Trichomoniasis treatment: a dual approach

Dr Raina Fichorova is leading the way in the study of *Trichomonas vaginalis*, a sexually transmitted infection with serious consequences. With the help of a wealth of experts, inroads are being made towards finding an effective therapy

Your results have shown that pathogenicity of the sexually transmitted protozoan parasite T. vaginalis – the cause of trichomoniasis – is fuelled by a viral invader. How does this disrupt common understandings of parasites?

Current diagnostic methods do not distinguish between virus-free and virus-invaded parasites. Moreover, routine anti-parasitic therapy is completely oblivious to the potentially harmful effects of protozoan viruses, which may explain why this therapy has failed to prevent complications associated with trichomoniasis. For example, metronidazole treatment, even if effective in clearing the protozoan infection, does not reduce and can even increases the risk of preterm birth attributable to T. vaginalis infection. Thus, our results open the door to a novel treatment-prevention paradigm. A combination therapy targeting the parasite and the virus or viral signaling would prevent the inflammatory complications.

Why is the double-stranded (ds) nature of the Trichomonas vaginalis viruses (TVVs) important?

The genome structure of some viruses, including the TVVs, is made of dsRNA. This dsRNA is quite stable, and the diagnostic methods that can be devised for detecting them are different from those for detecting single-stranded RNA or DNA. That is the practical importance of knowing that the TVVs have a dsRNA genome. From a mechanistic biological standpoint, dsRNA is one of the known 'pathogen-associated molecular patterns' (PAMPs), that our cells can detect and respond to for mounting anti-pathogen responses such as production of interferons and other cytokines. Thus, the dsRNA genome in TVVs provides one such type of PAMP that contribute to our bodies' responses to T. vaginalis infections. The trouble is that since TVV does not multiply in human cells but only within protozoa, the cytokine responses are not efficient in clearing the TVV infection and we are left to deal with only the pro-inflammatory harmful side effects of the PAMP recognition.

Are you collaborating with other researchers in order to ensure the success of your investigations? How has a multidisciplinary approach aided the project?

My success is ensured by an amazing interdisciplinary team that complements my expertise in immunology and reproductive tract biology, and my many years of experimentalist experience. To unveil the multifaceted aspects of the T. vaginalis infection I partnered with:

- Max Nibert, Professor at Harvard Medical School, who directs a basic virology lab and performs molecular-genetic characterisation of dsRNA viruses
- Bibhuti Singh, Associate Professor at The State University of New York, Upstate University, and Cathy Costello, Professor at Boston Medical Center, who both provide expertise in defining the biochemical composition and structure of TV surface domains
- Andrew Onderdonk, Harvard Medical School Professor and Director of clinical microbiology at Brigham and Women's Hospital, who brings over 30 years experience in vaginal microbiology
- Jill Huppert, University of Cincinnati Associate Professor, a clinical scientist devoted to sexually transmitted disease prevention, currently holding Chief position at the Centers for Disease Control and Prevention (CDC)
- Academic clinicians Susan Cu-Uvin, Professor at Brown University, and Kenneth Mayer, Professor at Harvard School of Public Health, who provides the HIV/AIDS related aspects of TV pathogenesis
- Several scientists at the Harvard-MIT Division of Health Sciences and Technology

I also owe a great deal of my team's success to the young researchers in my lab, who have contributed to all my studies and publications, and whose tireless passion and creativity are the true fuel of the discovery engine.

Could you expand on how the knowledge about T. vaginalis-host interactions could aid preventative methods and treatment?

Understanding the molecular pathways and cell biology of this infection, as well as its interaction with the vaginal microbiome, can lead to a combination therapy simultaneously targeting the parasite and the virus and restoring the healthy microbial communities, thus preventing harmful inflammatory damage.

With impending budget cuts in many areas of research alongside aggressive investment in this sector by other nations, how can the US ensure it does not fall behind on the global stage?

In the US, research has always been driven by an extremely competitive funding mechanism. Not for the first time in modern history scientists throughout the world face the challenges of a global economic recession. I am confident that these challenges can only make us stronger and force us to be more cost-efficient and more industrious in adopting the technological advances of the 21st Century. Scientists are still to learn to better share and translate experience across fields and disciplines.

Why is dissemination so important to your discipline?

Raising the next generation of young scientists is the key to a better future for our planet. Hopefully by sharing successful and inspiring research stories like ours, reports such as *International Innovation* can contribute to attracting young people to science careers, which we now need more than ever.

DR RAINA FICHOROVA

Sexual parasitology

Trichomoniasis remains understudied despite its devastating effects. Now, a group of scientists, headed by **Brigham and Women's Hospital**, is investigating its mechanics to improve the knowledgebase of this sexually transmitted infection

RESEARCHERS FROM HARVARD Medical School, Brigham and Women's Hospital, and SUNY Upstate Medical University have revealed that a common parasitic sexually transmitted infection (STI), Trichomoniasis, is exacerbated by a virus that prays on the parasite.

To date, trichomoniasis remains one of the top neglected parasitic diseases, despite there being ~249 million new cases in adults each year according to 2005 WHO figures. This amounts to more cases than chlamydia, gonorrhoea and syphilis combined. Cases are spread roughly equally between the sexes, with 143 million cases reported in males and 106 million in females. Although sexual contact is the primary cause of transmission, the parasite can exist for several hours in water on toilet seats, on bath towels and in urine. The infection moves quietly between continents, classes and ethnicities indiscriminately and the effects can be devastating.

HARROWING CONSEQUENCES

Although the infection can be symptom-free initially, it quickly establishes within the vagina, or the urethra in both men and women. Eventually it manifests itself as the leading cause of vaginitis and nongonococcal urethritis, a source of inflammation, tissue damage and considerable pain. Inflammation in the female genital tract can cause significant problems during pregnancy such as preterm delivery, low birth weight and developmental delay.

Infants born to women with trichomoniasis are at risk of mortality, infection and morbidity. Suffering the infection or carrying the parasite also leaves women more susceptible to the human papillomavirus, which raises the risk of contracting cervical cancer and pelvic inflammatory disease, which can lead to infertility. Men do not escape the scourge of trichomoniasis as infection, which has been shown as possible contributor to prostate cancer and infertility. Carrying the parasite can even raise the risk of contracting HIV, thus increasing the likelihood of developing AIDS.

INVESTIGATING AN UNDER-RESEARCHED PATHOGEN

Dr Raina Fichorova, an Associate Professor of Obstetrics, Gynaecology and Reproductive Biology at Brigham and Women's Hospital, has been leading the multidisciplinary team of researchers responsible for identifying the virus and reducing the knowledge gap of trichonomiasis.

Explaining the rudiments of the single cell Trichomonas vaginalis parasite responsible, Fichorova extols its abilities: "It moves in a zigzag fashion thanks to its tail apparatus and adheres to the epithelial cells, taking advantage of the anaerobic environment and feeding on mucosal nutrients".

The virus that prays on the parasite in turn is called T. Vaginalis Virus (TVV) and is a double-stranded (ds)RNA endosymbiont. Unlike its parasitic counterpart that causes so much distress and damage, the virus has seemingly no detrimental effects on its own protozoan host. In fact, the researchers believe its presence may even enable the parasite to thrive. Until recently, scientists did not know the effects of (ds)RNA viruses on the human body. "It is unfortunate that a human pathogen of such worldwide significance has been neglected to such a degree," notes Fichorova's co-author Dr Max Nibert of Harvard Medical School.

ENLIGHTENING RESULTS

The study involved testing a collection of Trichomonas samples, some infected with the virus and others TVV-free, to investigate the effects on human lab-grown cells. The aim was to investigate the mechanisms of T. vaginalis pathogenicity, with a view to transferring the knowledge gained into innovative clinical solutions. So far, the research has made several key discoveries that could determine the most effective way to tackle the trichomoniasis infection.

The group's studies have found that the molecule lipophosphoglycan (LPG), which is commonly found on the parasite surface, carries a sugar-coated functional cell surface domain that enables the parasite to attach

MEMBERS OF THE RESEARCH TEAM

to vaginal epithelial cells (www.ncbi.nlm.nih.gov/pubmed/16988255, www.ncbi.nlm.nih.gov/pubmed/18604640). This is the trigger for inflammation but also for suppression of a natural microbicidal reaction. Fichorova's recent publications showed that as a direct effect of LPG signalling (www.ncbi.nlm.nih.gov/pubmed/23903808), the vaginal secretions of women infected with trichomoniasis contained significantly reduced levels of the secretory leukocyte protease inhibitor (SLPI), the anti-inflammatory mechanism that should revert the infection's crippling symptoms (www.ncbi.nlm.nih.gov/pubmed/23355743).

Further to this, T. vaginalis has an ability to dampen the body's microbial response, suppressing the good lactobacillus bacteria colonies in the epithelial tissue while simultaneously encouraging the survival of *Prevotella bivia* and *Atopobium vaginae*, which are both opportunistic pathogens (www.ncbi.nlm.nih.gov/pubmed/23903808).

But the discovery that shook the current clinical paradigm the most, explains Fichorova, is identifying a novel culprit for the TV-induced inflammation and the failure of standard antibiotic therapy to prevent trichomoniasis complications (www.ncbi.nlm.nih.gov/pubmed/23144878). A number of diverse protozoan parasites carry dsRNA endosymbiont viruses, and among them, T. vaginalis leads the parade by simultaneously carrying more than one species of the TVV family (www.ncbi.nlm.nih.gov/pubmed/21345965). Nibert explains: "Unlike flu viruses, for example, this virus can't spread by jumping out of the cell into another one. It just spreads between protozoa when they [the hosts] divide or mate". In this respect, the parasite carries its own STI.

The prevalence of the virus cannot be underestimated. It was found in 80 per cent of the trichomonasis isolates during the clinical tests. Bacterial vaginosis is very common in women of reproductive age, and the experimentally-recorded inflammatory response to TVV was increased even further during this type of infection (www.ncbi.nlm.nih.gov/pubmed/23903808).

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Worryingly, the study discovered that Metronidazole, the current medication used to treat trichomoniasis, does not effectively combat the infection because of a nasty side effect. When the protozoans die, they release unharmed virions that signal the cells to increase inflammatory symptoms (www.ncbi.nlm.nih.gov/pubmed/23144878). It is this reaction that may harm expectant mothers and potentially their unborn babies.

PLANS FOR THE FUTURE

The findings in Fichorova and Nibert's paper (www.ncbi.nlm.nih.gov/pubmed/23144878) have already caught the attention of a number of science websites and publications including *Live Science*, *Science Daily* and *Microbe World*. A recent podcast sponsored by the American Society for Microbiology also highlighted the research. The challenge now is to develop new treatments that will combat both the parasite and the virus. Understanding the cell biology and molecule behaviour of the infection is the next necessary step ahead. The ongoing study will now investigate the mucosal aspect of the parasite's feeding, and what part of the virus' structure or lifecycle may be most vulnerable to drugs, with combination therapy being a viable option for affected pregnant women and young women at risk of developing STIs or cervical cancer.

As with all STIs, knowledge and dissemination through general practitioners and family planning clinics will be valuable in equipping sexually active adults to seek urgent treatment for trichomoniasis and its accompanying complications, but the study will also be of value in developing new diagnostic tools to catch both bugs before the damage is done.

INTELLIGENCE

MOLECULAR *T. VAGINALIS*-HOST INTERACTIONS IN RELEVANCE TO INFLAMMATORY SEQUELAE

OBJECTIVES

To identify novel mechanisms of T. vaginalis virulence and modifiable host factors that can be translated into improvement of clinical practice and prevention of T. vaginalis-attributable illness in women and newborns.

KEY COLLABORATORS

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RAINA FICHOROVA, a Harvard Medical School Faculty member since 1997, earned her MD and PhD from the Medical University of Sofia, Bulgaria. She is the Founding Director of the Laboratory of Genital Tract Biology, a division of the Brigham and Women's Hospital's Department of Obstetrics, Gynaecology and Reproductive Biology, focusing on inflammation and infection in the female reproductive tract.