLP Types	Adjuvant	Production	Licensed
Cervarix	GSK	16,18	AS04*	Insect Cells	2007
Gardasil	Merck	16,18, 6,11	Alum	Yeast	2006
Gardasil-9	Merck	16,18,31, 33,45,52,58 6,11	Alum	Yeast	2014

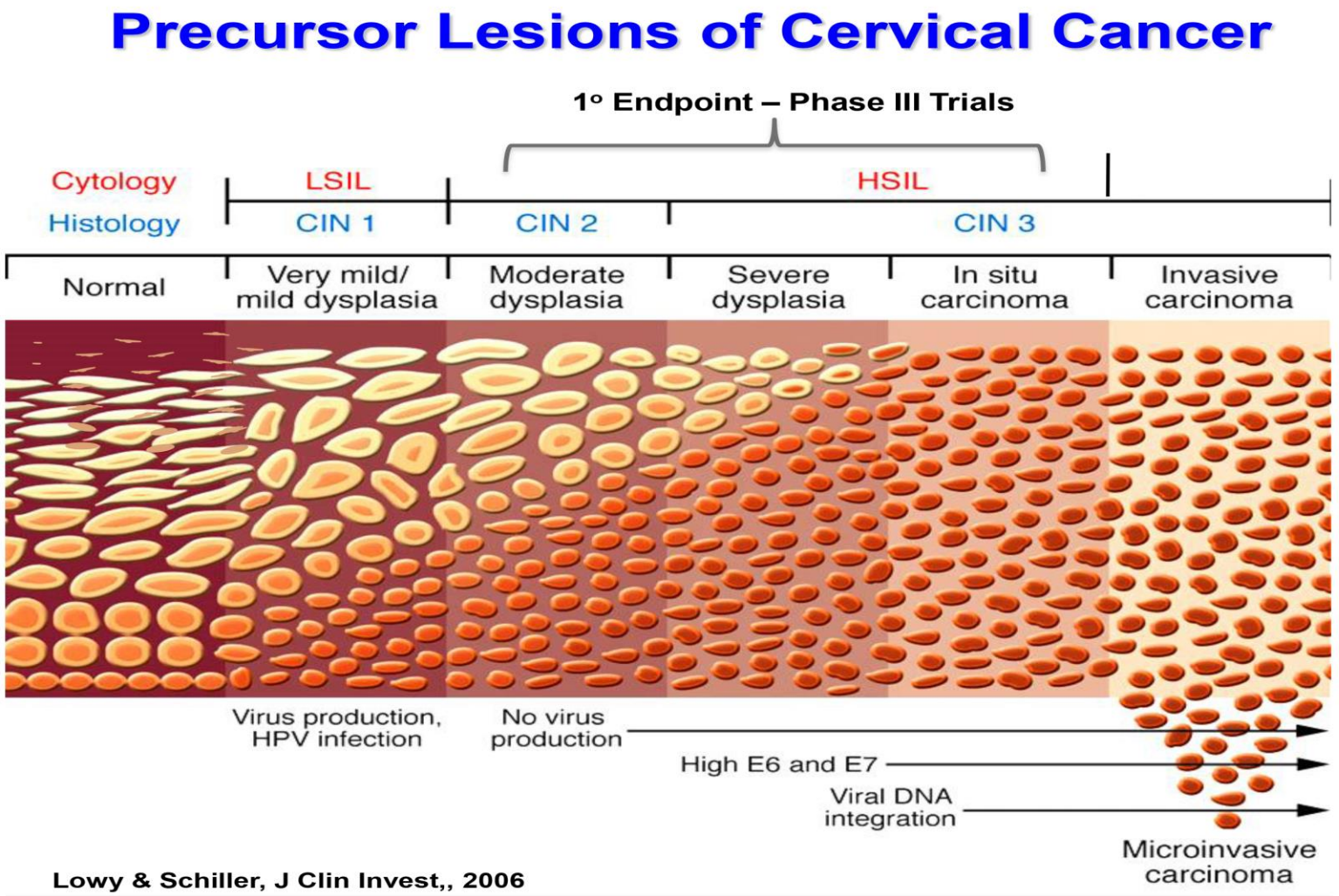
IM Injections at 0, 1 or 2, and 6 months
1, 6 months for <15 yrs in EU, and now in U.S.

* MPL First TLR Agonist Adjuvant to be FDA Approved

Timeline of HPV Association



Precursor Lesions



Efficacy of HPV Vaccine

Efficacy of HPV VLP Vaccines Against Incident Disease By *Vaccine-Targeted Types* in Randomized Trials

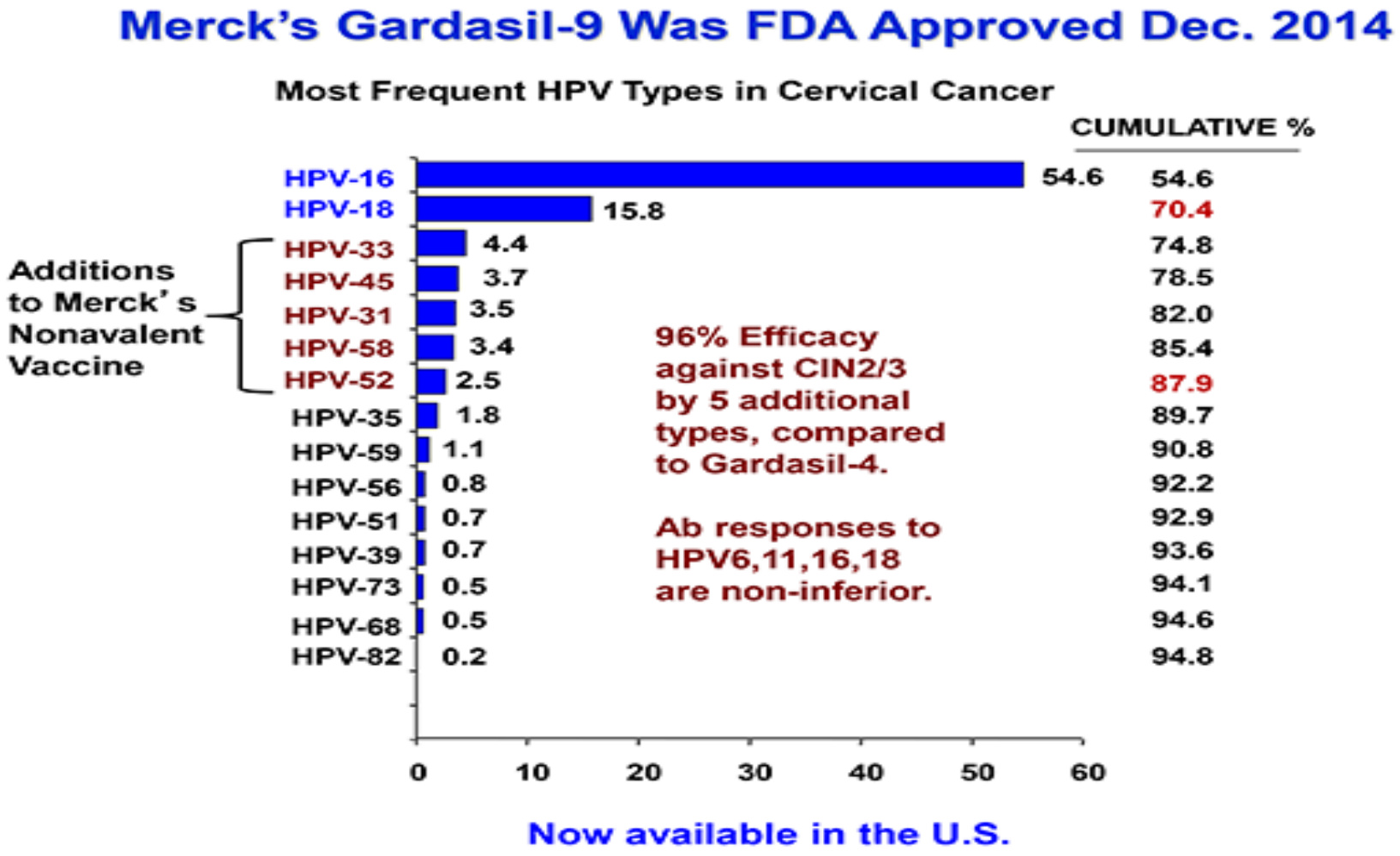
No gential HPV infection detected in at entry

End Point	Sex	Age	Vaccine	Efficacy (95% CI)
CIN III	Female	15-25	Cervarix	100% (90.5-100)
CIN III	Female	15-26	Gardasil	100% (85.5-100)
Genital Warts	Female	15-26	Gardasil	96.4% (91.4-98.4)
AIN	Male	16-26	Gardasil	77.5% (39.6-93.3)
Genital Warts	Male	16-26	Gardasil	89.4% (65.5-97.9)

Data from Lehtinen Lancet Oncol 2011; Munoz JNCI 2010; Palefsky NEJM 2011; Giuliano NEJM 2011

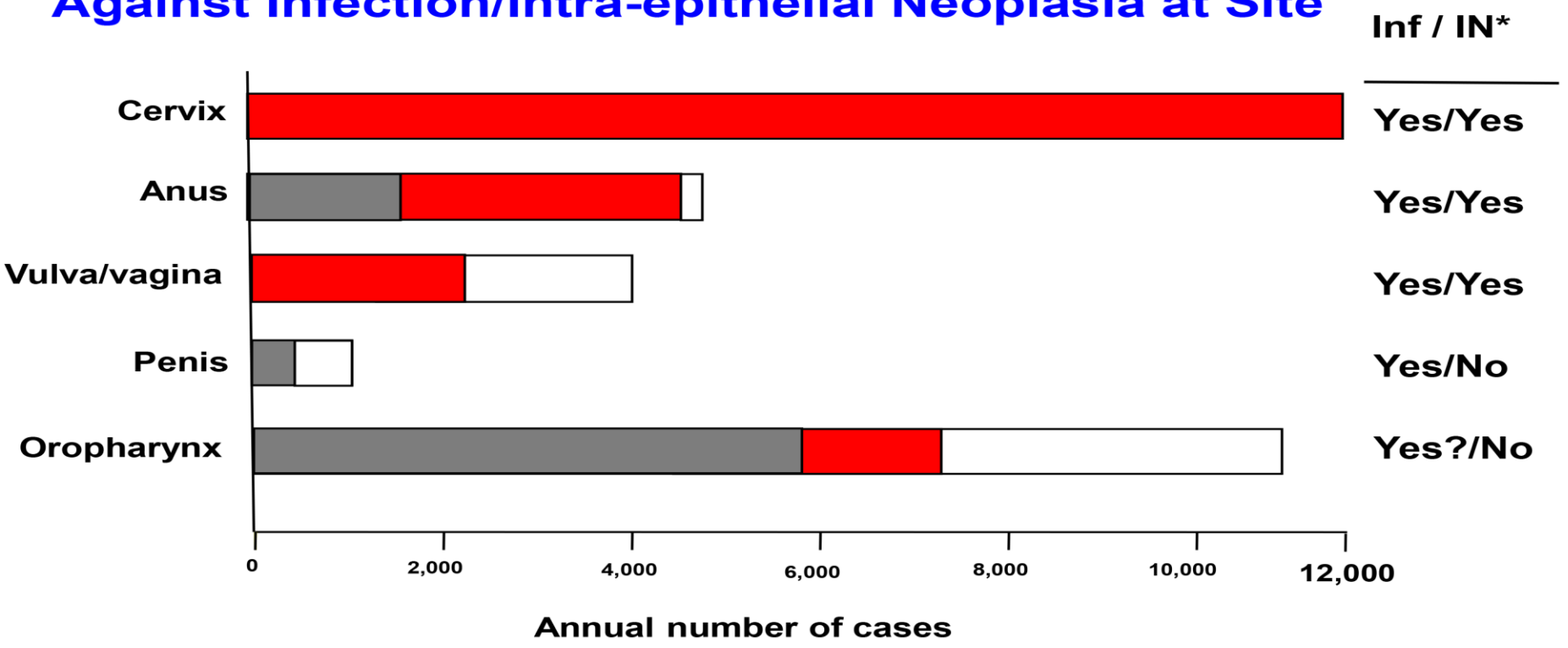
CIN III: Cervical Intraepithelial Neoplasia Grade 3
AIN: Anal Intraepithelial Neoplasia of any grade

Gardasil-9


Merck's Gardasil-9 Was FDA Approved Dec. 2014
Most Frequent HPV Types in Cervical Cancer
CUMULATIVE %
HPV-16 54.6 54.6
HPV-18 15.8 70.4
HPV-33 4.4 74.8
HPV-45 3.7 78.5
HPV-31 3.5 82.0
HPV-58 3.4 85.4
HPV-52 2.5 87.9
HPV-35 1.8 89.7
HPV-59 1.1 90.8
HPV-56 0.8 92.2
HPV-51 0.7 92.9
HPV-39 0.7 93.6
HPV-73 0.5 94.1
HPV-68 0.5 94.6
HPV-82 0.2 94.8
Additions to Merck's Nonavalent Vaccine
96% Efficacy against CIN2/3 by 5 additional types, compared to Gardasil-4.
Ab responses to HPV6,11,16,18 are non-inferior.
0 10 20 30 40 50 60
Now available in the U.S.

Clinical Trial Evidence

Clinical Trial Evidence for Vaccine Efficacy Against Infection/Intra-epithelial Neoplasia at Site



* Against Vaccine Targeted Types

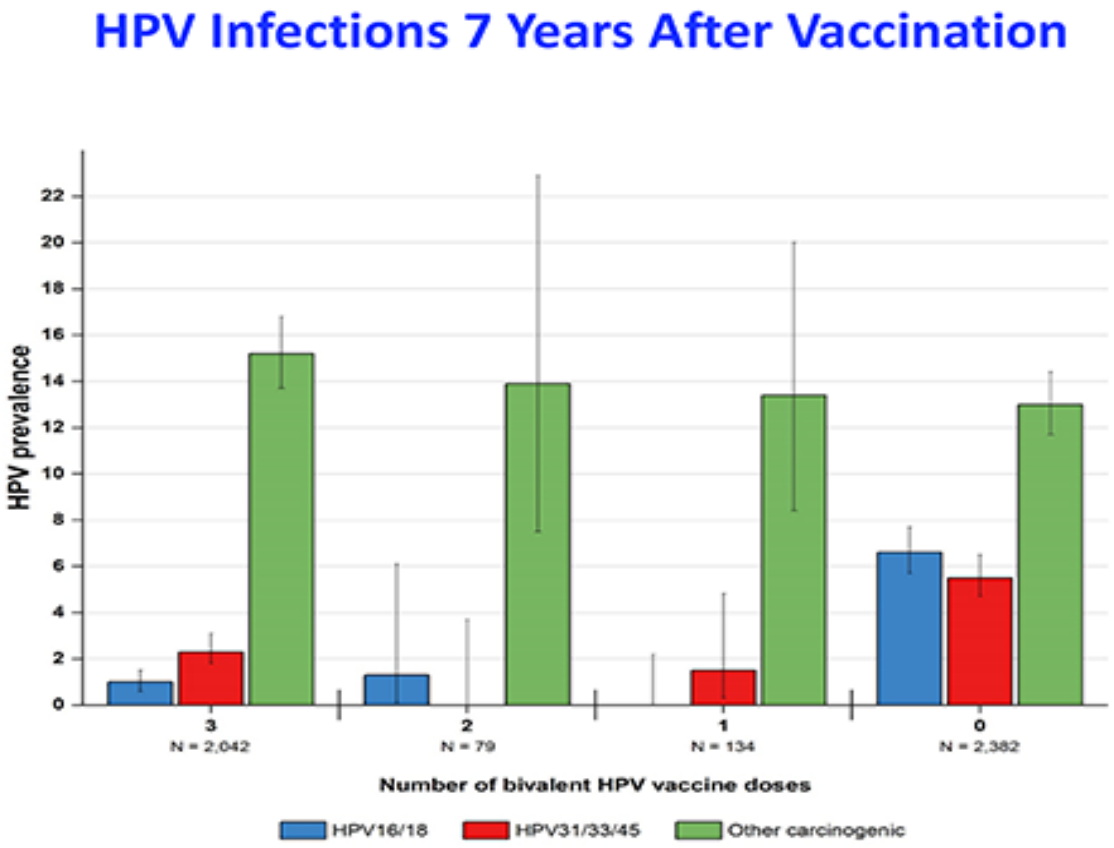
MMWR Weekly 61:253-80, 2012

What the HPV Vaccines Don't Do

They don't prevent infection or disease caused by most of the other HPV types that cause 30% of cervical cancer not caused by HPV16/18.

They don't induce regression of established HPV infections or prevent progression of HPV-induced lesions.

7 Years after vaccination



Safaeian M J Natl Cancer Inst 2017; 110 (2)

Protection from Initial Infection

Protection From Initial Infection

- Most Vaccinees never tested positive for HPV infection as measured by sensitive PCR Assays.
- "Breakthrough" infection tended to appear early in the trials suggesting that most were emergence of prevalent infection.
- Results imply that sterilizing immunity normally generated.

HPC vaccine

What the HPV Vaccines Don't Do

- They don't prevent infection or disease caused by most of the other HPV types that cause cervical cancer.
- They don't induce regression of established HPV infections or prevent progression of HPV-induced lesions.

Safety record

HPV VLP Vaccines Have an Excellent Safety Record

- Low grade and transient injection site reactions, particularly pain, are common.
- Systemic reactions, when they occur, are mild and self-limiting.
- Synocope (fainting) is sometimes observed (needle related).

Serious Adverse Events Following HPV Vaccination

Study	Vaccine	% Vaccine	% Control	Relative Risk (95% CI)
Future I	Gardasil	1.8%	1.7%	1.07 (0.71-1.60)
Future II	Gardasil	0.7%	0.9%	0.83 (0.56-1.24)
PATRICIA	Cervarix	7.5%	7.5%	1.00 (0.91-1.11)

No patterns of serious adverse events following immunization in trials or post-licensure surveillance that would suggest a causal relation to the vaccine.

Reviewed in: Macartney, Drug Saf 2013; 36:393-412; Sheller JAMA 2015; 313:54-61; Arnheim-Dahlstrom, BMJ 2013; 347, f5906.

Vaccine effectiveness

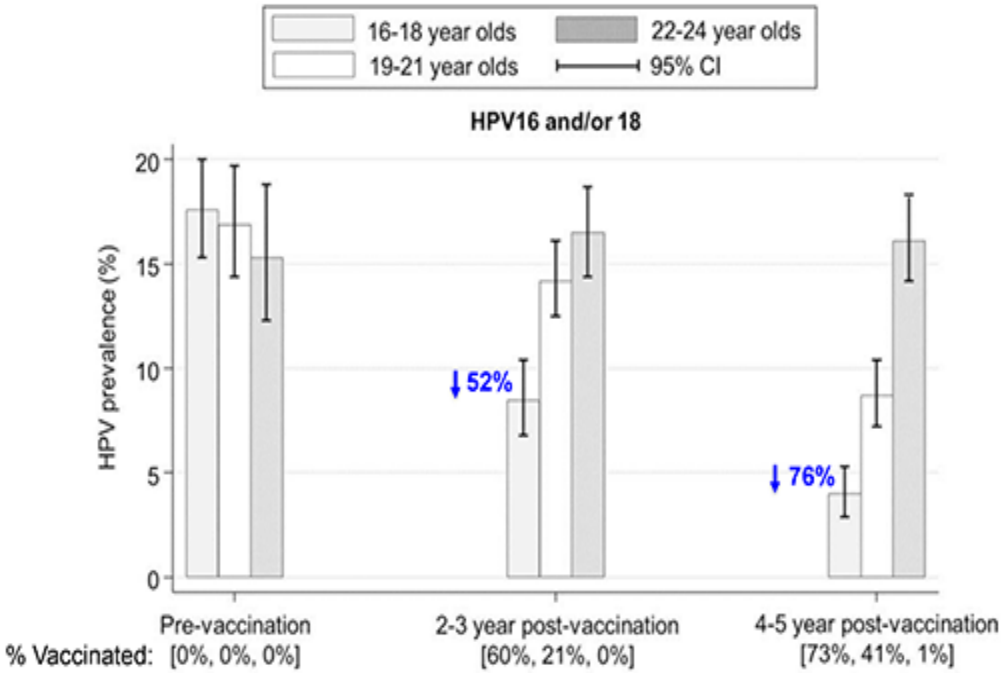
Vaccine Effectiveness: Evidence From National Immunization Programs

Country	Type-Specific Infection		Genital Warts		Cervical Lesions
	Female	Male	Female	Male	Female
Australia	+	+	+	+	+
Britain	+		+		+
USA	+	+	+		+
Canada			+		+
Denmark			+	+	+
Sweden			+	+	+
France	+				
Spain			+		
Italy			+	+	
Israel			+	+	

Drolet, Lancet Infect Dis 2015;15:565-580; Brotherton, Best Pract Res Clin Obst & Gyn, 2017

Vaccination

Prevaccination and Postvaccination Prevalence of HPV Types By Age: Cervarix in England

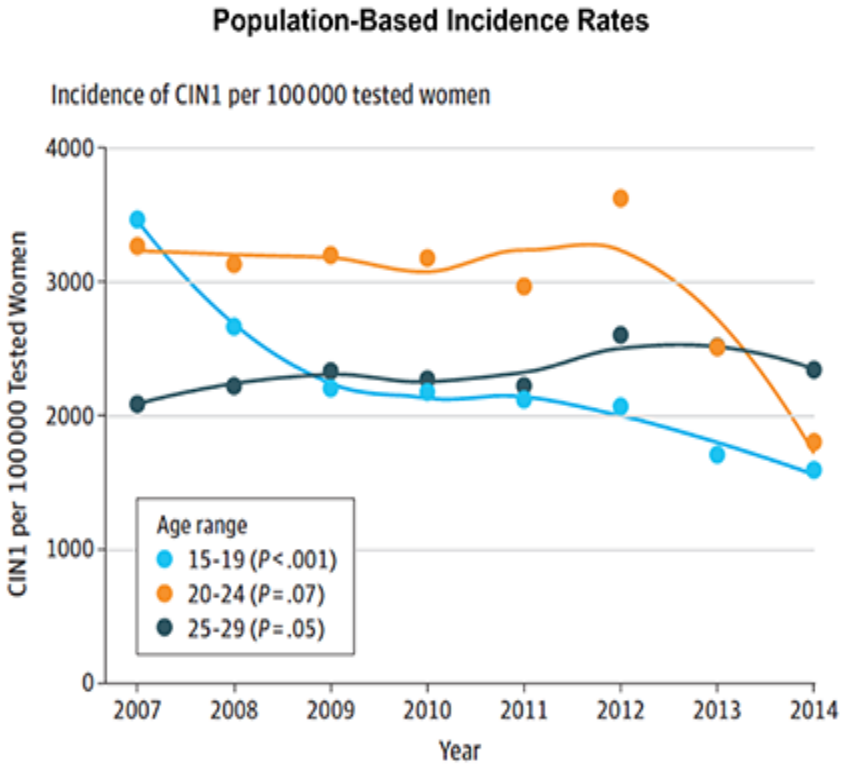


In young women attending Chlamydia screening

David Mesher et al. BMJ Open 2016;6:e009915

CIN1 reduction

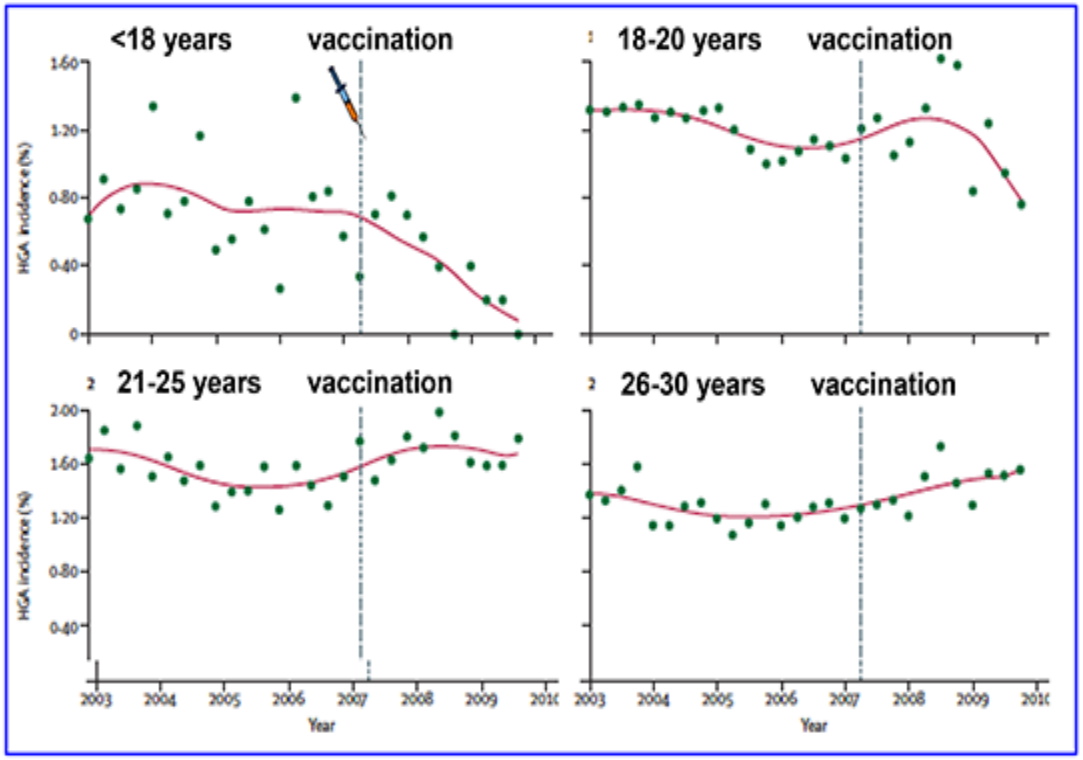
Reduction in CIN1 in New Mexico



Benard et al. JAMA Oncology, epub Sept 29, 2016

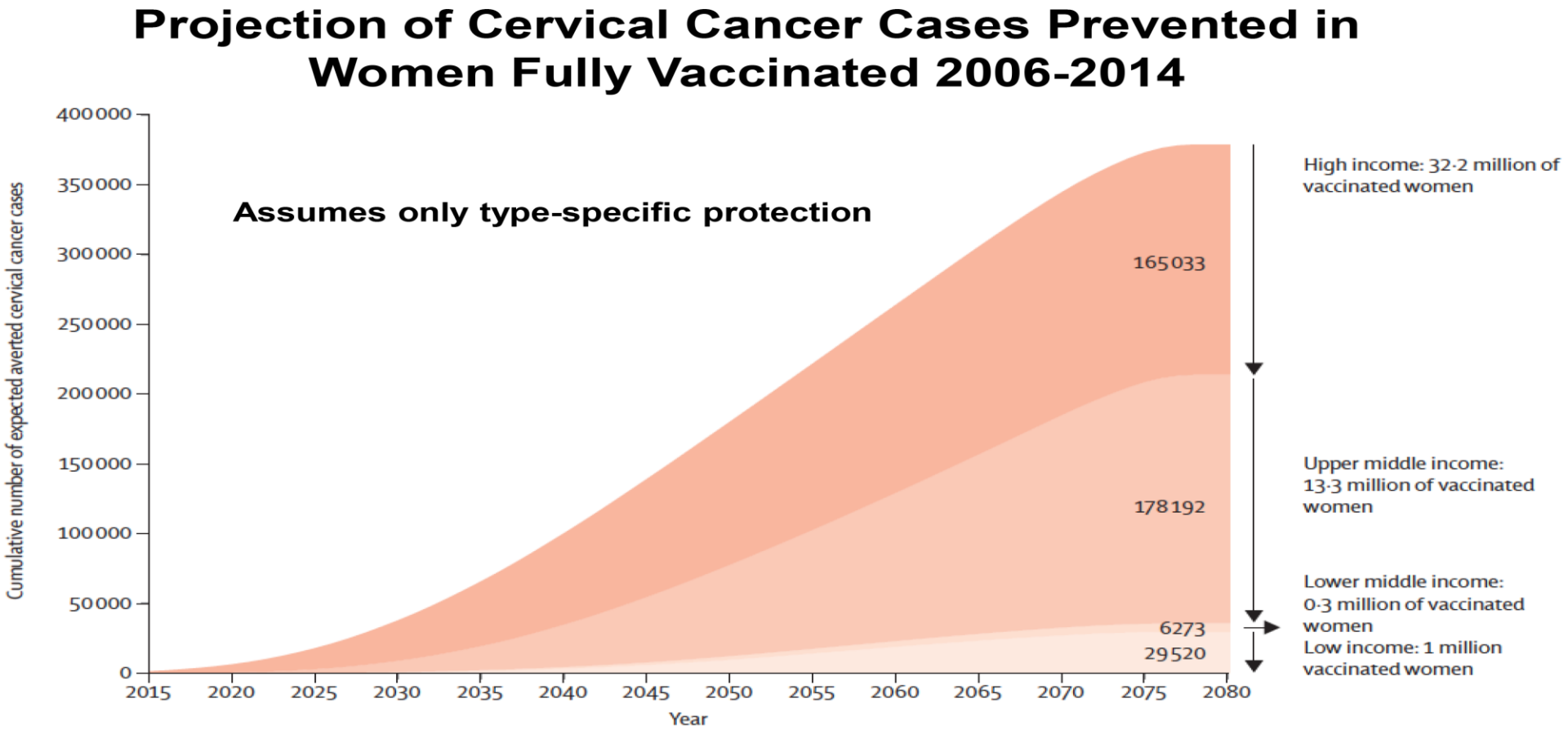
Effectiveness

Effectiveness: Reduction in CIN2+ Cervical Dysplasia by Gardasil in Australia



Brotherton et al, Lancet 377: 2085-92, 2011

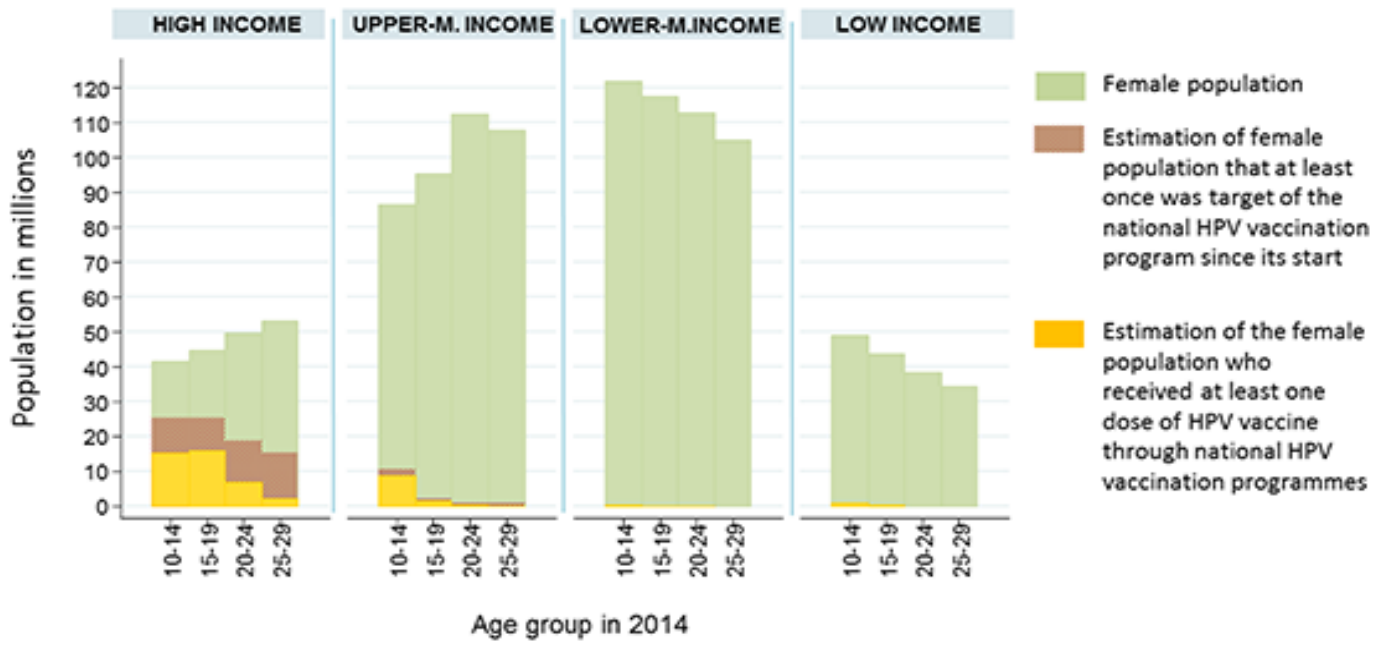
Cervical cancer prevention



We have already prevented 400,000 future cases of cervical cancer

Bruni et al. Lancet Global Health 2016; 4:3453-63

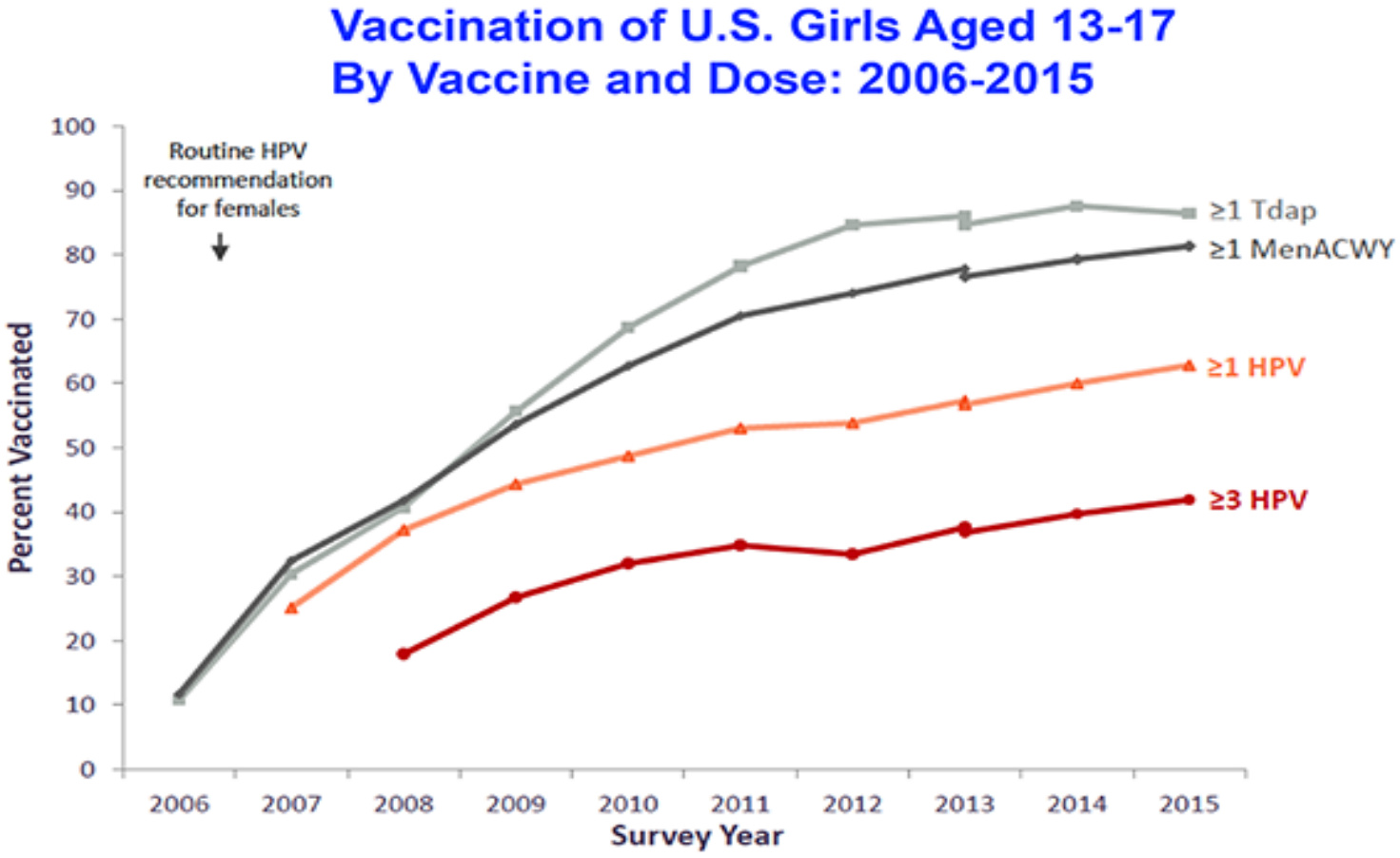
Worldwide HPV vaccine uptake



Only 3% of girls in lower and lower-middle income countries have been vaccinated

Bruni et al. Lancet Global Health2016; 4:3453-63

US girls vaccination rate



Reagan-Steiner et al. MMWR 2016

Increasing Uptake, Particularly in Low Resource Settings

Increasing Uptake, Particularly in Low Resource Settings

- Both companies are committed to sale to GAVI at less than $5 per dose.
- Vaccine manufacture in emerging countries.
- Address vaccination hesitancy by education programs aimed families and health care providers.
- Deliver fewer than three doses.

One dose efficacy of Cervarix

Surprising One Dose Efficacy of Cervarix After 4 Yrs

NCI-sponsored Costa Rica Vaccine Trial
***Post Hoc* Analyses of Incident HPV16/18 of 6+ mo. persistent infection**
Women ages 18-25 years

# of Doses	Vaccine Arm	# Women	# (%) with endpoint	HPV16/18 VE (95%CI)
3	Control	3010	229 (7.6%)	84% (77% to 89%)
	HPV	2957	37 (1.3%)	
2	Control	380	24 (6.3%)	81% (53% to 94%)
	HPV	422	5 (1.2%)	
1	Control	188	15 (8.0%)	100% (79% to 100%)
	HPV	196	0 (0%)	

p trend= 0.2

Kreimer AR et al *JNCI, 2011*

Cervarix after 4 years

Surprising One Dose Efficacy of Cervarix After 4 Yrs

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	HPV	196	0 (0%)	

p trend= 0.2

Kreimer AR et al *JNCI, 2011*

One dose clinical trials

Other One Dose Clinical Trial Results

Cervarix:

4 year *post hoc* dose stratified efficacy results for the GSK-sponsored PATRICIA trial showed similar efficacy for 1, 2 and 3 dose recipients.

Kreimer et al. Lancet Oncol 16:e424-5, 2015

Gardasil:

In an interrupted Indian cluster randomized trial, after 3 years, there was similar protection in young women receiving 1, 2, or 3 doses.

Sankaranarayanan et al. Lancet Ocol, 2015

Single dose efficacy trial

NCI-Sponsored* Single Dose Efficacy Trial

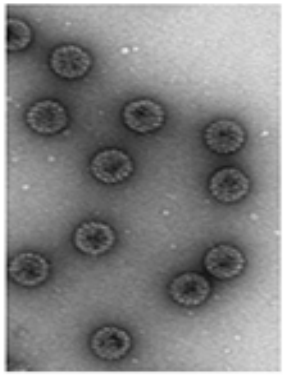
- 4 Arms RCT: 1 vs 2 dose Cervarix
  1 vs 2 dose Gardasil-9
  In addition: Survey of HPV prevalence in region
- 5000 12-16 yr old Costa Rican females per arm.
- 4 year primary trial; long term follow up.
- Primary Endpoint: 6 mo. Persistent infection by the types in the vaccine.
- The trial has just begun.

* With help from the Gates Foundation

HPV vaccines

Why Do HPV VLP Vaccines Work So Well?

- The vaccines are exceptionally good at inducing neutralizing antibodies.
- Infection mechanism make HPVs exceptionally susceptible to neutralizing antibodies.



HPV16 L1 VLPs

- Virus evolution: HPVs have DNA genomes so can't evolve rapidly to evade nAb responses.

Immune mediators of protection

Why Antibodies Are Likely to be the Immune Mediators of Protection

- High levels of virus-neutralizing antibodies are routinely generated by VLP vaccination.
- Cross type-protection in clinical trials mirrors antibody-mediated cross-neutralization in vitro.
- Protection can be passively transferred in serum from vaccinated to naïve individuals in animal challenge models.
- Cell mediated effectors generally function only after infection has occurred and sterilizing immunity is observed.
- L1 is not detectably expressed in basal epithelial cells.

Antibody responses to VLPs

Consistency of Antibody Response to VLPs

Percent of Women Serocoverting to Individual HPV VLPs in Merck VLP Vaccine Gardasil*

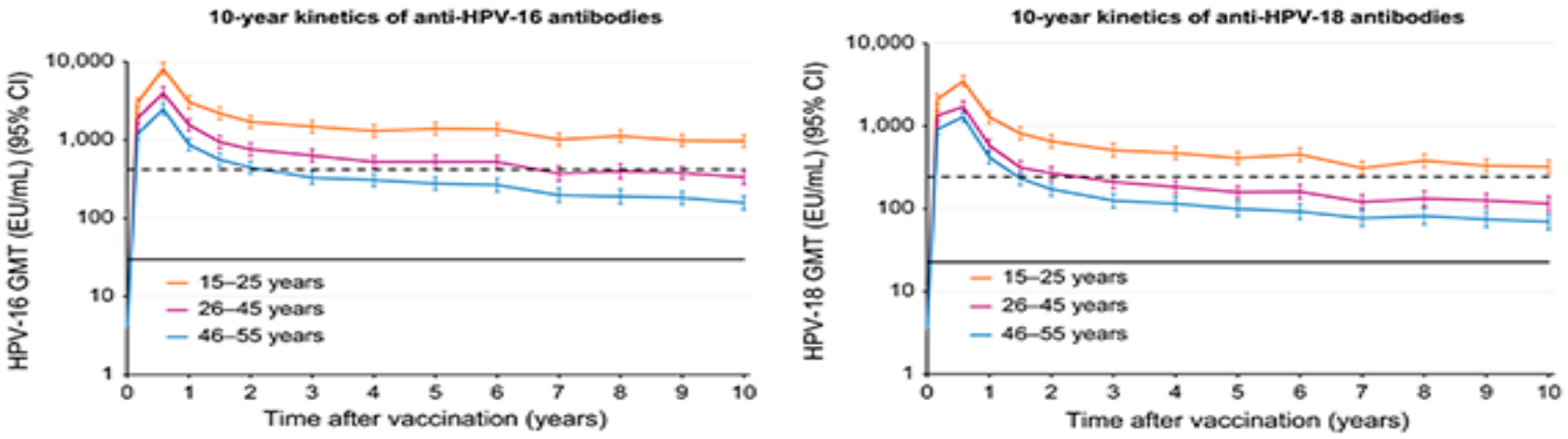
HPV6	99.8%
HPV11	99.8%
HPV16	99.8%
HPV18	99.5%

*4666 women vaccinated 3 times by intramuscular injection

Persistence of antibodies

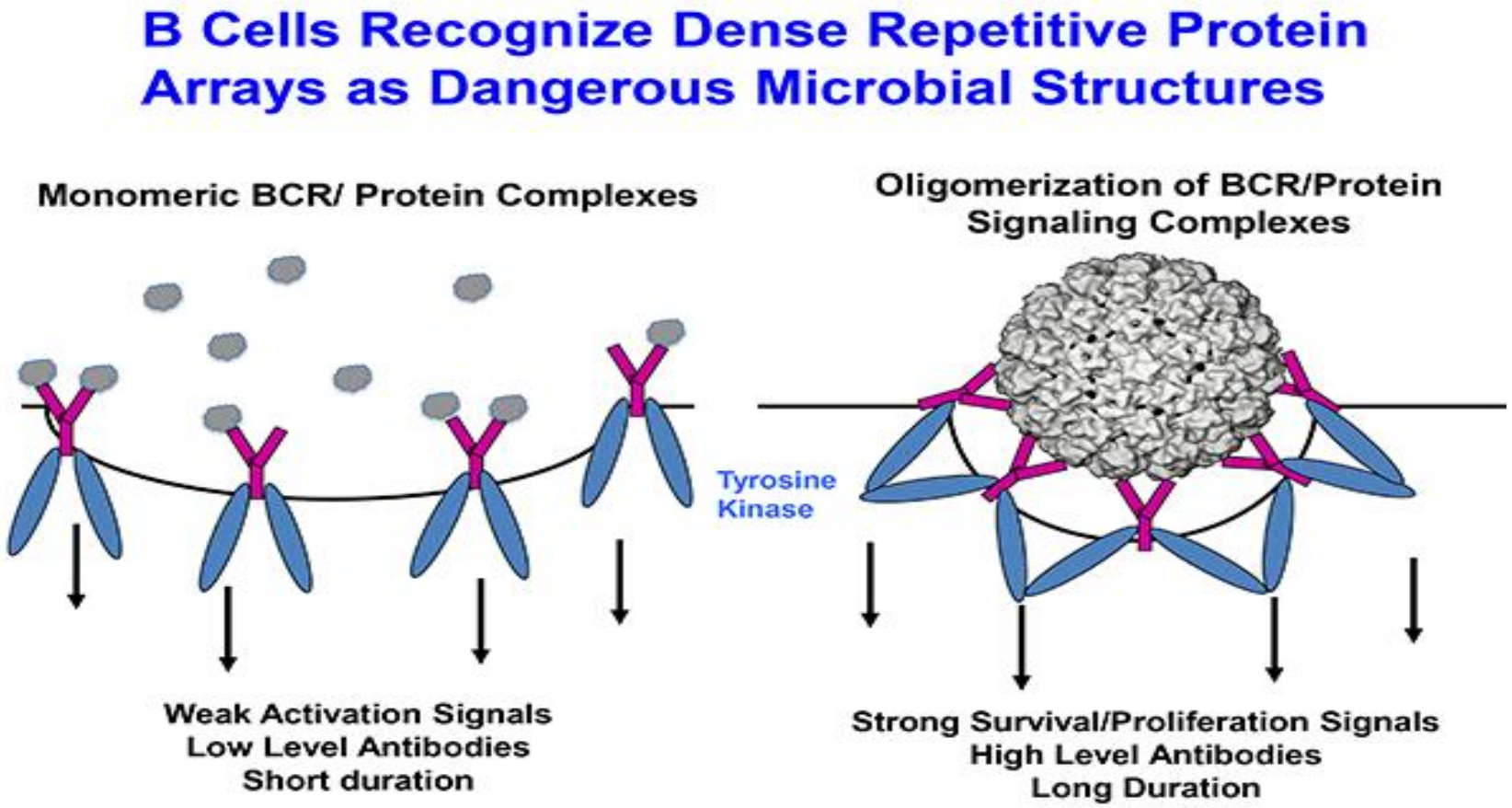
Persistence of Antibodies to Cervarix in Females Vaccinated at 15–55 Years of Age

Received Three Doses



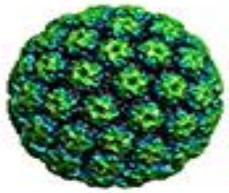
Cancer Medicine
5 OCT 2017 DOI: 10.1002/cam4.1155

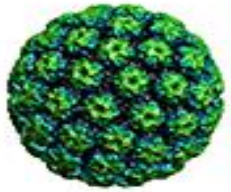
B cells recognize dense repetitive protein arrays



Repetitive antigen display

VLPs Have Highly Repetitive Antigen Display





B cells specifically recognize particulate antigens with epitope spacing of 50-100Å as foreign.

This epitope spacing is commonly found on microbial surfaces, e.g. virus major capsid protein or bacterial pili.

Protein complexes with this spacing rarely occur in vertebrate animals.

So BCRs have evolved as antigen specific pattern recognition receptors.

Bachmann et al. Science 1993; 262: 1448

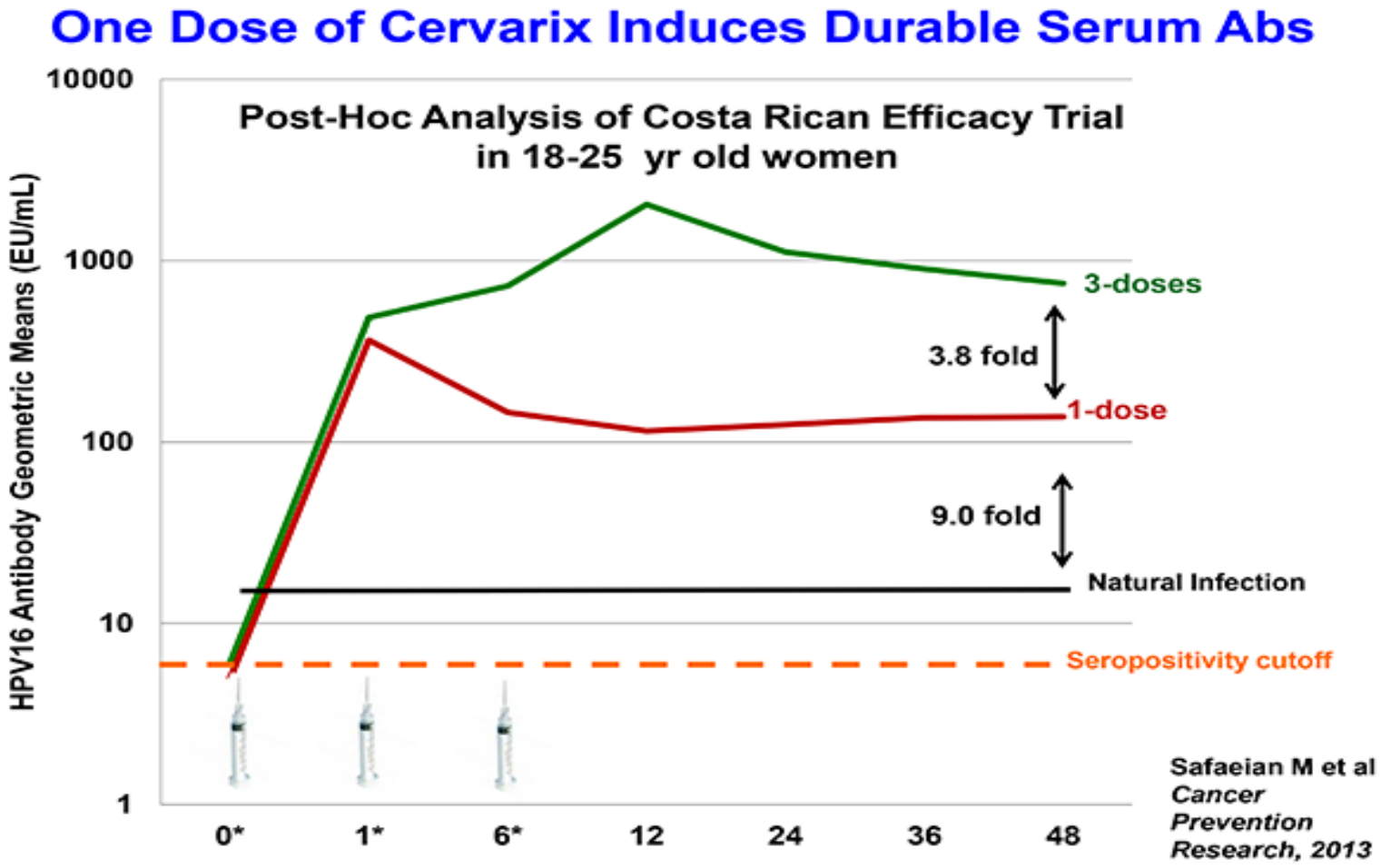
Virus like particles

Other Good Things That VLPs Do

- They have the right particle size for efficient trafficking to lymph nodes.
- They are readily phagocytized and so induce strong T helper responses.
- Their poly-valency leads to stable binding of natural low-avidity IgM and Complement which promotes their acquisition by follicular dendritic cells.*

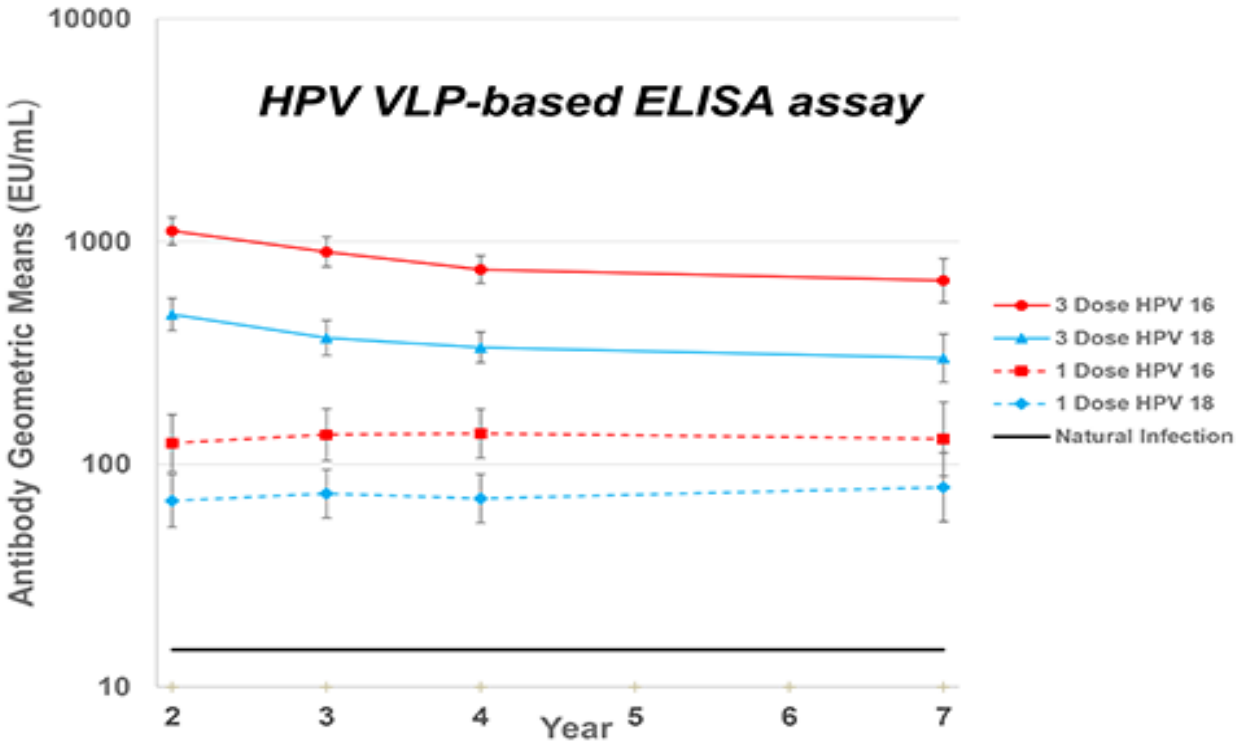
*A Link JI 188:3724-33, 2012

Durable serum Abs



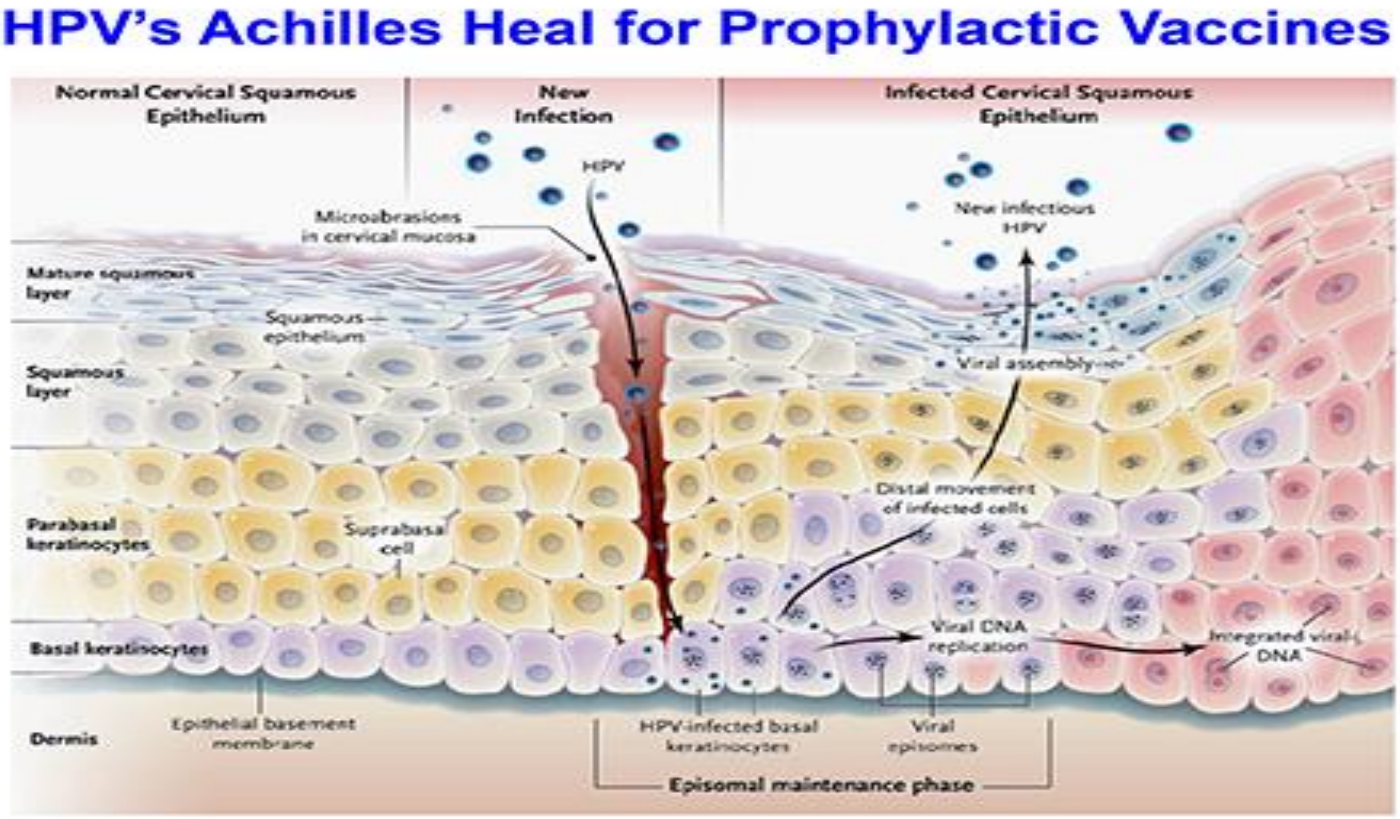
Antibody response

Durability of VLP Antibody Responds To 7 Years



Safaeian M et al, J Nat Cancer Inst 110 (2), 2017.

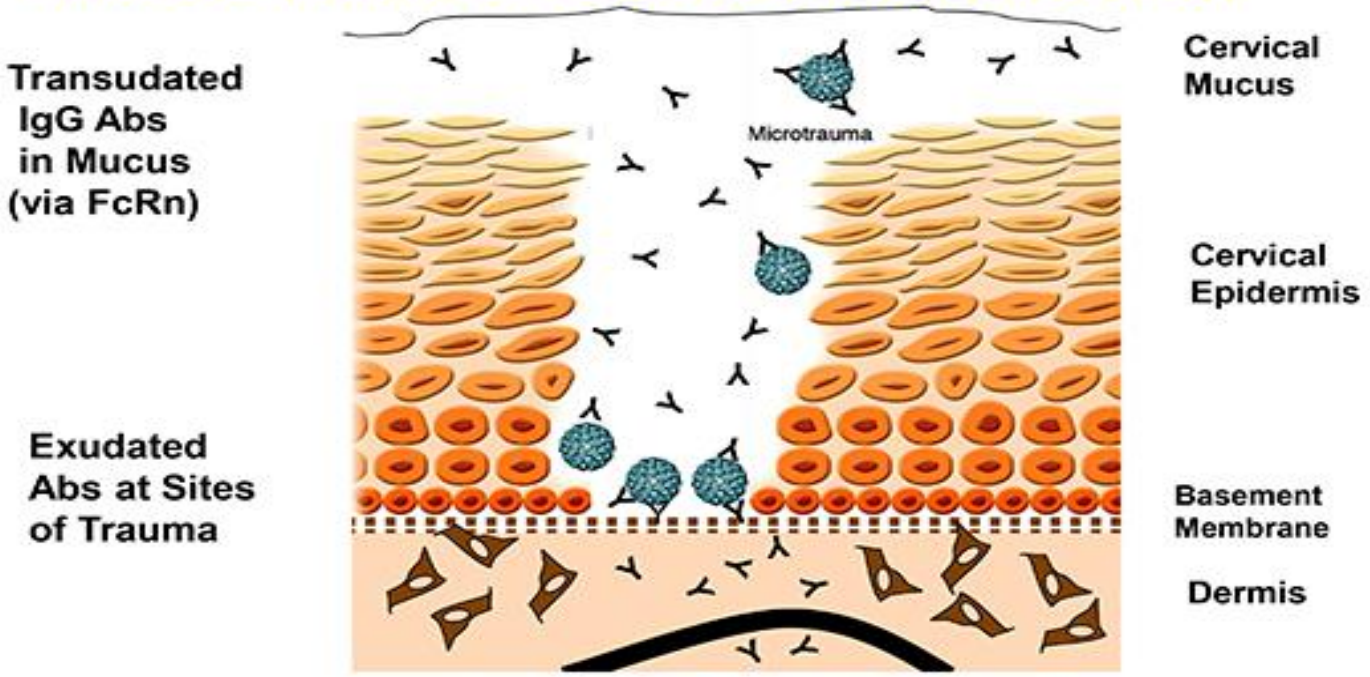
HPV achilles heal



HPVs shed their virion on external surfaces and so virions are not routinely exposed to system immunity. So the virus hasn't evolved defenses against vaccines, like IM injection of VLPs, that do.

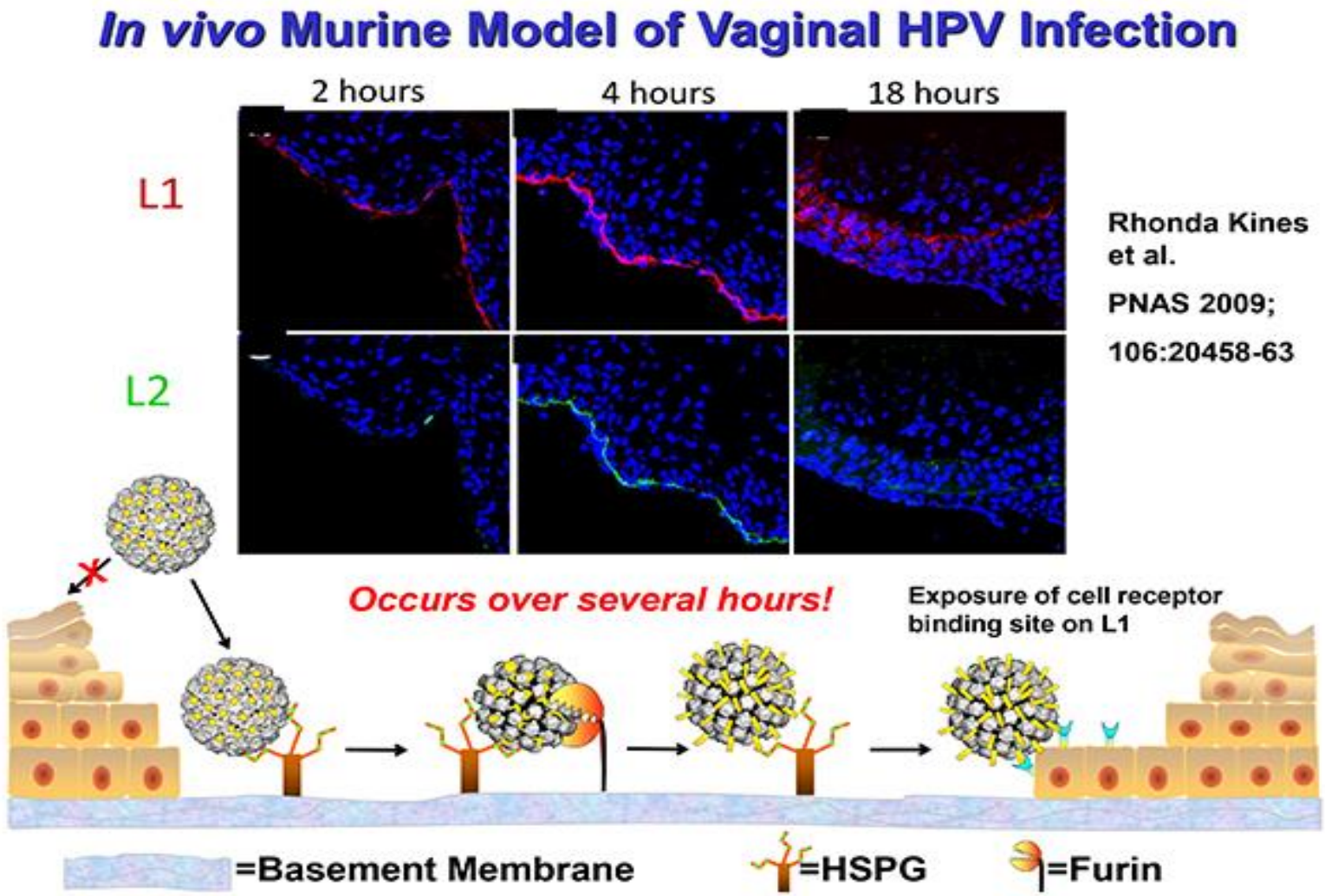
Cervix Ab response

How Could IM Injection of a VLP Vaccine Induce a Protective Ab Response at the Cervix?

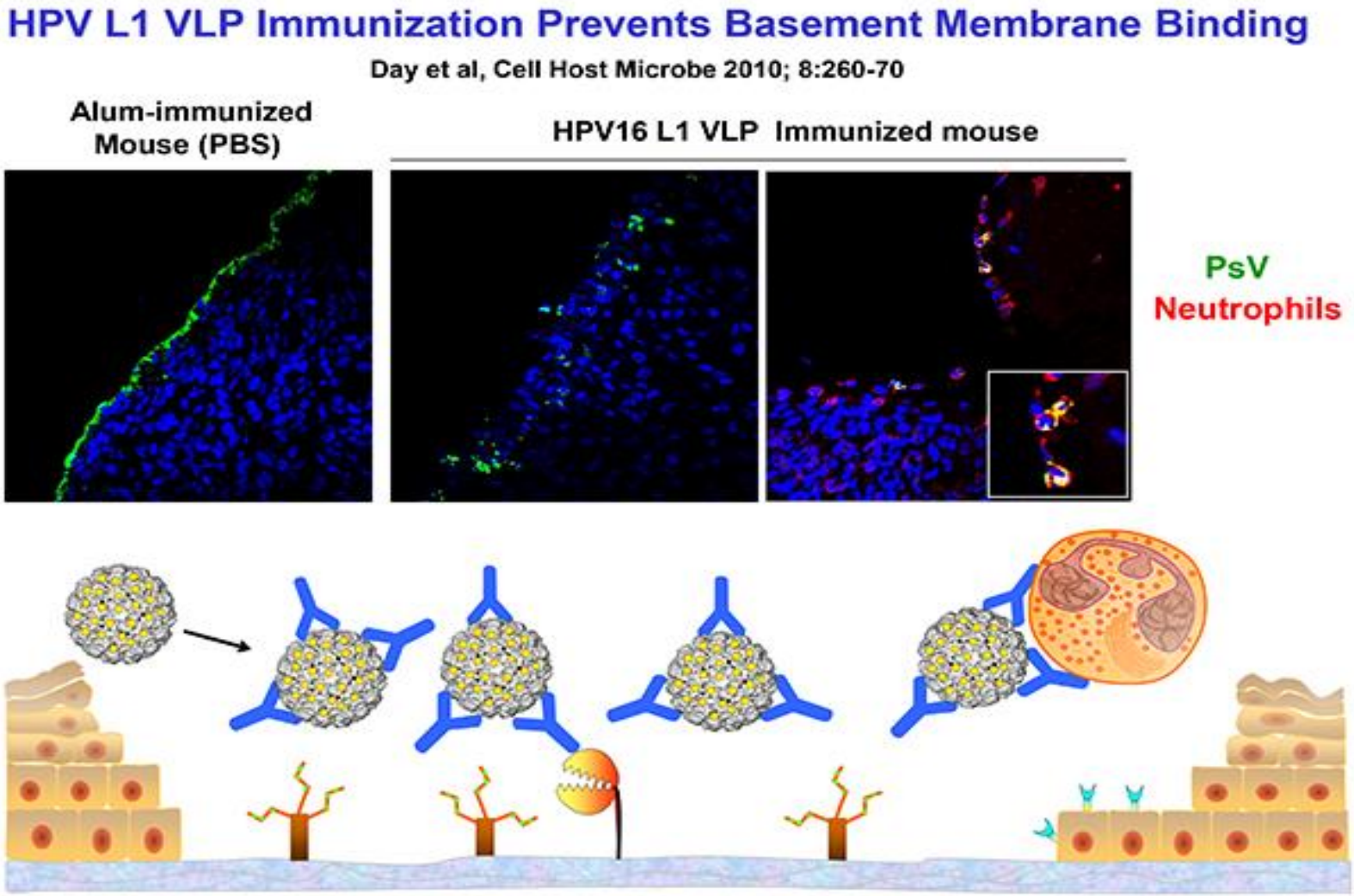


- VLP-specific IgG in women's cervical mucus after IM vaccination: but 10-100X less than in serum - *Nardelli et al. JNCI, 2003*
- Cervicovaginal HPV infection in a mouse model requires epithelial trauma: *Roberts et al., Nat Med, 2007*

Murine model



Basement membrane



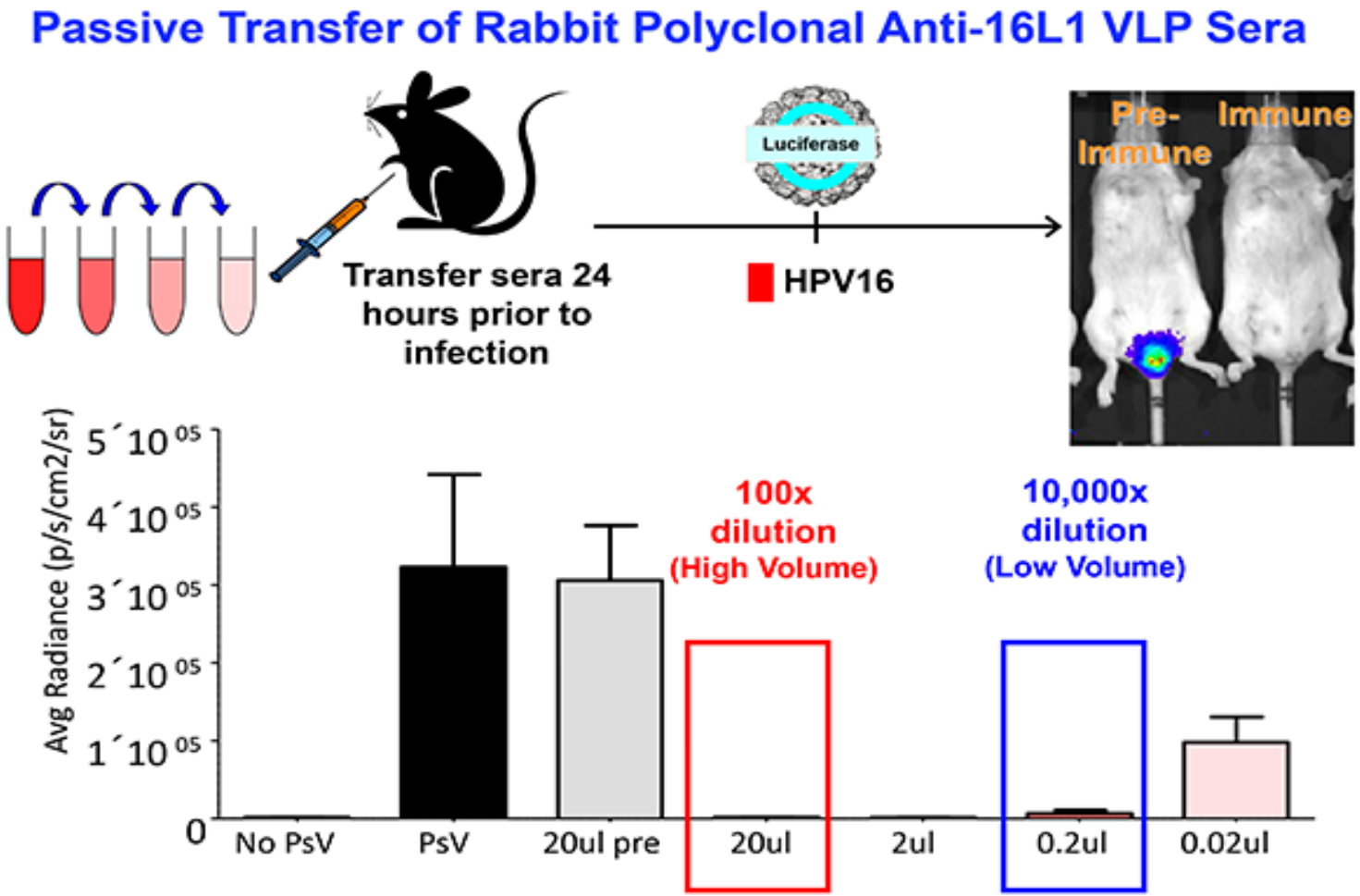
Antibody titers and protection

Antibody Titers and Protection

Are the plateau titers after vaccination near the minimum needed for protection?

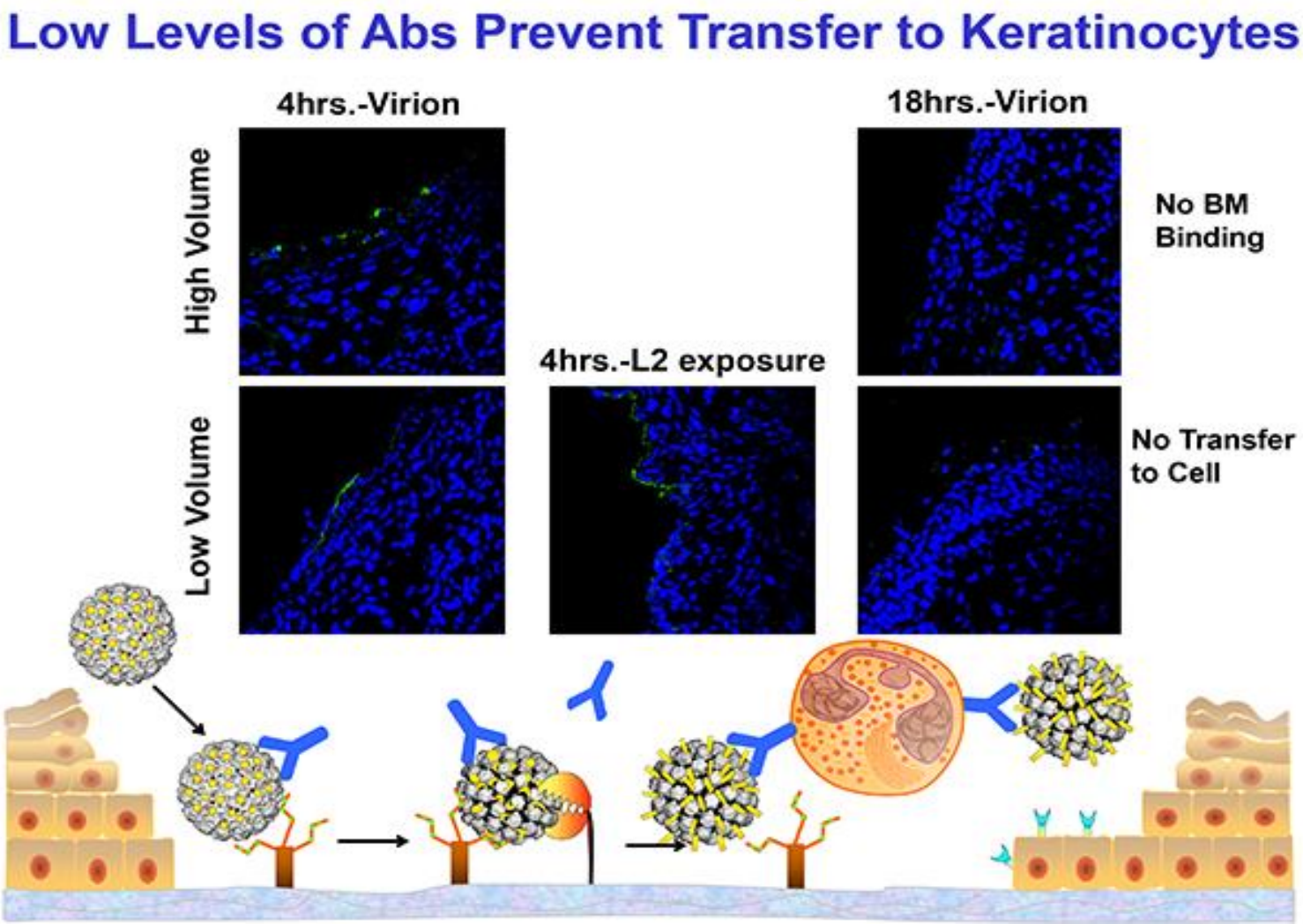
Will the 4-fold difference between Ab titers after three vs one dose influence long-term protection?

Passive transfer



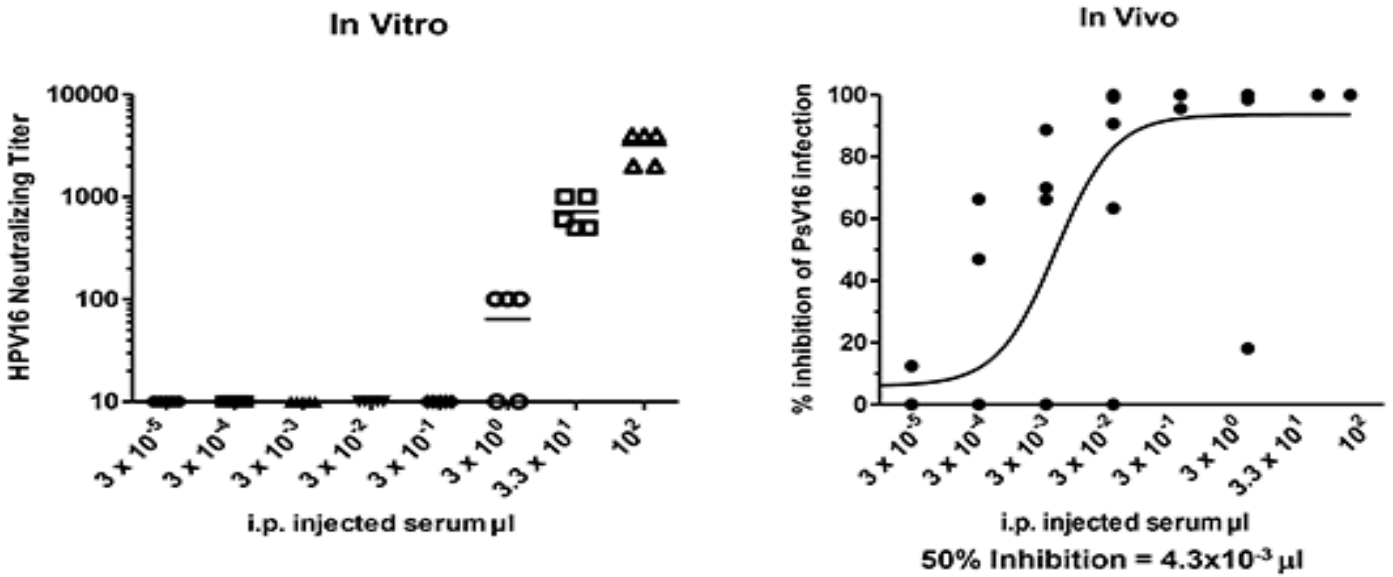
* Challenged with HPV16. See no protection from infection when challenged with HPV45

Keratinocyte transfer



Gardasil sera protection

In vitro vs In Vivo Protection of Gardasil Sera Against HPV16 Pseudovirus Infection



Protection detected with 500-fold less sera in vivo than in vitro!

The in vitro assay is missing some potent mechanism of infection inhibition.

Longet et al, J Virol 2011

Conclusions

Conclusions

- The HPV VLP vaccines are very effective at preventing incident infection and disease by the vaccine types.
- Because the VLPs are exceptionally potent induces of neutralizing antibodies and the virus is exceptionally susceptible to inhibition by antibodies.
- The vaccines have great potential for reducing the burden of HPV-induced cancer worldwide.
- The primary challenge now is to see that the vaccines reach the individuals most in need of them.
- Demonstrating sustained efficacy of a single dose in a RCT could transform implementation programs.

Collaborators

Key Collaborators

Present Members of the Lab:

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