

Microbicide research: current and future directions

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Purpose of review

Microbicide research has been in the forefront of scientific literature in recent months. Results of large-scale clinical trials have been announced with resultant investigations into the factors that may have contributed to the disappointing outcomes of the most promising candidates. This review takes into consideration clinical, basic scientific and behavioural research published on microbicides in the past 12–18 months.

Recent findings

Two trials testing PRO 2000, a sulphated polymer, suggested that it has no effect on HIV. Basic science research revealed several facts such as the loss of antiviral activity of microbicides in the presence of seminal plasma. Methodological models suggested that dilution factors might impact on measures of efficacy. Advancement into safety testing of highly specific antiretroviral products such as tenofovir and UC781 for both rectal and vaginal use shows promise. Development of drug delivery systems such as intravaginal rings may alleviate some of the adherence challenges faced when using coitally dependant products.

Summary

In the recent past, microbicide research has had disappointing outcomes. However, it has also provided a better understanding of factors that may reduce effectiveness of promising products, enabling the field to be better equipped to select and test new products.

Keywords

clinical trials, efficacy, microbicides, modelling, safety

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Introduction

The disproportionately high HIV infection rates among young women in many developing countries indicate an urgent need for HIV prevention options initiated by women. Condoms, when correctly and consistently used, have been shown to prevent HIV transmission; however, this method may not be available to women who are unable to negotiate condom use with their male partners [1–3].

Microbicides are products designed to be applied to the vagina or rectum for the purpose of reducing the acquisition of sexually transmitted infections (STIs) including HIV [4]. In the last year, we have seen new developments in this field ranging from basic and social/behavioural science research to results of large-scale clinical trials. Some of the published work is reviewed here.

Microbicide gels: recent clinical trials and research

To date, six candidate microbicides have been tested in clinical trials to assess their efficacy. These included N9

(sponge, film and gel formulation) [5–7], Savvy (C31G; Cellegy Pharmaceuticals, Huntington Valley, PA, USA) [8,9], cellulose sulphate [10,11], Carraguard (PC-515; Clean Chemical Sweden [CCS], Borlänge, Sweden) [12], PRO 2000 (Endo Pharmaceuticals, Chadds Ford, PA, USA) [13] and BufferGel (ReProtect LLC, Baltimore, Maryland, USA) [14**]. All products were shown to have no effect on HIV, with two – cellulose sulphate and N9 – showing a trend towards increased risk of HIV infection.

After a decade of research, a glimmer of hope for a possible proof of concept was raised in the trial testing PRO 2000, a sulphated polymer [14**]. The HPTN 035 trial, funded by the National Institutes of Health, was designed to test the safety and effectiveness of the BufferGel and 0.5% PRO 2000 microbicide gels for the prevention of male-to-female HIV transmission. The phase II/Ib, four-arm randomized, placebo-controlled trial was conducted in Malawi, South Africa, Zambia, Zimbabwe and the USA. A total of 3087 women were followed up for an average of 20.4 months with a retention rate of over 90% and adherence to product use of 81.1%. The results suggested that 0.5% PRO 2000 reduced the rate of HIV acquisition in women by 30%

but the results were not statistically significant [hazard ratio vs. no gel 0.7, 95% confidence interval (CI) 0.4–1.0, $P=0.06$]. PRO 2000 had no effect on STIs or prevention of pregnancy. BufferGel had no effect on transmission of HIV or STIs and did not prevent pregnancy. Both products were safe to use [14**].

Another trial testing PRO 2000 in two concentrations (0.5 vs. 2%) against a placebo (MDP 301) was conducted by the UK-based Microbicides Development Programme. The trial was conducted among 9380 women from Uganda, Zambia, Tanzania and South Africa. The women were followed up for an average of 12 months with a retention rate of over 80%, whereas reported adherence was more than 80%. In early 2008, a recommendation was made by the Data and Safety Monitoring Committee to drop the 2% arm due to futility. The final data released in December 2009 suggested that although both concentrations of PRO 2000 were well tolerated, 0.5% had no effect on HIV acquisition [hazard ratio vs. placebo (95% CI) 1.05 (0.82–1.34)] [15]. These results eclipsed the glimmer of hope raised by the HPTN 035 trial.

These disappointing results raised concerns about the relevance and adequacy of preclinical data used to advance products into large-scale clinical trials. As expected, several groups analysed various factors ranging from basic science to study design and adherence to products to better understand the clinical trial outcomes of what seemed like promising products.

Although products such as PRO 2000, cellulose sulphate and Carraguard are all classified as polyanions, members of this class differ chemically from each other, with variations in pharmacological, toxicological and antiviral properties [4]. Given that all the mentioned polyanions have failed to show efficacy against HIV in clinical trials and cellulose sulphate appears to increase risk, Sonza *et al.* [16*] conducted studies to determine whether SPL7013 and PRO 2000 enhanced HIV-1 replication in human peripheral blood mononuclear cells *in vitro*. They concluded that SPL7013 and PRO 2000 did not enhance HIV replication *in vitro*, and that any enhancement observed *in vitro* may be related to assay conditions and is not an intrinsic property of the compounds tested. These authors suggest that adherence to product use by study participants and its antiviral potency is more relevant for its in-vivo efficacy relative to the extent to which the product compromises the integrity of the genital tract, increases inflammation and interferes with immune function.

In keeping with research on polyanions or entry inhibitors, a phase II extended safety study [17] of two Invisible Condom (Universite Laval, Quebec, Canada) formulations was conducted among 220 Cameroonian women. Invisible Condom is formulated in the following two

ways: as a polymer gel, which acts as a physical barrier that blocks entry of pathogens, and as a polymer gel with added sodium lauryl sulphate, which provides an additional chemical barrier. The authors report that both formulations and applicators were well tolerated and acceptable for twice-daily application for 8 weeks.

von Mollendorf *et al.* [18] published the results of a safety and feasibility study of the cervical diaphragm used with ACIDFORM gel or K-Y Jelly. ACIDFORM is an acid-buffering gel, which helps to maintain the acidic vaginal pH, and is spermicidal. The objective was to use a microbicide gel in combination with a physical barrier to determine the safety and feasibility of a dual method of protection. Pelvic safety assessments included assessment of vaginal symptoms, abnormal pelvic examination findings or noniatrogenic colposcopy findings. Overall, there were more safety events in the ACIDFORM gel arm compared with the K-Y Jelly arm, although this difference was not statistically significant. The authors concluded that on the whole, women liked the use of dual methods of protection against HIV. Although ACIDFORM may not display the high specificity to HIV required for HIV prevention, the use of the diaphragm together with a highly active antiretroviral (ARV) product may contribute to prevention of certain STIs [19].

Research into factors influencing the outcomes of trials

A recently published paper by Keller *et al.* [20**] discusses a possible explanation of why promising preclinical data on PRO 2000 could not be replicated in large-scale human clinical trials. A study in which cervicovaginal lavage samples were collected pre- and postcoitally from heterosexual couples suggested that PRO 2000 significantly inhibited HIV-1 and HSV2 (both $P=0.01$) precoitally. However, in the postcoital sample, the antiviral activity of PRO 2000 was reduced and no significant protective effect was observed for HIV ($P=0.45$) and HSV2 ($P=0.56$), suggesting that seminal plasma may interfere with the anti-HIV activity of PRO 2000. This was confirmed by laboratory cell culture techniques [20**]. The authors recommended testing of postcoital efficacy of new products prior to embarking on large-scale clinical trials.

Kyeong-Ae *et al.* [21**] showed that fragments of prostatic acid phosphatase, which form amyloid-like aggregates (also known as 'semen-derived enhancer of viral infection' or SEVI [22]), are the factor in semen, which decreases the effectiveness of microbicides. SEVI binds to both HIV and target cells and thus facilitates viral fusion; only certain anionic polymers were found to inhibit this activity [23**]. The identification of microbicides that are active in the presence of semen/SEVI would be a positive step towards the development of effective products.

Others provide further explanations as to why cellulose sulphate and N9, for example, may have contributed to the increased risk of HIV acquisition. Mesquita *et al.* [24**] used a dual chamber culture system to measure transepithelial electrical resistance (TER) to evaluate the effect of microbicides on epithelial barriers, expression of tight and adherens junctional proteins and subsequent ability of HIV to cross this barrier and infect target cells cultured in the lower chamber of the model. They showed that exposure to N9 or cellulose sulphate – but not tenofovir or PRO 2000 – resulted in a rapid and sustained reduction in TER and a marked increase in HIV infection. They concluded that epithelial barrier disruption resulted in the increased risk of HIV acquisition observed in the N9 and cellulose sulphate trials, and that cellulose sulphate may also have enhanced HIV replication. The authors recommended that more comprehensive in-vitro safety tests are needed, with more stringent preclinical assessment of moderate safety, which could be more predictive of clinical outcome.

Mâsse *et al.* [25**] used a statistical model to provide further explanations as to why the products tested to date have not been found to be effective. They propose that ‘diluting’ factors such as adherence, time off product due to pregnancy, acquisition of HIV infection through anal intercourse and the lubricating/physical barrier effect of the placebo may render an efficacious product apparently less effective in clinical trials. For example, in completed trials, the true efficacy of an active gel might be 50%, but dilution effects could reduce the overall effectiveness to the range of 24–33%. When estimating the efficacy of a product during protocol development, consideration should be given to the impact of dilution factors. This option will allow for more accurate estimation of the overall efficacy one is likely to see.

Given that accurate reporting of sexual behaviour data is important to mitigate adherence as a dilution factor (as described above), Norris Turner *et al.* [26*] used audio computer-assisted self interviews (ACASIs) in a small number of women to determine accuracy of reporting. In their study of 132 women, 104 women reported that they had misinformed trial interviewers at least once for several reasons: politeness, to avoid criticism or seek praise or to avoid embarrassment. The desirability bias is high in clinical trials and further research is needed on strategies that encourage participants to report accurately on their sexual behaviour, practices and adherence to product use.

Tolley *et al.* [27**] reviewed adherence measures in all the trials conducted to date. Analysis of adherence to product use is critical in clinical trials of microbicides in order to determine effectiveness, provide additional evidence to support trial results, understand safety and

understand acceptability. Several mechanisms for measuring adherence have been used in past microbicide clinical trials; these have included both direct and indirect measures. Direct measures could facilitate the collection of unbiased data, which would improve the determination of product effectiveness. Indirect measures fall into two categories: objective measures and self-reported measures. Self-reported measures are generally considered the most susceptible to error; Tolley *et al.* [27**] suggest that triangulation techniques employing more than one measurement method may resolve such errors and contribute to greater accuracy. Successful development of methods to enhance both adherence and its measurement will enable researchers to interrogate data on a far more sophisticated level than is now possible [27**].

Drug delivery systems

Selection of an appropriate drug delivery system is important if the intervention is to be acceptable and user friendly. Rohan and Sassi [28] outline some of the challenges of designing drug delivery systems. They propose that prior to identifying the drug delivery system, the manufacturers need to clearly understand the chemical and physical characteristics of the potential drug candidate and its mechanisms of action. Coupled with this should be a thorough understanding of the vaginal environment, tissues and the natural defence mechanisms present. Although the majority of the products tested to date have been formulated as vaginal gels, new developments include vaginal tablets, films and rings. An efficacious microbicide should be formulated in a delivery system which is acceptable to users and accords with their potential social and cultural preferences.

The new generation of microbicide products

Given the low specificity to HIV indicated in all the microbicide products tested in large-scale trials to date, the logical extension to finding novel and highly effective strategies for prevention of acquisition of HIV was to use potent but well tolerated ARV agents, which have good safety and tolerability data and are already used in the treatment of HIV infection. Preexposure prophylaxis (PrEP) with ARVs has been effective in preventing mother-to-child transmission of HIV [29]. The concept is now extended to determine the role of oral and vaginally administered ARV prophylaxis for HIV prevention in heterosexuals and homosexuals [30*].

Evidence derived from studies [31,32] on mice and macaques suggests that systemic ARVs can provide protection from HIV infection, and supports both daily and intermittent use. ARV-based microbicides developed as

gels are now undergoing testing in ongoing phase I, II and IIb trials [33]. Tenofovir and emtricitabine/tenofovir (Truvada; Gilead Sciences Inc., Foster City, California, USA) are the products of choice based on their good safety and tolerability data.

A novel delivery system for microbicides is the intravaginal ring (IVR). Dapivirine (TM120), a lead candidate nonnucleoside reverse transcriptase inhibitor (NNRTI), has been incorporated into such a ring for sustained mucosal delivery. Use of an IVR for drug delivery is expected to increase adherence and efficacy over that achieved with the conventional direct application of a microbicide gel. A phase I trial [34[•]] compared a matrix IVR (25 mg dapivirine uniformly dispersed) and reservoir IVR (25 mg dapivirine and 25 mg radio-opaque barium sulphate) against a placebo IVR. Reservoir type rings have a central core containing the active drug surrounded by an unmedicated outer ring, whereas the active ingredient is dispersed throughout the ring in the matrix type [35]. The authors reported that both IVRs were safe and well tolerated [34[•]].

Chief among concerns regarding the use of ARVs for PrEP have been the possibility of systemic uptake and development of viral resistance. Data from clinical trials [36–38] have shown that ARVs from microbicides may be absorbed through the vagina, but that the concentration of the compounds in the blood was several orders of magnitude smaller than that found for oral administration [36,37]. This suggests that the likelihood of systemic toxicity may be lower than that experienced with oral formulations. Were a microbicide with a high propensity to cause resistance to be widely used, the risk could be outweighed by decreased transmission if the product is moderately efficacious [39]. Eight countries have implemented the WHO methodology for surveillance of HIV drug resistance mutations and all reported less than 5% transmitted resistance to all drugs and drug classes [40]. However, further research on the possibility of development of drug resistance in HIV-positive microbicide users and transmission of resistant virus to users from partners is needed. This is particularly pertinent for the use of tenofovir-containing microbicides in Africa, as subtype C virus has been shown to rapidly develop the K65R mutation in culture [41], although the diminished rate of transmission and lowered replicative capacity of viruses with this mutation must be weighed against this ability [42,43[•]]. In order to detect seroconversion as early as possible and minimize the development of resistance, stringent HIV testing regimes have been included in current trials of tenofovir-containing microbicides – product use is halted as soon as seroconversion is detected. Licensure and roll-out of an effective ARV-containing microbicide will necessitate the development of adequate infrastructure

and human capacity for frequent testing and counselling of users.

Safety studies in men

Very few studies have assessed the safety of topical application of microbicide gels directly to the penis. Schwartz *et al.* [44[•]] undertook a study among circumcised ($n = 18$) and uncircumcised ($n = 18$) men randomized to receive either tenofovir gel or K-Y Jelly. The gels were applied to the penis at bedtime and washed off 6–10 h later. Reported genital symptoms were mild and resolved by the end of the study, with no difference observed in the circumcised vs. uncircumcised group. The authors concluded that men exposed to tenofovir gel during unprotected intercourse with gel users would be unlikely to experience significant genital or systemic toxicities.

A similar study [45] was conducted with candidate vaginal gel SPL7013 (VivaGel; Starpharma Holdings Ltd., Melbourne, Victoria, Australia) to determine the safety of the gel among uncircumcised and circumcised men. The results suggested that SPL7013 was well tolerated [45].

These findings are important, as women prefer to inform their primary partner about gel use; this is in contrast to the previous belief that the ability to use microbicides covertly was important. However, a study [46] in the USA reported that women in the trial preferred products that could not be noticed by the primary partner, but that added lubrication and provided sexual pleasure without generating opposition from partners. The value placed upon the possibility of covert use is likely to be context and location specific.

Rectal microbicides

Heterosexual anal sex is widely practiced among couples in Africa and elsewhere. The use of N9-containing lubricants as an anti-HIV measure among men who have sex with men was noted before it was tested for the same purpose among women in clinical trials [47]. It is generally assumed that if effective, microbicides would be used for vaginal and rectal application by both men and women.

Research on rectal microbicides has gained momentum in recent years. Vaginal candidate UC781 (Conrad, Arlington, Virginia, USA), an NNRTI, was tested for acceptability among HIV-1-uninfected adults [48[•]]. The phase I blinded, placebo-controlled trial enrolled 10 women and 26 men into three groups of 12, randomized to receive 0.1% UC781 gel, 0.25% UC781 gel or a placebo gel (hydroxyethyl cellulose). The gels were similar in volume and to touch, but had variable viscosity and colour. Gels were

administered in a single dose after a preparatory enema. Participant product acceptability questionnaires were administered via web-based ACASI. Overall 56% of the participants liked the gel (irrespective of which gel group), whereas the rest of the participants felt neutral. Formulation-associated factors such as colour, smell, taste and consistency were well accepted. Application-associated factors such as the process of applying the product were neutrally rated; on the contrary, the applicator was not well liked.

Given that safety of the microbicide for vaginal or rectal use is of the utmost importance, Patton *et al.* [49] developed a standardized protocol for preclinical rectal safety and (chlamydial) efficacy assessment of topical microbicide candidates in a nonhuman primate model. Their study suggests that product development should continue if the candidate meets the following safety criteria in macaques: rectal microflora remained stable, pH remained stable and changed only transiently and there was no evidence of gross tissue damage. Reformulation was recommended if any of the following were observed: presence of grossly identifiable blood/red blood cells, epithelial sheets of at least 3 mm in dimension in quantities greater than those observed in the control arm, resistant changes in pH, significant decrease in H₂O₂-producing microorganisms, increase in pathogenic organisms, suppression of microflora or all.

Of the 12 products that underwent safety evaluation, two formulations of one product showed severe negative effects on the rectal environment (combination formulations of SPL7013 and BufferGel). The authors caution against extrapolating rectal safety evaluations to the vagina, as these are anatomically and histologically distinct. They point out that the fragile nature of the rectal compartment means it is likely to be more vulnerable to irritation, sloughing and infection. Preclinical evaluation in an animal model such as theirs can provide significantly more detailed information and early warning of products that may have harmful effects, assisting us to steer more promising products into clinical trials [49].

Conclusion

For the past 12–18 months, publications on microbicides have primarily focused on results of clinical trials and investigations into preclinical, methodological and social factors that may have contributed to disappointing outcomes. From lessons learned in past trials, the field is beginning to unpack the complexity of microbicide research. In this way, it is becoming increasingly equipped to overcome the existing challenges, with renewed commitment, hope and determination to find a women-initiated HIV prevention option.

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There are no conflicts of interest.

References and recommended reading

Papers of particular interest, published within the annual period of review, have been highlighted as:

- of special interest
- of outstanding interest

Additional references related to this topic can also be found in the Current World Literature section in this issue (pp. 352–353).

- 1 Elias C, Coggins C. Female-controlled methods to prevent sexual transmission of HIV. *AIDS* 1996; 10:S43–S51.
- 2 Elias CJ, Heise LL. Challenges for the development of female-controlled vaginal microbicides. *AIDS* 1994; 8:1–10.
- 3 Stone A, Hitchcock P. Vaginal microbicides for preventing the sexual transmission of HIV. *AIDS* 1994; 8 (Suppl 1):S285–S293.
- 4 Balzarini J, Van Damme L. Microbicide drug candidates to prevent HIV infection. *Lancet* 2007; 369:787–797.
- 5 Kreiss J, Ngugi E, Holmes K, *et al.* Efficacy of Nonoxynol 9 contraceptive sponge use in preventing heterosexual acquisition of HIV in Nairobi prostitutes. *JAMA* 1992; 268:477–482.
- 6 Van Damme L, Ramjee G, Alary M, *et al.* Effectiveness of COL-1492, a nonoxynol-9 vaginal gel, on HIV-1 transmission in female sex workers: a randomised controlled trial. *Lancet* 2002; 360:971–977.
- 7 Roddy RE, Zekeng L, Ryan KA, *et al.* A controlled trial of nonoxynol 9 film to reduce male-to-female transmission of sexually transmitted diseases. *N Engl J Med* 1998; 339:504–510.
- 8 Peterson L, Nanda K, Opoku BK, *et al.* SAVVY (C31G) gel for prevention of HIV infection in women: a phase 3, double-blind, randomized, placebo-controlled trial in Ghana. *PLoS One* 2007; 2:e1312.
- 9 Feldblum P, Adeiga A, Bakare R, *et al.* SAVVY vaginal gel (C31G) for prevention of HIV infection: a randomized controlled trial in Nigeria. *PLoS One* 2008; 3:e1471.
- 10 Halpern V, Ogunsola F, Obunge O, *et al.* Effectiveness of cellulose sulfate vaginal gel for the prevention of HIV infection: results of a phase III trial in Nigeria. *PLoS One* 2008; 3:e3784.
- 11 Van Damme L, Govinden R, Mirembe F, *et al.* Lack of effectiveness of cellulose sulfate gel for the prevention of vaginal HIV transmission. *N Engl J Med* 2008; 359:463–472.
- 12 Skoler-Karhoff S, Ramjee G, Ahmed K, *et al.* Efficacy of carraguard for prevention of HIV infection in women in South Africa: a randomised, double-blind, placebo-controlled trial. *Lancet* 2008; 372:1977–1987.
- 13 Nunn A, McCormack S, Crook A, *et al.* Microbicides Development Programme: design of a phase III trial to measure the efficacy of the vaginal microbicide PRO 2000/5 for HIV prevention. *Trials* 2009; 10:99.
- 14 Abdool Karim S, Coletti A, Richardson B, *et al.* Safety and effectiveness of vaginal microbicides BufferGel and 0.5% PRO 2000 gel for the prevention of HIV infection in women: results of the HPTN 035 trial [abstract #48LB]. In: 16th Conference on Retroviruses and Opportunistic Infections; 8–11 February 2009; Montreal, Canada; 2009.
- This abstract was the first publication to provide some proof of concept for microbicides. PRO 2000 at 0.5% appeared to reduce the rate of HIV acquisition by 30% as compared with the control gel, although this was not statistically significant.
- 15 HIV 'prevention' gel PRO 2000 proven ineffective. <http://www.mdp.mrc.ac.uk/archive.html>.
- 16 Sonza S, Johnson A, Tyssen D, *et al.* Enhancement of human immunodeficiency virus type 1 replication is not intrinsic to all polyanion-based microbicides. *Antimicrob Agents Chemother* 2009; 53:3565–3568.
- Some recent publications have claimed that polyanion microbicides enhance HIV infection; Sonza *et al.* [16] show that this effect may be due to assay conditions, and is not an intrinsic property.
- 17 Mbopi-Keou F-X, Trottier S, Omar RF, *et al.* A randomized, double-blind, placebo-controlled phase II extended safety study of two Invisible Condom formulations in Cameroonian women. *Contraception* 2010; 81:79–85.
- 18 von Mollendorf CE, Van Damme L, Moyes JA, *et al.* Results of a safety and feasibility study of the diaphragm used with ACIDFORM Gel or K-Y Jelly. *Contraception* 2010; 81:232–239.

- 19 Ramjee G, van der Straten A, Chipato T, *et al.* The diaphragm and lubricant gel for prevention of cervical sexually transmitted infections: results of a randomized controlled trial. *PLoS One* 2008; 3:e3488.
- 20 Keller MJ, Mesquita PMM, Torres NM, *et al.* Postcoital bioavailability and ●● antiviral activity of 0.5% PRO 2000 gel: implications for future microbicide clinical trials. *PLoS One* 2010; 5:e8781.
- Keller *et al.* [20**] provide important evidence for the influence of coitus on microbicide activity. Coital activity appears to reduce the effectiveness of PRO 2000; this has important implications for the formulation and testing of future microbicides. New formulations should retain anti-HIV activity in the presence of semen.
- 21 Kyeong-Ae K, Jiang S, Kirchoff F, Muench J. SEVI and semen impair the anti- ●● HIV-1 activity of drugs and microbicides. *Antiviral Res* 2009; 82:A64–A164. This abstract provides further support for the influence of semen on the antiviral activity of microbicides.
- 22 Münch J, Rücker E, Ständker L, *et al.* Semen-derived amyloid fibrils drastically enhance HIV infection. *Cell* 2007; 131:1059–1071.
- 23 Roan NR, Munch J, Arhel N, *et al.* The cationic properties of SEVI underlie its ●● ability to enhance human immunodeficiency virus infection. *J Virol* 2009; 83:73–80.
- Roan *et al.* [23**]'s investigations provide data supporting the development of combination microbicides, as at least some anionic polymers are capable of blocking the activity of SEVI.
- 24 Mesquita PMM, Cheshenko N, Wilson SS, *et al.* Disruption of tight junctions ●● by cellulose sulfate facilitates HIV infection: model of microbicide safety. *J Infect Dis* 2009; 200:599–608.
- A very interesting paper that outlines an important method that could be used in preclinical assessment of microbicides. The effects of some microbicides, which have previously failed to reduce HIV infection in clinical trials on transepithelial resistance of cell cultures, are described.
- 25 Mâsse B, Boily M-C, Dimitrov D, Desai K. Efficacy dilution in randomized ●● placebo-controlled vaginal microbicide trials. *Emerg Themes Epidemiol* 2009; 6:5.
- Mâsse *et al.* [25**] outline the dilution factors that researchers have to consider in the design of clinical trials. Dilution factors could mask the effectiveness of microbicial products, making an effective product appear less so.
- 26 Norris Turner A, De Kock A, Meehan-Ritter A, *et al.* Many vaginal micro- ●● bicide trial participants acknowledged they had misreported sensitive sexual behavior in face-to-face interviews. *J Clin Epidemiol* 2009; 62: 759–765.
- Many microbicide clinical trials rely on self-reports of adherence and sexual behaviour. These authors used ACASI to evaluate the difference in face-to-face vs. computer-assisted reports. ACASI could be an important tool for the assessment of adherence in microbicide trials, as participants may be more comfortable using a computer than responding to interviewer questions.
- 27 Tolley EE, Harrison PF, Goetghebeur E, *et al.* Adherence and its measurement ●● in phase 2/3 microbicide trials. *AIDS Behav* 2009. DOI: 10.1007/s10461-009-9635-x. [Epub ahead of print]
- This review provides a summary of possible mechanisms for measuring adherence and gives a set of recommendations.
- 28 Rohan LC, Sassi AB. Vaginal drug delivery systems for HIV prevention. *AAPS J* 2009; 11:78–87.
- 29 Jackson JB, Musoke P, Fleming T, *et al.* Intrapartum and neonatal single-dose nevirapine compared with zidovudine for prevention of mother-to-child transmission of HIV-1 in Kampala, Uganda: 18-month follow-up of the HIVNET 012 randomised trial. *Lancet* 2003; 362:859–868.
- 30 Garcia-Lerma JG, Paxton L, Kilmarx PH, Heneine W. Oral preexposure ●● prophylaxis for HIV prevention. *Trends Pharmacol Sci* 2010; 31:74–81.
- A useful summary of preclinical PrEP data; the study also provides details of PrEP trials currently in progress around the world.
- 31 Garcia-Lerma JG, Otten RA, Qari SH, *et al.* Prevention of rectal SHIV transmission in macaques by daily or intermittent prophylaxis with emtricitabine and tenofovir. *PLoS Med* 2008; 5:e28.
- 32 Denton PW, Estes JD, Sun Z, *et al.* Antiretroviral preexposure prophylaxis prevents vaginal transmission of HIV-1 in humanized BLT mice. *PLoS Med* 2008; 5:e16.
- 33 Microbicide and PrEP candidates in ongoing clinical trials. <http://www.avac.org/ht/a/GetDocumentAction/i/3109>.
- 34 Nel A, Smythe S, Young K, *et al.* Safety and pharmacokinetics of dapivirine ●● delivery from matrix and reservoir intravaginal rings to HIV-negative women. *J Acquir Immune Defic Syndr* 2009; 51:416–423.
- IVRs are a novel form of microbicide delivery; evidence reported here suggests that IVRs may be an important delivery vehicle for microbicides or ARVs in the future.
- 35 Garg A, Nuttall J, Romano J. The future of HIV microbicides: challenges and opportunities. *Antivir Chem Chemother* 2009; 19:143–150.
- 36 Nel AM, Coplan P, van de Wijgert JH, *et al.* Safety, tolerability, and systemic absorption of dapivirine vaginal microbicide gel in healthy, HIV-negative women. *AIDS* 2009; 23:1531–1538.
- 37 Mayer KH, Maslankowski LA, Gai F, *et al.* Safety and tolerability of tenofovir vaginal gel in abstinent and sexually active HIV-infected and uninfected women. *AIDS* 2006; 20:543–551.
- 38 Schwartz JL, Kovalevsky G, Lai J-J, *et al.* A randomized six-day safety study of an antiretroviral microbicide candidate UC781, a nonnucleoside reverse transcriptase inhibitor. *Sex Transm Dis* 2008; 35:414–419.
- 39 Wilson DP, Coplan PM, Wainberg MA, Blower SM. The paradoxical effects of using antiretroviral-based microbicides to control HIV epidemics. *Proc Natl Acad Sci U S A* 2008; 105:9835–9840.
- 40 Surveillance of transmitted HIV drug resistance. <http://www.who.int/hiv/topics/drugresistance/surveillance/en/print.html>.
- 41 Brenner BG, Oliveira M, Doualla-Bell F, *et al.* HIV-1 subtype C viruses rapidly develop K65R resistance to tenofovir in cell culture. *AIDS* 2006; 20:F9–F13.
- 42 Martinez J, Coplan P, Wainberg MA. Is HIV drug resistance a limiting factor in the development of anti-HIV NNRTI and NRTI-based vaginal microbicide strategies? *Antiviral Res* 2006; 71:343–350.
- 43 Xu H-T, Martinez-Cajas J, Ntemgwa M, *et al.* Effects of the K65R and K65R/ ●● M184 V reverse transcriptase mutations in subtype C HIV on enzyme function and drug resistance. *Retrovirology* 2009; 6:14.
- Xu *et al.* [43*] show that K65R reverse transcriptase mutations in subtype C viruses behave in a similar manner to those in subtype B HIV.
- 44 Schwartz JL, Poindexter A, Wheelless A, *et al.* Safety evaluation of 1% ●● tenofovir gel in healthy men. *Int J STD AIDS* 2009; 20:384–386.
- The new generation of ARV-containing vaginal microbicides must be shown to be safe to male sexual partners. Schwartz *et al.* [44*] have shown that 1% tenofovir gel was not toxic when applied to the penis overnight.
- 45 Chen MY, Millwood IY, Wand H, *et al.* A randomized controlled trial of the safety of candidate microbicide SPL7013 gel when applied to the penis. *J Acquir Immune Defic Syndr* 2009; 50:375–380.
- 46 Hoffman S, Morrow KM, Mantell JE, *et al.* Covert use, vaginal lubrication, and sexual pleasure: a qualitative study of urban U.S. women in a vaginal microbicide clinical trial. *Arch Sex Behav* 2010; 39:748–760.
- 47 Gross M, Buchbinder SP, Celum C, *et al.* Rectal microbicides for U.S. gay men: are clinical trials needed? Are they feasible? *Sex Transm Dis* 1998; 25:296–302.
- 48 Ventuneac A, Carballo-Diequez A, McGowan I, *et al.* Acceptability of UC781 ●● gel as a rectal microbicide among HIV-uninfected women and men. *AIDS Behav* 2009. DOI: 10.1007/s10461-009-9611-5. [Epub ahead of print]
- Research on the applicability of microbicides to the rectal compartment is an important new direction, particularly as it is suspected that anal intercourse is an underreported risk factor for HIV acquisition in vaginal microbicide trials.
- 49 Patton DL, Sweeney YT, Paul KJ. A summary of preclinical topical microbicide rectal safety and efficacy evaluations in a pigtailed macaque model. *Sex Transm Dis* 2009; 36:350–356.