

REVIEW

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Combining biomedical preventions for HIV: Vaccines with pre-exposure prophylaxis, microbicides or other HIV preventions

Janet M. McNicholl

Division of HIV/AIDS, Laboratory Branch, Centers for Disease Control and Prevention, Atlanta, GA, USA

ABSTRACT

Biomedical preventions for HIV, such as vaccines, microbicides or pre-exposure prophylaxis (PrEP) with antiretroviral drugs, can each only partially prevent HIV-1 infection in most human trials. Oral PrEP is now FDA approved for HIV-prevention in high risk groups, but partial adherence reduces efficacy. If combined as biomedical preventions (CBP) an HIV vaccine could provide protection when PrEP adherence is low and PrEP could prevent vaccine breakthroughs. Other types of PrEP or microbicides may also be partially protective. When licensed, first generation HIV vaccines are likely to be partially effective. Individuals at risk for HIV may receive an HIV vaccine combined with other biomedical preventions, in series or in parallel, in clinical trials or as part of standard of care, with the goal of maximally increasing HIV prevention. In human studies, it is challenging to determine which preventions are best combined, how they interact and how effective they are.

Animal models can determine CBP efficacy, whether additive or synergistic, the efficacy of different products and combinations, dose, timing and mechanisms. CBP studies in macaques have shown that partially or minimally effective candidate HIV vaccines combined with partially effective oral PrEP, vaginal PrEP or microbicide generally provided greater protection than either prevention alone against SIV or SHIV challenges. Since human CBP trials will be complex, animal models can guide their design, sample size, endpoints, correlates and surrogates of protection. This review focuses on animal studies and human models of CBP and discusses implications for HIV prevention.

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Global HIV epidemic and the prevention tool box



Since the beginning of the HIV/AIDS epidemic, almost 80 million people have become infected with HIV¹, predominantly through heterosexual intercourse. In 2014 alone, 2 million new infections occurred worldwide, with most occurring in sub-Saharan Africa, although concentrated epidemics occur in many risk groups.¹ In the US, 40,000 infections per year are estimated to occur, particularly in young men who have sex with men (MSM).^{1,2} Although the lifetime risk of being diagnosed with HIV is dropping, without scale up of US national HIV prevention targets, an estimated 1 in 6 MSM will be diagnosed with HIV in their lifetime in the US, including 1 in 2 black MSM.³

Existing preventive measures against HIV infection include behavioral, barrier and biological methods. The latter include treatment of HIV infected mothers to prevent mother-to-child transmission, treatment of other HIV infected individuals or populations, post-exposure prophylaxis, male circumcision and most recently, pre-exposure prophylaxis (PrEP) with antiretroviral drugs.⁴

Pre-exposure prophylaxis

In clinical trials (Fig. 1), oral daily PrEP with Truvada (a combination of the viral reverse transcriptase inhibitors, tenofovir

disoproxil fumarate [TDF] and emtricitabine) had HIV-prevention efficacy* ranging from high to none. Efficacy rates of 44%, 62%, and 75% were observed in MSM, heterosexual men and women, and HIV-1 serodiscordant couples, respectively.^{5,6,7} Oral TDF alone reduced HIV infection by 62% in serodiscordant couples⁷ and by 49% in injecting drug users.⁸ High efficacy (86%) of oral Truvada has also been seen in MSM in the PROUD⁹ and IPERGAY studies.¹⁰ However in some studies of women, no or minimal efficacy of oral PrEP was observed.^{11,12} In 2012 oral PrEP with Truvada was approved in the United States in combination with safer sex practices to reduce the risk of sexually-acquired HIV infection in HIV-negative high-risk men and women¹³ and later the US CDC and the WHO recommended oral PrEP be integrated into HIV prevention strategies for high risk individuals.^{14,15} Oral PrEP is being prescribed more widely, usually as a daily pill. It is also sometimes being offered as part of the prevention package given to participants in HIV-prevention clinical trials. PrEP, along with other strategies can significantly impact the HIV/AIDS epidemic. In the US, it is estimated that if national targets for HIV prevention, including expanded HIV testing and treatment and increased use of daily PrEP, were met there would be a 70% reduction in new HIV infections by 2020.¹⁶

CONTACT Janet M. McNicholl  jkm7@cdc.gov  Laboratory Branch, Division of HIV/AIDS Prevention, MS A25, Centers for Disease Control and Prevention, Atlanta, GA, 30329, USA.

The findings and conclusions in this report are those of the author and do not necessarily represent the views of the Centers for Disease Control and Prevention (CDC).

*Footnote: in this review, efficacy is used to describe the reduction in HIV incidence reported in the trial regardless of adherence, and usually refers to the modified intention to treat data.

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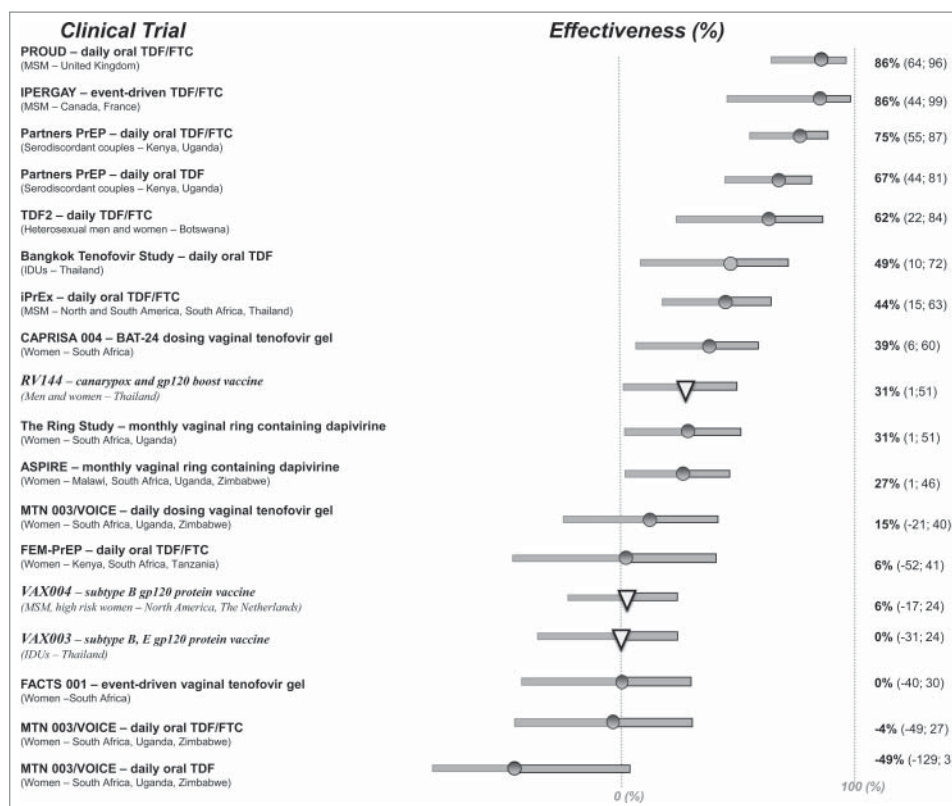


Figure 1. Biomedical HIV preventions are ranked from highest to lowest effect size. Confidence intervals are shown in (). MSM, men who have sex with men. Adapted from AVAC (www.avac.org/sites/default/files/resource-files/evidence_HIVprevention_feb2016.pdf) with permission, adapted from original figure in Ref. 55, p. 2060. Three vaccine trials are identified by italic text and an inverted triangle in the effectiveness bar. Two HIV-vaccine trials, the STEP and Phambili trials of an Ad5 subtype B gag/pol/nef vaccine, are not shown as there was no efficacy and HIV acquisition was enhanced in some subgroups.

There has been less success with topical PrEP. A vaginal microbicide gel containing 1% tenofovir (TFV) had a 39% risk reduction in the CAPRISA 004 clinical trial¹⁷ but was not effective in 2 others.^{12,18} Monthly intravaginal rings (IVR) delivering the non-nucleoside reverse transcriptase inhibitor, dapivirine had 27% and 31% efficacy in 2 separate trials.^{19,20} Higher efficacy was seen in women over 21 y of age.¹⁹ Poor adherence in oral and topical PrEP studies, with drug levels below a known or likely protective threshold, has generally explained low PrEP efficacy. Adherence is influenced by many factors including characteristics of the product or the user. The PrEP field is developing strategies to overcome adherence including, better and longer-lasting intravaginal rings, more covert products such as vaginal films or tablets, long-acting injections or implants and novel PrEP drugs or biomedical approaches including broadly neutralizing antibodies.⁴ PrEP or microbicide approaches for rectal delivery are also being developed.

HIV vaccines

Despite the optimism surrounding PrEP use and efforts to get more HIV-infected people on anti-retroviral treatment, modeling indicates that even with ideal use and roll out of PrEP and treatment, the most significant impact on epidemic control will be the availability of an effective HIV vaccine.²¹ Harmon et al. modeled multiple HIV epidemic scenarios from the present to 2070 in low and middle income countries, including many sub-Saharan African countries where the greatest burden of the

epidemic is concentrated.⁴ With current trends, if a vaccine with 70% efficacy was introduced in 2027, the number of annual infections would decline from 1.7 million to 257,000 in 2070. Even an HIV vaccine with 30% efficacy could have significant impact. The authors conclude that combining an HIV vaccine with PrEP, other HIV preventions, testing, treatment and care offers provides the greatest possible impact to curb the epidemic.⁴

Six human HIV vaccine efficacy trials have been conducted to date, evaluating 4 different vaccine concepts (reviewed in refs.²²⁻²⁴). The Vax003 and Vax004 trials immunized with alum-adsorbed HIV-1 gp120 envelope protein and although antibodies were raised, the vaccines did not protect against HIV acquisition.^{25,26} The Step and Phambili trials used recombinant adenovirus serotype 5 expressing gag, pol and nef proteins. The vaccine did not protect against HIV acquisition and enhanced infection in some subgroups.^{27,28} The HVTN 505 trial evaluated a DNA prime expressing gag, pol, env and nef followed by immunizations with recombinant adenovirus serotype 5 expressing gag, pol and env. The trial was halted for futility to prevent HIV acquisition.²⁹ Only one trial (Fig. 1), the RV144 trial, set in Thailand in a low HIV incidence population and using a combination of a canarypox viral vector expressing gag, pol and env and alum-adsorbed gp120 protein boosts has prevented HIV acquisition.³⁰ At one year, efficacy was 60% but protection waned and was 31% overall.³⁰ This vaccine was not licensed due to the low overall efficacy and other factors.^{31,32} However, the trial provided the first data on any

immune correlates of protection, including antibodies to regions of the V1V2-region of the HIV envelope.^{33,34} Based on these findings, next generations of this canary-pox based vaccine strategy are being evaluated with novel adjuvants^{22,24,35} and an efficacy trial, known as HVTN 702, of a clade C version of this canarypox-protein boost vaccine will start in Southern Africa in 2016.^{22,23,24,35-37}

To date, over 100 HIV-vaccines candidates have been tested in safety and immunogenicity trials. Newer HIV vaccines are being designed^{22,24,35,36} and include DNA and viral vectors (e.g. adenovirus serotype 26, modified vaccinia Ankara, cytomegalovirus), purified proteins and peptides and novel adjuvants to drive appropriate, long-lasting immune responses.^{22,24,36,37} Newer clinical trial designs and statistical approaches are being considered.^{35,38} An effective HIV vaccine would have enormous advantages over drug-based preventions such as PrEP or microbicides, as once a vaccine series is complete, immune memory should be long-lasting, perhaps only requiring periodic boosts. This would overcome the adherence problem of current PrEP, i.e. taking daily or peri-coital product and needing renewed prescriptions. Even as long-acting forms of PrEP are introduced, these likely would not be administered for life and would only be given to the highest risk individuals. A safe and effective HIV vaccine could be given more universally, perhaps to pre-teens, teens and young adults.

Prevention of HIV with combined biomedical preventions

From the perspective of controlling the HIV-epidemic, combining a partially effective HIV vaccine with partial effective PrEP, microbicides or other non-vaccine preventions would seem logical. This is currently only a theoretical concept in humans: no trials of such combinations have been conducted. Obstacles include that to date, no HIV-vaccine has been licensed or approved, that vaccines and PrEP or microbicides have not been co-developed and that the path to licensure of such a combination is likely to be challenging. Efficacy trials could be complex, require large sample sizes^{39,40} and have challenges related to informed consent and testing for HIV. Non-human primate (NHP) studies can avoid some of these challenges, provide proof-of concept data regarding synergistic, additive or other interactions and determine drug or immune correlates of protection that could guide clinical trial design.

NHP studies of biomedical preventions

NHP studies provided key proof-of-concept data that led to human PrEP trials⁴¹ and are being used to evaluate microbicides and next generation PrEP. NHP studies have also been used to evaluate candidate HIV vaccines. While their predictive value for HIV-vaccine protection in humans has been less certain than in most studies of PrEP, the finding of immune correlates in the RV144 trial^{33,34} is influencing NHP HIV vaccine studies. For example, canarypox-SIV vaccine constructs have been developed, and with gp120 boosts, the vaccine schedule models the RV144 trial⁴² and is providing guidance for evaluating other candidate HIV-1 vaccines.

NHP studies of HIV vaccines combined with PrEP or microbicides

Five NHP studies evaluated combined candidate HIV vaccines with drugs (or microbicides) delivered orally or topically as candidate HIV preventions (Table 1).^{43,44,45,47} Four modeled vaginal HIV acquisition and one evaluated rectal acquisition. When combined as preventions, overall efficacy tended to increase in 4 of the 5 studies and where increased protection was not seen, beneficial effects on post-infection viral load was sometimes noted. The studies suggested either additive or synergistic effects and allowed dissection of mechanisms.

Cheng-Mayer et al. reported the first NHP study of CBP (Table 1).⁴³ A DNA prime recombinant adenovirus boost T cell-based vaccine was administered to rhesus macaques followed by a vaginal microbicide gel with a suboptimal concentration of an HIV-1 nucleocapsid zinc finger inhibitor. Animals received 20 repeated vaginal challenges with escalating doses of SHIV_{162P3}. Three of 15 animals in the CBP group compared to 2 of 8 in the vaccine group were protected indicating no increased protection by combining gel and vaccine. However, 2 CBP breakthrough animal had transient viremia and never seroconverted. If these animals were considered protected the efficacy of the CBP would have been more than 30%. The CBP delayed SHIV acquisition in the breakthrough animals and viral load was reduced compared to controls, suggesting an interaction between the preventions. The authors suggested the vaccine established immunity that controlled SHIV infection, while the microbicide delayed virus replication and spread. A limitation of the study is that the class of zinc finger inhibitor evaluated has not been evaluated for HIV prevention in humans. Nevertheless, this study provided proof-of-concept that combining vaccines with microbicides could be beneficial over either prevention alone.

Barouch et al. evaluated two primarily T-cell inducing adenovirus-based vaccines in combination with vaginal gels containing the fusion inhibitor T-1249 or the CCR5-targeted entry inhibitor maraviroc (Table 1).⁴⁴ Neither of the vaccines was protective alone but when combined with the gels, efficacy appeared to increase, to 30% against vaginal SIV challenge and to 67% against vaginal SHIV_{162P3} challenge. The study was done in rhesus macaques treated with the hormone Depo-Provera to produce vaginal thinning. Animals received one high dose SIV challenge. The sample size (n = 6) in the individual arms of this study was smaller than in the Cheng-Mayer study, but the CBP significantly reduced acquisition compared to controls. The authors hypothesized that the microbicides delayed virus expansion, allowing vaccine-raised immune responses to prevent or control infection. Adenovirus-based vaccines and CCR5 directed drugs continue to be evaluated in humans as possible HIV-prevention strategies.

Le Grand et al. evaluated a tenofovir-containing vaginal gel combined with a protein-based HIV-vaccine candidate designed to induce T- and B-cell immunity (Table 1).⁴⁵ The vaccine had proteins from clades B and C, adjuvanted with MF59, which is being used in the next phase III canarypox-gp120 HIV vaccine efficacy trial in Southern Africa, or a novel toll-like receptor 7/8 agonist. It was given intramuscularly and intranasally to cynomolgous macaques to induce mucosal immunity. The vaccine

Table 1. Non-human primate studies modelling combinations of candidate HIV vaccines with PrEP or microbicides.

Author	Year Ref.	Macaques	Vaccine route (Efficacy)	Other biomedical prevention, route (Efficacy)	Viral challenge, route, timing	CBP effect (Efficacy)	Viral load effect in CBP	Comments
Cheng-Mayer	2011 ⁴³	46 female rhesus	DNA with SIVmac ₂₃₉ G/P prime rAd5 virus boost, i.m. T-cell inducing(25%)	SAMT-247 Zn finger inhibitor, 0.1% vaginal gel, 30 min before challenges (6%)	SHIV _{162P3} , 20 escalating vaginal challenges 12 weeks after last vaccination	Probable (20%)	Reduced peak and AUC vs vaccine, gel or naïve controls	Acquisition delayed vs vaccine or gel alone; 2 CBP animals had transient lowseronegative viremia, if included CBP efficacy was higher in CBP group; Ad-based vectors in human trials, SAMT-247 not; vaccine raised cellular immunity; large sample size
Barouch	2012 ⁴⁴	53 female rhesus Depo-Provera treated 30 d before challenge	Ad26/Ad5HVR48 with SIV G/P/E/N prime-boost, i.m. T-cell inducing(0%) Ad35/Ad26 with SIVsm _{ES43} G/P/E/N prime-boost, i.m. (0%)	T-1249 fusion inhibitor, 200 ug/ml vaginal gel(0%) Maraviroc entry inhibitor vaginal gel(43%)	SIVmac ₂₅₁ Single vaginal challenge 8 months after last vaccination SHIV _{162P3} Single vaginal challenge 6 months after last vaccination	Probable(30%) Probable(67%)	No Reduced at peak and day 28 vs placebo and naïve controls	Acquisition not delayed; Ad-based vectors in human trials; Depo-Provera allowed single challenge; immune responses not reported Acquisition not delayed; heterologous env challenge; Depo-Provera used; immune responses not reported; one control infected, hence efficacy <50%; maraviroc being evaluated in humans for PrEP and in use for treatment
Le Grand	2016 ⁴⁵	52 female cynomolgous	Gp140 (Clades B,C) i.n. prime with R848, TLR 7/8 adjuvant, i.m. boosts with MF59 adjuvant; T and B-cell inducing (0%)	Tenofovir, 1% vaginal gel 1h before challenge(46% after 614% after 12 challenges)	SHIV _{162P3} < 22 vaginal challenges 11 weeks after last vaccination	Probable (81% after 6 63% after 12 challenges)	Not noted	CBP efficacy was 38% at 22 challenges with no gel for last 10; vaccine raised cellular and humoral immunity; TDF and TDF-DP reported in blood and vaginal tissues; phase III TFV gel trials complete; MF59 in human use; large sample size.
Ross	2014 ⁴⁷	13 male rhesus	DNA with SIVmac ₂₃₉ G prime, poly clade B VLP-alum- adjuvanted boosts i.m., i.n.; B-cell inducing (n/a, likely > 50%)	FTC (22 mg/kg), TDF (20mg/kg), orally 2h before and 22h after challenge (~50%, historical)	SHIV _{162P3} 14 weekly rectal challenges 8 weeks after last vaccination	Probable (87.5%)	Yes, but only one animal	CBP interaction likely additive; vaccine raised humoral responses; challenges boosted G and E antibodies; CBP breakthrough animal had high antibody avidity and protective TFV-DP and FTC-TP levels; one CBP protected animal had transient viremia; only rectal study; only study with approved PrEP; small sample size.

Abbreviations (by column): Ad, adenovirus; G/P/E/N, Gag, Pol, Env, Nef, i.m., intramuscular; i.n., intranasal; VLP, virus like particle; CBP, combined biomedical prevention; interaction on acquisition or viral load; AUC, area under curve; Ab, antibody, PrEP, pre-exposure prophylaxis. Efficacy is as reported by the authors.

had no protection against repeated SHIV_{162P3} challenges when given alone. The 1% tenofovir gel alone had 46% efficacy after 6 vaginal challenges, mimicking the protection seen in the CAPRISA 004 trial of this gel.¹⁷ When vaccine and gel were combined, efficacy increased, as at 6 challenges protection was 81%. The CBP also provided protection at 12 and 22 challenges.⁴⁵ The gel may have limited mucosal viral burden, providing time for vaccine-raised immunity to expand and amplify the PrEP efficacy. While next generation human HIV vaccines are unlikely to include a protein alone, the fact that the protein-based vaccine appeared to amplify protection provided by the tenofovir gel suggests that this CBP may hold promise for humans.

None of these previous studies evaluated oral PrEP with Truvada, the only approved PrEP for HIV-1 prevention, and all modeled vaginal acquisition of HIV. Rectal acquisition of HIV-1 is a key driver of the MSM HIV-1 epidemic and a significant mode of HIV infection in women.⁴⁶

Ross et al. determined whether combining a polyvalent humoral-based vaccine with a human-equivalent dose of oral PrEP, would protect against rectal SHIV challenge in rhesus macaques.⁴⁷ The DNA/virus-like particle vaccine encoded HIV-1 Env and SIVmac239 Gag, was given intramuscularly and was boosted with intramuscular and intranasal injections of alum-adjuvanted Gag and Env particles. This combination had been shown to partially protect against vaginal SHIV challenge.⁴⁸ Intermittent treatment with oral PrEP (TDF and FTC in doses equivalent to human oral Truvada) was given 2 hours before and 22 hours after rectal SHIV_{162P3} challenge. This regimen was known to have partial efficacy against rectal SHIV.⁴⁹ The combination of vaccine and PrEP protected 7/8 animals. Although the study used historical efficacy data for PrEP alone, modeling approaches³⁹ applied to the data suggested that given an additive model, the CBP would have been predicted to be 83.5% protective, which is similar to the 87.5% protection observed. The model also suggested that negative or subtractive interactions of vaccine and PrEP did not occur. Interestingly, SHIV exposures during PrEP amplified vaccine-raised antibody titers in protected animals and the one CBP animal that became infected had a reduced peak VL compared to controls and early, high avidity antibodies to env and gag. These results suggest that combining oral PrEP with HIV vaccines could enhance protection against HIV-1 infection. Mechanistically, the oral PrEP may have allowed HIV exposures to amplify vaccine-raised immune responses providing immunologic and pharmacologic synergy.

Potential impact of CBP on post-infection viremia

Analysis of breakthrough infections in animals who have received CBP can be very informative, depending on the study design. For example, PrEP only or CBP animals may continue to receive PrEP for a period of time following infection since there is an eclipse of ~1 week between when infection occurs and peripheral blood RNA becomes positive. Moreover, by convention, most researchers require 2 consecutive viral RNA tests be positive and some studies continue PrEP until all the controls become infected (as done in the Ross et al study). The presence of drug following infection in the NHP models

mimics the human situation where individuals are intermittently tested for HIV infection and are likely to continue to receive drug or microbicide for some time after HIV infection. This along with vaccine-raised immune responses may provide an added benefit by limiting the size of the viral reservoir. The NHP studies of CBP support that this is likely. First, in the 5 studies evaluated, peak and set point viremia were frequently lower in the breakthrough CBP animals. Second, the 3 animals with transient, seronegative viremia (2 in the Cheng-Meyer et al. study and one in the Ross et al. study^{43, 47} suggest that CBP may have controlled and cleared local infections. Amplification of vaccine-raised antibody and T-cell responses by SHIV exposures in uninfected animals as seen in the Ross et al. and Cheng-Meyer et al. studies respectively,^{43, 47} supports the possibility of transient, cleared infections. More intensive evaluation of mucosal samples for viral reservoirs, drug levels or mucosal immune responses could elucidate these interactions in future CBP studies.

Theoretical human study designs for evaluating combined, partially effective biomedical preventions

Discussions about combining an HIV vaccine with other biomedical preventions in humans have been ongoing. Key questions, as in the NHP studies, are the sample size and type of study design required. Two publications summarize some of these deliberations. One group³⁹ provided information based on the number of incident cases required to determine an effect and the other group⁴⁰ based the estimates on assumed efficacies of the preventions. One example from each of the publications is shown in Table 2.

Excler et al.³⁹ were the first to provide designs to estimate efficacy of combined biomedical preventions. They provide expected numbers of incident infections in 4 different scenarios where combined vaccine and PrEP (VAXPREP) effects could be compared to vaccine, PrEP or no prevention. The scenarios are 2×2 factorial designs evaluating HIV incidence with or without an effect on viral load in participants who became infected. One of these scenarios (scenario B in³⁹ and described in Table 2A) was selected for discussion since with increased early HIV treatment today, it may be impossible to determine effects on viral load. Table 2A shows 4 study arms: placebo, vaccine, PrEP and VAXPREP (here termed CBP) and the outcome of interest is HIV incidence. Vaccine and PrEP efficacy were each set at 30%, additive and synergistic effects of vaccine and PrEP were modeled and background HIV incidence in the trial population was assumed to be 2%. In this model, if synergy of the CBP occurred, 80 incident infections and a sample size of ~4900 would provide adequate power to detect the effect on reduced HIV incidence. However, if vaccine and PrEP effects were additive, 200 incident infections and a much larger sample size would be needed. Given that enhancement of HIV-incidence by an HIV vaccine has been observed, the authors discuss designs to detect enhancement and note that this 4 arm design could possibly detect enhancing effects during interim monitoring³⁹. In their report, Excler et al. illustrate the impact of varying incidence and prevention efficacies on sample size and trial duration.

Table 2. Possible human combined biomedical prevention trial designs.

Prevention	Theoretical Efficacy (%)	Theoretical Incidence (%)	Study arms	Sample size	Comments
None		2	Nothing or double placebo	1200+	May not be possible if PrEP or vaccine are standard of care
HIV vaccine	30	1.4	Vaccine +/- placebo PrEP	1200+	
Other prevention	30	1.4	PrEP +/- placebo vaccine	1200+	
CBP, model	?		Vaccine + PrEP	1200+	
*CBP modelling estimates on efficacy and incidence					
Additive	51	0.98		May increase	May need to detect > 200 infections, increasing sample size Detecting 80 incident infections may suffice, overall sample size ~ 4900 for 80% power Effect may be determined during interim monitoring
Synergistic	> 51	0.67		May increase	
Enhancing [^]	?	2.6		No estimate	
B.					
Prevention	Theoretical Efficacy (%)		Study arms	Sample size	Comments re number of incident infections, sample size and power
None			Double placebo	1900	May not be possible if PrEP or vaccine are standard of care
HIV vaccine	40		Vaccine + PrEP placebo	1900	
Other prevention	60		PrEP + vaccine placebo	1900	
CBP, model	?		Vaccine + PrEP	1900	
**CBP modelling estimates on efficacy					
Additive	76				Can be evaluated
Synergistic	88				Can be evaluated
Antagonistic	64				Can be evaluated

Designs are presented for illustrative purposes only and may not be possible if an HIV vaccine or PrEP are standard of care or in widespread use. Then, alternative designs would be necessary.

A. Incidence-based 4 arm test-of-concept trial design to evaluate CBP. This is described in "scenario B" in Exler et al. 2011.³⁹ Assumptions are: background incidence of 2%; no viral load in model; vaccine and PrEP effects, both = 30%; vaccine and PrEP occur sequentially; 18 months of follow up. The design does not allow comparison of CBP to vaccine alone or PrEP alone unless the synergistic effect is large, but might allow ranking of preventions. Sample sizes are available (not shown) for 30 months in trial with 10% loss to follow up. * Additive calculation: $1.4 - (1.4 \times 0.3) = 0.98$, i.e. a 51% reduction. Synergistic incidence assigned as 0.67 empirically, assuming an effect > 51%.
^ Enhancing effect set at 30%.

B. Efficacy based 4 arm study design to evaluate CBP. This is described in trial design "A" in Janes et al. 2013.⁴⁰ Assumptions are: background incidence of 4%; 2 sided, 0.05 level likelihood ratio test under Cox proportional hazard model. Powered to detect: 40% vaccine efficacy rejecting null hypothesis of < 0% vaccine efficacy; 76% efficacy of CBP; rejecting null hypothesis that CBP efficacy is < 30% and to compare vaccine to CBP groups and rejecting null hypotheses that vaccine efficacy is not superior to CBP efficacy. ** CBP calculations: additive, $1 - (1 - 0.4) \times (1 - 0.6) = 1 - 0.24 = 0.76$, (Relative risk [RR] vaccine + PrEP); synergistic with decreased RR of 50, $1 - (0.24 - 0.12) = 0.88$, (RR vaccine + PrEP < RR vaccine x RR PrEP); antagonistic with increased RR of 50%, $1 - (0.24 + 0.12) = .64$, (RR vaccine + PrEP > RR vaccine x RR PrEP). Power to evaluate interactions varies.

Janes et al.⁴⁰ subsequently described 2, 3 and 4 arm CBP designs where the main outcome of interest is the efficacy of combination prevention. For illustrative purposes design A, a 4 arm study (double placebo, vaccine + PrEP placebo, PrEP (or other biomedical prevention) + vaccine placebo and the CBP) is described here (Table 2B). Note that both prevention arms have the alternative placebo, reducing the chance of unblinding. Several assumptions are different than the example from the Excler paper described in Table 2A³⁹ including a higher background incidence of 4%, PrEP efficacy of 60% and vaccine efficacy of 40%. With these assumptions, 1900 persons per arm are needed, giving an overall sample size of 7600 persons. Janes et al. describe two-stage randomizations that may apply where the willingness of vaccinees to take PrEP determines which arm they are randomized into.

If PrEP is approaching the standard of prevention, an HIV-vaccine alone is not justified and high CBP efficacy is predicted, different study designs will be required. Janes et al.⁴⁰ describe a 2-arm study (design D, not shown) requiring 4600 per arm comparing CBP to PrEP alone. In another scenario, where vaccine and PrEP (or other CBP) have been co-developed and are likely to have high CBP efficacy, Janes et al. estimate that only 1400 trial participants would be required in a 2 arm design (E, not shown, with 700 persons per arm where the CBP is compared to placebo).

Case study of PrEP use impacting HIV-vaccine trial redesign

Trial designs may need to evolve as the prevention landscape changes. An informative example of this, from the HVTN 505 DNA-Ad5 HIV-vaccine trial, is presented by Janes et al.⁴⁰ In 2010, data from the MSM oral Truvada PrEP trial, iPrEx⁵ became available indicating 44% reduction in HIV acquisition risk with PrEP. At this time, the HIV vaccine trial known as HVTN 505²⁹ (using DNA and Adenovirus-based HIV-vaccines) was ongoing and 50% of the proposed 1350 participants had been enrolled. Although, at this time, oral Truvada as PrEP had not been approved in the US or other countries, questions arose as to whether it should be provided as part of the prevention package and how use might impact HIV-incidence. The HVTN 505 trial had not been powered to detect a reduction in HIV incidence, only a reduction in viral load in vaccine recipients who became infected. The HVTN study team considered several options including that illustrated in Table 2B. After much discussion among trial investigators and community, the HVTN 505 trial sample size was increased to 2200 to detect a vaccine efficacy of 50% reduction in incidence and PrEP use by trial participants was monitored. The trial was stopped for efficacy futility in 2013.^{29,40}

Non-inferiority designs and placebo arms

As more preventions emerge, studies without placebo arms and non-inferiority designs may be needed to compare next-generation preventions to those with already proven moderate or greater efficacy or to compare combinations of biomedical preventions against a single prevention alone.

One situation may be when low PrEP efficacy is anticipated. Here, if CBP with PrEP and an HIV vaccine are being evaluated, a non-inferiority design might be needed to evaluate CBP compared to HIV-vaccine alone as discussed by Janes et al.⁴⁰ Non-inferiority designs may also be needed when high PrEP efficacy is anticipated, either for novel PrEP against a “gold standard” or CBP against established PrEP. The PrEP efficacy in the population may be risk-group dependent. Indeed, in a first for the HIV prevention field, a non-inferiority study is launching in 2016 in MSM. Known as HPTN 083, the study compares a new injectable form of PrEP (the integrase inhibitor cabotegravir, also known as CAB LA or GSK744) to oral PrEP with Truvada for HIV prevention.⁵⁰ MSM and transgender women, expected to have incidence >2%, will receive intramuscular CAB and oral PrEP placebo pills or oral PrEP with placebo injections in a double blind, double-dummy design. Given certain assumptions, a sample size of 4500, with 1:1 randomization into the 2 arms was selected to have 90% power to determine non-inferiority.⁵⁰ Such sample sizes may be required to compare a CBP to an effective existing prevention such as PrEP. Future trials of biomedical preventions for HIV may not include placebo or double placebo arm. This may depend on the likely efficacy of the individual or combined preventions, availability of the products, the population, country, age, gender, likely adherence of study participants and other relevant factors. Designs could include those similar to the HPTN 083 study or other 2 or 3 arm designs (such as designs C and D of Janes et al.⁴⁰). Trials will also need to consider the different impact of transiently used preventions (e.g. PEP, PrEP) or permanent preventions (e.g., voluntary male circumcision) as discussed.³⁵

Ethical and other considerations

Combinations of preventions may introduce ethical, regulatory and laboratory challenges that are more complex than a trial of one product alone. Institutional review boards, drug regulatory and other such bodies will need to assess the standard of care that will be provided during the trial and following the trial if the CBP is effective. Unique features of each prevention may also influence aspects of design and counseling. For example, candidate HIV vaccines, particularly those containing Env and Gag may induce false reactive HIV-tests, requiring specialized approaches to testing vaccinees for HIV infection. Persons receiving oral PrEP may face stigma related to a perception they are HIV infected if they are receiving pills usually used to treat HIV infection. Acceptability and likely adherence to particular PrEP products may vary depending on the population of trial participants, including age, gender, cultural factors and the like.

Logical selection of combinations

Trials of combined biomedical preventions should be informed by the mechanism of action and likely interactions of the preventions. For example, a non-vaccine prevention that prevents virus replication or integration after HIV enters target cells (as do Truvada, dapivirine and CAB) might be best combined with a prevention that blocks HIV entry into target cells (such as neutralizing antibodies or a vaccine that induces strong antibody responses). Or an entry-blocking drug such as maraviroc

or another drug directed at the entry-receptor CCR5, might best be combined with a T-cell-based vaccine that targets infected cells for killing after they become infected. These effects would likely be additive. Synergy might be expected if a vaccine induced both entry-blocking and post-entry effects (e.g. B-cell and T-cell responses) and the drug, or other prevention had entry or post-entry effects. Mucosal and other interactions should be anticipated. For example, activation of HIV-specific immune cells on HIV-exposure of an HIV vaccine recipient will result in a local influx into vaginal or rectal tissues of targets for HIV entry. Moreover, increased levels of dATP in activated CD4⁺ T-cells could reduce tenofovir efficacy on viral reverse transcription by competing as a substrate.⁵¹ Conversely, tenofovir shifts the IL-10/IL-12 balance of stimulated cells in vitro and therefore could modulate in vivo immune responses.⁵² While evaluating these tissue-level interactions cannot be done in humans, in vivo studies in animals, particularly NHP can more easily predict the magnitude and type of interaction. These data can guide selection of combinations and human trial design.

What efficacy should be expected for combined biomedical preventions?

There is no universally agreed on “bar” or “cut-off” for HIV prevention efficacy that may lead to a recommendation by a national or international body for implementation or approval of that prevention. Generally, clinical trials of HIV vaccines, microbicides and PrEP are powered to detect efficacy of at least 50%. Reproduction of efficacy in one or more trials of the same product is a key factor in leading to recommendations for implementation. This was the case for oral PrEP with Truvada and tenofovir, where multiple trials⁵⁻¹⁰ showed efficacies close to or above 50%.

However, even when efficacy is less than 50%, if a product shows promise and is replicated in a second trial, steps toward approval may begin. As an example, in the ASPIRE and The Ring studies^{19,20} intravaginal rings delivering dapivirine as topical PrEP reduced HIV incidence by 27–31%. Although this efficacy is only moderate, the concurrent positive results, along with higher efficacy in older women has led to an open label study⁵³ which, if successful, may lead to recommendations for approval and use.

In the only efficacious HIV-vaccine trial, the RV144 trial of canarypox expressing HIV antigens and gp120 protein boosts, efficacy was 31%.³⁰⁻³² There has not yet been a second trial of the RV144 platform, although the trial of a clade C version of the vaccine slated to begin in 2017 will hopefully have higher efficacy that may lead to recommendations for approval and use of this class of HIV vaccines.

What efficacy bar should individual preventions reach to be considered for a combination prevention trial? In the example shown in Table 2A, Exler et al.³⁹ modeled HIV vaccine and PrEP efficacies of 30%. If an additive effect was predicted, combination of an RV144-like vaccine with efficacy of ~30% with a dapivirine-like intravaginal ring with ~30% efficacy would yield overall efficacy of ~51% but if synergy was observed, efficacy would likely be higher (Table 2A). Combination of a moderately effective vaccine with oral PrEP, which in many populations has high efficacy, might yield almost complete protection against HIV, as modeled in the Ross et al. macaque study⁴⁷. Decisions

about the efficacy which a CBP should reach for a clinical trial or for consideration for recommendations for use may vary based on the prevention and the intended population.

The future of combined biomedical preventions

Today, since oral PrEP is effective and increasingly available, and an HIV vaccine is not, PrEP is the cornerstone of new biomedical preventions. As HIV-vaccines and other preventions continue to be evaluated, one situation where CBP may occur is in HIV vaccine or other trials where oral PrEP may be taken or offered as part of a prevention package for trial participants. When a protective HIV vaccine becomes available, vaccine might be administered to pre-teens or young adults prior to onset of sexual activity as for human papilloma virus vaccine, but PrEP may still have a role. In catch-up scenarios in sexually active individuals, PrEP (or other biomedical prevention) may be recommended until HIV vaccinations are complete, particularly if the HIV vaccine was known to be only partially effective. PrEP use may precede vaccination, if a person has missed vaccination, is sexually active, and then being referred for vaccination. PrEP may also be useful in a clinical trial if there is a concern regarding transient enhancement of HIV risk by an HIV vaccine since concurrent PrEP may allow immunity to be raised without the risk of enhancement. In the future, if HIV vaccines need periodic updating to deal with strain variation, as is the case with influenza vaccines, PrEP could be used for at-risk persons when immunity was being raised, or if they missed new vaccinations. On the other hand, HIV-vaccines may provide added benefit if circulating HIV strains have PrEP-resistant drug-mutations.

In the field of biomedical prevention there is a blurring of the boundaries between different types of preventions. While PrEP currently refers to prevention with systemically delivered anti-retroviral drugs, topical microbicides historically contained HIV-inhibitory compounds that were not typical anti-retroviral drugs. Now, topical HIV prevention includes many of the anti-retroviral drugs used for systemic PrEP. HIV-neutralizing antibodies are being evaluated as systemic PrEP, if given as injections but as topical PrEP/microbicides when being delivered mucosally in gels or vaginal rings. These antibodies or other novel HIV-blocking agents may be more akin to a vaccine or gene-therapy if delivered by viral vectors.

Several HIV preventions may be combined in one product (e.g., a tablet, vaginal ring, long acting injection, implant may deliver several anti-retroviral drugs, HIV-neutralizing antibodies, HIV microbicides or combinations thereof). Multipurpose preventions technologies (MPT) go one step further, adding approaches to prevent HSV-2, other STI and pregnancy in addition to HIV.⁵⁴ Could MPT also deliver vaccine components, such as gp120, to boost immunity from a prior vaccine series? Could vaccine immunizations be given concurrently with periodic intramuscular injections of a long-acting anti-retroviral PrEP modality?

Conclusion

With more than 5,000 new HIV infections occurring each day worldwide, it is important to discuss the obstacles to combining

biomedical preventions modalities to accelerate an end to HIV/AIDS. Researchers in the PrEP, microbicide and HIV-vaccine fields could discuss ideal CBP combinations and their possible co-development or co-administration. Animal models could evaluate CBP concepts prior to clinical testing and guide clinical trial design.

Disclosure of potential conflicts of interest

No potential conflicts of interest were disclosed.

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References

- [1] UNAIDS fact sheet. http://www.unaids.org/sites/default/files/media_asset/20150901_FactSheet_2015_en.pdf
- [2] CDC HIV/AIDS statistics. <http://www.cdc.gov/hiv/statistics/overview/ata glance.html>
- [3] CDC news release. <http://www.cdc.gov/nchhstp/newsroom/2016/croi-2016.html>
- [4] Gray GE, Laher F, Doherty T, Abdool Karim S, Hammer S, Mascola J, Beyrer C, Corey L. Which new health technologies do we need to achieve an end to HIV/AIDS? *PLoS Biol* 2016; 14:e1002372; PMID:26933962; <http://dx.doi.org/10.1371/journal.pbio.1002372>
- [5] Grant R, Lama JR, Anderson PL, McMahan V, Liu AY, Vargas L, Goicochea P, Casapia M, Guanira-Carranza JV, Ramirez-Cardich ME, et al. Preexposure chemoprophylaxis for HIV prevention in men who have sex with men. *N Engl J Med* 2010; 363:2587-99; PMID:21091279; <http://dx.doi.org/10.1056/NEJMoa1011205>
- [6] Thigpen M, Kebaabetswe P, Paxton L, Smith D, Rose C, Segolodi T, Henderson F, Pathak S, Soud F, Chillag K, et al. Antiretroviral preexposure prophylaxis for heterosexual HIV transmission in Botswana. *N Engl J Med* 2012; 367:423-34; PMID:22784038; <http://dx.doi.org/10.1056/NEJMoa1110711>
- [7] Baeten J, Donnell D, Ndase P, Mugo N, Campbell J, Wangisi J, Tappero J, Bukusi E, Cohen C, Katabira E, et al. Antiretroviral prophylaxis for HIV prevention in heterosexual men and women. *N Engl J Med* 2012; 367:399-410; PMID:22784037; <http://dx.doi.org/10.1056/NEJMoa1108524>
- [8] Choopanya K, Martin M, Suntharasamai P, Sangkum U, Mock PA, Leethochawalit M, Chiamwongpaet S, Kitisin P, Natrujirete P, Kittimunkong S, et al. Antiretroviral prophylaxis for HIV infection in injecting drug users in Bangkok, Thailand (the Bangkok Tenofovir Study): a randomised, double-blind, placebo-controlled phase 3 trial. *Lancet* 2013; 381:2083-90; PMID:23769234; [http://dx.doi.org/10.1016/S0140-6736\(13\)61127-7](http://dx.doi.org/10.1016/S0140-6736(13)61127-7)
- [9] McCormack S, Dunn D, Desai M, Dolling D, Gafos M, Gilson R, Sullivan A, Clarke A, Reeves I, Schembri G, et al. Pre-exposure prophylaxis to prevent the acquisition of HIV-1 infection (PROUD): effectiveness results from the pilot phase of a pragmatic open-label randomised trial. *Lancet* 2015; 387:53-60; PMID:26364263; [http://dx.doi.org/10.1016/S0140-6736\(15\)00056-2](http://dx.doi.org/10.1016/S0140-6736(15)00056-2)
- [10] Molina JM, Capitant C, Spire B, Pialoux G, Cotte L, Charreau I, Tremblay C, Le Gall JM, Cua E, Pasquet A and the ANRS IPERGAY Study Group. On-demand preexposure prophylaxis in men at high risk for HIV-1 infection. *N Engl J Med* 2015; 373:2237-46; PMID:26624850; <http://dx.doi.org/10.1056/NEJMoa1506273>
- [11] Van Damme L, Corneli A, Ahmed K, Agot K, Lombaard J, Kapiga S, Malahleha M, Owino F, Manongi R, Onyango J, et al. Preexposure prophylaxis for HIV infection among African women. *N Engl J Med* 2012; 367:411-22; PMID:22784040; <http://dx.doi.org/10.1056/NEJMoa1202614>
- [12] Marrazzo J, Ramjee G, Richardson B, Gomez K, Mgodini N, Nair G, Palanee T, Nakabiito C, van der Straten A, Noguchi L, et al. Tenofovir-based preexposure prophylaxis for HIV infection among African women. *N Engl J Med* 2015; 372:509-18; PMID:25651245; <http://dx.doi.org/10.1056/NEJMoa1402269>
- [13] FDA. FDA approves first drug for reducing the risk of sexually acquired HIV infection. <http://www.fda.gov/NewsEvents/Newsroom/PressAnnouncements/ucm312210.htm>
- [14] CDC PrEP guidelines. <http://www.cdc.gov/hiv/pdf/PrEPguide lines2014.pdf>
- [15] WHO HIV guidelines. <http://www.who.int/hiv/pub/guidelines/key populations/en/>
- [16] CDC media release. <http://www.cdc.gov/nchhstp/newsroom/2016/croi-2016.html>
- [17] Abdool Karim Q, Abdool Karim S, Frohlich J, Grobler A, Baxter C, Mansoor L, Kharsany A, Sibeko S, Mlisana K, Omar Z, et al. Effectiveness and safety of tenofovir gel, an antiretroviral microbicide, for the prevention of HIV infection in women. *Science* 2010; 329:1168-74; <http://dx.doi.org/10.1126/science.1193748>
- [18] Rees H, Delany-Moretlwe S, Lombard C, Baron D, Panchia R, Myer L, Schwartz J, Doncel GF, Gray G. FACTS 001 Phase III trial of pericoital tenofovir 1% gel for HIV prevention in women. Conference on Retroviruses and Opportunistic infections 2015, Seattle, WA.
- [19] Baeten JM, Palanee-Phillips T, Brown ER, Schwartz K, Soto-Torres LE, Govender V, Mgodini NM, Matovu Kiweewa F, Nair G, Mhlanga F, et al. and MTN-020-ASPIRE Study Team. Use of a vaginal ring containing dapivirine for HIV-1 prevention in women. *N Engl J Med* 2016; [epub ahead of print]; <http://dx.doi.org/10.1056/NEJMoa1506110>
- [20] Nel A, Kapiga S, Bekker LG, Devlin B, Borremans M, Rosenberg Z, for the IPM 027/Ring Study Research Center Teams. Safety and efficacy of dapivirine vaginal ring for HIV-1 prevention in African women. Conference on Retroviruses and Opportunistic infections 2016, Boston. LB 110
- [21] Harmon T, Fisher K, McGlynn M, Stover J, Warren M, Teng Y, Naveke A. Exploring the potential health impact and cost-effectiveness of AIDS vaccine within a comprehensive HIV/AIDS response in low- and middle-income countries. *PLoS One* 2016; 11:e0146387; PMID:26731116; <http://dx.doi.org/10.1371/journal.pone.0146387>
- [22] Barouch DH, Michael NL. Accelerating HIV-1 vaccine efficacy trials. *Cell* 2014; 159:969-72; PMID:25416935; <http://dx.doi.org/10.1016/j.cell.2014.10.046>
- [23] Excler JL, Robb ML, Kim JH. Prospects for a globally effective HIV-1 vaccine. *Am J Prev Med* 2015; 49:S307-18; PMID:26590431; <http://dx.doi.org/10.1016/j.amepre.2015.09.004>
- [24] Sheets R, Zhou T, I K. Review of efficacy trials of HIV-1/AIDS vaccines and regulatory lessons learned: A review from a regulatory perspective. *Biologicals* 2015; 44:73-89; PMID:26776940; <http://dx.doi.org/10.1016/j.biologicals.2015.10.004>
- [25] Pitisuttithum P, Gilbert P, Gurwith M, Heyward W, Martin M, van Griensven F, Hu D, Tappero JW, Choopanya K. Randomized, double-blind, placebo-controlled efficacy trial of a bivalent recombinant glycoprotein 120 HIV-1 vaccine among injection drug users in Bangkok, Thailand. *Infect Dis* 2006; 194:1661-71; <http://dx.doi.org/10.1086/508748>
- [26] Flynn NM, Forthal DN, Harro CD, Judson FN, Mayer KH, Para MF. Placebo-controlled phase 3 trial of a recombinant glycoprotein 120 vaccine to prevent HIV-1 infection. *J Infect Dis* 2005; 191:654-65; PMID:15688278; <http://dx.doi.org/10.1086/428404>
- [27] Buchbinder SP, Mehrotra DV, Duerr A, Fitzgerald DW, Mogg R, Li D, Gilbert PB, Lama JR, Marmor M, Del Rio C, et al. Efficacy assessment of a cell-mediated immunity HIV-1 vaccine (the Step Study): a double-blind, randomised, placebo-controlled, test-of-concept trial. *Lancet* 2008; 372:1881-93; PMID:19012954; [http://dx.doi.org/10.1016/S0140-6736\(08\)61591-3](http://dx.doi.org/10.1016/S0140-6736(08)61591-3)
- [28] Gray GE, Allen M, Moodie Z, Churchyard G, Bekker LG, Nchabeleng M, Mlisana K, Metch B, de Bruyn G, Latka MH, et al.

- Safety and efficacy of the HVTN 503/Phambili study of a clade-B-based HIV-1 vaccine in South Africa: a double-blind, randomised, placebo-controlled test-of-concept phase 2b study. *Lancet Infect Dis* 2011; 11:507-15; PMID:21570355; [http://dx.doi.org/10.1016/S1473-3099\(11\)70098-6](http://dx.doi.org/10.1016/S1473-3099(11)70098-6)
- [29] Hammer SM, Sobieszczyk ME, Janes H, Karuna ST, Mulligan MJ, Grove D, Koblin BA, Buchbinder SP, Keefer MC, Tomaras GD, et al. Efficacy trial of a DNA/rAd5 HIV-1 preventive vaccine. *N Engl J Med* 2013; 369:2083-92; PMID:24099601; <http://dx.doi.org/10.1056/NEJMoa1310566>
- [30] Rerks-Ngarm S, Pitisuttithum P, Nitayaphan S, Kaewkungwal J, Chiu J, Paris R, Prensri N, Namwat C, de Souza M, Adams E, et al. Vaccination with ALVAC and AIDSVAX to prevent HIV-1 infection in Thailand. *N Engl J Med* 2009; 361:2209-20; PMID:19843557; <http://dx.doi.org/10.1056/NEJMoa0908492>
- [31] Gilbert PB, Berger JO, Stablein B, Becker S, Essex M, Hammer SM, Kim JM, DeGruttola VG. Statistical interpretation of the RV144 HIV vaccine efficacy trial in Thailand: A case study for statistical issues in efficacy trials. *J Infect Dis* 2011; 203:969-75; PMID:21402548; <http://dx.doi.org/10.1093/infdis/jiq152>
- [32] Kim JH, Excler JL, Michael NL. Lessons from the RV144 Thai phase III HIV-1 vaccine trial. *Ann Rev Med* 2015; 66:1275-1286.
- [33] Haynes BF, Gilbert PB, McElrath MJ, Zolla-Pazner S, Tomaras GD, Alam SM, Evans DT, Montefiori DC, Karnasuta C, Sutthent R, et al. Immune-correlates analysis of an HIV-1 vaccine efficacy trial. *N Engl J Med* 2012; 366:1275-86; PMID:22475592; <http://dx.doi.org/10.1056/NEJMoa1113425>
- [34] Corey L, Gilbert PB, Tomaras GD, Haynes BF, Pantaleo G, Fauci AS. Immune correlates of vaccine protection against HIV-1 acquisition. *Sci Transl Med* 2015; 310rv7:1-10
- [35] Sheets R, Zhou T, Knezevic I. Scientific and regulatory challenges in evaluating clinical trial protocols for HIV-1/AIDS vaccines - A review from a regulatory perspective. *Biologicals* 2015; 44:90-110; PMID:26732973; <http://dx.doi.org/10.1016/j.biologicals.2015.11.003>
- [36] AIDS Vaccine Advocacy Coalition. <http://www.avac.org/trial/hvtn-702>
- [37] HIV Prevention Trial Network. http://www.hptn.org/research_studies
- [38] Huang Y, DiazGranados C, Janes H, Huang Y, deCamp AC, Metch B, Grant S, Sanchez B, Phogat S, Koutsoukos M, et al. Selection of HIV vaccine candidates for concurrent testing in an efficacy trial. *Curr Opin Virol* 2016; 17:57-65; PMID:26827165; <http://dx.doi.org/10.1016/j.coviro.2016.01.007>
- [39] Excler J, Rida W, Priddy F, Gilmour J, McDermott A, Kamali A, Anzala O, Mutua G, Sanders E, Koff W, et al. AIDS vaccines and pre-exposure prophylaxis: is synergy possible? *AIDS Res Hum Retroviruses* 2011; 27:669-80; PMID:21043994; <http://dx.doi.org/10.1089/aid.2010.0206>
- [40] Janes H, Gilbert P, Buchbinder S, et al. In pursuit of an HIV vaccine: designing efficacy trials in the context of partially effective non vaccine prevention modalities. *AIDS Res Hum Retroviruses* 2013; 11:1513-23; <http://dx.doi.org/10.1089/aid.2012.0385>
- [41] Veazey RS. Animal models for microbicide safety and efficacy testing. *Curr Opin HIV AIDS* 2013; 8:295-303; PMID:23698560
- [42] Pegu P, Vaccari M, Gordon S, Keele B, Doster M, Guan Y, Ferrari G, Pal R, Ferrari M, Whitney S, et al. Antibodies with high avidity to the gp120 envelope protein in protection from simian immunodeficiency virus SIV(mac251) acquisition in an immunization regimen that mimics the RV-144 Thai trial. *J Virol* 2013; 87:1708-19; PMID:23175374; <http://dx.doi.org/10.1128/JVI.02544-12>
- [43] Cheng-Mayer C, Huang Y, Gettie A, Tsai L, Ren W, Shakirzyanova M, Sina S, Trunova N, Blanchard J, Jenkins L, et al. Delay of simian human immunodeficiency virus infection and control of viral replication in vaccinated macaques challenged in the presence of a topical microbicide. *AIDS* 2011; 25:1833-41; PMID:21750420; <http://dx.doi.org/10.1097/QAD.0b013e32834a1d94>
- [44] Barouch DH, Klasse PJ, Dufour J, Veazey RS, Moore JP. Macaque studies of vaccine and microbicide combinations for preventing HIV-1 sexual transmission. *Proc Natl Acad Sci U S A* 2012; 109:8694-8; PMID:22586094; <http://dx.doi.org/10.1073/pnas.1203183109>
- [45] LeGrand R, Bosquet N, Dispinseri S, Hopewell N, Gosse L, Desjardins D, Shen X, Tolazzi M, Ochsenbauer C, Saidi H, et al. Microbicide-vaccine combination provides significant protection against vaginal SHIV 162P3 challenge in cynomolgous monkeys. HIV Research for Prevention Conference, Durban, SA, October 2014. Abstract A08.06 LB.
- [46] Baggaley R, Dimitrov D, Owen B, Pickles M, Butler A, Masse B, Boily M. Heterosexual anal intercourse: a neglected risk factor for HIV? *Am J Reprod Immunol* 2013; 69:95-105; PMID:23279040; <http://dx.doi.org/10.1111/aji.12064>
- [47] Ross TM, Pereira LE, Luckay A, McNicholl JM, García-Lerma JG, Heneine W, Eugene HS, Pierce-Paul BR, Zhang J, Hendry RM, Smith JM. A polyvalent clade B VLP HIV vaccine combined with partially protective Truvada pre-exposure prophylaxis prevents SHIV infection in macaques and primes for virus-amplified immunity. *AIDS Res Hum Retroviruses* 2014; 30:1072-81; PMID:24914761; <http://dx.doi.org/10.1089/aid.2014.0030>
- [48] McBurney S, Landucci G, Forthal D, Ross T. Evaluation of heterologous vaginal SHIV SF162p4 infection following vaccination with a polyvalent Clade B virus-like particle vaccine. *AIDS Res Hum Retroviruses* 2012; 28:1063-72; PMID:22214267
- [49] García-Lerma J, Cong M, Mitchell J, Youngpairoj A, Zheng Q, Masciotra S, Martin A, Kuklenyik Z, Holder A, Lipscomb J, et al. Intermittent prophylaxis with oral Truvada protects macaques from rectal SHIV infection. *Sci Transl Med* 2010; 2:14ra4
- [50] HPTN 083 trial. http://www.hptn.org/research_studies/hptn083.asp
- [51] García-Lerma J, Aung W, Cong M, Zheng Q, Youngpairoj A, Mitchell J, Holder A, Martin A, Kuklenyik S, Luo W, et al. Natural substrate concentrations can modulate the prophylactic efficacy of nucleotide HIV reverse transcriptase inhibitors. *J Virol* 2011; 85:6610-7; <http://dx.doi.org/10.1128/JVI.00311-11>
- [52] Melchjorsen J, Risør MW, Sogaard OS, O'Loughlin KL, Chow S, Paludan SR, Ellermann-Eriksen S, Hedley DW, Minderman H, Østergaard L, Tolstrup M. Tenofovir selectively regulates production of inflammatory cytokines and shifts the IL-12/IL-10 balance in human primary cells. *J Acquir Immune Defic Syndr* 2011; 57:265-75; PMID:21471820; <http://dx.doi.org/10.1097/QAI.0b013e328185276>
- [53] NIH news release. <http://www.niaid.nih.gov/news/newsreleases/2016/Pages/HOPE-trial.aspx>
- [54] Initiative for Multipurpose Prevention Technology. <http://mpts101.org>
- [55] Karim SSA. HIV pre-exposure prophylaxis in injecting drug users. *Lancet* 2013; 381(9883):2060-2062; [http://dx.doi.org/10.1016/S0140-6736\(13\)61140-X](http://dx.doi.org/10.1016/S0140-6736(13)61140-X)