PREVENT
Griffithsin-based Rectal Microbicide Development Program

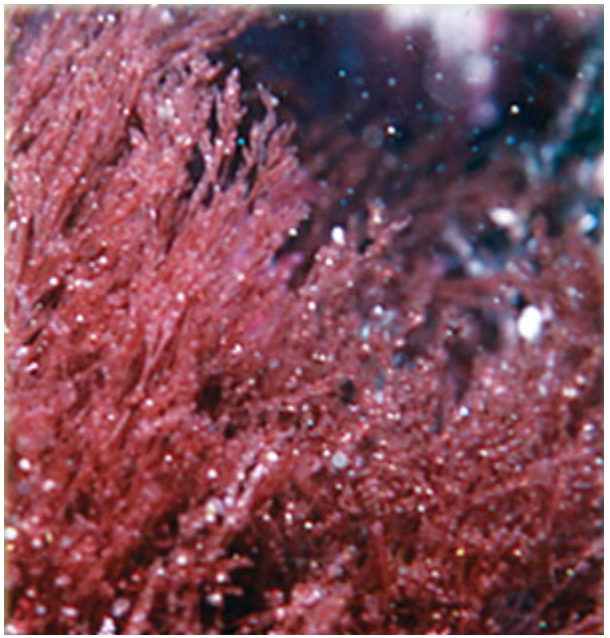
Kenneth E. Palmer, Ph.D.

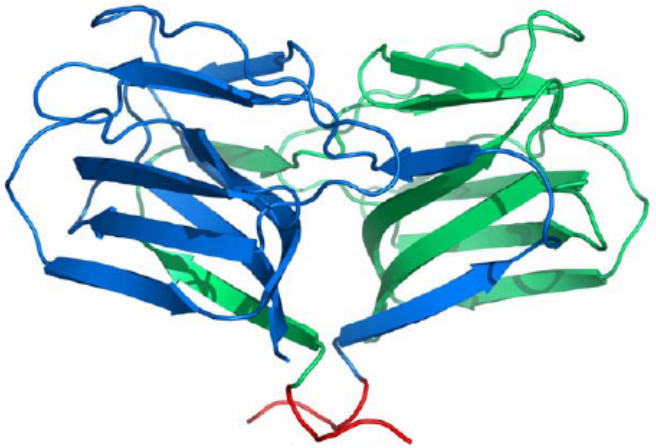
UofL SCHOOL OF MEDICINE

GRFT PREVENT

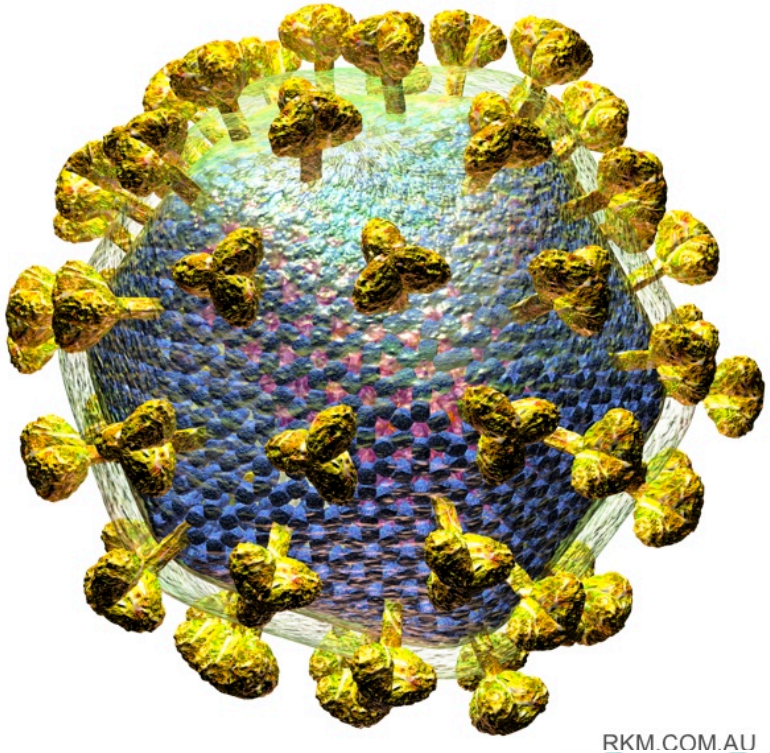
Griffithsin (GRFT)

- *Griffithsia,* red alga originally collected off Chatham Island New Zealand
  - Used for nutritive and traditional medicine
- Aqueous extract displayed potent anti-HIV activity
- Active constituent appeared to be a protein, one of the most potent HIV-1 entry inhibitors
- Active component is a lectin that targets the dense clusters of sugars (glycans) present on the surface of HIV





HIV-1 Envelope Glycosylation



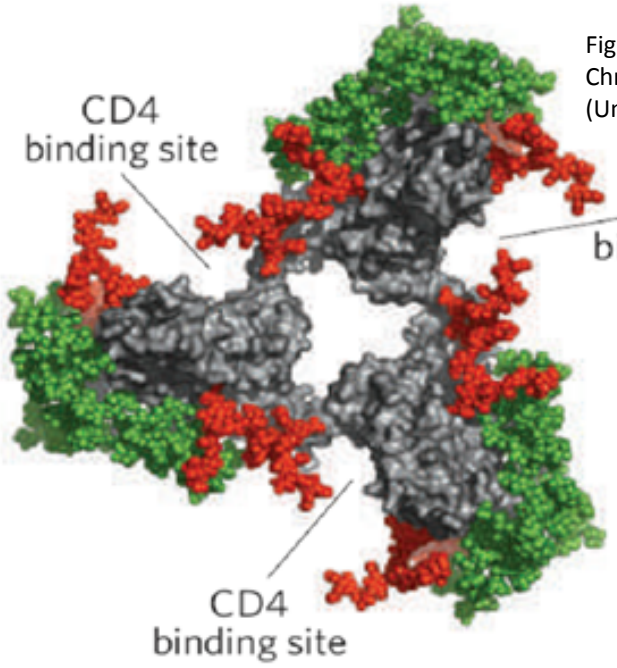
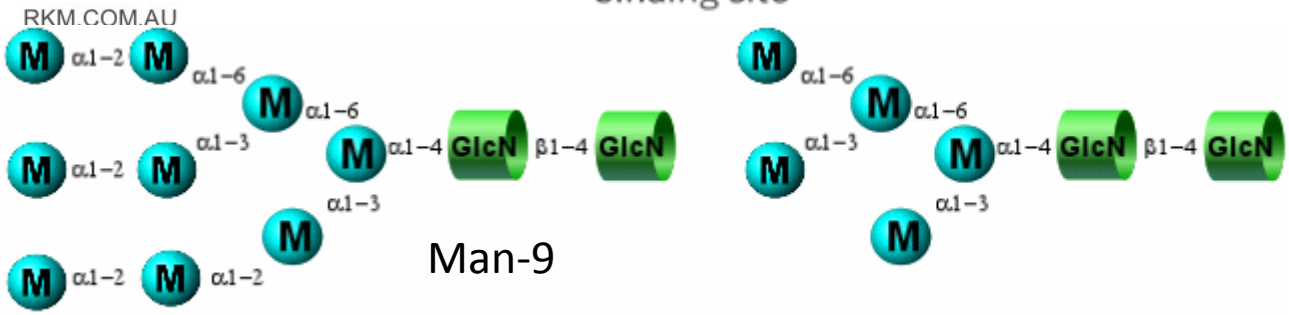


Figure courtesy of
Chris Scanlan
(University of Oxford)

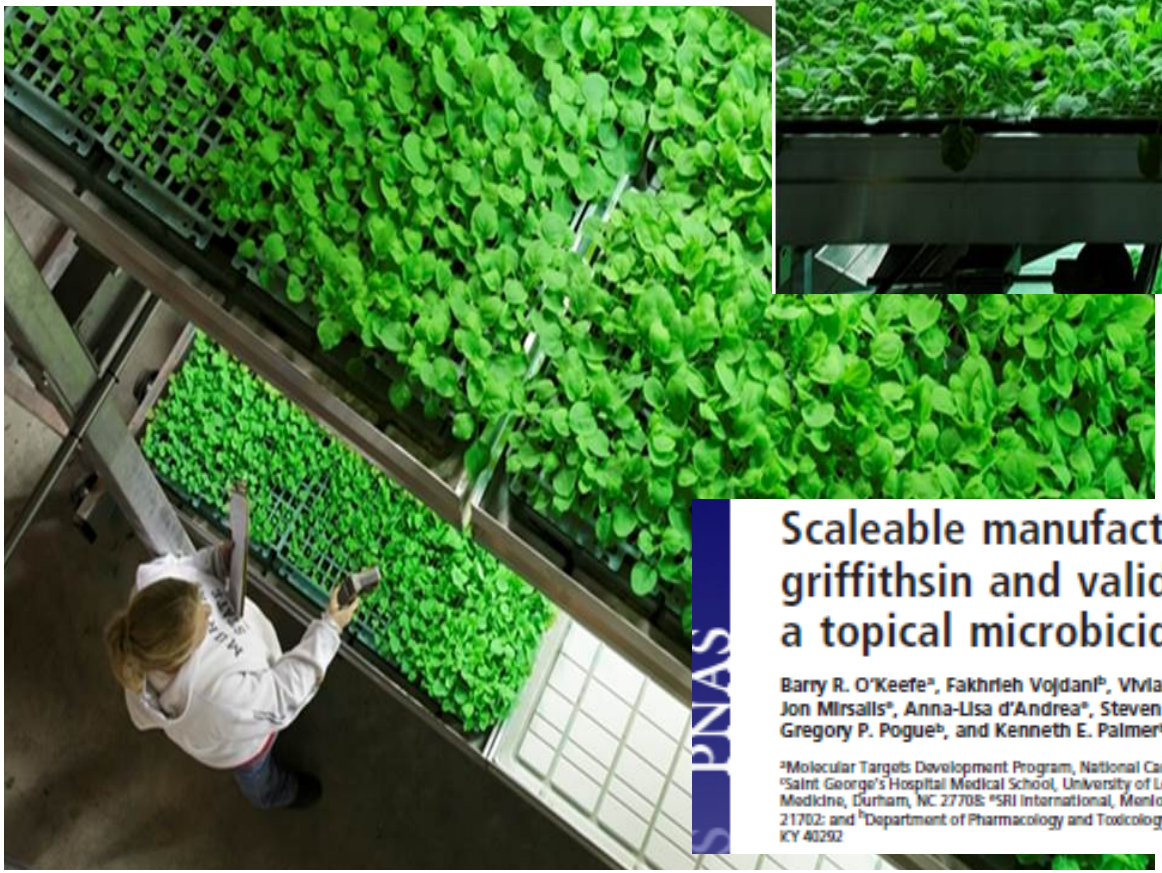
HIV-1 Oligomannose Glycans



Man-9

Manufacturing of GRFT

KBP
KENTUCKY BIOPROCESSING



PNAS

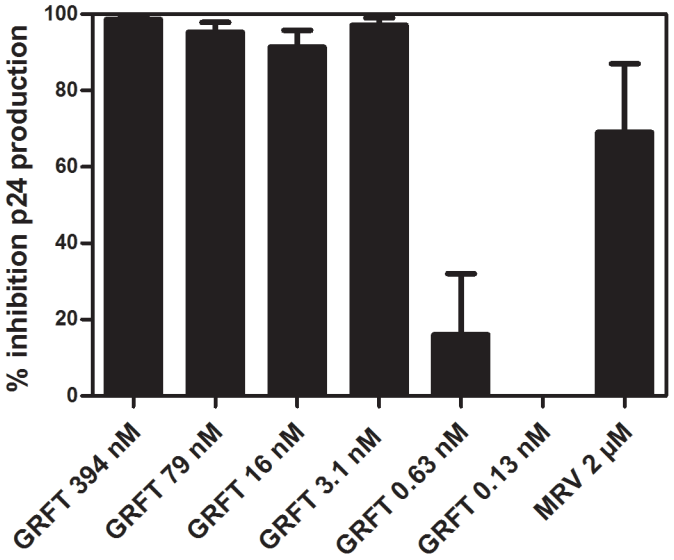
Scaleable manufacture of HIV-1 entry inhibitor griffithsin and validation of its safety and efficacy as a topical microbicide component

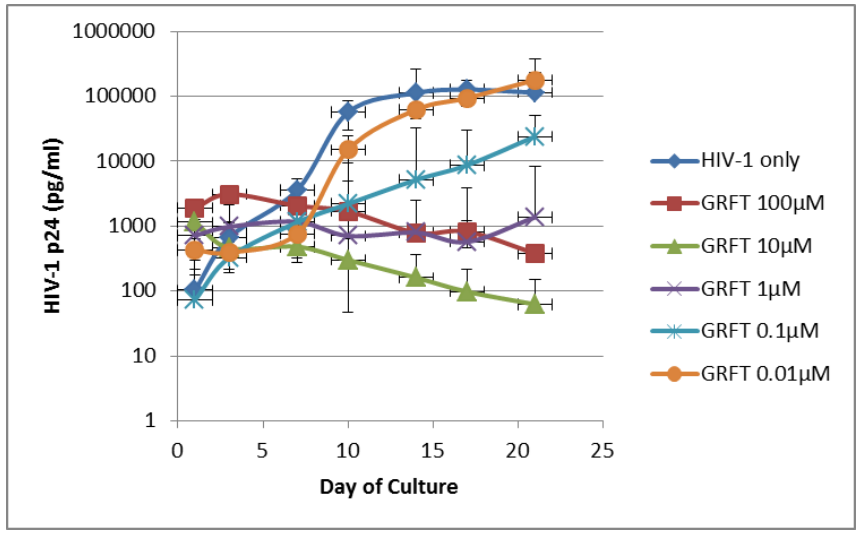
Barry R. O'Keefe[a], Fakhrieh Vojdani[b], Viviana Buffa[c], Robin J. Shattock[c], David C. Montefiori[d], James Bakke[e], Jon Mirsalis[e], Anna-Lisa d'Andrea[e], Steven D. Hume[f], Barry Bratcher[f], Carrie J. Saucedo[a,g], James B. McMahon[a], Gregory P. Pogue[b], and Kenneth E. Palmer[b,h,1]

[a]Molecular Targets Development Program, National Cancer Institute at Frederick, Frederick, MD 21702; [b]Intrucept Biomedicine, Owensboro, KY 42301; [c]Saint George's Hospital Medical School, University of London, London SW17 0RE, United Kingdom; [d]Department of Surgery, Duke University School of Medicine, Durham, NC 27708; [e]SRI International, Menlo Park, CA 94025; [f]Kentucky Bioprocessing, Owensboro, KY 42301; [g]SAIC-Frederick, Frederick, MD 21702; and [h]Department of Pharmacology and Toxicology and James Graham Brown Cancer Center, University of Louisville School of Medicine, Louisville, KY 40292

Griffithsin Activity Against HIV-1

- *In vitro,* Griffithsin has mid picomolar to low nanomolar entry inhibitor activity against a broad range of primary HIV-1 isolates from all clades tested to this point.
- GRFT shows good synergy *in vitro* with ARV from other classes e.g. TFV, RAL, MVC





- Persistent HIV inhibitory activity in pre-treated PBMC suggests that surface-bound GRFT retains antiviral activity.

OPEN ACCESS Freely available online PLoS one

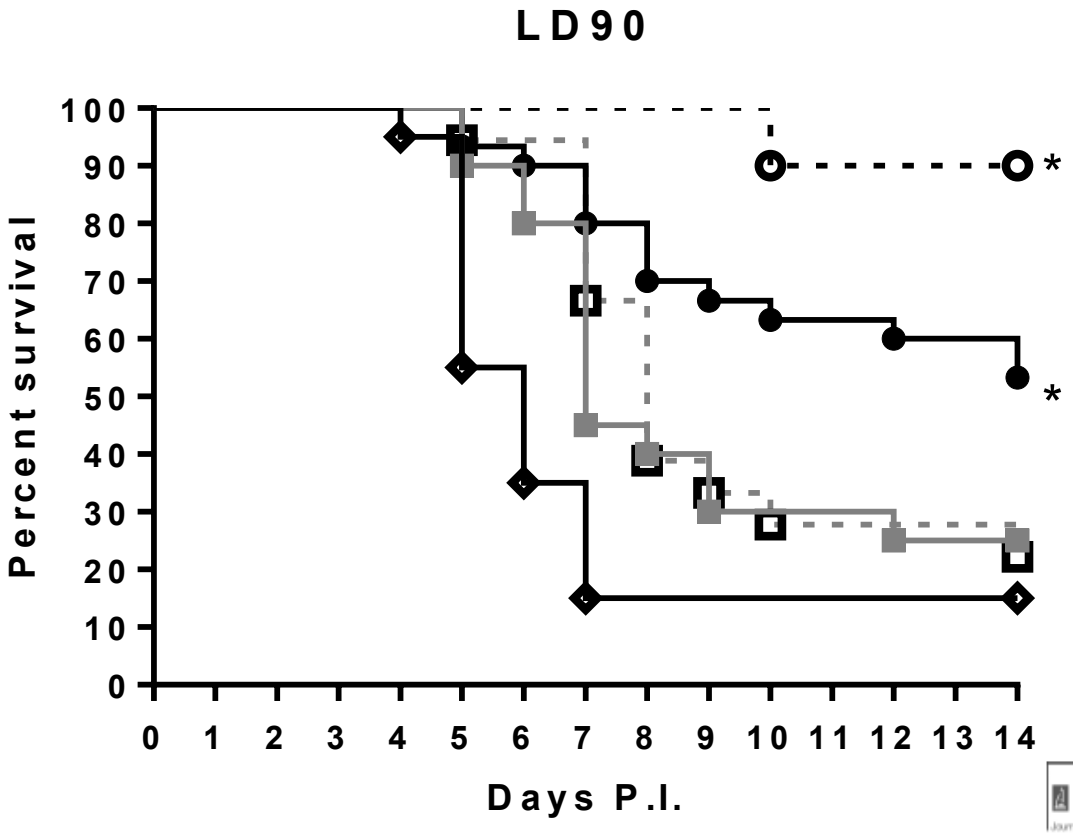
Investigation of Griffithsin's Interactions with Human Cells Confirms Its Outstanding Safety and Efficacy Profile as a Microbicide Candidate

Joseph Calvin Kouokam[1], Dana Huskens[2], Dominique Schols[2], Andrew Johannemann[1], Shonna K. Riedell[1], Wendye Walter[1], Janice M. Walker[1], Nobuyuki Matoba[1], Barry R. O'Keefe[3], Kenneth E. Palmer[1]*

1 Owensboro Cancer Research Program, James Graham Brown Cancer Center and Department of Pharmacology and Toxicology, University of Louisville School of

- **GRFT prevents infection of polarized colorectal tissue to 1 µM.** C. Dezzutti, unpublished.

Griffithsin Carbopol Gel Protects Mice from Genital Herpes (HSV-2) Infection



- ***Note enhanced efficacy in presence of SP***

JVI

Griffithsin Protects Mice from Genital Herpes by Preventing Cell-to-Cell Spread

Briana Nixon,[a] Martha Stefanidou,[a] Pedro M. M. Mesquita,[a] Esra Fakioglu,[a] Theodora Segarra,[a] Lisa Rohan,[b] William Halford,[c] Kenneth E. Palmer,[d,e] Betsy C. Herold[a]

Departments of Pediatrics and Microbiology and Immunology, Albert Einstein College of Medicine, Bronx, New York, USA[a]; Department of Pharmaceutical Sciences, University of Pittsburgh, Pittsburgh, Pennsylvania, USA[b]; Department of Microbiology, Southern Illinois University School of Medicine, Springfield, Illinois, USA[c]; Department of Pharmacology and Toxicology and James Graham Brown Cancer Center, Louisville, Kentucky, USA[d]; Owensboro Cancer Research Program, Owensboro, Kentucky, USA[e]

Multipurpose Prevention Applications for GRFT-Based Microbicides

- GRFT has potent activity *in vitro* against HIV-1; HIV-2; hepatitis C virus
- HCV inhibitory activity suggests applications for prevention in HIV positive MSM at risk for HCV and HIV-1 superinfection

ANTIMICROBIAL AGENTS AND CHEMOTHERAPY, Nov. 2011, p. 5159–5167 Vol. 55, No. 11
0066-4804/11/$12.00 doi:10.1128/AAC.00633-11

Griffithsin Has Antiviral Activity against Hepatitis C Virus[▽]

Philip Meuleman,[1,2*] Anna Albecka,[1] Sandrine Belouzard,[1] Koen Vercauteren,[2] Lieven Verhoye,[2] Czeslaw Wychowski,[1] Geert Leroux-Roels,[2] Kenneth E. Palmer,[3] and Jean Dubuisson[1*]

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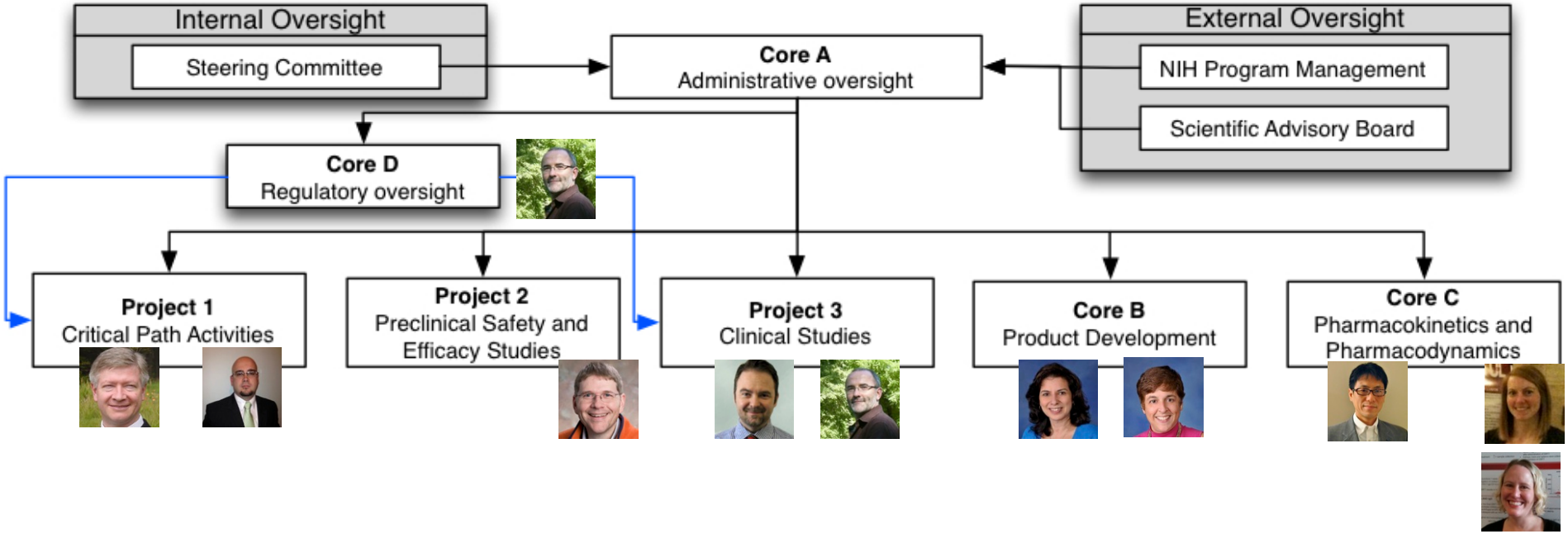
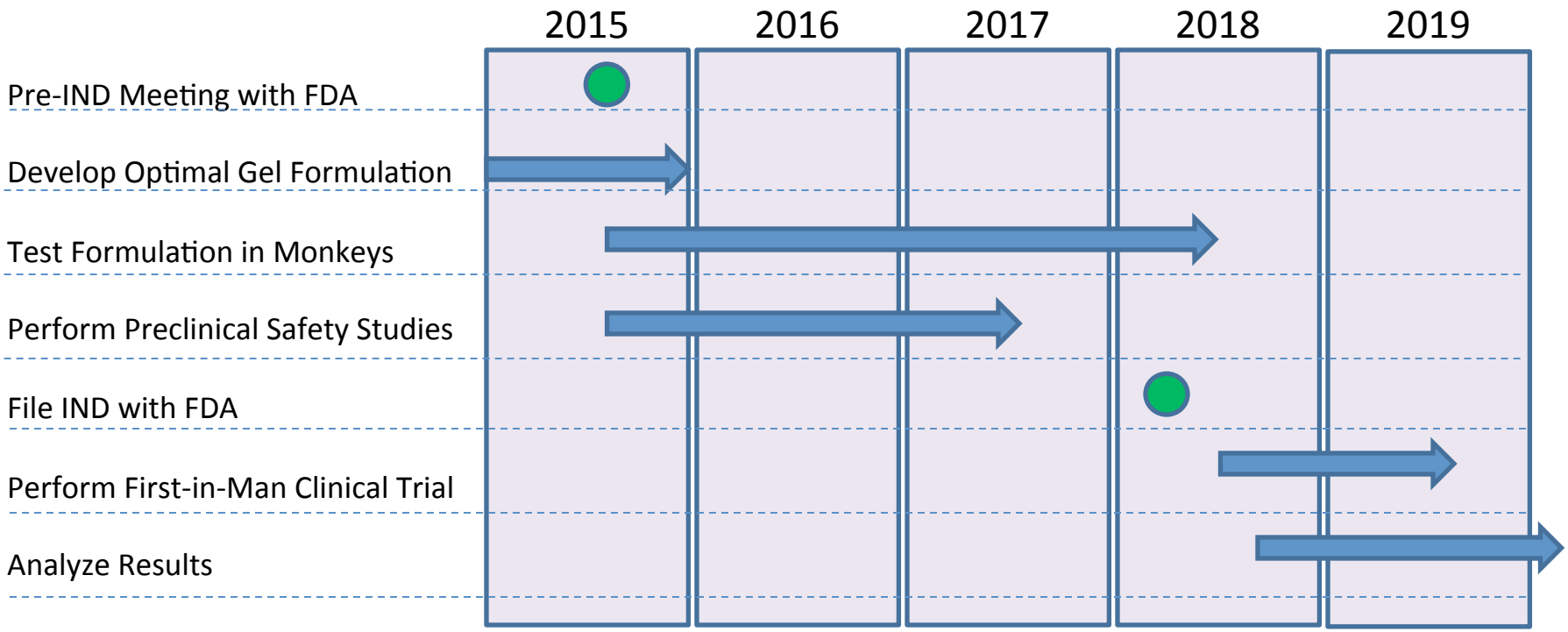
PREVENT PROGRAM STRUCTURE



GRFT PREVENT RECTAL GEL

- Rectal Gel
- First generation product:
  - Single active ingredient GRFT
  - Prevention of HIV transmission
    - Prevention of HCV and HSV-2 transmission
- Second generation product: co-formulation with ARV
- First in man clinical trial will enroll 18 healthy MSM volunteers to test first generation gel product
  - Single dose administration for safety assessment
  - Multiple dose safety assessment

PREVENT PROGRAM TIMELINE



ACKNOWLEDGEMENTS

- PREVENT Team
  - Daniel Tusé, Ph.D. — Intrucept Biomedicine
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  - Ross Cranston, M.D. — University of Pittsburgh
  - Lisa Rohan, Ph.D. — University of Pittsburgh
  - Charlene Dezzutti, Ph.D. — University of Pittsburgh
  - Nobuyuki Matoba, Ph.D. — University of Louisville
  - Ian McGowan, M.D., Ph.D. — University of Pittsburgh
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Development of a Rectal Enema (Douche) as Microbicide (D.R.E.A.M.): PrEP that People will Enjoy Using

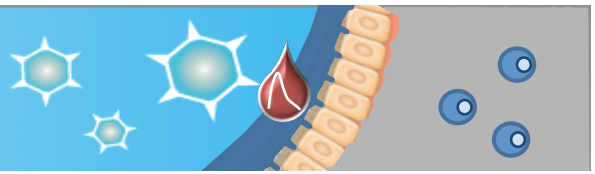
Craig W. Hendrix, MD

Johns Hopkins University

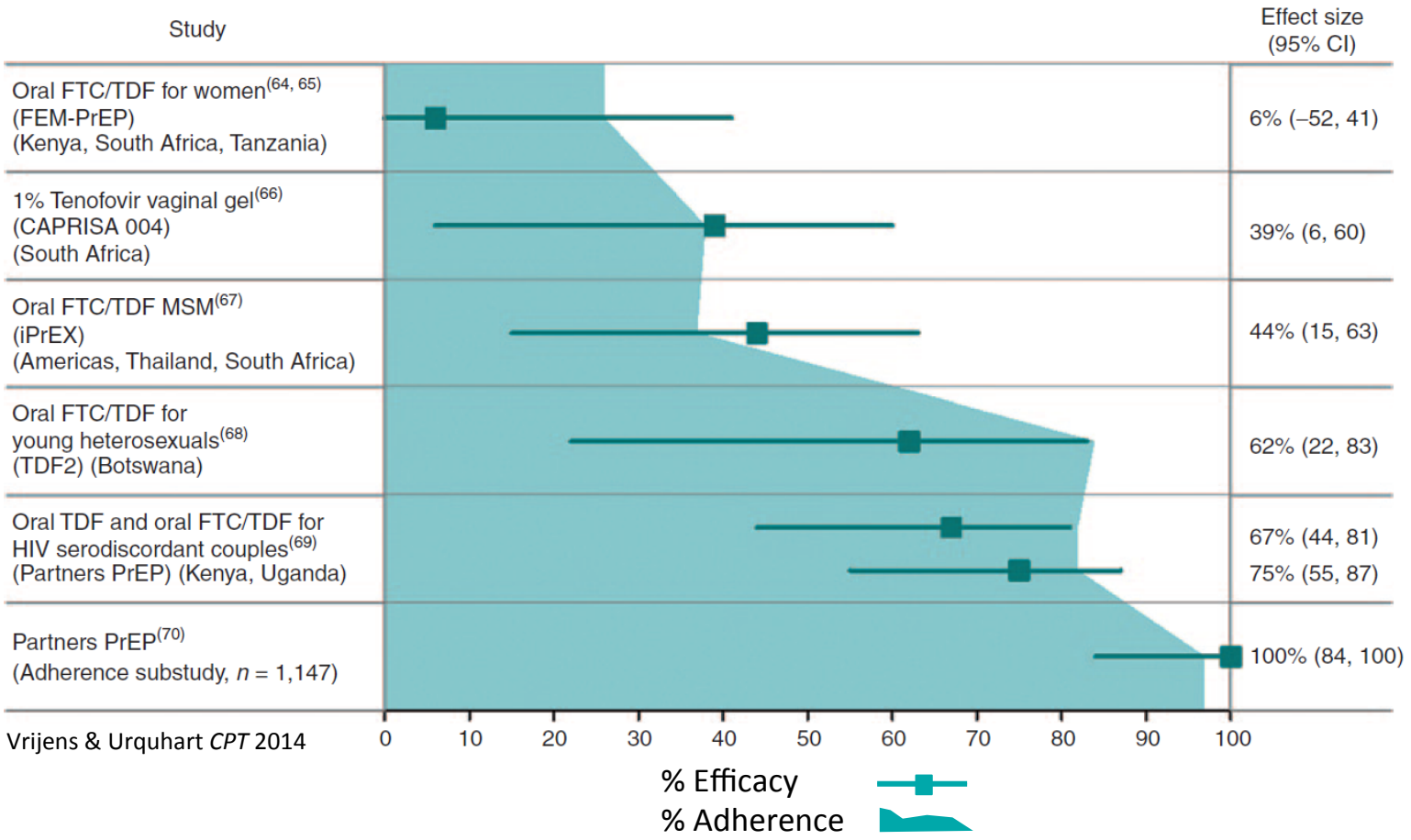
DREAM Program Objective

Develop a TFV prodrug enema/douche to provide HIV protection for one week after receptive anal intercourse

Exploit behaviorally-congruent PrEP formulation to mitigate adherence concerns



Adherence Biggest Cause of PrEP Failure



Vrijens & Urquhart *CPT* 2014

Douching is commonly part of anal sex

- Hylton J, **Fuchs EJ, Hendrix CW**. Lubricant and enema use among MSM: factors affecting the development of a rectal microbicide for clinical trials. XVI International AIDS Conference. Toronto, 2006. Abstract no. CDC0649.
- **Carballo-Dieguez A**, Bauermeister J, Ventuneac A, Dolezal C, Balan I, Remien RH. The use of rectal douches among HIV-uninfected and infected men who have unprotected receptive anal intercourse: implications for rectal microbicides. AIDS Behav 2008 Nov;12(6):860-6.
- **Carballo-Dieguez A**, Bauermeister J, Ventuneac A, Dolezal C, Mayer K. *Why rectal douches may be acceptable rectal-microbicide delivery vehicles for men who have sex with men*. Sex Trans Dis 2010 Apr;37(4):228-33.
- Galea JT, Kinsler JJ, Imrie J, Nureña CR, Sánchez J, Cunningham WE. *Rectal douching and implications for rectal microbicides among populations vulnerable to HIV in South America: a qualitative study*. Sex Transm Infect 2014 Feb;90(1):33-5.
- Javanbakht M, Stahlman S, **Pickett J**, LeBlanc M-A, Gorbach PM. *Prevalence and types of rectal douches used for anal intercourse: results from an international survey*. BMC Infectious Diseases 2014 Feb 21;14:95.
- Noor SW, Rosser BRS. *Enema use among men who have sex with men: A behavioral epidemiologic study with implications for HIV/STI prevention*. Arch Sex Behav 2014 May;43(4):755-69.

Positive Medicated Douche Marketing

Positive Medicated Douche Marketing

What formulation do we need?

- People use a variety of applicators
  - Bottles, bulbs, "sinkers," bidets, shower attachments, etc.
  - Are these *all* suitable for delivery of a medicated douche?
- What applicator/bottle will we use?
- Will the product be portable?

- or -

or

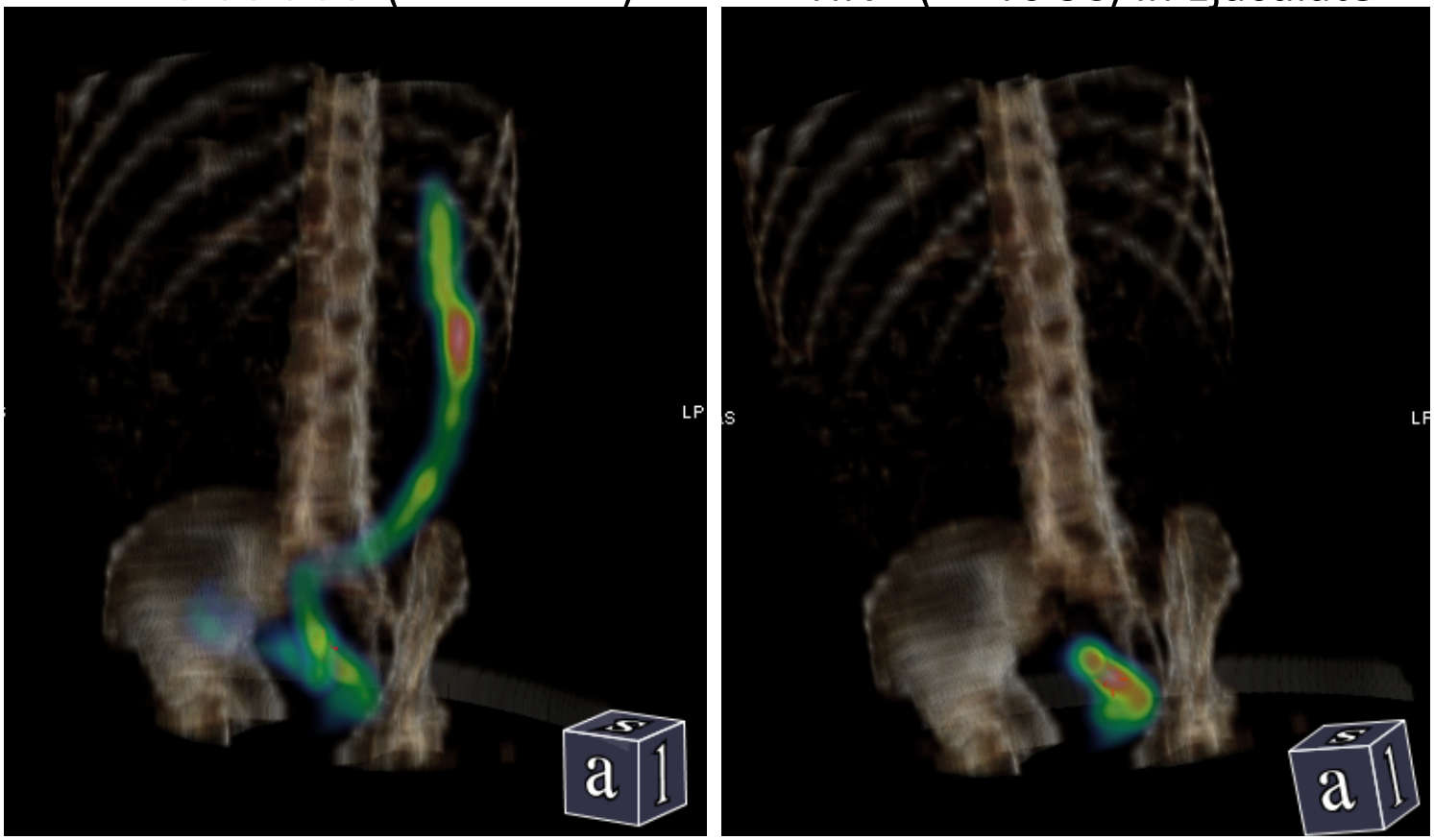
+

We should develop a product accepted by users

Will a Douche "Cover" HIV in the colon?

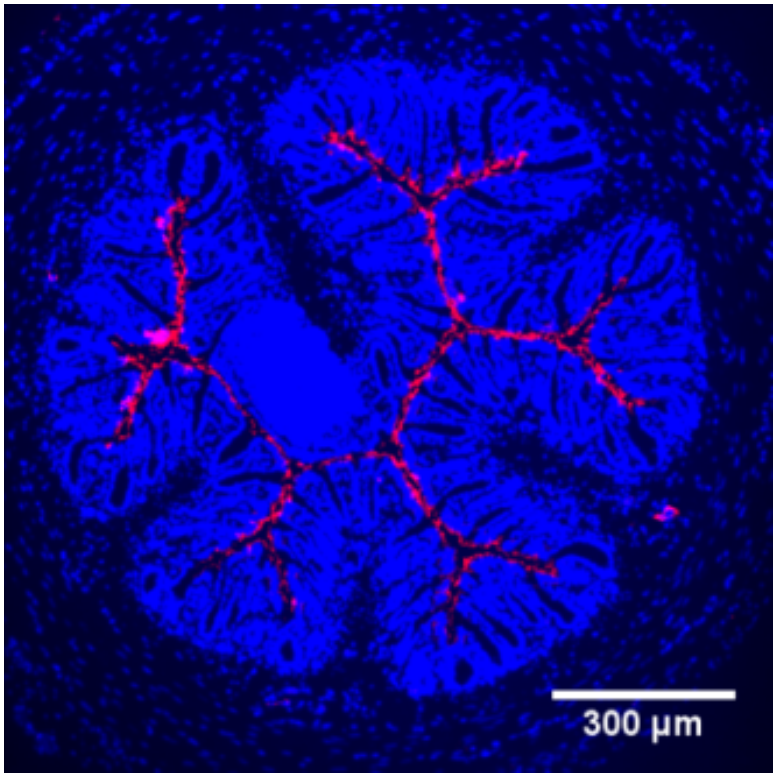
"Microbicide"(^{111}In-DTPA)

"HIV" (^{99m}Tc-SC) in Ejaculate



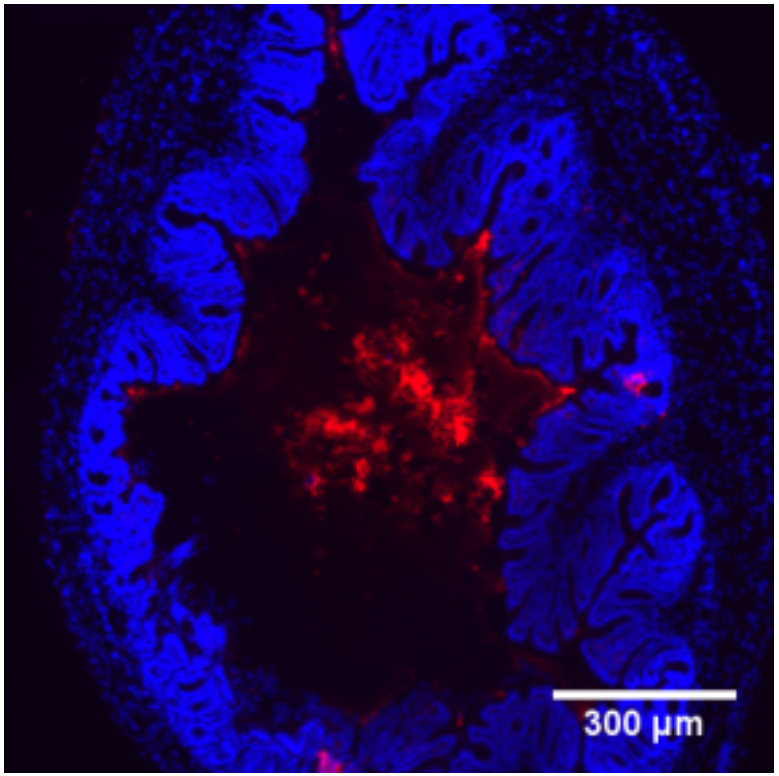
Rectal TFV gel (0h), simulated sex/ejaculation (1h), SPECT/CT (2h)

Does the drug contact the mucosa?



Hypotonic Saline (NaCl)

Excellent drug-mucosal contact

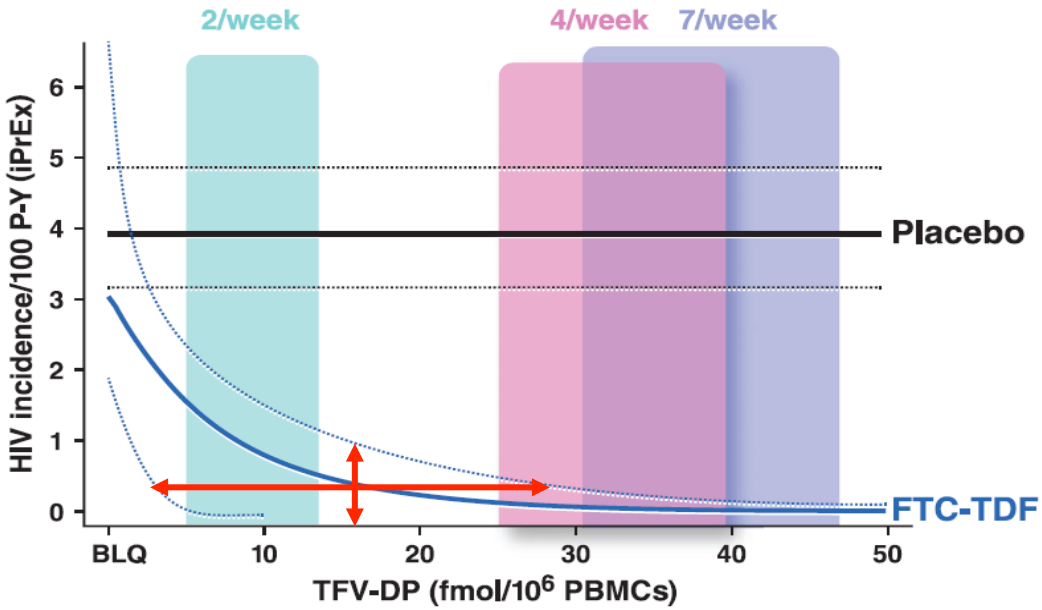


Hypertonic Saline (~Fleet)

Poor drug-mucosal contact

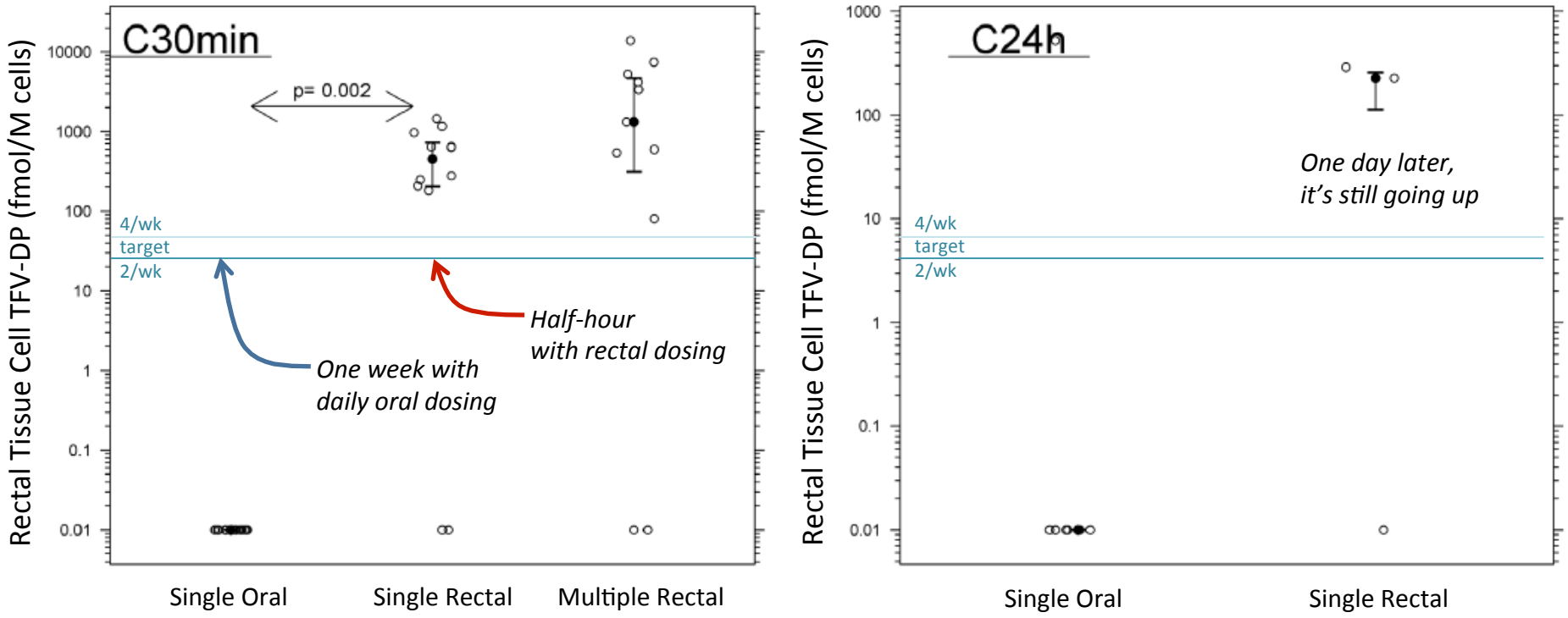
Maisel K. J Control Rel 2015

What Concentration is Protective?



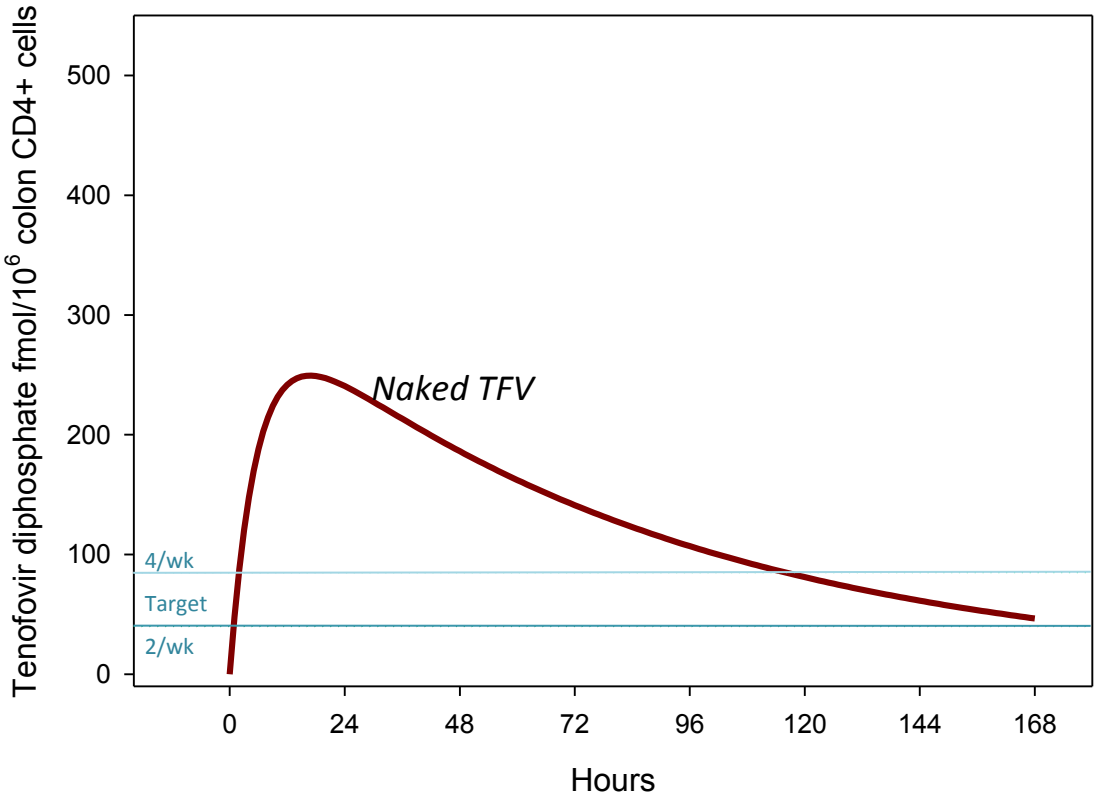
- iPrEx EC_{90} 16 fmol/10^6 cells (3-28 95% CI) (Anderson *STM* 2012)
- Colored panels, adherence benchmarks (STRAND DOT IQRs)
- Other studies relate protective PBMC TFV-DP to estimate colon tissue TFV-DP tissue concentrations (Anton *ARHR* 2012, Hendrix CROI 2012, Louissaint ARHR 2013)

How soon is HIV protection achieved?



RMP-02/MTN-006 Yang, et al. PLOS One 2014

Planned Product Enhancement



TFV enema PK Enhancements

- Absorption
  - TFV analogs
  - Hypotonic vehicle
- Sustained release
  - Nanoparticle
  - Gelling agent

Reference Targets

- Colon CD4+ cell TFV-DP
- Bridging RCT-PK studies

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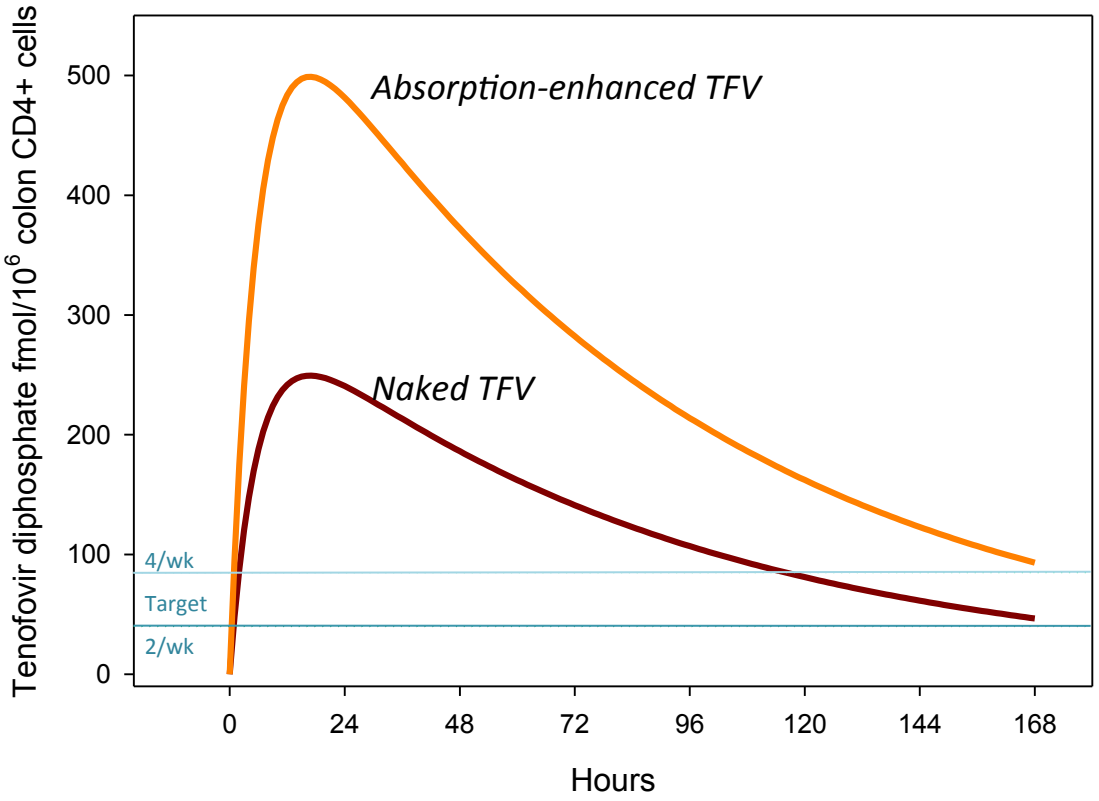

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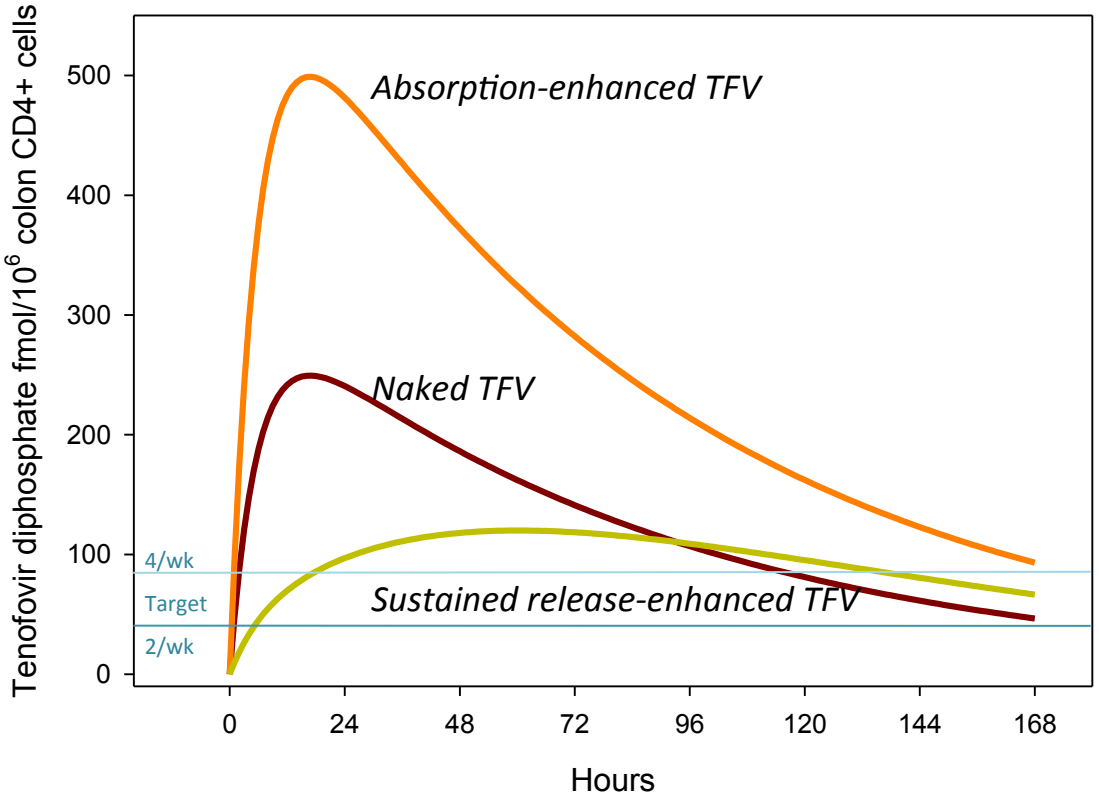

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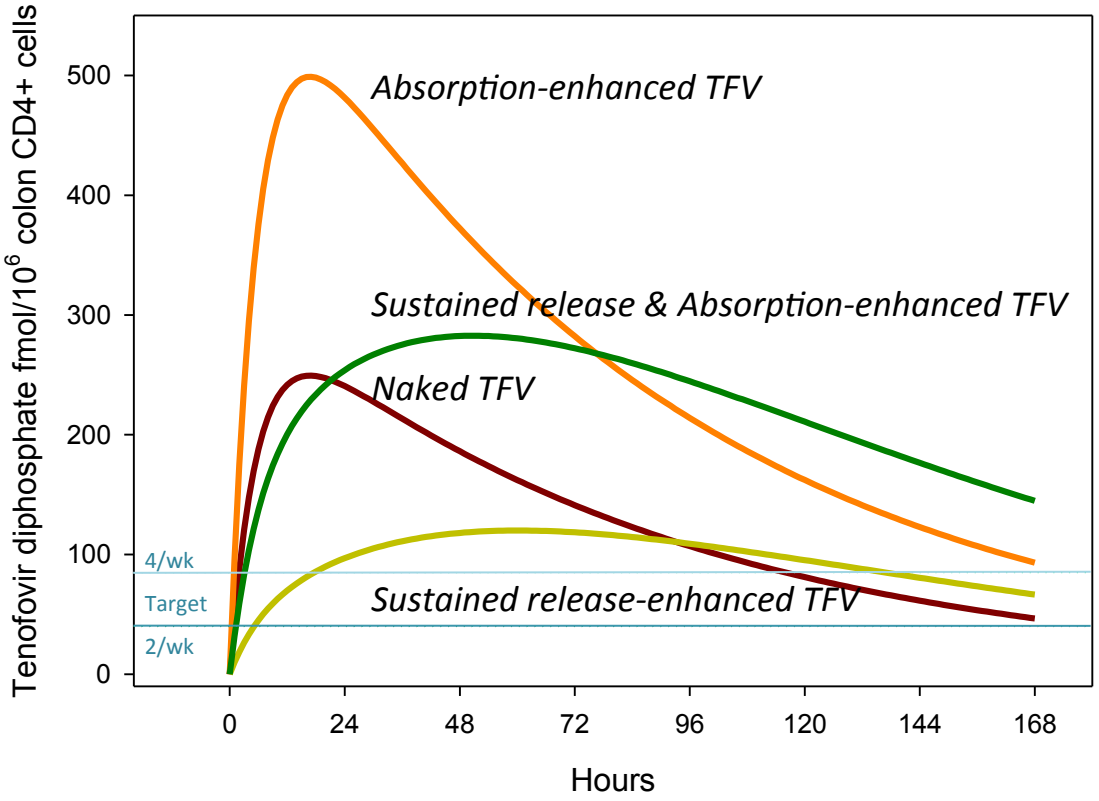

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DREAM Program Studies

- Clinical
  - DREAM 01: TFV dose escalation, hypotonic saline
  - DREAM 02: Sex-enema/enema-sex distribution effect
  - DREAM 03: Compare optimized TFV vs. TFV prodrug
- Pre-Clinical (mice, macaques)
  - Select optimal saline for clinical studies
  - Select optimal TFV prodrug for TFV comparison
  - Compare TFV v. TFV analog nano/thermoreversible gel

DREAM 01 Study Design Overview

- Open label, sequential 3 product, single dose
- 18 healthy volunteers across 3 sites (JHU, UCLA, Pitt)
- Products
  - 1x (~TFV 1%), 3x (<5 day TFV 1%), 6x (hypotonic 3x)
  - 125 mL + NaCl (normal saline or half-normal saline)
  - 1 dose in clinic, 3 doses at home with RAI
- Assessments
  - Safety: clinical, histology, transcriptomics/proteomics
  - Acceptability: questionnaire, interview
  - Drug concentration in blood and rectal tissue
  - Drug activity: *ex vivo* HIV explant challenge

Thank you!

Can rectal TFV protect as well as oral?

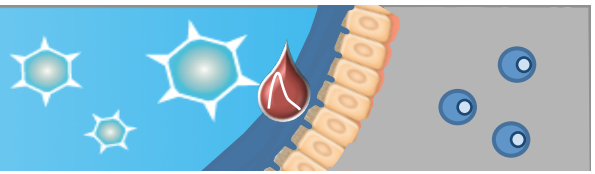
	^{14}C-TDF*	RMP-02/MTN-006				Rectal vs. Oral	
Matrix	**Single oral**	**Single oral**	**Single Rectal**	**QD x 5 Rectal**	**D5/D1****	**D1 PR/ PO**	**D5 PR/ PO**
Plasma TFV ng/mL (LLOQ 0.31)	40 (24, 51)	35.8(21.4 - 54.9)	0.31 (BLQ - 1.2)	0.32 (BLQ-2.80)	1	0.01	0.01
Tissue homog. TFV ng/mg (LLOQ 0.14)	0.03 (BLQ, 0.21)	BLQ (BLQ-14.6)	BLQ (BLQ-12.6)	13.0 (1.7-430.4)	93	-	-
Tissue homog. TFV-DP fmol/mg (LLOQ 17)	7.5 (3.5, 60.9)	BLQ (BLQ - 991)	285 (BLQ - 490)	789 (56-7188)	3	-	105
Colon Total Cell TFV-DP fmol/M cells (LLOQ 160)	25 (15, 88)	BLQ (BLQ - 227)	124 (BLQ-412)	1,324 (BLQ-13,880)	11	5	53
Colon CD4+ TFV-DP fmol/M cells (LLOQ 229)	1 (BLQ, 4)	BLQ (BLQ – BLQ)	266 (BLQ – 3950)	1,083 (BLQ-31,153)	4	266	1083

*LLOQ do not apply to 14C-TDF study which used AMS

**Ratio of median C_{24} Day 5 rectal gel to median C_{24} Day 1 rectal gel

5 rectal doses may not yet have achieved at steady-state based on PBMC TFV-DP concentrations.

- *Rectal dose achieves 266 x greater colon CD4+ TFV-DP than oral*
- *5 daily rectal gel doses achieves 4x single dose colon CD4+ TFV-DP*
- *Rectal dose only 1% of oral dose systemic exposure*



Would you like an Enema or Douche?

- Enema
  - Constipation
  - Preparation for medical procedures
  - Illness

"How many enemas did you give him ?"

- Douche
  - Cleansing
  - Eliminating odor
  - Getting ready for sex

Are Douches safe?

- Rectal douching associated with increased risk for HIV transmission *(Coates 1988, Moss 1988)*
- Tap water & hyper-osmolar enemas show colonic epithelium damage *(Meisel 1977, Schmelzer 2004)*
- Rectal hyper-osmolar gels induce greater epithelial loss than iso-osmolar gels *(Fuchs 2007)*

MTN-026/IPM 038

Ross D. Cranston MD FRCP
Associate Professor
University of Pittsburgh

MTN-026/IPM-038

- A Randomized, Double Blind, Placebo-Controlled, Phase 1 Safety and Pharmacokinetic Study of Dapivirine Gel (0.05%) Administered Rectally to HIV-1 Seronegative Adults

Products

- Dapivirine 0.05%
- HEC Placebo Gel

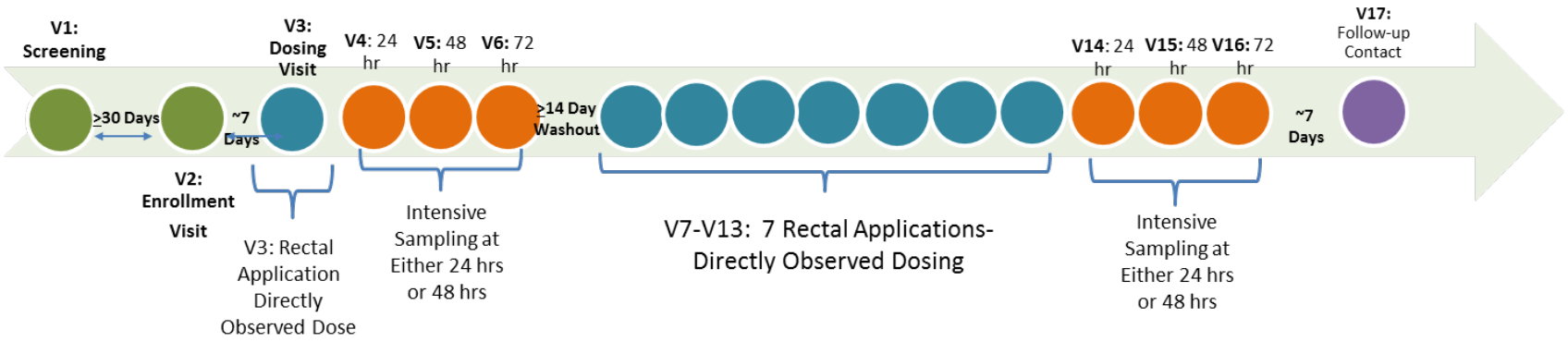
Dapivirine

- NNRTI
- Increased potency compared to tenofovir in *in vitro* testing
- Safe and acceptable in MTN-012 (penile tolerance study)
- API in the ASPIRE Ring study

Study Summary

- Phase 1, multi-site, randomized (2:1), double-blind, placebo-controlled trial
- 24 evaluable HIV-uninfected men and women aged 18-45 years inclusive
- Approximately 42 days of follow-up per participant is planned with a projected accrual period of 6-8 months
- Participants will be randomized to receive either a single dose of dapivirine gel or universal HEC placebo gel rectally, followed by 7 daily doses of the same product to be administered under direct observation in the clinic

Study Visit Schedule



Primary Objectives/Endpoints

- Safety: To evaluate the safety of dapivirine gel formulation when applied rectally
  - Grade 2 or higher AEs
- Pharmacokinetics: To characterize the systemic and compartmental pharmacokinetics of dapivirine gel following rectal application
  - Dapivirine concentrations
    - Blood,
    - Rectal fluid
    - Rectal mucosal tissue homogenates

Secondary Objectives/Endpoints

- Acceptability: To identify product attributes considered likely to challenge and facilitate future sustained use of rectally applied dapivirine gel
  - Product attributes considered likely to challenge future sustained use.
- Mucosal Safety: To evaluate the mucosal safety of dapivirine gel when applied rectally
  - Mucosal Safety (rectal proteomics/transcriptome/ microflora/histology) and rectal tissue flow cytometry

Timeline

- Protocol development meeting Dec 2014
- PSRC Feb 2015
- PSRC approval Mar 2015
- Projected Version 1.0 May 2015
- Projected start date Oct 2015

Acknowledgements

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- IPM

Thank You