

*Review*

# New ways of preventing HIV infection: thinking simply, simply thinking

R. V. Short\*

*Department of Obstetrics and Gynaecology, University of Melbourne, 132 Grattan Street, Carlton Vic. 3053, Australia*

HIV infection is the greatest health crisis in human history. It continues to spread unchecked among the poor in the developing world because we have failed to design simple preventative methods that are available and affordable to those living on under \$2 a day. Five new methods are discussed. (i) *A natural microbicide*. Intravaginal lime or lemon juice has been used for centuries as a traditional contraceptive. The juice can also kill HIV in the laboratory, but clinical trials are needed to see if vaginal application is acceptable, safe and effective. (ii) *Intravaginal oestrogen*. Monkeys can be protected from Simian immunodeficiency virus (SIV) infection by keratinizing the vagina with topical oestrogen. If women take the oral contraceptive pill vaginally it retains its contraceptive efficacy, and the oestrogen it contains should thicken the vagina and protect against HIV infection. Clinical trials are needed. (iii) *Male circumcision*. Removal of the inner foreskin removes the main site of HIV entry into the penis, resulting in a sevenfold reduction in susceptibility to infection. The practice needs to be promoted. (iv) *Post-coital penile hygiene*. Wiping the penis immediately after intercourse with lime or lemon juice or vinegar should kill the virus before it has had a chance to infect. A clinical trial of efficacy is needed. (v) *PhotoVoice*. Asking schoolchildren in developing countries to photograph their impressions of HIV/AIDS is a powerful way of getting them to discuss the subject openly, and develop their own preventative strategies.

**Keywords:** HIV/AIDS prevention; natural microbicides; lemon and lime juice; topical vaginal oestrogen; male circumcision; post-coital penile hygiene

## 1. INTRODUCTION

The global AIDS pandemic gives increasing cause for concern. A recent consensus conference of the world's leading economists, including three Nobel laureates, concluded that HIV/AIDS was the greatest problem facing the world (Lomborg 2004). Controlling HIV/AIDS is also listed as one of the United Nations' Millennium Development Goals (Sachs 2005). Currently, there are 35–42 million adults and children living with the infection, with around 5 million new infections and 3 million deaths a year (UNAIDS 2004). Sub-Saharan Africa is the epicentre of the pandemic, with 25 million currently infected and 3 million new infections annually. Next comes South and South-East Asia, with 6.5 million current infections. There is no doubt that the pandemic is still in its exponential growth phase, and that the situation will inevitably get much worse.

But in spite of these grim statistics, there is reason for hope; relatively simple interventions could bring the infection under control, even though we may never be able to eradicate the disease completely. The virus is a fragile one, with low infectivity. Over 80% of infections occur during heterosexual intercourse, but the relative reproductive rate,  $R_0$ , is only slightly in excess of 1 in most populations (May *et al.* 2001). For

the infection to spread,  $R_0$  must be greater than one; each infected individual must infect more than one other person on average. The probability of transmission per act of heterosexual intercourse between otherwise healthy individuals may be as low as 1 in 1000 (Royce *et al.* 1997), although it can be much higher if other sexually transmitted diseases (STD) are present, or in the early days post-infection when viral titres in the ejaculate are at their highest (Pilcher *et al.* 2004). Behavioural changes, such as reducing the number of sexual partners, and reducing the age difference between partners, could greatly reduce the rate of transmission. But there is one major obstacle; of all aspects of human behaviour, it is our sexual behaviour that is least amenable to change. By parasitizing our sexuality, the virus has ensured its propagation.

The most widely advocated programme of prevention which is acceptable, at least in part, to most of the world's political and religious leaders is the ABC approach—Abstinence, Be Faithful, and Condoms. But each component has its inherent weaknesses:

*Abstinence:* 'Tis a consummation devoutly to be wished. In practice, promoting abstinence does not work.

*Be Faithful:* Alas, we are not by nature a monogamous species. If we were, then STD would not exist.

\*r.short@unimelb.edu.au

*Condoms:* We can never rely on male compliance to control a disease that is increasingly female-orientated.

The old adage that prevention is better than cure is never more true than in the case of HIV/AIDS, where there is no cure in sight. The advent of antiretroviral drugs can now offer significant improvements in the quality of life and increase in life expectancy to those who are infected. Even if they only doubled life expectancy, they would be a priceless gift. But the drugs are costly, the daily treatment regimen may be difficult to comply with, there can be serious side effects, and ultimately the high mutation rate of the virus ensures that resistance is inevitable. These drugs are emphatically not a cure; you will probably still die of an AIDS-related disease. However, as treatments improve and become more widely available, more people are likely to volunteer for HIV testing, and this alone will greatly help HIV prevention.

The problem with antiretroviral drugs is that the virus can mutate far faster than the pharmaceutical companies can develop new therapies, so the virus will always be ahead of the game. This high viral mutation rate also bedevils attempts to produce an anti-HIV vaccine, which is not yet on the horizon, and may never be. After all, we still do not have a vaccine against malaria, after many more years of research. And we have recently uncovered a nasty twist to HIV. If somebody becomes infected with two different strains of the virus, this greatly accelerates disease progression, and the two strains may even recombine to form a new, resistant strain (Gottlieb *et al.* 2004).

So almost a quarter of a century after the first recognition of AIDS, the infection continues to run rampant throughout the globe. We have spent billions of dollars on the development of treatments and the search for vaccines, and we understand the genetic makeup of the virus in minute detail. But we have ignored the development of simple new preventative measures. The condom was first developed in Italy in the sixteenth century as a defence against the syphilis pandemic then sweeping the country, syphilis being a Greek word meaning ‘companion of love’ (Potts & Short 1999). Surely we should not still be relying on sixteenth century technology as our sole defence against HIV infection?

We need a new ethic to stem the global spread of HIV infection. We must give the highest priority to the development of simple preventative methods that are designed first and foremost for women, and especially for the poor living in the developing world. These methods must be acceptable, available and affordable for that quarter of the world’s population who are living on under two dollars a day (Potts & Short 2003).

In order to do this, we need to go back to first principles and ask ourselves four very simple questions. How does the virus enter the female reproductive tract, and how might a woman’s natural resistance be increased? How does the virus enter the male reproductive tract, and how might a man’s natural resistance be increased?

Let us examine five possible new approaches to HIV prevention that address these questions.

## 2. LEMON OR LIME JUICE: A NATURAL VAGINAL MICROBICIDE?

The principal route of entry of HIV into the female reproductive tract appears to be via the Langerhans cells and other dendritic cells in the vaginal epithelium. The vagina is well endowed with Langerhans cells, and seems to be a far more important route for HIV infection than the cervix or uterus (Li & Short 2002; Miller & Shattock 2003). It therefore makes sense to try and kill the virus before it can become attached to the specific HIV receptors in the vaginal epithelium.

There is general agreement that we urgently need to develop new ways of protecting women from HIV infection that do not depend on the compliance of their male partners (Pilcher 2004), and the American-based Microbicides Alliance, with over 100 million dollars at its disposal, is going down the conventional route of drug discovery, patent protection and pharmaceutical company development to test more than 40 new compounds as potential vaginal microbicides (Coplan *et al.* 2004). But the Microbicides Alliance is faced with a major hurdle to overcome before any of their compounds can be licensed for use. The American Food and Drug Administration (FDA) has insisted that efficacy needs to be tested in a 3-arm clinical trial, comparing the active product with a placebo and with condoms only. These trials will probably have to be done outside the United States, in countries with high rates of female HIV infection, and it is estimated that 5600 volunteers would be needed per trial to demonstrate superior efficacy for one female microbicide with 50% efficacy relative to the placebo group. None of the new compounds under development can meet these stringent requirements before 2010 at the earliest.

In the meantime, it is possible that we could take a short-cut by studying traditional compounds that women have been using intra-vaginally in an attempt to regulate their fertility. We have opted for this approach, studying a natural contraceptive that women have been using for centuries—lemon juice. Norman Himes (1963) has given a comprehensive review of the history of contraceptive development throughout the world. He describes how Casanova advocated the use of half a lemon, partially squeezed out, as a cervical cap to protect women from pregnancy—the forerunner of today’s cervical caps. And he also describes how Turkish women in the eighteenth century regularly used a piece of sponge soaked in dilute lemon juice and placed in the vagina just before coitus as a contraceptive. He concludes that this practice ‘is theoretically not surpassed in reliability by any modern clinical contraceptive’.

It has long been known that dilute lemon juice is a very effective spermicide, because of the low pH produced by its citric acid content (Himes 1963). Since the human immunodeficiency virus is also extremely susceptible to low pH (Martin *et al.* 1985), we were excited by the possibility that this age-old contraceptive might be a new natural microbicide.

We have been able to confirm the spermicidal effect of both lemon and lime juice (Clarke *et al.* in press). Fresh lime juice, with a pH of 2.1, added to a fresh human ejaculate (pH 8.4) to give a final concentration of 20% lime juice in the mixture reduced the pH to 4.1.

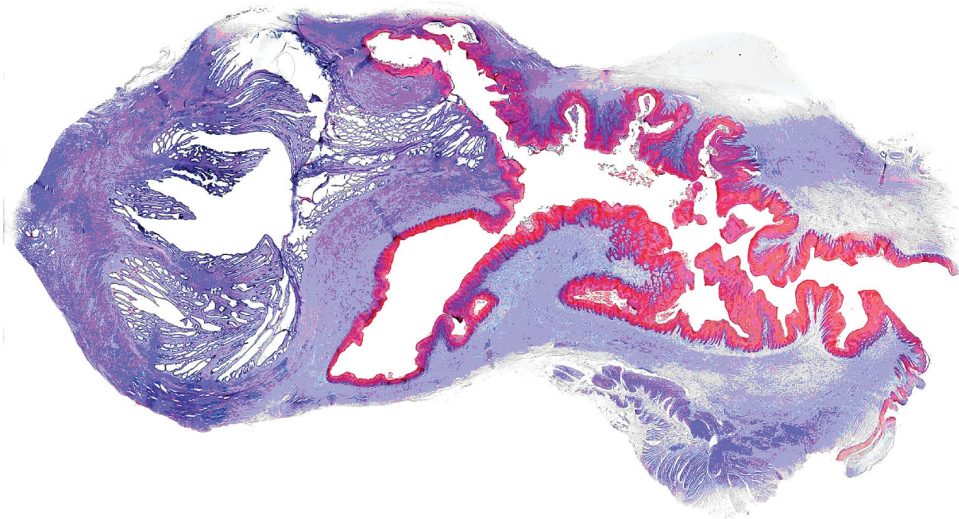


Figure 1. Histological appearance of the cervix and vagina of a macaque monkey exposed to daily intravaginal lime juice for a month. The cervical and vaginal epithelia appear normal. Mallory's Stain.

This produced 100% irreversible immotility of all sperm within 30 s, probably by inactivating the dynein ATPase in the sperm midpiece (Burgess *et al.* 2003). A 10% lime juice mixture, with a pH of 5.1, left 1% of sperm still motile after 5 min, although they could recover their motility if the pH of the mixture was subsequently returned to neutrality several hours later. So it is easy to see why intravaginal lime or lemon juice is such a very effective contraceptive. Since improved access to Family Planning is seen as one of the keys to poverty reduction in the developing world in the recently announced Millennium Development Goals (Sachs 2005), this simple contraceptive deserves to be much more widely known.

We have gone on to study the virucidal effects of lemon and lime juice (Short *et al.* 2004). Using filtered pools of lemon juice (pH 2.3) and lime juice (pH 2.4), we added various volumes to cultures of HIV-BaL grown in the laboratory. The presence of live virus was determined by its ability to infect peripheral human blood mononuclear cells cultured for 14 days. Viral replication was measured by viral reverse transcriptase activity in culture supernatants. The culture medium had little buffering capacity, so a 20% concentration of lime or lemon juice reduced the pH to 2.9, inactivating 90% of the virus in 2 min; a 10% concentration (pH 3.4–3.7) inactivated up to 50% in 2 min. We are currently studying the time course of inactivation when fresh human seminal plasma is 'spiked' with HIV and exposed to different concentrations of lemon juice. In order to maximize the virucidal effect, it will be necessary to lower the pH of the ejaculate to below 4. A pH of 4 is also known to kill other STD in culture, such as *HSV-2*, *Neisseria gonorrhoeae*, *Treponema pallidum* and *Chlamydia*, while leaving *Lactobacillus acidophilus* unharmed. Since the mean ejaculate volume of fertile men is 3.7 ml, at least 3 ml of juice needs to be present in the vagina if the virus in an ejaculate is to be killed quickly.

Would undiluted citrus juice placed in the vagina before sex be painful to either partner? A simple clinical trial was carried out in Mexico City in 2004 (Gutierrez *et al.* 2005, unpublished work). At the end of a public

lecture on HIV/AIDS the audience was asked to volunteer to self-administer fresh lime juice to the penis or vagina and vulva and to score the degree of discomfort on a scale from 0 to 10 in an anonymous questionnaire. By way of comparison, they were asked to rank the pain score when the juice was applied to the tip of the tongue. They were also asked to record sex, age, and number of prior sexual partners, and men were asked to record their circumcision status. Sixty one men and 64 women took part in the study. The results showed that the median pain score on application of neat lime juice to the penis or vagina was 0, no different from the pain when applied to the tip of the tongue. Male circumcision was without effect, and there was no effect of age, or number of prior sexual partners. A small number of women did experience moderate discomfort, apparently associated with the onset of menstruation, but in no case was it severe. This is a convincing demonstration of the potential acceptability of intravaginal citrus juice to both partners.

How safe would it be to repeatedly administer neat citrus juice to the vagina? We undertook a clinical trial in 12 cynomolgus monkeys (*Macaca fascicularis*), six treated and six controls, each housed in isolation. The treated animals had a small cotton wool ball soaked in about 0.8 ml fresh, undiluted lime juice placed in the vagina daily for 28 days, starting on the first day of menstruation. The animals were killed at the end of the treatment and the reproductive tracts fixed in 10% formol saline. The six control animals were not killed, but had vaginal biopsies taken on the first day of menstruation. The vaginal epithelium of the treated animals did not differ histologically from the control animals, and the cervical epithelium appeared normal (see figure 1). Both treated and control animals showed occasional foci of sub-epithelial vaginal infection. This is a common finding in this species (Miller & Shattock 2003), and was unrelated to treatment.

These results, although extremely encouraging, might at first sight seem rather surprising. Why would not the repeated vaginal administration of a highly acidic solution, pH around 2.3, with no male ejaculate

to neutralize it, produce some histological reaction in the epithelium? Perhaps it is because the vagina is normally acidic. Human vaginal pH changes during the course of the menstrual cycle, falling to 4.2 at the time of ovulation (Wagner & Ottesen 1982). Citric acid, which is one of the body's essential intermediary metabolites, is also the major chemical constituent of human semen, being secreted by the prostate (Mann 1964), so the female reproductive tract is well adapted to it. Nevertheless, human clinical trials are still needed to assess the long-term safety of repeated administration of pharmacological doses of citric acid, in the form of lemon or lime juice, to the human vagina.

But perhaps these trials have already been done. In Jos, Nigeria over 80% of a group of 200 female commercial sex workers routinely douche with lime or lemon juice immediately before or after sex, in the firm belief that this protects them from pregnancy, and from STD; they do not know their HIV status (Imade *et al.* 2005). Some women have been doing this for 10 years. By carrying out a prospective trial of these women we can determine the safety and efficacy of citrus juice as a contraceptive and as a microbicide directed against HIV and other STD. We may also be able to optimize the procedure; for example, it may be better to douche with undiluted juice before sex, rather than using diluted juice after sex. In view of the 61% incidence of HIV infection in Nigerian female commercial sex workers, we will not need large numbers of volunteers to get significant results. We can also answer the critically important question of whether the juice has any adverse effects in those who are already HIV positive, with compromised immune defences. We have learned a sobering lesson from nonoxynol 9, which was widely used throughout the world as a microbicide until studies in African commercial sex workers showed that frequent use damaged the vaginal epithelium and increased their susceptibility to HIV infection (Van Damme *et al.* 2002). We do not want to repeat that mistake with citrus juice.

If follow-up studies of the Nigerian commercial sex workers can demonstrate that intravaginal lime or lemon juice is a safe and effective contraceptive and microbicide, how readily available would it be to poor women in developing countries? The wild ancestor of today's cultivated limes and lemons was indigenous to China, India, and South East Asia. It was Alexander the Great who brought the first wild fruits to Europe, where the Romans subsequently developed the lemon and the Arabs the lime (Mabberley 1997). Lemons and limes are now found throughout all tropical and temperate regions of the globe, but will not withstand prolonged frost. Thus there is production plant—citrus trees—strategically located in all the tropical and temperate regions of the world where the HIV pandemic is most severe. In the supermarkets of Capetown, you can buy five large lemons, capable of yielding at least 200 ml of juice, for the price of one condom. The juice is stable at room temperature for at least a month, and does not damage either latex or plastic condoms.

Phase 1 clinical trials of the acceptability and safety of intravaginal lime juice will begin in Thailand in 2006. Hopefully, prospective clinical trials of the safety

and efficacy of lime and lemon juice douching in Nigerian commercial sex workers can also begin in 2006, subject to the availability of funding.

### 3. TOPICAL VAGINAL OESTROGEN

The vagina is exposed to potential pathogens from the male every time sexual intercourse takes place. It has therefore had to develop specific local defence mechanisms to protect itself both from the trauma of intercourse and the presence of pathogens. Since female mammals, with one notable exception, only permit intercourse when in behavioural oestrus at around the time of ovulation, they have made use of the oestrogen produced by the developing Graafian follicle in the ovary to thicken and keratinize the vaginal epithelium at this time, making this the first line of defence against potential pathogens. The first person to observe this phenomenon of cyclical vaginal thickening and keratinization was the renowned French biologist from Bordeaux, Fernand Lataste, who produced a 500 page monograph on the subject (Lataste 1886, 1887). Working with rats, mice, hamsters and several species of wild rodents from neighbouring North Africa, he noted that these cyclical changes in the vaginal epithelia of rodents were in marked contrast to the much less pronounced changes in the human vaginal epithelium. He wisely concluded that:

On peut voir, dans ce fait, la justification anatomique de l'aptitude constante de la Femme au coit

The fact that humans are polygamous and uniquely have sex for pleasure more frequently than for procreation, and at any stage of the menstrual cycle, may have made us particularly prone to sexually transmitted infections (Potts & Short 1999). Perhaps this is why we have maintained a partially oestrogenized vagina throughout the menstrual cycle, thanks to the oestrogen also produced by the human corpus luteum in the second half of the cycle.

Since the vagina, rather than the cervix, appears to be the principal route of HIV infection in women, it might be possible to reinforce this first line of defence against HIV entry by using topical vaginal oestrogen treatment to increase the hypertrophy and keratinization of the vaginal epithelium. Topical vaginal oestriol cream (Ovestin: Organon) or vaginal oestradiol pills (Vagifem: Novo Nordisk) have been used by millions of postmenopausal women in the Western world to treat the symptoms of oestrogen deficiency, such as atrophy of the vaginal epithelium. It seems highly significant that postmenopausal women also have a four to eightfold increase in the chance of being infected with HIV compared to premenopausal women (European Study Group on heterosexual transmission of HIV 1992). A very important study in ovariectomized Rhesus monkeys (Smith *et al.* 2004) has recently shown that Ovestin vaginal cream thickened and cornified the vaginal epithelium when administered at a dose of 1 mg oestriol twice weekly for five weeks. When the animals were then challenged with an intravaginal inoculum of pathogenic Simian immunodeficiency virus (SIV), six of the eight untreated controls became infected, but only one of the twelve

oestrogen-treated animals, a highly significant difference ( $p=0.0044$ ). Systemic oestriol levels fell during the course of treatment, probably because of reduced absorption through the thickened vaginal epithelium. The duration of the effect lasted for 12 days after the cessation of treatment. There was no evidence of systemic oestrogenic effects, and the circulating LH levels did not differ between treatment and control groups. Oestriol is such a weak natural oestrogen that it seems unlikely that twice weekly vaginal administration to premenopausal women could have any systemic contraceptive effect by suppressing pituitary gonadotrophin secretion.

Many women of reproductive age might welcome a contraceptive that also offered them protection against HIV infection, and an exciting opportunity exists for achieving this objective. If the oestrogen-gestagen combined oral contraceptive pill is taken vaginally rather than orally, this should maximize the local protective oestrogenic effects on the vaginal epithelium, while still retaining a systemic contraceptive effect. The clue came from a study conducted by the South to South Cooperation in Reproductive Health, funded by the Rockefeller Foundation (Coutinho *et al.* 1993), although the investigators were unaware of the significance of their observations at the time.

The objective of the trial was to see if daily vaginal administration of the oral contraceptive pill would reduce the incidence of adverse side effects, such as headaches, nausea, and weight gain, that might deter women in developing countries from using it. Although the trial failed to demonstrate any such reduction, it did demonstrate very convincingly that when given vaginally, the oral pill retains its full contraceptive efficacy.

The trial was carried out in Brazil, the Dominican Republic, Mexico, Cuba, Kenya, Nigeria, Zambia, Egypt and China, and involved 1055 women studied for 12 630 months. Two brands of oral contraceptive pill were used, a Schering product containing 250 µg levonorgestrel and 50 µg ethinyl oestradiol, and an Organon product containing 150 µg desogestrel and 30 µg ethinyl oestradiol. The participants were instructed to insert one pill in the vagina daily for 21 days, beginning on the fifth day of the menstrual cycle, followed by a 7 day pill-free period to allow withdrawal bleeding. One year pregnancy rates for the Schering pill were 2.78%, and 4.54% for the Organon preparation, values that are similar to those following oral administration. No attempt was made to study the vaginal histology, but it seems almost certain that repeated vaginal exposure to such a high local concentration of ethinyl oestradiol would result in significant vaginal thickening and cornification. If this was the case, then vaginal pills could protect women against both pregnancy and HIV infection. This would be a major breakthrough, and prospective clinical trials should begin without delay. The pills, now off patent, could easily be manufactured in countries like India, China, Thailand or Indonesia at minimal cost.

#### 4. MALE CIRCUMCISION

How does HIV enter the human penis? It appears to be through the inner aspect of the foreskin. The foreskin,

like the vagina, is richly supplied with Langerhans and dendritic cells, but unlike the vagina, the inner foreskin epithelium is relatively thin and poorly keratinized at all times. By contrast, the glans penis has a highly keratinized epithelium to protect it from trauma during intercourse. Unless gross lesions are present, it is a most unlikely site for HIV entry (Szabo & Short 2000; Short 2004a; McCoombe & Short submitted). If the inner foreskin is the predilection site for HIV entry, it is hardly surprising that removal of the foreskin by circumcision greatly reduces a man's susceptibility to HIV infection.

Male circumcision is one of the oldest surgical procedures known. It is depicted in a wall relief from Saqqara, in Lower Egypt, dated at about 2300 BC (Szabo & Short 2000). The practice seems to have been almost universal among all strata of society in Ancient Egypt, and in the indigenous populations of the deserts of Saudi Arabia, Mexico, the Kalahari and central Australia, probably because in a hot, dry, sandy environment it would be easy for sand to get under the foreskin and set up a balanitis, which could lead to urethritis, cystitis, nephritis and death. In the Second World War, it was necessary to send urologists to circumcise all the Australian troops fighting in the North African campaign for this very reason. When the Jews were held in bondage in Egypt, they were all circumcised; so when Moses led them out of Egypt into their promised land, ritual neonatal circumcision on the seventh day after birth became their covenant with God. Christ was circumcised, and New Year's Day, the Feast of the Circumcision in the Christian calendar, is 7 days after Christmas Day. The Prophet Mahomed, born in hot, dry, sandy Saudi Arabia, was also circumcised, as are all Muslims to this day. Looking at the number of HIV infections in the world, the very small number of reported cases in large Muslim nations like Egypt, Iran, Pakistan, Bangladesh and Indonesia is striking; this is likely to be due at least in part to the high-prevalence of male circumcision in these countries (figure 2).

The magnitude of the protective effect of male circumcision against HIV infection becomes more impressive by the day (Reviewed in Short 2004a). Weiss *et al.* (2000) carried out a detailed systematic review and meta-analysis of 27 reports from different countries in sub-Saharan Africa, and showed that male circumcision appeared to reduce the relative risk of HIV infection to 0.56; among high-risk groups, such as STD clinic attendees, the relative risk was reduced to 0.29. A 30-month prospective study of HIV-discordant couples in Uganda where the woman was HIV positive showed zero seroconversions in 50 circumcised men over the 30 months, compared to 40 seroconversions in 137 uncircumcised men, a highly significant difference ( $p=0.004$ ; Quinn *et al.* 2000). A recent Cochrane Review of male circumcision and HIV infection concluded that there was a strong association between male circumcision and prevention of HIV, especially among high-risk groups. However, it stated that there was insufficient evidence to support an intervention until carefully controlled randomized prospective clinical trials have been carried out; these are currently underway in Kenya, Uganda and South Africa (Siegfried *et al.* 2003). The South African trial

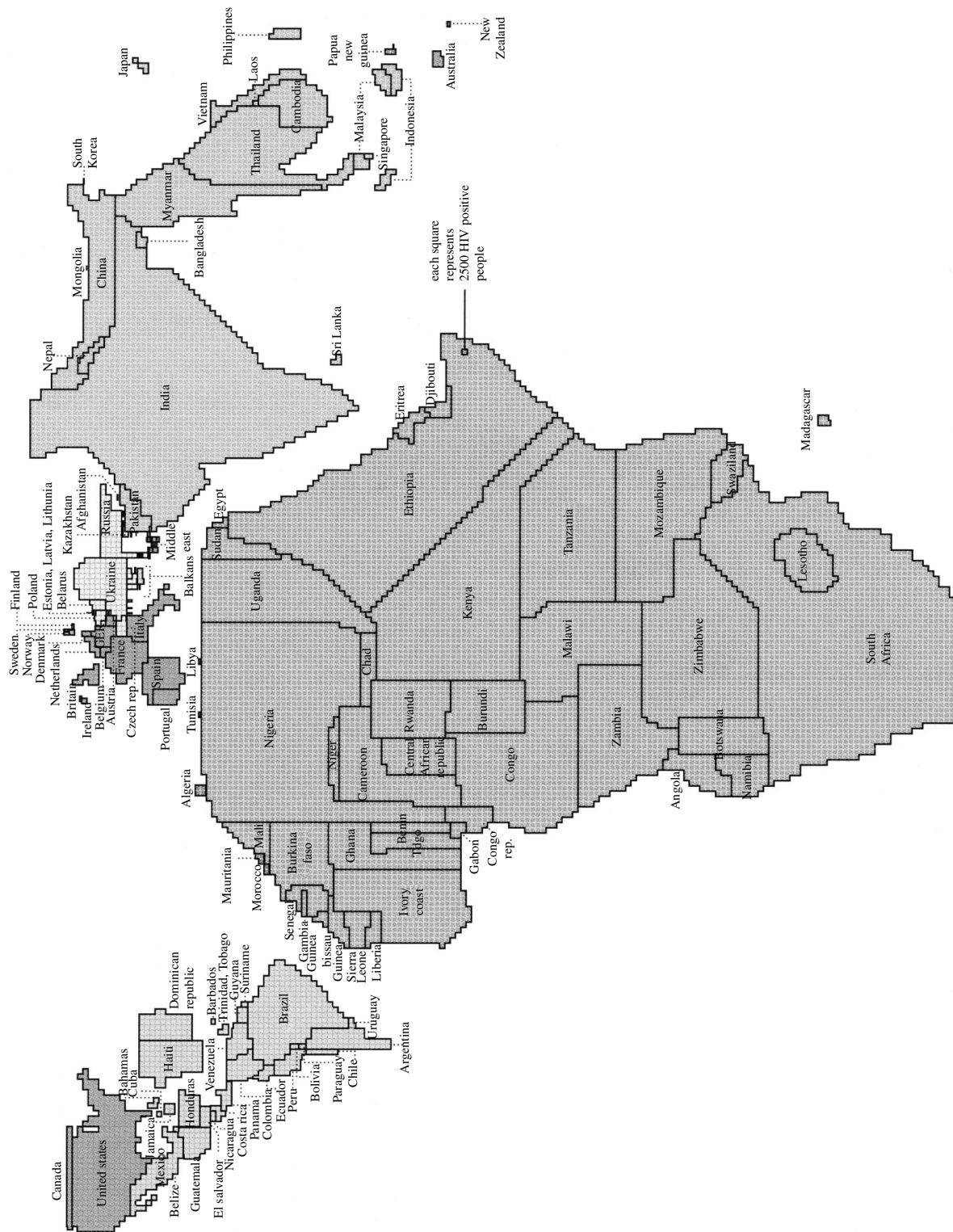


Figure 2. The global HIV-AIDS pandemic, 20 years to the day after the first recognition of the disease in the USA. Each small square represents 2500 people who are HIV positive. Note the small number of cases in large Islamic nations such as Egypt, Iran, Pakistan, Bangladesh and Indonesia, probably due at least in part to the protective effect of male circumcision. Modified from The New York Times, June 5 2001.

involving 3273 uncircumcised men aged 18–24 randomized into treatment and control groups and followed prospectively showed 65% HIV protection following circumcision, making it unethical to withhold circumcision from the control group (Auvert *et al.* 2005; Cohen 2005). A recent large prospective study of 2298 HIV-negative men attending STD clinics in Pune, India showed that male circumcision had a sevenfold protective effect against HIV infection, whereas there was no protective effect against HSV-2, syphilis or gonorrhoea (Reynolds *et al.* 2004).

Another important advantage of male circumcision is that it significantly protects men from human papilloma virus infection (HPV), and their wives from carcinoma of the cervix, caused by HPV acquired from their husbands (Castellsagué *et al.* 2002). Since cervical carcinoma is the second commonest female cancer worldwide, male circumcision also has significant benefits for female reproductive health.

Although male circumcision has been practised for over 4 millennia, the techniques used today are still relatively crude. Nelson Mandela gives a graphic account of his own circumcision, traditionally carried out by the Xhosa during adolescence in an elaborate ceremony as a preparation for manhood:

The old man was kneeling in front of me. I looked directly into his eyes. He was pale, and though the day was cold, his face was shining with perspiration. His hands moved so fast they seemed to be controlled by an otherworldly force. Without a word, he took my foreskin, pulled it forward, and then, in a single motion, brought down his assegai. I felt as if fire was shooting through my veins; the pain was so intense that I buried my chin in my chest. Many seconds seemed to pass before I remembered the cry, and then I recovered and called out 'Ndiyindoda' (I am a man). (Mandela 1994).

Probably the simplest, safest and most painless way of performing male circumcision today is to apply some local anaesthetic cream to the penis and then use the disposable PlastiBell device, in which a plastic bell is placed over the end of the penis beneath the foreskin and a piece of thread is then tied tightly around a groove at the base of the bell, cutting off the blood and nerve supply to the whole of the foreskin that can then be painlessly trimmed off with scissors (Alanis & Lucidi 2004). The device is made by Hollister in the United States, but is off-patent, and could be made for a fraction of a cent. The technique is currently being promoted throughout Botswana, a country in which male circumcision was once universally practised, but was abandoned during colonial rule (Modise *et al.* 2004). Currently, Botswana has one of the highest rates of HIV infection in the world.

If we had a cheap vaccine or drug that only needed to be administered once in a lifetime to confer a 65% protective effect against HIV infection, this would make headlines around the world. But the Western world has turned against routine male circumcision, regarding it as unnecessary male mutilation with no health benefits. If we believe in evidence-based medicine, we should think again, learn from Islam, and promote the practice as a first line of defence against male HIV infection, and to protect women from cervical cancer.

## 5. MALE POST-COITAL HYGIENE

How long following first contact with HIV does it take for a man's penis to become infected? The virus must become attached to specific HIV receptors in the penile epithelia, and studies in Rhesus monkeys in which SIV has been applied to the foreskin have suggested that it may take an hour or more before the virus is internalized (Miller 1998). Thus post-coital penile hygiene, wiping the penis, and in particular the inner aspect of the foreskin, with a potent microbicide within minutes of penile withdrawal from the vagina (or rectum) may significantly reduce a man's chances of becoming infected (Short 2004a).

Male post-coital hygiene is a seriously neglected area of research (Bailey *et al.* 2001), and it is interesting to recall that this was how Gabriel Fallopio, the great Italian anatomist, first advocated that men should protect themselves from syphilis. In his 1564 book *De morbo Gallico*, he states that 'As often as a man has intercourse, he should (if possible) wash the genitals, or wipe them with a cloth; afterward he should use a small linen cloth made to fit the glans, and draw forward the prepuce over the glans; if he can do so, it is as well to moisten it with saliva or a lotion. .... I tried the experiment on eleven hundred men, and I call immortal God to witness that not one of them was infected' (Potts & Short 1999). If the glans cap beneath the foreskin could be medicated with a microbicide, this might be a very effective way of preventing HIV infection post-coitally.

There are many readily available microbicides that can kill HIV, starting with soap and water, but perhaps the simplest is to rely on the virus' susceptibility to low pH. So once again, neat lemon or lime juice or vinegar might prove to be particularly useful, especially since it is painless, cheap, and readily available.

One situation that is crying out for such a simple solution is the case of migrant mine workers in South Africa's gold and diamond mines (Jackson 2002). Away from their homes and families for months at a time, living in crowded all-male dormitories, they soon start to have sex with local female commercial sex workers, many of whom are HIV positive.

Although the mines may provide free condoms, there is considerable prejudice against condom use in Southern Africa where skin-to-skin sexual contact is preferred, especially if you are paying for it. Thus many of the men will never use condoms. Within a week or two of first infection with HIV the men develop high temperatures, and this can cause them to collapse from heat stroke in the hot, humid and confined conditions of the mine shaft. The company employing them therefore has to discharge them as unfit for work, and has to repatriate them to their town or country of origin and recruit a replacement—a costly process. Meanwhile the man returns to his community with a high viraemia, at a time when he is most infectious to others (Pilcher *et al.* 2004). Thus the giant mining corporations that are employing hundreds of thousands of mineworkers are inadvertently propagating the HIV pandemic in Southern Africa. It should be a simple matter to carry out a trial, providing the mineworkers not only with free condoms but also with free lemons in

their dormitories, and monitoring whether this reduces the incidence of HIV infection.

Another important aspect of post-coital penile hygiene involves male homosexual behaviour. Those who practise mutual masturbation without condoms in the belief that this is safe sex have failed to realize that getting the infected ejaculate of your partner onto your penis is a dangerous procedure, particularly if you are uncircumcised. Some gay men also indulge in the practice of 'docking', where one man's penis is inserted inside the foreskin of his partner, followed by mutual masturbation; this is likely to be a very risky procedure. For those indulging in such practices, post-ejaculatory hygiene, washing the penis with a potent virucide, should be strongly recommended. The simple public health message that getting the semen of your potentially infected partner in contact with your penis is unsafe seems to have been largely ignored.

## 6. EDUCATING TEENAGERS

The key to all forms of HIV prevention is education, and the theory needs to be taught before sexual activity commences. Parents are notoriously bad at educating their children about this sensitive subject, so the school classroom is the obvious alternative. But who should do the teaching, and what should be taught? Peer group education has many advantages over conventional didactic teaching to a set curriculum, but it immediately raises the question of how to train the peer group teachers. A recent nationwide attempt to develop peer group sex education in English schools was a failure (Short 2004b).

We have therefore pioneered an alternative approach in Botswana, using PhotoVoice. Teenage school children aged between 13 and 17 in a school in Gaborone, Botswana were lent cheap disposable cameras, and asked to go out into the community and take photographs of their daily lives, illustrating the theme of HIV/AIDS prevention. After all the photographs had been developed, the pupils selected the ones they thought were the best, and divided them up into various HIV/AIDS themes. This led on to intense group discussions about why each particular photograph had been taken, with the photographer in question explaining the theme to the rest of the group. The photographs were then given captions and pinned up in a photographic exhibition which students from other schools were invited to attend. The whole process was monitored by a young Botswanan medical student, who was able to supply all the relevant background scientific information if requested (Modise *et al.* 2004). By using this PhotoVoice technique, it is possible to develop extremely effective HIV/AIDS education of the students, by the students, for the students. They have pride of ownership of their project, and the news can be rapidly transmitted to other schools. Perhaps the key message to get across is the very high risk of HIV infection for a young girl if she has unprotected sex with an older man, who may well be HIV-positive.

The success of this pilot PhotoVoice project in Botswana has already resulted in a request to demonstrate the technique in neighbouring Malawi.

Since it costs so little to organize, hopefully it can spread throughout sub Saharan Africa, and give young people the best protection of all against HIV infection—knowledge.

## 7. IN CONCLUSION

Of the five new ways of HIV prevention discussed here, some are unproven but seem worthy of immediate clinical trials, such as intravaginal lime or lemon juice, topical vaginal oestrogen, and post-coital penile hygiene. The protective effect of male circumcision is already staring us in the face; developing better ways of informing young teenagers about how to protect themselves from HIV infection must become a top priority for all nations. At the end of the day prevention is better than cure, and is certainly far more cost-effective. We *can* stop AIDS, especially if we think simply, and simply think.

I would like to express my deep personal thanks to Senator Mechai Viravaidya for his constant encouragement from the first beginnings of this project, to Dr Rob Moodie and VicHealth for providing the initial funding, and to my scientific colleagues Prof. Malcolm Potts, Dr Suzanne Crowe, Dr Daniel Stefanski, Dr Robert Szabo, Dr Mingjia Li, Dr Carlos Guterrez, Godwin Imade, Scott McCoombe, Dr Gary Clarke, Modise Modise and Max Nhlatho for doing all the hard work, and to my long-suffering Secretary, Anne Brockman. We are all indebted to the devotion of Brian Hail, who maintains our website, [www.aids.net.au](http://www.aids.net.au), which is an invaluable and up-to-date source of information about all our current activities.

## REFERENCES

- Alanis, M. C. & Lucidi, R. S. 2004 Neonatal circumcision: a review of the world's oldest and most controversial operation. *Obstet. Gynaecol. Surv.* **59**, 379–395. (doi:10.1097/00006254-200405000-00026)
- Auvert, B., Taljaard, D., Lagarde, E., Sobngwi-Tambekou, J., Sitta, R. & Puren, A. 2005 Randomised, controlled intervention trial of male circumcision for reduction of HIV infection risk: the ANRS 1265 trial. *PLoS Medicine* **2**, 1112–1122.
- Bailey, R. C., Plummer, F. A. & Moses, S. 2001 Male circumcision and HIV prevention: current knowledge and future research directions. *Lancet Infect. Dis.* **1**, 223–231. (doi:10.1016/S1473-3099(01)00117-7)
- Burgess, S. A., Walker, M. L., Sakakibara, H., Knight, P. J. & Oiwa, K. 2003 Dynein structure and power stroke. *Nature* **421**, 715–718. (doi:10.1038/nature01377)
- Castellsagué, X. *et al.* 2002 Male circumcision, penile human papillomavirus infection, and cervical cancer in female partners. *N. Engl. J. Med.* **346**, 1105–1112. (doi:10.1056/NEJMoa011688)
- Clarke, G. N., McCoombe, S. G. & Short, R. V. In Press. The sperm immobilizing properties of lemon juice. *Fertil. Steril.*
- Cohen, J. 2005 Male circumcision thwarts HIV infection. *Science* **309**, 860. (doi:10.1126/science.309.5736.860)
- Coplan, P. M., Mitchnik, M. & Rosenberg, Z. F. 2004 Regulatory challenges in microbicide development. *Science* **304**, 1911–1912. (doi:10.1126/science.1100441)
- Coutinho, E. M. *et al.* 1993 Comparative study on the efficacy and acceptability of two contraceptive pills administered by the vaginal route: an international multicenter clinical trial. *Clin. Pharmacol. Ther.* **53**, 65–75.
- European Study Group 1992 European study group on heterosexual transmission of HIV. *Br. Med. J.* **304**, 809–813.

- Gotlieb, G. S. *et al.* 2004 Dual HIV-1 infection associated with rapid disease progression. *Lancet* **363**, 619–622. (doi:10.1016/S0140-6736(04)15596-7)
- Himes, N. E. 1963 *Medical history of contraception*. New York: Gamut Press Inc.
- Imade, G. E., Sagay, A. S., Onwuliri, V., Egah, D. Z., Potts, M. & Short, R. V. 2005 Use of lemon or lime juice douches in women in Jos, Nigeria. *Sexual Health*, **2**, 237–239. (doi:10.1071/SH04055/1448-5028/05/040237)
- Jackson, H. 2002 *AIDS Africa: continent in crisis*. Zimbabwe: SFAIDS.
- Lataste, F. 1886 Notes prises au jour le jour sur différentes espèces de l'ordre des Rongeurs observées en captivité. *Act. Soc. Linn. Bordeaux* **40**, 293–466.
- Lataste, F. 1887 Notes prises au jour le jour sur différentes espèces de l'ordre des Rongeurs observées en captivité. *Act. Soc. Linn. Bordeaux* **41**, 201–536.
- Li, M. & Short, R. V. 2002 How oestrogen or progesterone might change a woman's susceptibility to HIV-1 infection. *Aust. NZ J Obstet. Gynaecol.* **42**, 472–475. (doi:10.1111/j.0004-8666.2002.00472.x)
- Lomborg, B. (ed.) 2004 *Global crises, global solutions*. Cambridge, UK: Cambridge University Press.
- Mabberley, D. J. 1997 A classification for edible *Citrus*. *Telopea* **7**, 167–172.
- Mandela, N. 1994 *Long walk to freedom*. London: Little, Brown and Co.
- Mann, T. 1964 *The biochemistry of semen and of the male reproductive tract*. London: Methuen.
- Martin, L. S., McDougal, J. S. & Lososki, S. L. 1985 Disinfection and inactivation of the human thymotrophic virus type III/lymphadenopathy-associated virus. *J. Infect. Dis.* **152**, 400–403.
- May, R. M., Gupta, S. & McLean, A. R. 2001 Infectious disease dynamics: what characterises a successful invader? *Phil. Trans. R. Soc. B* **356**, 901–910. (doi:10.1098/rstb.2001.0866)
- McCoombe, S. G. & Short, R. V. Submitted. How HIV-1 infects the human penis. *AIDS*.
- Miller, C. J. 1998 Localization of Simian immunodeficiency virus-infected cells in the genital tract of male and female Rhesus macaques. *J. Reprod. Immunol.* **41**, 331–339. (doi:10.1016/S0165-0378(98)00069-2)
- Miller, C. J. & Shattock, R. J. 2003 Target cells in vaginal HIV transmission. *Microbes Infect.* **5**, 59–67. (doi:10.1016/S1286-4579(02)00056-4)
- Modise, M., Nhlatho, M. & Short, R. 2004 Stopping AIDS in Africa. *Melbourne Univ. Mag.*, 21–23.
- Pilcher, H. 2004 Starting to gel. *Nature* **430**, 138–140. (doi:10.1038/430138a)
- Pilcher, C. D., Tien, H. C., Eron, J. J., Vernazza, P. L., Leu, S. Y., Stewart, P. W., Goh, L. E. & Cohen, M. S. 2004 Brief but efficient: acute HIV infection and the sexual transmission of HIV. *J. Infect. Dis.* **189**, 1785–1792. (doi:10.1086/386333)
- Potts, M. & Short, R. 1999 *Ever since Adam and Eve*. Cambridge, UK: Cambridge University Press.
- Potts, M. & Short, R. V. 2003 Using microbicides to fight the spread of HIV. *Science* **300**, 431. (doi:10.1126/science.300.5618.431a)
- Quinn, T. C. *et al.* 2000 Viral load and heterosexual transmission of human immunodeficiency virus type 1. *N. Engl. J. Med.* **342**, 921–929. (doi:10.1056/NEJM200003303421303)
- Reynolds, S. J., Shepherd, M. E., Risbud, A. R., Gangakhedkar, R. R., Brookmeyer, R. S., Divekar, R. S., Mehendale, S. M. & Bollinger, R. C. 2004 Male circumcision and risk of HIV-1 and other sexually transmitted infections in India. *Lancet* **363**, 1039–1040. (doi:10.1016/S0140-6736(04)15840-6)
- Royce, R. A., Sena, A., Cates, W. & Cohen, M. S. 1997 Current concepts: sexual transmission of HIV. *N. Engl. J. Med.* **336**, 1072–1078. (doi:10.1056/NEJM199704103361507)
- Sachs, J. D. 2005 Achieving the millennium development goals—the case of malaria. *N. Engl. J. Med.* **352**, 115–117. (doi:10.1056/NEJMp048319)
- Short, R. V. 2004a The HIV/AIDS pandemic: new ways of preventing infection in men. *Reprod. Fertil. Dev.* **16**, 555–559. (doi:10.1071/RD03109)
- Short, R. V. 2004b Teaching safe sex in English schools. *Lancet* **364**, 307–308. (doi:10.1016/S0140-6736(04)16735-4)
- Short, R., McCoombe, S. G., Maslin, C., Naim, E. & Crowe, S. 2004 Lemon and lime juice as potent natural microbicides. Abstract TuPeB4668, XV Int. AIDS Conf., Bangkok, Thailand.
- Siegfried, N. *et al.* 2003 Male circumcision for prevention of heterosexual acquisition of HIV in men (Cochrane Review). *The Cochrane Library* **3**, 1–39.
- Smith, S. M., Mefford, M., Sodora, D., Klase, Z., Singh, M., Alexander, N., Hess, D. & Marx, P. A. 2004 Topical estrogen protects against SIV vaginal transmission without evidence of systemic effect. *AIDS* **18**, 1637–1643. (doi:10.1097/01.aids.0000131393.76221.cc)
- Szabo, R. & Short, R. V. 2000 How does male circumcision protect against HIV infection? *Br. Med. J.* **320**, 1592–1594.
- UNAIDS 2004 *UNAIDS report on the global AIDS epidemic*. Geneva: UNAIDS.
- Van Damme, L. *et al.* 2002 Effectiveness of COL-1492, a nonoxynol-9 vaginal gel, on HIV-1 transmission to female sex workers: a randomised controlled trial. *Lancet* **360**, 971–977. (doi:10.1016/S0140-6736(02)11079-8)
- Wagner, G. & Ottesen, B. 1982 Vaginal physiology during menstruation. *Ann. Intern. Med.* **96**, 921–923.
- Weiss, H. A., Quigley, M. A. & Hayes, R. J. 2000 Male circumcision and risk of HIV infection in sub Saharan Africa: a systemic review and meta-analysis. *AIDS* **14**, 2361–2370. (doi:10.1097/00002030-200010200-00018)

## Discussion

The Royal Society's Committee on the Public Understanding of Science (COPUS) invited me to give one of its Lunch Time Science Lectures in the National Portrait Gallery, Trafalgar Square, on September 20, 1995. I was asked to talk about Dr Marie Stopes, the pioneer of Family Planning in Britain, in front of a magnificent portrait of her, painted by Sir Gerald Kelly in 1953 when she was 73 years old.

My lecture was entitled 'Marie Stopes and Married Love', in recognition of her ground-breaking book 'Married Love', first published in 1918, which eventually sold over a million copies. A 1935 survey of American academics rated it among the 25 most influential books of the previous 50 years, ahead of Einstein's *Relativity*, Freud's *Interpretation of Dreams*, Hitler's *Mein Kampf* and Maynard Keynes' *Economic Consequences of Peace* (Hall 1977).

Reading Ruth Hall's excellent biography of Marie Stopes in preparation for my lecture, I was amazed to learn that when Marie Stopes established her first Birth Control Clinic in Marlborough Road, Holloway in 1921, one of the only contraceptives being used by women in London at that time was half a lemon, partially squeezed out and then inserted in the vagina to cover the cervix like a cap.

When I related this story to the audience attending my lecture, it aroused an embarrassed giggle from many of the elderly women in attendance. After the lecture was over, a number of them drew me aside, one at a time, and politely informed me that they had relied on lemons to regulate their fertility. Those ladies taught me an invaluable lesson, not about the Public Understanding of Science, but about the need for Scientists to understand the Public.

That 1995 encounter was the inspiration for much of the research described in this article.

***Additional references***

Stopes, M. C. 1918 *Married love: a new contribution to the solution of sex difficulties*. Fifeild.

Hall, R. 1977 *Marie Stopes: a biography*. London: Andre Deutsch.