Frederick National Laboratory
for Cancer Research

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HPV Serology at FNL: Progress to Date and Future Directions

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Director, Vaccine, Immunity and Cancer Program

October 24, 2019

DEPARTMENT OF HEALTH AND HUMAN SERVICES • National Institutes of Health • National Cancer Institute

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Vaccine, Immunity and Cancer Program: Who We Are

Clinical Group
Barry Gause, MD

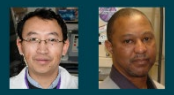
Applied and Development Research Directorate (ADRD)
Michael Baseler, PhD

Vaccine, Immunity and Cancer Program (VIC)
Ligia Pinto, PhD, Director

HPV Immunology Laboratory (HPVIL)

DCEG-sponsored

2 Research Associates



HPV Serology Laboratory (HSL)

CSSI/BMGF-sponsored

Troy Kemp, PhD

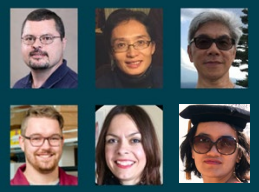
3 Research Associates
1 QA Specialist

Cancer Immunoprevention Laboratory (CIPL)

DCP-sponsored

Jason Marshall, PhD
Yurong Song, PhD
Scientific Project Manager

2 Research Associates
1 Postdoctoral Fellow



Mission: *To provide scientific leadership and laboratory infrastructure to study immune responses to Human Papillomavirus (HPV) vaccines and other cancer preventive strategies in the context of clinical and pre-clinical studies*

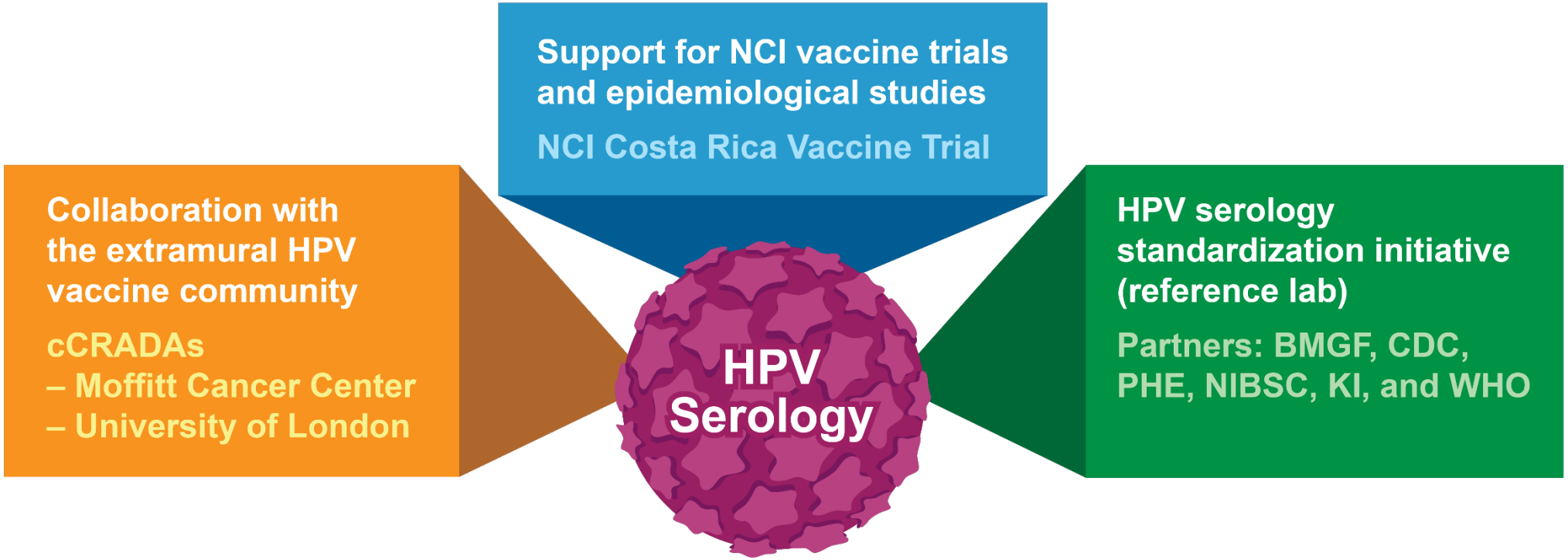
Vaccine, Immunity and Cancer Program: HPV Serology Efforts

Investigate immune responses to vaccines, infections, and cancer

Develop and validate new methods for laboratory markers of protection

Monitor immunity in clinical trials and pre-clinical studies

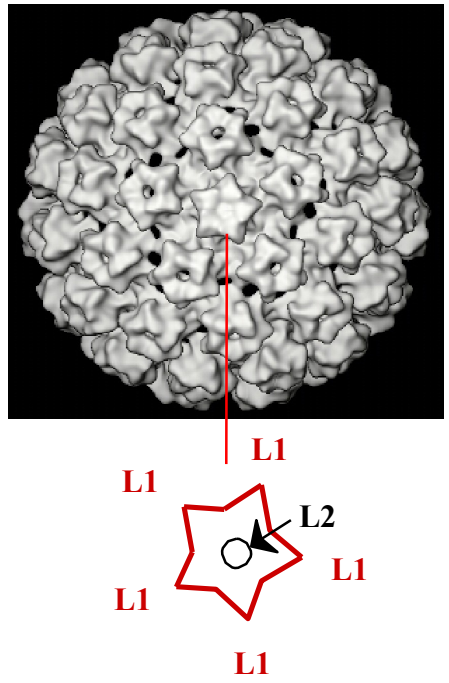
Provide evidence to inform new trials and create tools to enable decision-making and public health changes

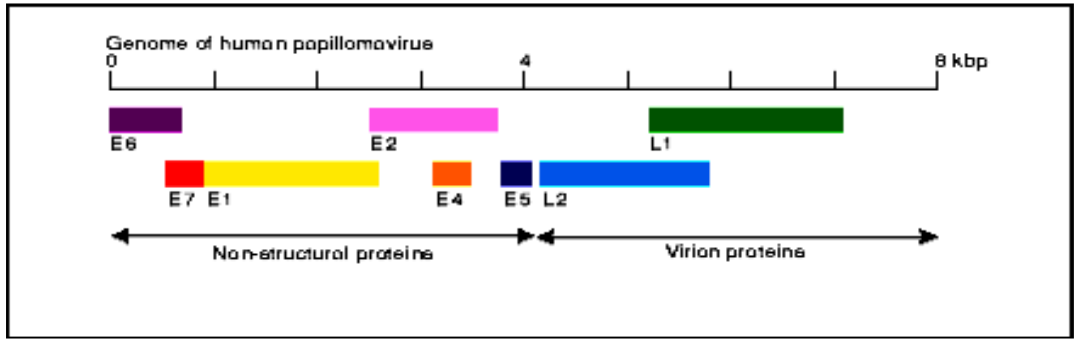


Human Papillomavirus (HPV)

- **Non-enveloped double-stranded DNA virus**
- **>200 types identified**
- **~40 types sexually transmitted**
  - 13 high-risk (oncogenic) types
    - HPV-16 and HPV-18 types account for 70% of cervical cancer cases
  - Low-risk (non-oncogenic) types
    - HPV-6 and HPV-11 associated with >90% of anogenital warts

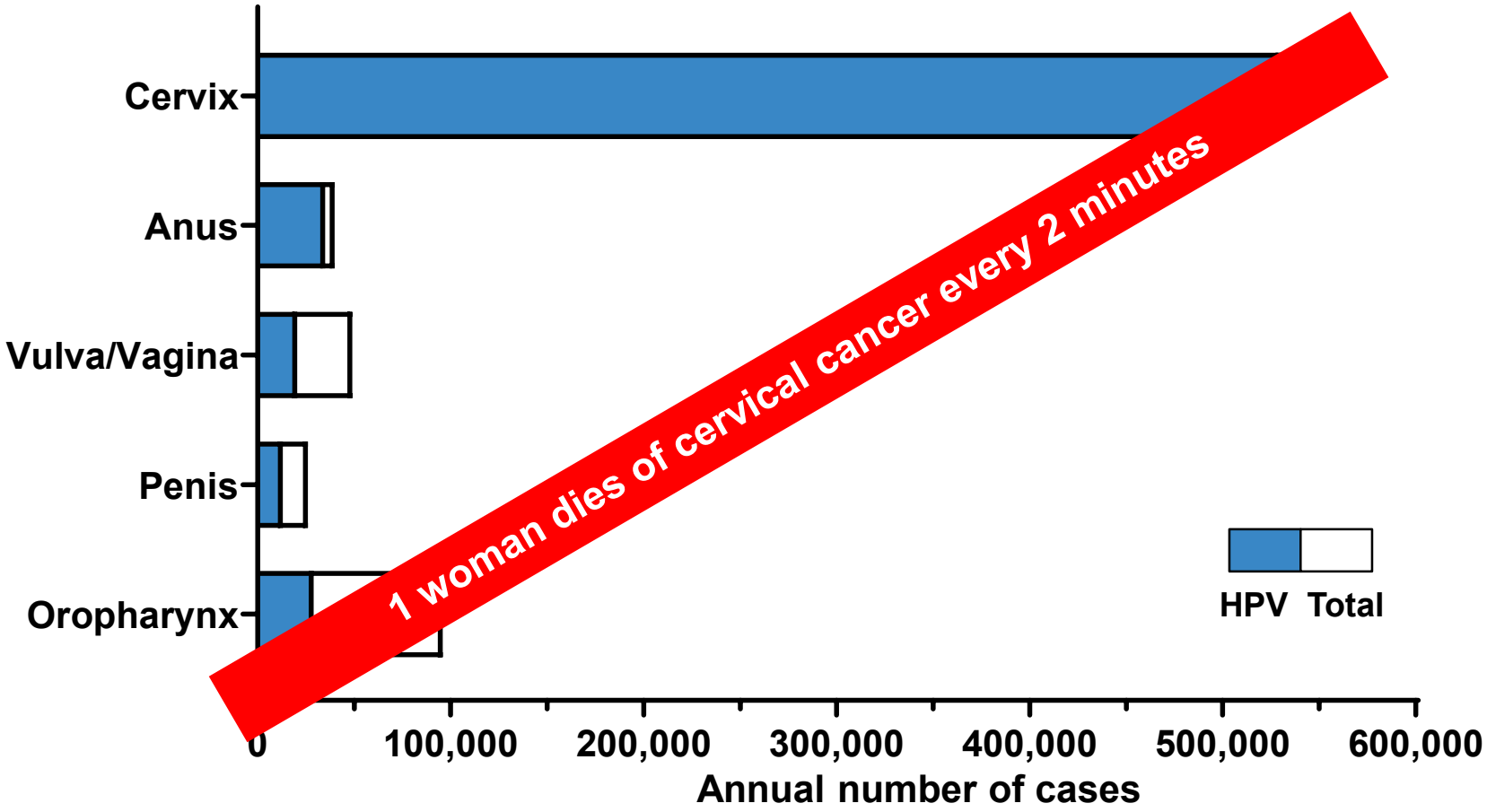
Most common sexually transmitted infection





Worldwide Incidence and Distribution of Cancers Attributable to HPV

Estimated cancer cases attributable to HPV ~ 5%

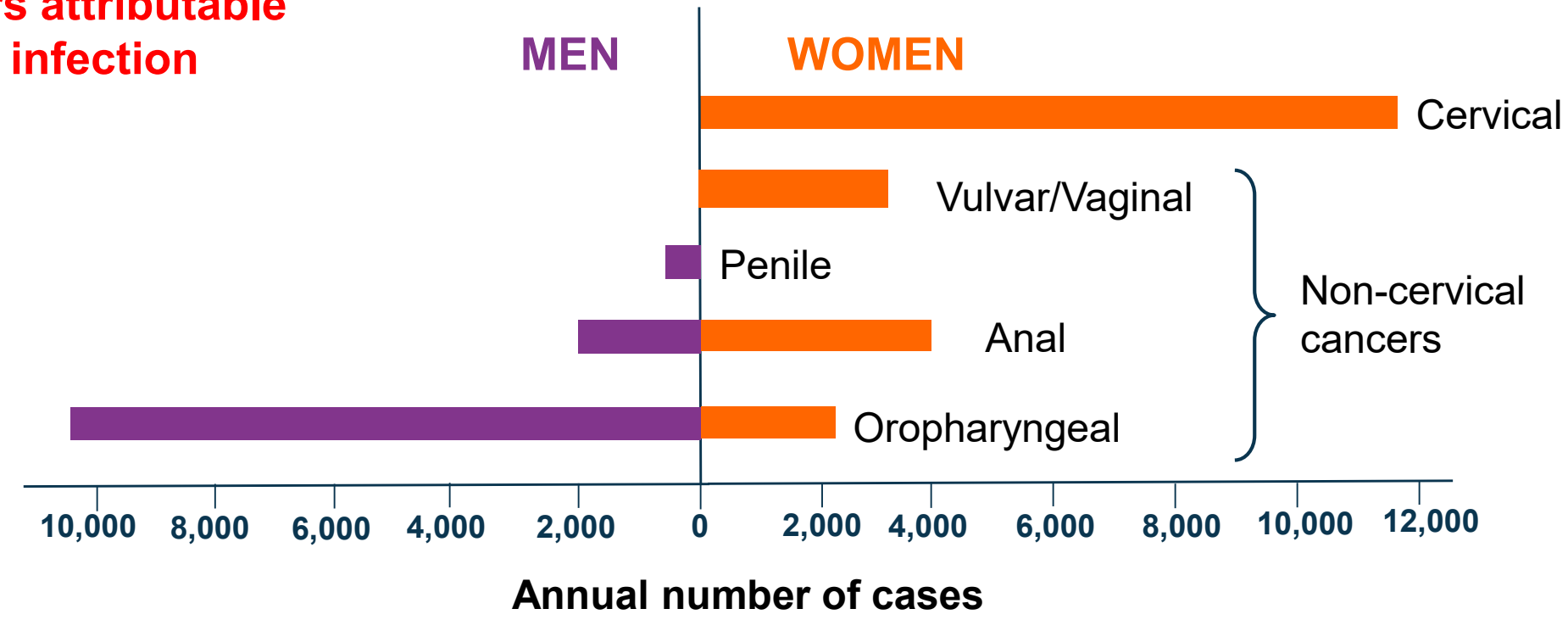


Cervical cancer accounts for ~ 83% of HPV attributable cancers globally

Adapted from de Martel et al., Int J Cancer 2017;141(4): 664, GLOBOCAN

USA: HPV-Associated Cancers Affect Both Sexes

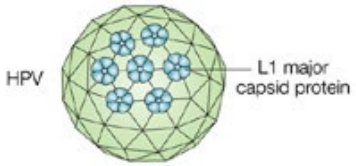
Cancers attributable to HPV infection



- **Total number of HPV-positive cancers = ~34,800, ~60% women; ~40% men**
- **HPV-16/18: Accounts for ~70% of cervical cancers, ~90% of non-cervical cancers**

Adapted from Senkomago et al., MMWR, Vol 68, #33, August 23, 2019

HPV and HPV-Associated Cancers Are Preventable: Three Licensed HPV Prophylactic Vaccines

	**Quadrivalent (Gardasil®) Merck**	**Bivalent (Cervarix®) GSK**	**Nonavalent (Gardasil®9) Merck**
HPV L1 VLP types	HPV-6/11/16/18	HPV-16/18	HPV-6/11/16/18/ 31/33/45/52/58
FDA Approval	2006	2009	2014
Sex/age groups	Females and males 9-26 yrs	Females 9-25 yrs	Females and males 9-26 yrs
Licensed* Schedule	3 doses	3 doses	3 doses
Expected Coverage	70% of Cervical Cancers 90% Genital Warts	70% of Cervical Cancers	90% of Cervical Cancers 90% Genital Warts

** New recommendation approved by FDA (Oct 2016): 2 doses (0 and 6 month), 9-14 yrs*

 # New approval by FDA (Oct 2018): 27-45 yrs for Gardasil®9

High Efficacy of HPV Prophylactic Vaccines Against New Cervical Precancer and Genital Warts in Randomized Trials

In women with no genital HPV infection detected at the start of each trial

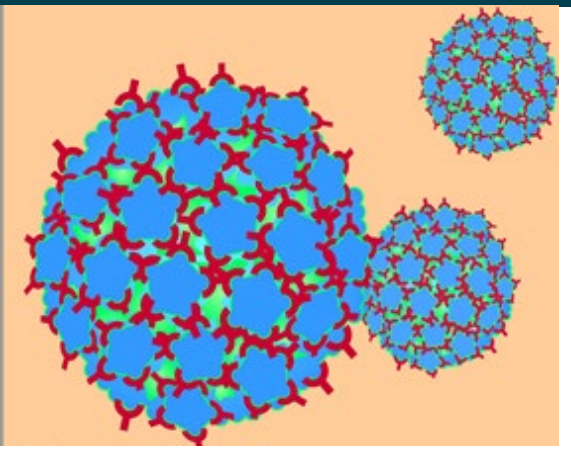
End Point	Sex	Age	Vaccine	Targeted HPV Types	Efficacy (95% CI)
Precancer	Female	15-26	Quadrivalent/ Gardasil	HPV-6/11/16/18	100% (85.5-100)
Genital Warts	Female	15-26	Quadrivalent/ Gardasil	HPV-6/11/16/18	96.4% (91.4-98.4)
Precancer	Female	15-25	Bivalent/ Cervarix	HPV-16/18	100% (90.5-100)
Precancer	Female	16-26	Nonavalent/ Gardasil 9	HPV-6/11/16/18/ 31/33/45/52/58	97.1% (83.5-99.9)

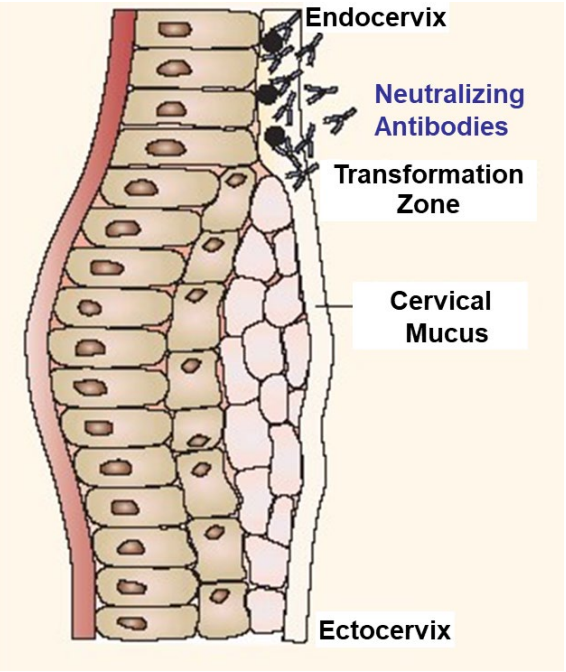
Precancer = Cervical Intraepithelial Neoplasia Grade 3

Lehtinen et al., Lancet Oncol 2011; Munoz et al., JNCI 2010; Huh et al., Lancet 2017

HPV Vaccine Immunogenicity: A Critical Parameter in HPV Vaccine Trials

- **HPV serology** is a critical tool for assessment of vaccine immunogenicity, which is a **key parameter** along with clinical efficacy (protection against infection/or disease) in vaccine trials
- **HPV neutralizing antibodies** are the main **mechanism of protection** against infection induced by prophylactic vaccines
- Vaccination in animal papillomavirus models have implicated **antibodies as immune effectors**
  - **Protection against infection** and subsequent lesion development **can be passively transferred** by serum antibodies





HPV Immunology Laboratory Support to HPV Vaccine Trials: Immunogenicity Studies

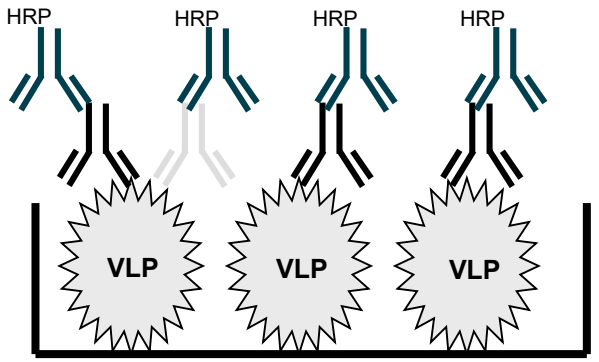
- **Phase III Trial of the Bivalent HPV Vaccine in Healthy Young Adult Women in Costa Rica (NCI Costa Rica Vaccine Trial) (PI: Dr. Allan Hildesheim, NCI)**
  - Women, ages 18-25 years (n=7,466) (NCT00128661, NCT00867464)
- **Safety and Immunogenicity Trial in Adult Volunteers of an HPV-16 L1 VLP Vaccine (PI: Dr. Clayton Harro, The Johns Hopkins University)**
  - Women, ages 18-29 years (n=72)
- **Phase II Trial of the Quadrivalent HPV Vaccine in Mid-Adult Males (MAM) (PI: Dr. Anna Giuliano, Moffitt Cancer Center)**
  - Men, ages 27-45 years (n=150) (NCT01432574)

- Developed and optimized **assays** for measuring markers of protection against infection, both in blood and at sites of infection
- Monitored **duration** of antibody responses to vaccination
  - ***Neutralizing antibodies*** *are the main mediators of protection*
- Demonstrated **long-term antibody responses even in single dose** vaccine recipients – suggesting a single dose may work

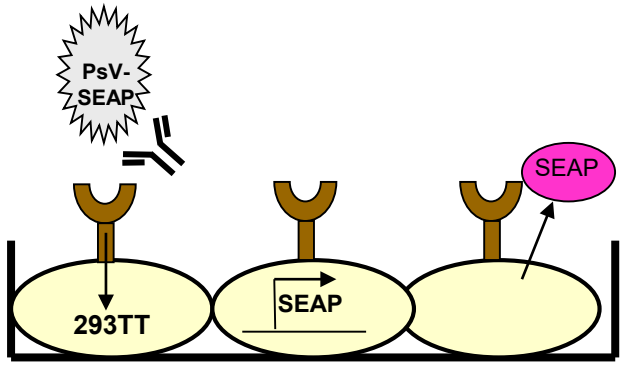
Validated Assays for Measurement of Antibodies (In Serum and at Mucosal Sites)

HPV Serology Assays

- **Quantity:**
  - **HPV IgG ELISA**
    - **HPV-16/18 L1 VLP**
  - **Luminex-based multiplex assay**



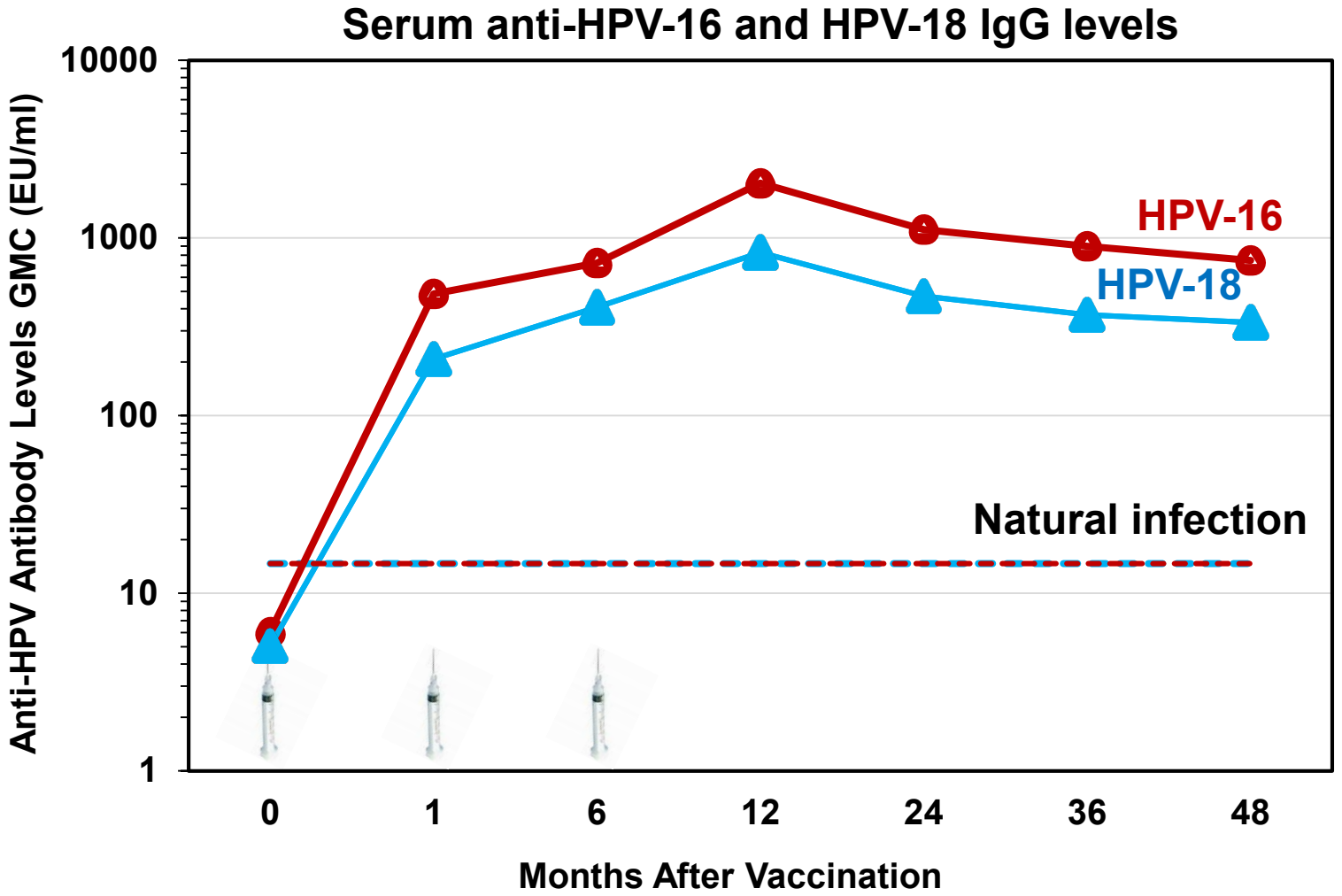
- **Quality/Function:**
  - **Pseudovirion neutralization assay**
    - **HPV-16/18/31/45/52/58**
  - **ELISA antibody avidity assay**
  - **Memory B cell ELISPOT**



All assays use in-house produced HPV VLPs, standards, and controls

HPV-16/18 L1 VLP Vaccine Immunogenicity: NCI Costa Rica Vaccine Efficacy Trial

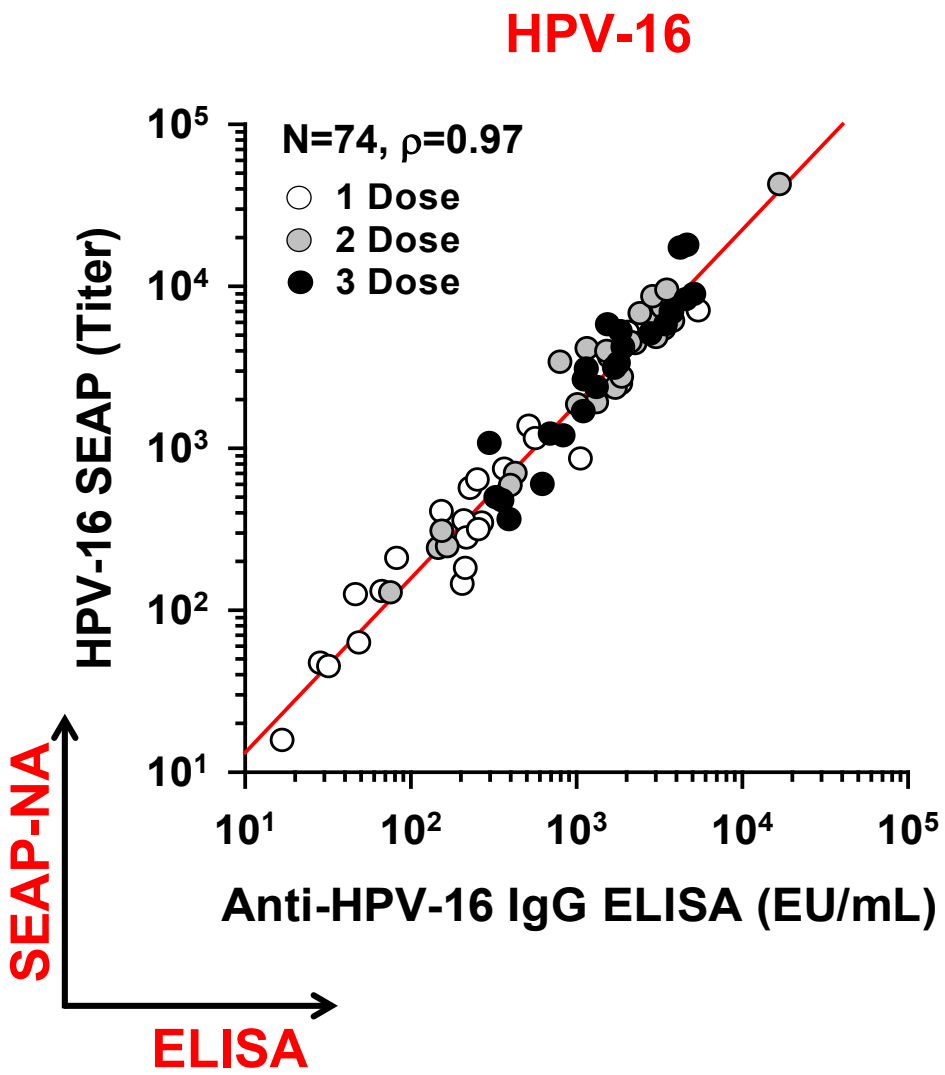
ELISA

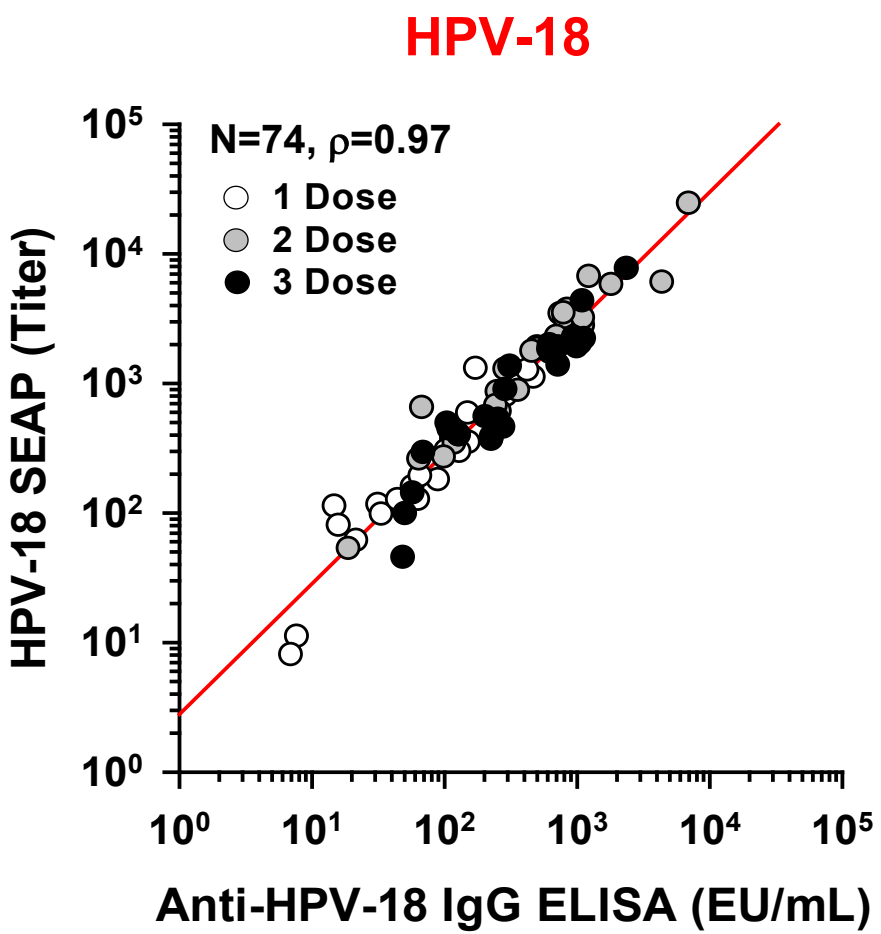


ELISA cutoff: HPV-16= 8 EU/ml
HPV-18= 7 EU/ml

Adapted from Safaeian et al., Cancer Prevention Research 2013; 6(11): 1242

Strong Correlations between ELISA and Neutralization Assays

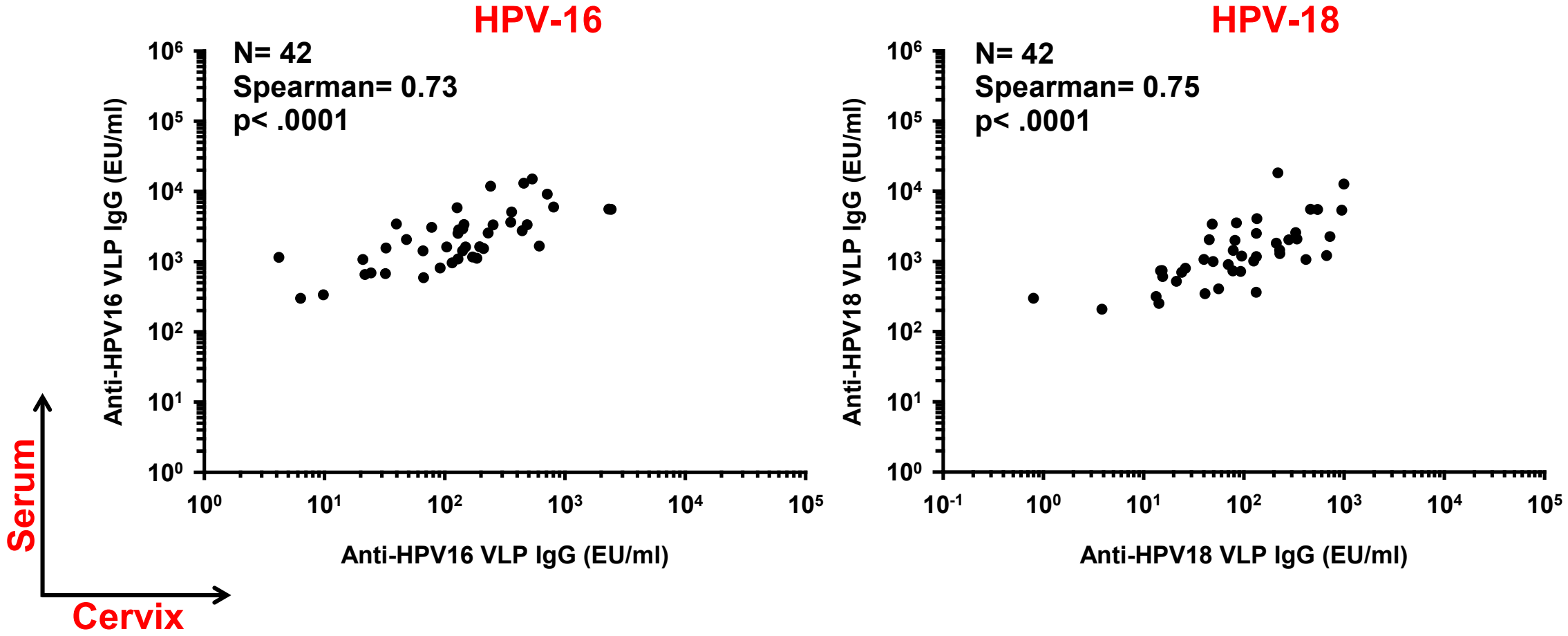




LaMontagne et al., Vaccine 2014; 32 (47):6303

Antibody Levels at the Cervix Correlate with Systemic Levels

NCI Costa Rica Vaccine Trial (CVT)

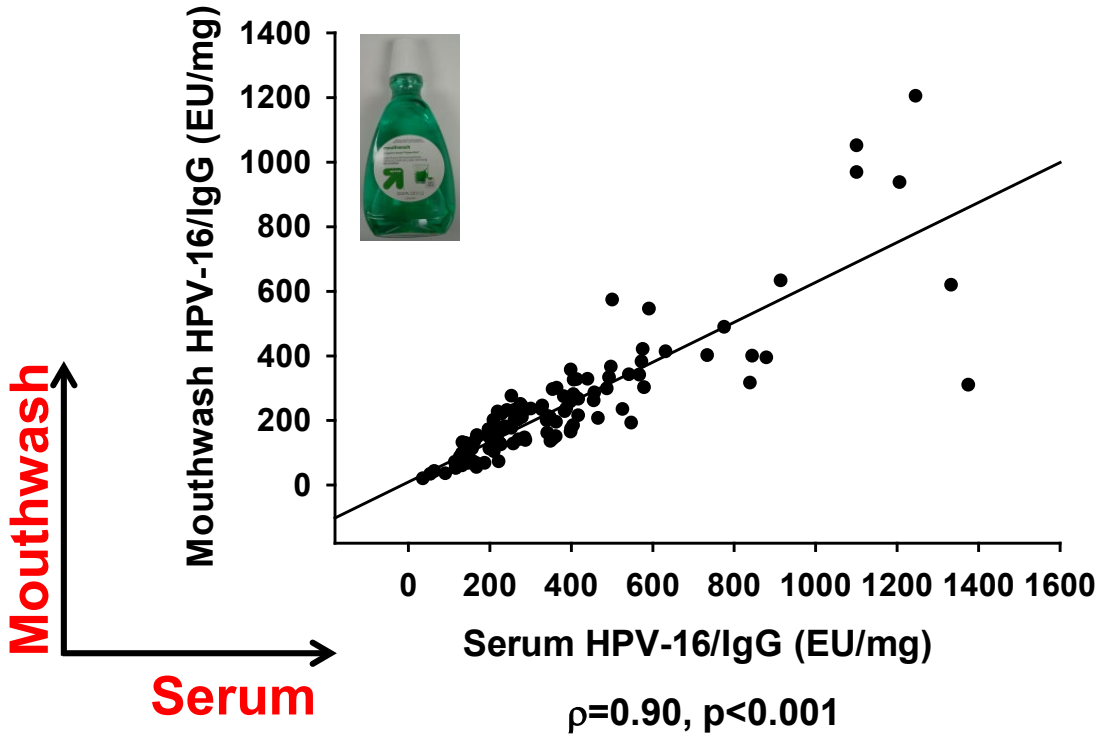


Kemp et al., Vaccine 2008; 26: 3608

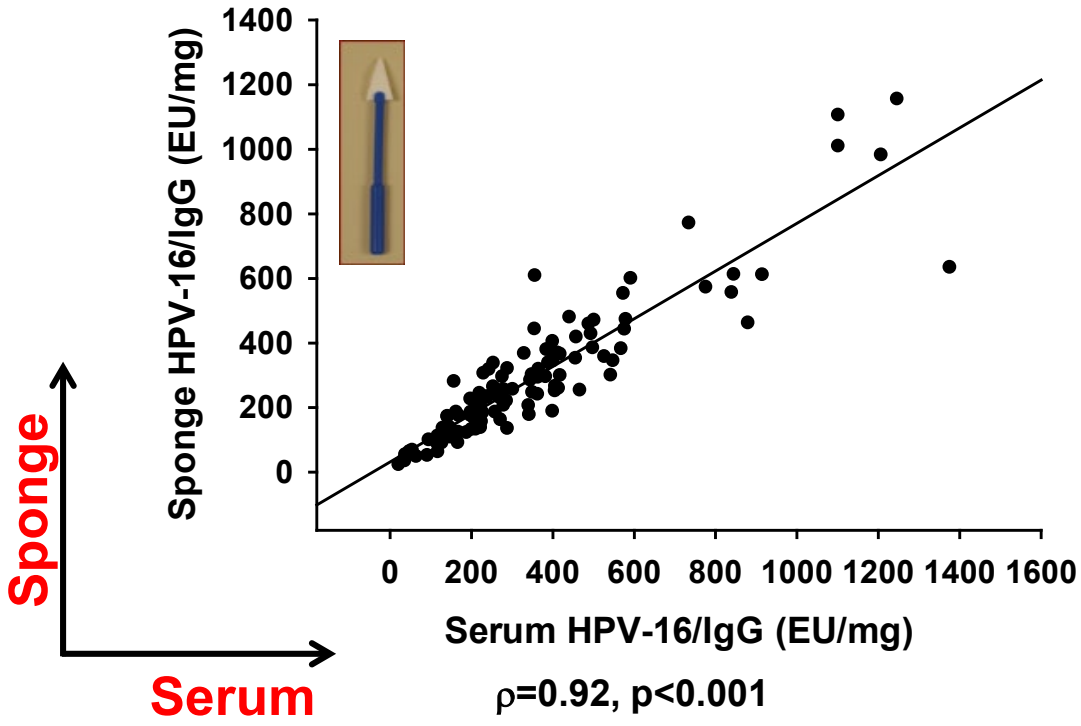
Antibody Levels in Saliva Correlate with Systemic Levels

HPV-16 antibody 1 month post-dose 3 of quadrivalent HPV vaccine
(Cohort: Mid-Adult Male Vaccine Trial, Moffitt Cancer Center)

HPV-16
Mouthwash



HPV-16
Sponge



Pinto et al., J Infect Dis. 2016, 214(8):1276

One Dose of the Bivalent HPV Vaccine Is Effective in Preventing HPV Infection Compared With Multiple Doses (48 Months After Vaccination)

Cohort: NCI Costa Rica Vaccine Trial, HPV-16/18 DNA negative at 1st vaccination
Endpoint: Incident HPV-16/18 infections that persist for >6 months

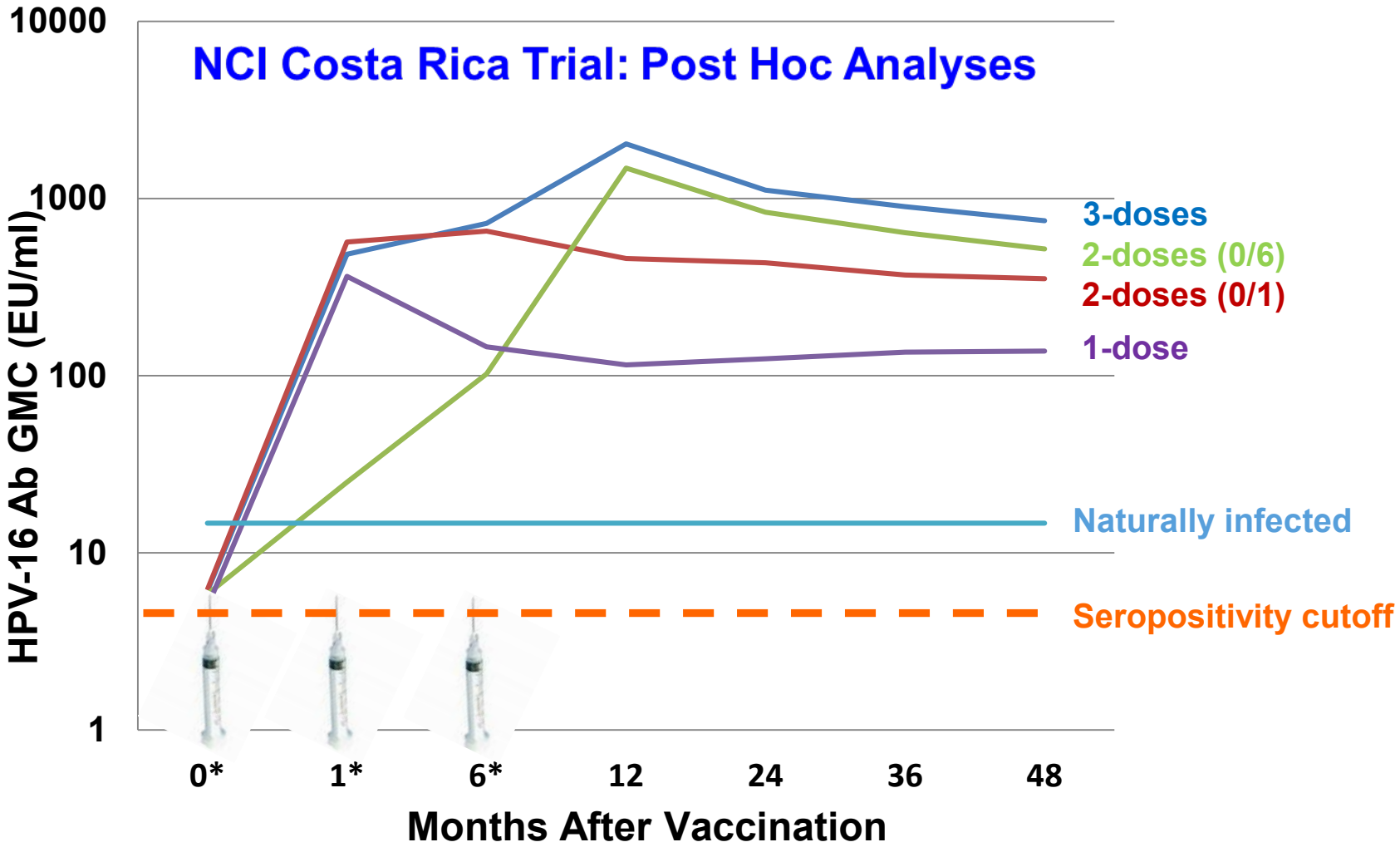
# of Doses	Arm	# of Women	# of Persistent HPV-16/18 Infections	HPV-16/18 VE (95%CI)
3	Control	3010	133	81% (71% to 88%)
	HPV	2957	25	
2	Control	380	17	84% (50% to 96%)
	HPV	422	3	
1	Control	188	10	100% (67% to 100%)
	HPV	196	0	

VE= Vaccine Efficacy

p trend= 0.2

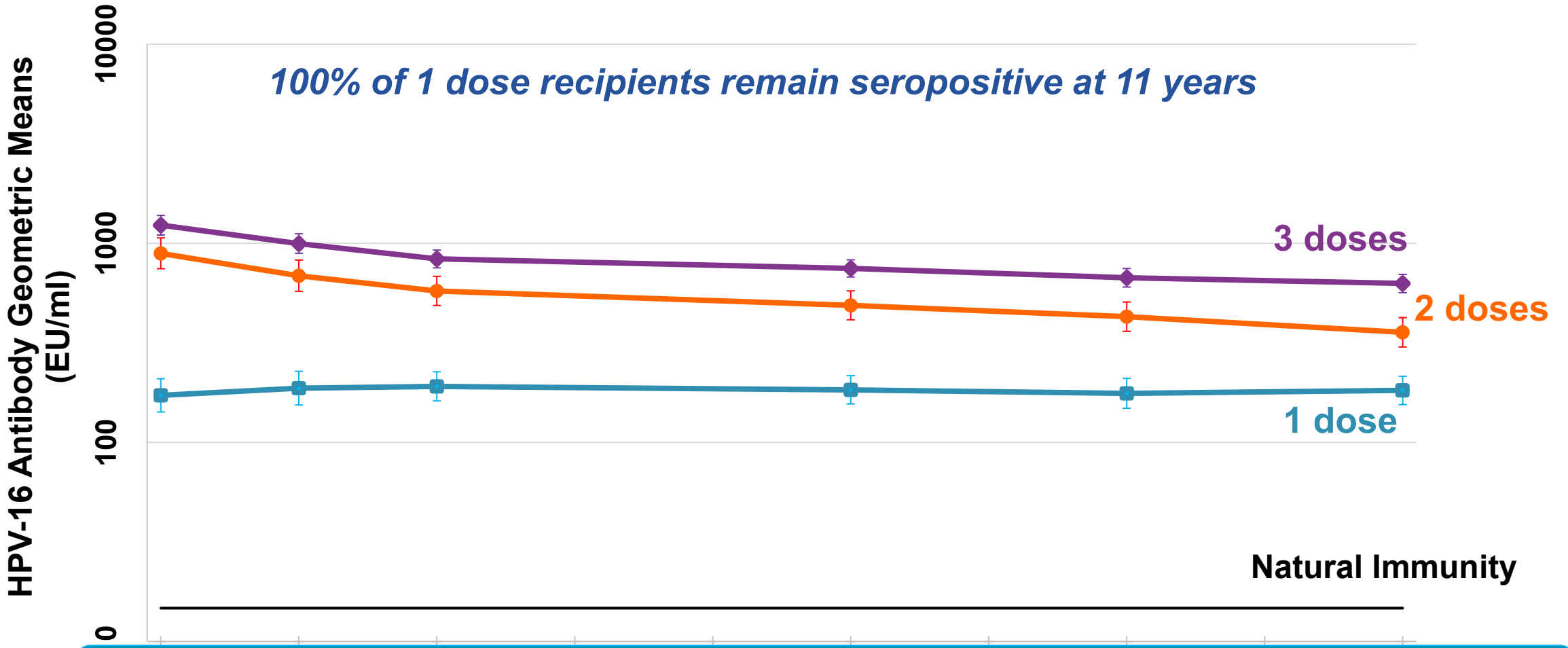
Kreimer et al., JNCI 2011; 103(19):1444

The Bivalent HPV Vaccine Induces Durable Antibody Responses in One Dose Recipients



Safaeian et al., Cancer Prevention Research 2013; 6(11): 1242

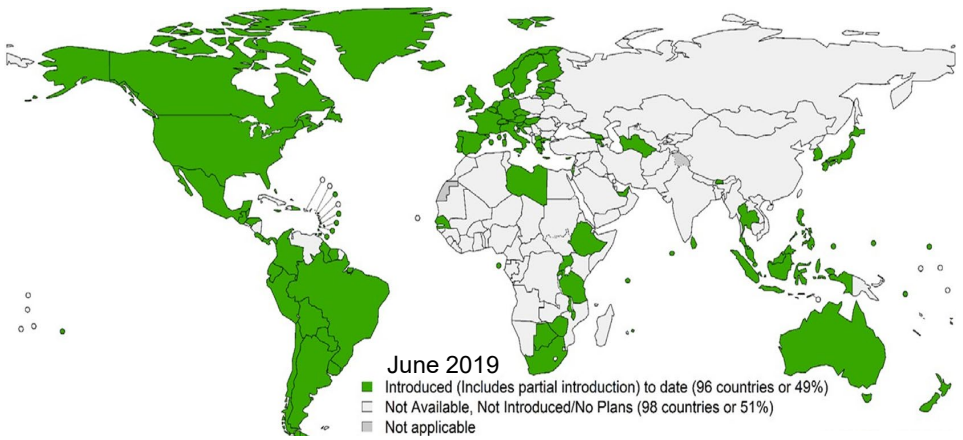
Stable HPV-16 Serum Antibodies a Decade After One Dose of the Bivalent HPV Vaccine (NCI Costa Rica Vaccine Trial)



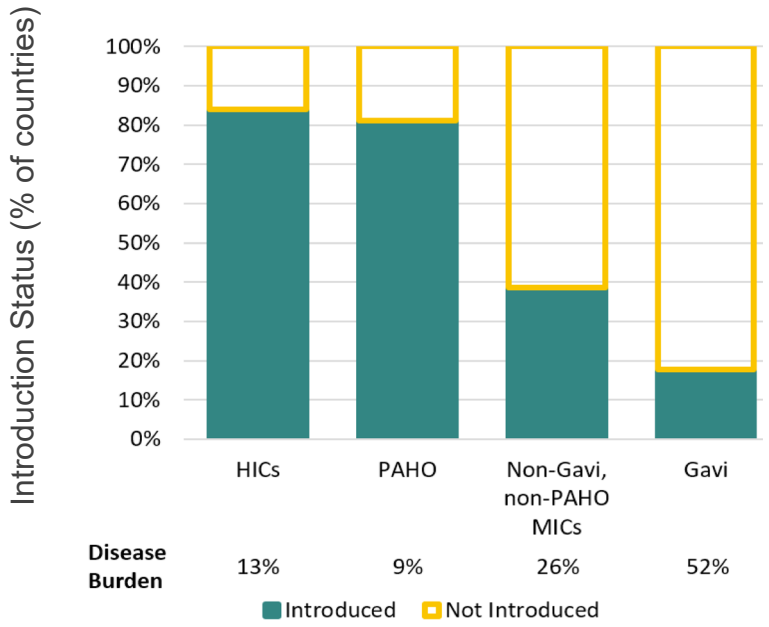
One dose schedules may help reduce the current programmatic and economic challenges

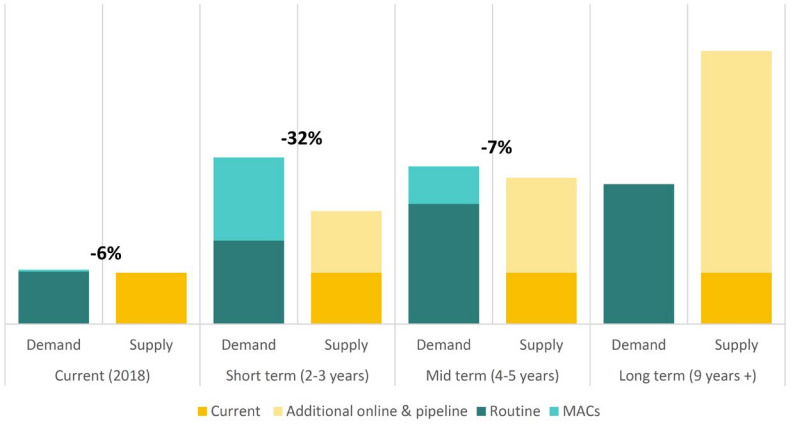
Kreimer et al., Submitted; Safaeian et al., JNCI 2018; 110 (2): 205

HPV Vaccination Status: Current Global HPV Vaccine Demand/Supply Imbalance



- Only ~50% of countries with national immunization programs
- Introductions are lowest in low- and middle-income countries (LMIC)
- Very low global vaccine uptake
- WHO call for action towards global cervical cancer elimination will increase demand over the next 10 years
- Imbalance projected to last 3-5 years





World Health Organization

MI4A
MARKET INFORMATION FOR ACCESS TO VACCINES

GLOBAL MARKET STUDY
HPV
09/2018

New One Dose Trials to Evaluate Immunogenicity and Non-Inferiority at FNL to Accelerate Policy Recommendations (2020-2025)

Scientific evaluation of one or two doses of the bivalent or nonavalent prophylactic HPV vaccines (ESCUDDO): (Dr. Aimee Kreimer, NCI), 20,000 girls, ages 12-16, NCT03180034 — **Costa Rica**

Kenya single dose HPV vaccine efficacy (KEN SHE): (Dr. Ruanne Barnabas, University of Washington), 2,250 women, ages 15-20, NCT03675256 — **Kenya**

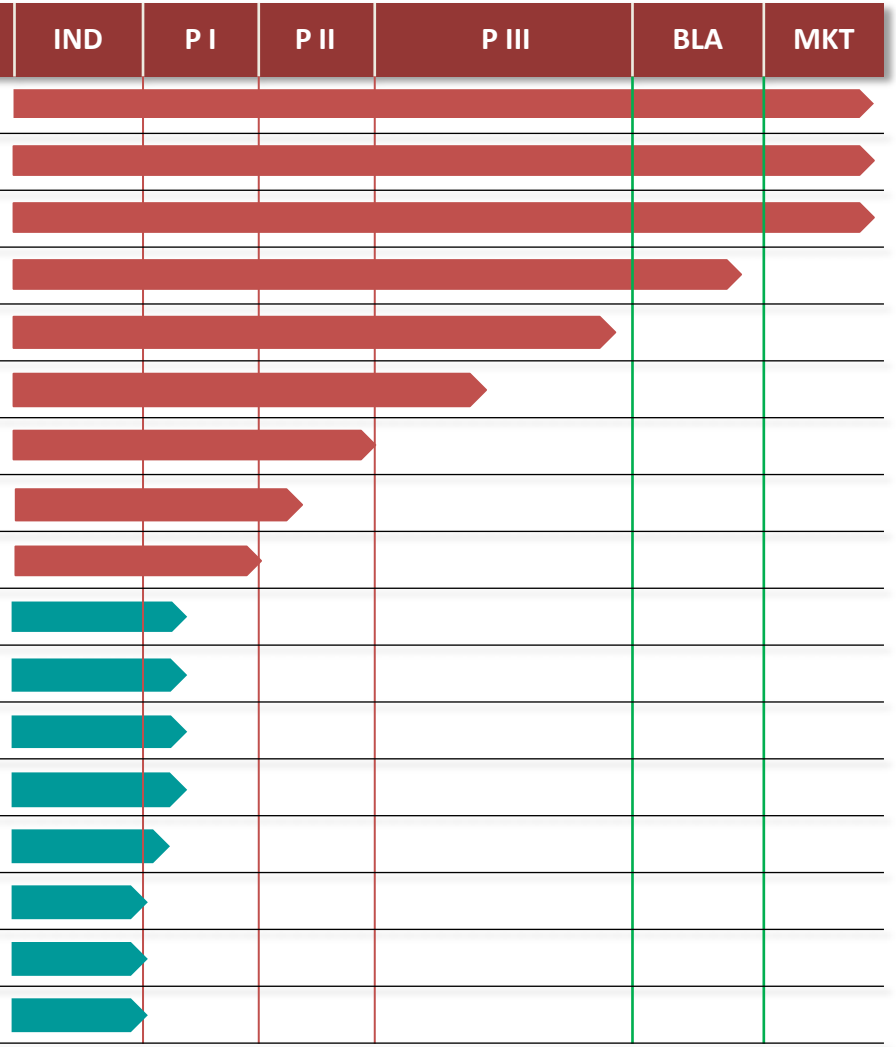
A dose reduction immunobridging and safety study of two HPV vaccines in Tanzanian girls (DoRIS): (Dr. Deborah Watson-Jones, University of London), 900 girls, ages 9-14, NCT02834637 — **Tanzania**

Non-inferiority trial comparing immunogenicity from 1 dose of bivalent HPV vaccine in girls to 3 doses of quadrivalent vaccine in women (PRIMAVERA): (Dr. Aimee Kreimer, NCI), 620 girls, ages 9-14 and 620 women, ages 18-25, NCT03728881 — **Costa Rica**

Phase IIA trial of a nonavalent prophylactic HPV vaccine to assess immunogenicity of a prime and deferred-booster dosing schedule among 9-11 year-old girls and boys: (Drs. Yi Zeng, Anna-Barbara Moscicki and Vikrant Sahasrabuddhe), 200 boys and girls, ages 9-11, NCT02568566 — **USA**

Increasing Number of Prophylactic HPV Vaccines in Development - China

	Company	Vaccine	Expression system
1	GSK	HPV-2 (16, 18)	*Insect cell*
2	Merck	HPV-4 (6,11,16,18)	*Yeast (S. cerevisiae)*
3	Merck	HPV-9 (6,11,16,18,31,33,45,52,58)	*Yeast (S. cerevisiae)*
4	Innovax	HPV-2 (16,18)	*E.coli*
5	Zerun	HPV-2 (16,18)	*Yeast (P. pastoris)*
6	CNBG/CDIBP	HPV-4 (6,11,16,18)	*Yeast (H. polymorpha)*
7	Innovax	HPV-2 (6,11)	*E.coli*
8	Kangleweishi	HPV-3 (16,18,58)	*E.coli*
9	Bovax	HPV-4 (6,11,16,18)	*Yeast (H. polymorpha)*
10	Bovax	HPV-9 (6,11,16,18,31,33,45,52,58)	*Yeast (H. polymorpha)*
11	Zerun	HPV-9 (6,11,16,18,31,33,45,52,58)	*Yeast (P. pastoris)*
12	Innovax	HPV-9 (6,11,16,18,31,33,45,52,58)	*E.coli*
13	CNBG/SIBP	HPV-4 (6,18,52,58)	*Yeast (P. pastoris)*
14	Kangleweishi	HPV-9 (6,11,16,18,31,33,45,52,58)	*E.coli*
15	Jiangsu Ruike	HPV-9 (6,11,16,18,31,33,45,52,58)	*Yeast (H. polymorpha)*
16	CNBG/CDIBP	HPV-11 (6,11,16,18,31,33,45,52,58,59,62)	*Yeast (H. polymorpha)*
17	Nuoning	HPV-14 (6,11,16,18,31,33,35,39,45,51,52,56,58,59)	*Insect cell*



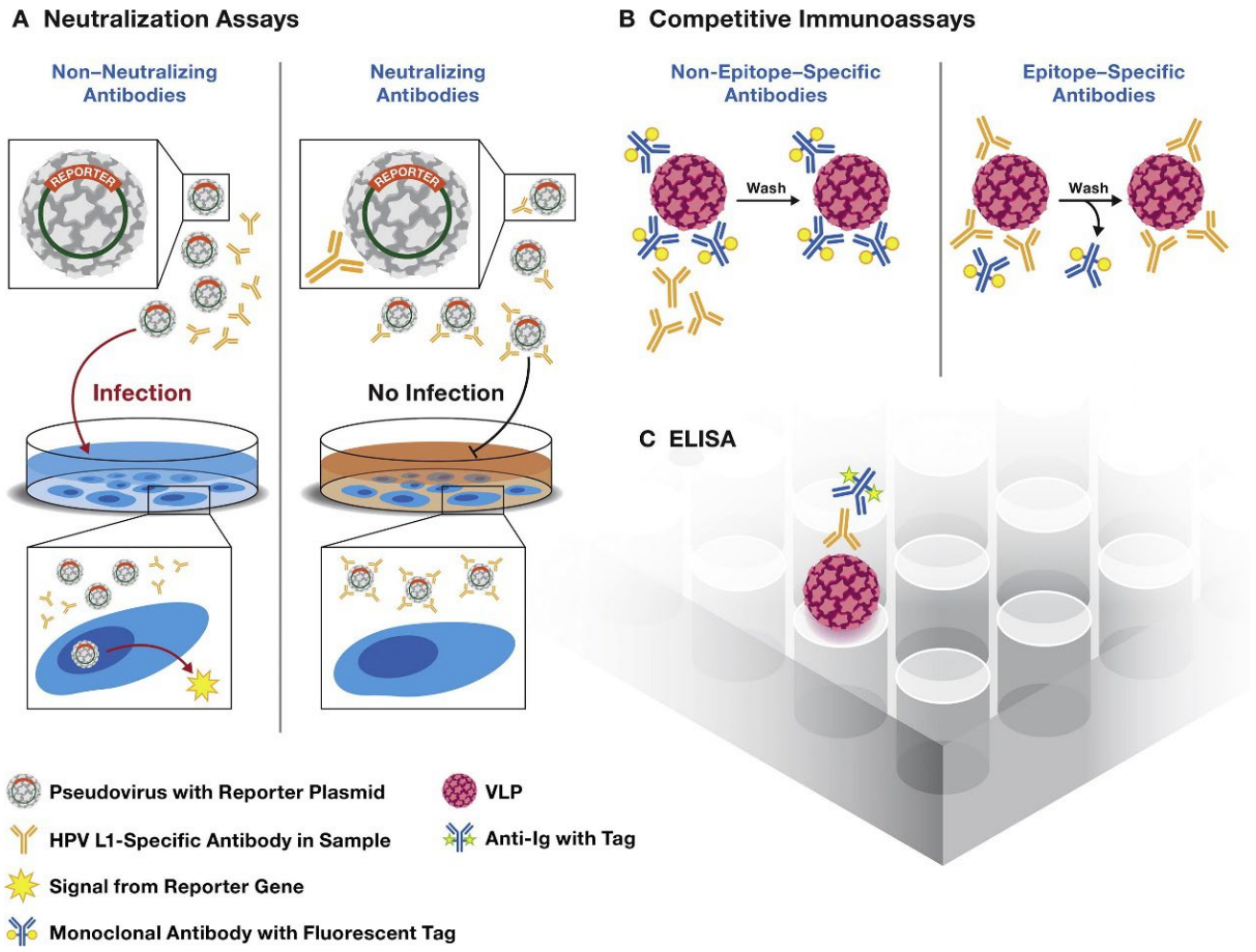
Early stage Late stage

Data source: NMPA data; clinicaltrial.gov. updated in April, 2019

Prepared by Peter Dull, BMGF

Transition from Clinical to Serology Endpoints in Vaccine Trials: Increased Demand for Standardized Serology Testing

- Increasing number of clinical trials are proposing to use serology as endpoints
- No commercially available validated assays
- Lack of uniform, standardized assays, procedures, and reagents accessible to the vaccine trial community
- Vaccine trials have used different serologic assays, different VLP production methods and different reporting units
- **Difficulty in comparing results across studies**



The HPV Serology Standardization Initiative - 2017
(Sponsored by NCI and The Bill & Melinda Gates Foundation)

HPV Serology Lab
Advancing HPV Assay Standardization

Mission:

- To work in **partnership** with the **international** HPV serology community to **promote further standardization, harmonization and proficiency of HPV serology assays** to assess vaccine immunogenicity in vaccine trials through:
  - development of **qualified assay standards**, **critical reagents** (HPV Virus-Like Particles), **multiplex assays and guidelines** available to the scientific community

Impact:

- Enable comparisons of data between different vaccines and studies
- Accelerate implementation of new vaccines and new vaccine recommendations

Partners:

Frederick National Laboratory: Ligia Pinto, Troy Kemp

NCI: Doug Lowy, John Schiller, Sean Hanlon

The Bill & Melinda Gates Foundation: Peter Dull

CDC: Elizabeth Unger

Karolinska Institute: Joakim Dillner

Public Health England: Simon Beddows

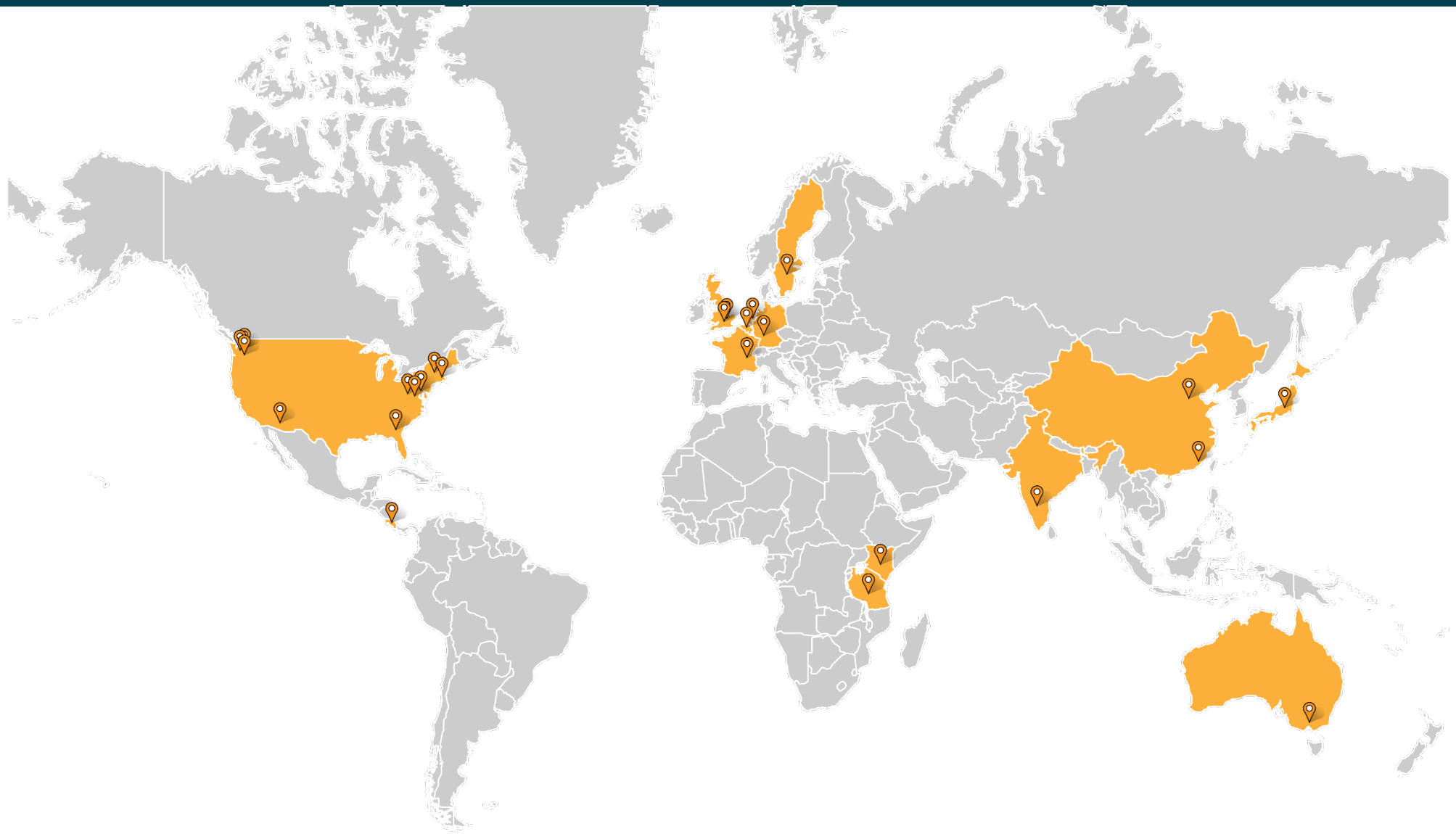
Biostat Consulting, LLC: Brian Plikaytis

https://frederick.cancer.gov/science/hpvserologylab/overview

HPV Serology Standardization: Progress to Date

- ✓ Generated **qualified secondary standards** and bank of specimens for use as **assay proficiency panels**
- ✓ Completed production of **qualified reference Virus-Like Particles** for the 9 HPV types included in the nonavalent HPV vaccine
- ✓ Developed and validated **Multiplex Serology Assays** for vaccine trial use
- ✓ **Promote use of standards** (meetings, publications, data, and protocol sharing)
  - ✓ FNL developed a **website** with all Standard Operating Procedures created in-house
  - ✓ Host **meetings** at the International Papillomavirus Conferences
  - ✓ **20 Material Transfer Agreements (MTA)**

HPV Serology Standardization: Collaboration Across the Globe (20 Material Transfer Agreements)



Main Achievements Working Together With NCI and Extramural Collaborators

- ✓ Contributed to more than **93 peer-reviewed publications** (52 in the area of HPV)
- ✓ Established four **Contractor Cooperative Research and Development Agreements (cCRADAs)** to study immune responses to vaccination in females and males
- ✓ Provided laboratory evidence that stimulated the ongoing large **NCI single dose efficacy trial of the HPV vaccine in Costa Rica** (20,000 participants) and other single dose HPV vaccine trials
- ✓ Provided preliminary evidence to support HPV vaccine **efficacy trials in adult men living with HIV**
- ✓ Established an **international HPV serology standardization initiative** to reliably measure immune responses to vaccination and enable comparisons of data between different studies

Impact of Our Work: How Are We Contributing to the Elimination of Cervical Cancer and Other HPV-Associated Cancers?

Investigate immune responses to vaccines, infections, and cancer

Develop and validate new methods for laboratory markers of protection

Monitor immunity in clinical trials and pre-clinical studies

Provide evidence to inform new trials and create tools to enable public health changes

Understanding how HPV vaccines work and mechanisms associated with protection

Providing immuno-epidemiological evidence to move forward with new trials for novel vaccine recommendations

Leading an international HPV serology standardization initiative to enable use of laboratory markers for demonstration of vaccine effectiveness

Supporting development and evaluation of new candidate vaccines

HPV Serology Standardization: Future Plans

- Establishment of a **Center of Excellence for HPV Serology**
- Implement **high throughput HPV antibody testing core unit, with Good Clinical Laboratory Practice (GCLP) capability** to support NCI trials and extramural vaccine trial community
- Build an **HPV Vaccine Trial Network** of laboratories involved in serological testing of HPV vaccines
  - Develop training capability
  - Develop an Assay Proficiency Panel Program
- Continue **working with NIBSC and WHO** for development of WHO International Standards as well as with all the other stakeholders

Acknowledgments

Frederick National Laboratory for Cancer Research

HPV Immunology Laboratory

- David Pan
- Marcus Williams

HPV Serology Laboratory

- Troy Kemp
- Angelina Richards

Moffitt Cancer Center

- Anna Giuliano

The Bill & Melinda Gates Foundation

- Peter Dull

Division of Cancer Epidemiology and Genetics, NCI

- Allan Hildesheim
- Mahboobeh Safaeian
- Aimee Kreimer

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