Editorial

Clinical safety issues in developing & testing of vaginal microbicides

Research on vaginal microbicides that could protect against sexual transmission of HIV and other pathogens is a priority and nearly 60 products are being evaluated as candidate microbicides worldwide. The products will be available in the market only after establishment of their safety and efficacy. International regulatory issues in pre-clinical and clinical microbicide development have been described previously. Serious public health concern was raised when COL-1492 containing nonoxynol-9 (marketed as a contraceptive for more than 30 yr worldwide) was found to be associated with increased risk of HIV-1 transmission in the phase III clinical trial in commercial sex workers. Therefore, identifying methods for screening out products in the pre-clinical stages of development and measuring their safety in clinical trials are important research challenges in this field.

Pre-clinical studies are done to ensure that the estimated risk/benefit profile of any new pharmaceutical product is reasonably in favour of the indicated purpose. Antiviral activity of microbicides may be due to disruption of the virus, blocking of viral attachment and/or membrane fusion, or inhibition of some other step in the virus life cycle such as reverse transcriptase or polyprotein processing (Fig.). Early steps in the pre-clinical development of microbicides include evaluation of their anti-HIV activity, cytotoxicity, mechanism of action, pathways to resistance, cross-resistance to approved drugs and effects on vaginal microflora and pathogens that cause sexually transmitted diseases (STDs).

Products screened through pre-clinical testing procedures enter phase I, II and III clinical trials for evaluation of their safety and efficacy in humans. Local and systemic safety assessment is the main objective of phase I trials; phase II trials evaluate expanded safety; and in phase IIb and III studies, efficacy and long term safety are assessed.

Phase I studies involve the introduction of investigational products in healthy human volunteers for the first time and hence include rigorous product safety evaluation. Local safety of a product is assessed by documenting self-reported symptoms associated with its use and clinical/colposcopic examination to observe changes in vaginal mucosa. Colposcopy was first introduced by Hans Hinselmann in Germany in 1950 for visualization of the uterine cervix for the early diagnosis of cancer. In the late 1980s and early 1990s, colposcopy was introduced to look for vaginal and cervical changes with the use of vaginal products such as spermicides and vaginal rings. The World Health Organization (WHO) published a manual for colposcopy for vaginally administered products in 1995. It was revised by the Contraceptive and Research Development (CONRAD) and WHO in 2000. The most recent version of the manual titled "Manual for the Standardization of Colposcopy for the Evaluation of Vaginal Products, Update 2004" is available on the websites of CONRAD and WHO (www.conrad.org and www.who.int/reproductive-health). Baseline colposcopy is recommended to decide on the eligibility of the participants. Macroscopic as well as microscopic inflammation of the female reproductive tract mucosa increases susceptibility to HIV and other viral infections. Colposcopic examination during or after vaginal product use aims at evaluating epithelial disruption, sub-epithelial...
Colposcopy has limitations and it is possible that all significant epithelial changes may not be detected visually. Therefore, laboratory-based methods are employed to measure the levels of proinflammatory and anti-inflammatory factors which are present in a state of physiological equilibrium in healthy women. Cervicovaginal lavage (CVL) is a non-invasive procedure for evaluating inflammatory biomarkers in clinical trials of microbicides. Proinflammatory cytokines (interleukin IL-1, IL-6, and tumour necrosis factor, TNF-α) are released in response to vaginal irritation/inflammation and play a critical role in HIV-1 pathogenesis. These cytokines recruit additional immune target cells into the area and may enhance HIV replication. Release of proinflammatory cytokines during the vaginal product use is considered suggestive of immunoinflammatory responses and may help to screen out the product. For example, repeated nonoxynol-9 exposure was shown to promote HIV-1 transmission through interleukin-1–mediated NF-kB activation, leading to chemokine-induced recruitment of HIV-1 host cells and increased HIV-1 replication in the infected cells.

Evidence of microhaemorrhage in the CVL has recently been described to be a marker for detecting disruption in the cervico-vaginal epithelium. Vaginal biopsy may be included in early phase I studies in sexually abstinent population to detect the inflammation of the genital tract. The inherent limitations of biopsy include this being an invasive procedure, limited feasibility, possibility of sampling error, infection and haemorrhage. These should be weighed against the potential benefits before taking a decision to incorporate biopsy in any safety assessment protocol.

Since microbicides are expected to act in a complex environment and an acidic pH is required for inactivation of HIV and other sexually transmitted pathogens, the effects of vaginal microbicides on the vaginal pH and microflora need to be closely monitored. Appropriate laboratory tests including complete blood cell count, coagulation profile and liver and renal function tests, before and after product use help in evaluating systemic safety of vaginal microbicides. Systemic absorption can be monitored by performing pharmacokinetic studies for the blood levels of the product or its active metabolite.

Some microbicides might not have contraceptive properties so that women and couples using such products can protect themselves against sexually transmitted infections (STIs) and yet have children.
It is essential to assess the incidence of pregnancy and the effect of new microbicide on the foetus. In phase I trials, product application is for shorter durations (up to 14 inter-menstrual days) and condom adherence is usually good. However, in long term phase III efficacy trials, incidence of pregnancy and possible effects on the foetus and the newborn must be closely monitored.

Though phase I studies are conducted among women having normal Papanicolau (Pap) smears, phase III efficacy trials have to be conducted in communities with high-risk behaviour and high HIV incidence. In developing countries, Pap smear screening may not be routinely available as a part of primary health care. It is important to obtain data on the safety of vaginal microbicide among women having abnormal Pap smears as well. Visual inspection of the cervix after application of acetic acid\textsuperscript{16,17} is being considered as an alternative approach for diagnosing cervical abnormalities as an easy and practical alternative where Pap smear facilities are not available for screening women before they start long-term use of the product.

Usually in phase I trials, participants who are not at risk or only at low risk of STIs, are enrolled. Participants having any STI at screening can be treated and then enrolled in the study. Those who are diagnosed with an STI while using the product should be treated appropriately and continued on the study after the infection is resolved. Therefore STIs should be closely monitored during the clinical studies and laboratory support for the diagnosis of incident STIs should be available. Presence of a vaginal product can interfere with the sensitivity of assays used to detect STIs. It is therefore necessary to select assays which are not altered by the product under evaluation.

In order to get adequate safety and efficacy data, it is necessary that participants adhere to the product regimen and also attend their regular follow up visits. Informed consent procedure and ongoing counselling should adequately stress the need for an informed decision to participate and adhere to both product use as well as the follow up schedule.

In men with heterosexual and homosexual orientation per act relative risk of HIV transmission is 100 times more with receptive anal sex compared to insertive fellatio\textsuperscript{18}. HIV is significantly more easily transmitted to a receptive partner and rectal microbicides could offer protection during anal intercourse. The pig-tailed macaque rectal infection model has been shown to be useful in evaluation of newly developed topical microbicides for rectal use\textsuperscript{19}. Studies are being carried out to assess the baseline levels of injury and inflammation that can occur in the rectum during typical anal intercourse with and without product usage. Parameters of safety assessment of microbicides for rectal use are different than those for vaginal use, and are still being evolved. Bleeding, inflammation, ulceration, erosion, sloughing and drug absorption are major co-ordinates to evaluate rectal safety, and presenting symptoms might include itching, fullness, bloating, numbness and diarrhoea\textsuperscript{20}.

Although condom promotion is an integral part of any HIV prevention clinical trial including vaginal/rectal microbicides research, it is likely that condoms may not be used during every sex act and the penis might get exposed to the microbicide under investigation. Therefore, ‘male tolerance’ trials need to be carried out to study whether potential microbicides cause any irritation of the penis or any injury to male urethra before they are taken in large-scale efficacy trials.

To conclude, safety should be carefully monitored during the pre-clinical as well as at various stages of clinical trials in humans and also during post-marketing surveillance. Regulatory authorities, ethicists, individuals, general population and programme managers should be convinced of the safety of the microbicides for broad range community-wide acceptance. Safety assessments are continuously undergoing revisions with the ultimate objective of having a ‘safe’ product especially intended for long term use for HIV prevention.

Smita Joshi & Sanjay Mehandale*
National AIDS Research Institute (ICMR)
G-73, MIDC, Bhosari
Pune 4110026, India
*For correspondence
e-mail : smehendale@nariindia.org
References


4. Stephenson J. Widely used spermicide may increase, not decrease, risk of HIV transmission. JAMA 2000; 284: 949.


