November 2013 Epidemics and AIDS Update

1. Hepatitis C, a Silent Killer, Meets Its Match
2. Rift widens over structure of HIV’s molecular anchor
3. Intestinal Bacteria Linked to Rheumatoid Arthritis
4. Discovery of HIV ‘Invisibility Cloak’ Reveals New Treatment Opportunities
5. Challenges to Assumptions About Bisexual Men, HIV Transmission
6. HIV/AIDS holy water cure put to the test
7. Sangamo BioSciences Presents Clinical Data From SB-728-T HIV Study Demonstrating Long-Term Immune Reconstitution and Reduction in the HIV DNA Reservoir In Subjects with Long-Term Infection
8. The HIV rebound nobody is discussing
9. The greatest allergy myths and misconceptions, debunked
10. Blocking the Active Site of Thiolase
11. Autoantibodies Found in Blood Years Before Symptoms of Autoimmune Disease
12. How Do Viruses Avoid Inhibition by Endogenous Cellular MicroRNAs?
13. An Unfolding Tragedy of Chagas Disease in North America (long)
14. Understanding the Persistence of Plague Foci in Madagascar (long)
15. Building Scenarios for Eliminating and Eradicating Onchocerciasis, Lymphatic Filariasis, and Human African Trypanosomiasis
16. HIV may be becoming less fit as it adapts to the immune system
17. Did the press comply with an HIV witch-hunt in Greece?
18. ‘People Think It’s Over’
20. In Rare Move, Uganda Leader Publicly Tests for HIV
21. NIH launches trial of investigational genital herpes vaccine
22. Another negative result in the quest for a shorter TB treatment course
23. Trial of game changing new anal condom begins in Boston
24. The other scandal in Sri Lanka—and 80% of Commonwealth countries: endemic homophobia
25. NIH Launched Trial of Investigational Genital Herpes Vaccine
26. How Zinc Starves Lethal Bacteria to Stop Infection
27. Molecular Interplay Explains Many Immunodeficiencies
28. Understanding Immune System Memory—In a Roundabout Way
29. New Cause Found for Muscle-Weakening Disease Myasthenia Gravis
30. ‘He’s Pretty Spunky’ — Anesthesia Comes of Age During Civil War
31. Did Inefficient Cellular Machinery Evolve to Fight Viruses and Jumping Genes?
32. Congress opens door to allowing HIV organ donations
33. Magic Johnson Dedicates Space for World AIDS Museum in Wilton Manors
34. HPV Rates up to Three Times Higher in American Indian Women Than US Population
35. CONRAD presents new technology combining contraception, HIV and herpes simplex virus-2 prevention
36. Putting the Brakes on Immunity
37. Anthrax toxin can lurk for days in cells as a lingering threat
38. New research reveals dengue fever mystery in 2 US cities both exposed to risk
39. Anthrax bacteria play hide and seek
40. New Treatment Discovered to Cure MRSA Infection
41. Harm reduction works: extremely low HIV incidence over almost 20 years among people who inject drugs in Australia
42. Novel microbicide gel for vagina and rectum shows potential for HIV prevention
43. Scientists report human dietary supplement cures lab animals infected with human intestinal parasite
44. AbbVie reports 96% cure rate for interferon-free hepatitis C regimen in phase 3 study
45. African Commission condemns coerced sterilisation of HIV+ women
46. The Lancet/Cell Conference Asks: What Will it Take to Achieve an AIDS-free World?
47. Halting and Reversing the Spread of HIV/AIDS in Uganda: President Museveni Publically Tests for HIV
48. Two Steps Obama Can Take to Defeat AIDS
Hepatitis C, a Silent Killer, Meets Its Match
By Andrew Pollack
Published: November 4, 2013
Determined to get rid of the hepatitis C infection that was slowly destroying his liver, Arthur Rubens tried one experimental treatment after another. None worked, and most brought side effects, like fever, insomnia, depression, anemia and a rash that “felt like your skin was on fire.”

New drugs could change the way Hepatitis C is treated; think that’s St. John’s Wort you’re buying?; Elizabeth Gilbert talks about her new novel, “The Signature of All Things.”
Tom Espinosa, of Oakland, Calif., is eagerly awaiting the new hepatitis C drugs. He has advanced cirrhosis and spots on his liver that might be cancer.
But this year, Dr. Rubens, a professor of management at Florida Gulf Coast University, entered a clinical trial testing a new pill against hepatitis C. Taking it was “a piece of cake.” And after three months of treatment, the virus was cleared from his body at last.

“I had a birthday in September,” Dr. Rubens, 63, said. “I told my wife I don’t want anything. It would take away from the magnitude of this gift.”

Medicine may be on the brink of an enormous public health achievement: turning the tide against hepatitis C, a silent plague that kills more Americans annually than AIDS and is the leading cause of liver transplants. If the effort succeeds, it will be an unusual conquest of a viral epidemic without using a vaccine.

“There is no doubt we are on the verge of wiping out hepatitis C,” said Dr. Mitchell L. Shiffman, the director of the Bon Secours Liver Institute of Virginia and a consultant to many drug companies.

Over the next three years, starting within the next few weeks, new drugs are expected to come to market that will cure most patients with the virus, in some cases with a once-a-day pill taken for as little as eight weeks, and with only minimal side effects.

That would be a vast improvement over current therapies, which cure about 70 percent of newly treated patients but require six to 12 months of injections that can bring horrible side effects.

The latest data on the experimental drugs is being presented at The Liver Meeting in Washington, which ends Tuesday.

But the new drugs are expected to cost from $60,000 to more than $100,000 for a course of treatment. Access could be a problem, particularly for the uninsured and in developing countries. Even if discounts or generic drugs are offered to poor countries, there are no international agencies or charities that buy hepatitis C medications, as there are for H.I.V. and malaria drugs.

And some critics worry that the bill will be run up when huge numbers of people who would have done fine without them turn to the drugs. That is because many people infected with hepatitis C never suffer serious liver problems.

“The vast majority of patients who are infected with this virus never have any trouble,” said Dr. Ronald Koretz, emeritus professor of clinical medicine at the University of California, Los Angeles. It is impossible to tell in advance whether an infected individual will go on to suffer serious consequences. For patients who can afford them, the temptation to take the new drugs before trouble arises will be powerful.

**A Heavy Toll**

An estimated three to four million Americans are infected with hepatitis C, and about 150 million worldwide — three to five times the number who have H.I.V. Most people who are infected do not know it, because it can take decades for the virus to damage the liver sufficiently to cause symptoms.

In the United States, the number of new infections has fallen to about 17,000 a year, from more than 200,000 per year in the 1980s, according to the Centers for Disease Control and Prevention. There has been a recent rise in cases among young people who inject pain medicines or heroin.

About 16,600 Americans had hepatitis C listed as a cause of death on death certificates in 2010, though that might vastly understate the mortality linked to the disease, according to the C.D.C. Although there are fewer new infections, the number of deaths is expected to keep rising as the infections incurred years ago increasingly take their toll.

Hepatitis C is spread mainly by the sharing of needles, though it can also be acquired during sex. The virus was transmitted through blood transfusions before testing of donated blood began in 1992. Dr. Rubens, the recently cured patient, believes he was infected when he worked as a paramedic long ago.

The main treatment has been interferon alfa, given in weekly injections for 24 or 48 weeks, combined with daily tablets of ribavirin. Neither drug was developed specifically to treat hepatitis C. The combination cures about half the patients, but the side effects — flulike symptoms, anemia and depression — can be brutal.

The new drugs, by contrast, are specifically designed to inhibit the enzymes the hepatitis C virus uses to replicate, the same approach used to control H.I.V. As with H.I.V., two or more hepatitis C drugs will be used together to prevent the virus from developing resistance.

One big difference is that H.I.V. forms a latent reservoir in the body, so H.I.V. drugs must be taken for life to prevent the virus from springing back. Hepatitis C does not form such a reservoir, so it can be eliminated permanently.

If no virus is detectable in the blood 12 weeks after treatment ends — a measure known as a sustained virologic response — there is almost no chance the virus will come back and the patient is considered essentially cured. The damaged liver can then heal itself somewhat, doctors say.
Yet even if the virus is cleared, people who were once infected may still have an increased risk of liver cancer, especially if cirrhosis, a scarring of the liver, has set in.

The new drugs now moving to market can achieve sustained viral responses in 80 to 100 percent of patients with treatment durations of 12 to 24 weeks, possibly shorter.

For Tom Espinosa, a building inspector in Oakland, Calif., the new treatments cannot arrive fast enough. Mr. Espinosa, 59, has advanced cirrhosis and some spots on his liver that might be cancer. He is so fatigued that he spends all weekend in bed. He has tried all available treatments and nothing worked, making him envious of other patients who were cured.

“I became resentful for a little while, but I got over it,” he said. With time possibly running out, he plans to try the first new drug to hit the market.

To be sure, many of the new drug combinations have not been extensively tested yet. Side effects might still show up. And the drugs are not expected to work as well for patients with severe cirrhosis or those co-infected with H.I.V.

“I just don’t think we know the answer until we get more widespread clinical experience,” said Charles M. Rice, a hepatitis C expert at Rockefeller University. “We may be in for some surprises still.”

New Direction
Researchers and patients have been disappointed before, when the first two direct-acting antiviral pills, telaprevir and boceprevir, reached the market in 2011. The drugs, which inhibited the virus’s protease enzyme, still required interferon and ribavirin, but they raised the cure rate to about 70 percent.

There was a huge rush to treatment. But doctors now say that side effects were worse than expected, in part because the sickest patients had been excluded from the clinical trials of the drugs.

“A lot of that didn’t come to light until after the drugs were approved,” said Dr. Brian R. Edlin, an associate professor of public health and medicine at Weill Cornell Medical College. “Then it turns out they were just horrible.”

Among the new drugs, the one garnering the most excitement is sofosbuvir, from Gilead Sciences, which is expected to be approved by the Food and Drug Administration by Dec. 8. It inhibits the virus's polymerase enzyme, which builds new genomes out of RNA so the virus can replicate.

Sofosbuvir is an evil decoy of sorts. It looks like a building block of RNA. But once it is mistakenly incorporated into the RNA chain, the chain cannot grow and the virus cannot reproduce.

The effectiveness of the new drugs can vary depending on which strain of hepatitis C, known as genotypes, the patient has.

People infected with hepatitis C genotypes 2 and 3 — which account for 20 to 25 percent of cases in the United States — will take sofosbuvir with ribavirin but without interferon, making this the first all-oral treatment for hepatitis C. Treatment for genotype 2 will be 12 weeks, but for genotype 3 it will probably be 24 weeks.

Genotype 1, which accounts for more than 70 percent of patients in the United States, will still require interferon and ribavirin along with sofosbuvir, but only for 12 weeks. In a clinical trial, about 90 percent of previously untreated patients taking this combination achieved a sustained virologic response. The combination is expected to be somewhat less effective in those for whom previous treatments did not work.

Gilead hopes to have an all-oral treatment for genotype 1 approved by the end of 2014. It would be a once-a-day pill containing both sofosbuvir and another experimental Gilead drug, ledipasvir. This combination, used along with ribavirin, is what cured Dr. Rubens.

Other companies, including AbbVie, Merck and Bristol-Myers Squibb, are in a heated race to also bring all-oral combinations to market in the next two years or so.

Liver specialists will be able to put together an all-oral regimen for genotype 1 very soon, however, by prescribing both sofosbuvir and simeprevir, a Johnson & Johnson protease inhibitor that is expected to win approval soon. One study has shown this combination to be extremely effective, though insurers may balk at paying for two expensive drugs.

Awaiting Better Options
These new drugs are likely to alter the calculus about who gets treated and when.

Many doctors are now “warehousing” their hepatitis C patients — urging them to forgo treatment until the new drugs are approved.

“There's no way I'm going to put them on an interferon regimen when we're a year away from having interferon-free regimens,” said Dr. Scott Friedman, the chief of liver diseases at the Icahn School of Medicine at Mount Sinai. “It's rare you have to pull the trigger and get them on treatment in that period of time.”
Gilead estimates that only 58,000 Americans with hepatitis C are now undergoing treatment, a small fraction even of those who know they are infected. Wanting to avoid interferon’s side effects, some patients without symptoms try to monitor their liver and start treatment only if it shows signs of deterioration.

But with the new more tolerable treatments, some experts say, it makes sense to treat early-stage disease to prevent cirrhosis and the accompanying risk of liver cancer.

And it is likely that more pre-symptomatic patients will be found through wider screening. Both the United States Preventive Services Task Force and the C.D.C. have recently begun to recommend that all baby boomers — people born from 1946 to 1964 — be tested for infection with hepatitis C, since they represent about three quarters of all cases.

“It will be test and treat,” said Dr. Eugene Schiff, the director of the liver diseases center at the University of Miami, who is a consultant to drug companies.

Pharmaceutical companies, of course, have a financial interest in seeing that more people get screened and treated, and they have been providing support for hepatitis C awareness campaigns and sponsoring studies on the benefits of screening and treatment.

The all-oral regimens also may make it more feasible to treat the people who are most likely to spread the virus — intravenous drug users, the homeless and prison inmates, many of whom also have mental health problems.

“I can’t treat an unstable patient safely with interferon,” said Dr. Diana Sylvestre, who runs a clinic in Oakland, Calif, that treats illicit drug users and former users. “But I can sure as hell give them a few pills.”

Rift widens over structure of HIV’s molecular anchor
Studies of a potential vaccine target bolster claims that an earlier paper was flawed.

Brendan Borrell
The structure of a protein known informally as the HIV trimer is among the most highly prized goals of structural biology.

To infect a human cell, HIV sports protrusions that hook on to the cell’s surface. If researchers could find a way to block this process, they might have a way to develop a long-awaited vaccine. But the architecture of this molecular harpoon, which is called envelope glycoprotein and known informally as the HIV trimer, has been the centre of controversy since the summer, when scientists questioned the most precise description of it ever published. Now three studies have been published, two of them today, all of which agree with one another and differ from that earlier analysis, leading to calls for the paper to be retracted.

“I give no weight to the previous paper,” says structural biologist Marin van Heel of Leiden University in the Netherlands. “They are throwing up a lot of smoke screens, not releasing the data, and not retracting the paper. The next step is due with these new structures.”

The lead author of the earlier study, structural biologist Youdong Mao of the Dana-Farber Cancer Institute in Boston, Massachusetts welcomes the new data but says it has no bearing on the validity of his study. “Our team is firmly standing behind our earlier work,” he says.

Small wonder
Refining the three-dimensional architecture of HIV’s surface proteins with increasing resolution has been a goal for the past decade, as such knowledge can help to guide vaccine design. Because these proteins are inherently unstable, structural biologists seeking to visualize them have had to introduce stabilizing mutations and to freeze the structures in place using liquid nitrogen. This imaging technique is called cryo-electron microscopy (cryo-EM).

The dispute over the proteins’ architecture began in early June, when Mao and his colleagues published cryo-EM reconstructions at a resolution of 6-ångströms in Proceedings of the National Academy of Sciences. According to their study, the tip of the spike seemed to have a cavity at its centre.

However, several scientists, including Sriram Subramaniam at the National Cancer Institute in Bethesda, Maryland, disagreed with the results, claiming that no virus particles were even present in the ten raw images that the team later shared. “There was nothing to see,” says Subramaniam, “It was like ‘Where’s Waldo?’ Where’s the particle?”

According to structural biologist Richard Henderson at the MRC Laboratory of Molecular Biology in Cambridge, Mao’s group seem to have obtained their results by averaging more than 5,000 grainy images and aligning them to a lower-resolution reference model. Although the reference method is widely used in cryo-EM, it has the potential to create the illusion of a particle where none exists.
In a Perspective subsequently published in PNAS, Henderson pointed out that one can reconstruct Einstein’s face by aligning 1,000 images of white noise. He suspects that such an unintentional error crept in here, although Mao, in his team’s published response, wrote that they “took specific measures to avoid reference bias”.

However, because Mao and colleagues have not shared their entire dataset and methods with their critics, it is impossible to know exactly what was going on. “We cannot yet prove categorically that their work is nonsense,” says Henderson, “but I personally have no doubt that it is.”

**New data**

On 23 October, Subramaniam and his colleagues published their own cryo-EM-based reconstruction of the surface proteins in Nature Structural and Molecular Biology, revealing a very different architecture.

Rather than finding a cavity at the centre of the spike, they identified three helices (corkscrew-shaped structures) concealed within the spike in its inactivated state. When the surface proteins make contact with the surface of a cell from the human immune system, the outer components of the spike swivel around these helices and prepare for invasion in a manner strikingly similar to that seen in influenza viruses.

Now two further studies, published online today in Science, have used cryo-EM and X-ray crystallography to elucidate the surface proteins in even greater detail. Their results bolster the findings by Subramaniam’s team.

“This is a frame shift in how we think about vaccine design,” says structural biologist Andrew Ward at the Scripps Research Institute in La Jolla, California, an author on both of the Science studies. “Now that we have a better view of what the immune system sees, we can make better hypotheses and designs.”

In an email to Nature, Mao wrote that he is glad to see the new data coming out, but argues that the differences in the structures are simply due to different ways the molecules were engineered for imaging, not a flaw in their technique. “A full understanding of the [glycoprotein] structure likely will require scientist to capture all different ‘snapshots’ under different contexts and ‘connect’ them together.”

As a consequence of the controversy, structural biologists hope that, as with genetic sequence data, the publication of raw micrographs will become standard. Subramaniam, for instance, has released all 4,713 of his images, an unprecedented move. “We need to learn from this,” says van Heel, “and see how we can improve deposition in databases for better quality control.”

**Intestinal Bacteria Linked to Rheumatoid Arthritis**

Nov. 5, 2013 — Researchers have linked a species of intestinal bacteria known as *Prevotella copri* to the onset of rheumatoid arthritis, the first demonstration in humans that the chronic inflammatory joint disease may be mediated in part by specific intestinal bacteria. The new findings by laboratory scientists and clinical researchers in rheumatology at NYU School of Medicine add to the growing evidence that the trillions of microbes in our body play an important role in regulating our health.

Using sophisticated DNA analysis to compare gut bacteria from fecal samples of patients with rheumatoid arthritis and healthy individuals, the researchers found that *P. copri* was more abundant in patients newly diagnosed with rheumatoid arthritis than in healthy individuals or patients with chronic, treated rheumatoid arthritis. Moreover, the overgrowth of *P. copri* was associated with fewer beneficial gut bacteria belonging to the genera Bacteroides.

"Studies in rodent models have clearly shown that the intestinal microbiota contribute significantly to the causation of systemic autoimmune diseases," says Dan R. Littman, MD, PhD, the Helen L. and Martin S. Kimmel Professor of Pathology and Microbiology and a Howard Hughes Medical Institute investigator.

"Our own results in mouse studies encouraged us to take a closer look at patients with rheumatoid arthritis, and we found this remarkable and surprising association," says Dr. Littman, whose basic science laboratory at NYU School of Medicine’s Skirball Institute of Biomolecular Medicine collaborated with
clinical investigators led by Steven Abramson, MD, senior vice president and vice dean for education, faculty, and academic affairs; the Frederick H. King Professor of Internal Medicine; chair of the Department of Medicine; and professor of medicine and pathology at NYU School of Medicine.

"At this stage, however, we cannot conclude that there is a causal link between the abundance of *P. copri* and the onset of rheumatoid arthritis," Dr. Littman says. "We are developing new tools that will hopefully allow us to ask if this is indeed the case."

The new findings, reported today in the open-access journal *eLife*, were inspired by previous research in Dr. Littman's laboratory, collaborating with Harvard Medical School investigators, using mice genetically predisposed to rheumatoid arthritis, which resist the disease if kept in sterile environments, but show signs of joint inflammation when exposed to otherwise benign gut bacteria known as segmented filamentous bacteria.

Rheumatoid arthritis, an autoimmune disease that attacks joint tissue and causes painful, often debilitating stiffness and swelling, affects 1.3 million Americans. It strikes twice as many women as men and its cause remains unknown although genetic and environmental factors are thought to play a role.

The human gut is home to hundreds of species of beneficial bacteria, including *P. copri*, which ferment undigested carbohydrates to fuel the body and keep harmful bacteria in check. The immune system, primed to attack foreign microbes, possesses the extraordinary ability to distinguish benign or beneficial bacteria from pathogenic bacteria. This ability may be compromised, however, when the gut's microbial ecosystem is thrown off balance.

"Expansion of *P. copri* in the intestinal microbiota exacerbates colonic inflammation in mouse models and may offer insight into the systemic autoimmune response seen in rheumatoid arthritis," says Randy S. Longman, MD, PhD, a post-doctoral fellow in Dr. Littman's laboratory and a gastroenterologist at Weill-Cornell, and an author on the new study. Exactly how this expansion relates to disease remains unclear even in animal models, he says.

Why *P. copri* growth seems to take off in newly diagnosed patients with rheumatoid arthritis is also unclear, the researchers say. Both environmental influences, such as diet and genetic factors can shift bacterial populations within the gut, which may set off a systemic autoimmune attack. Adding to the mystery, *P. copri* extracted from stool samples of newly diagnosed patients appears genetically distinct from *P. copri* found in healthy individuals, the researchers found.

To determine if particular bacterial species correlate with rheumatoid arthritis, the researchers sequenced the so-called 16S gene on 44 fecal DNA samples from newly diagnosed patients with rheumatoid arthritis prior to immune-suppressive treatment; 26 samples from patients with chronic, treated rheumatoid arthritis; 16 samples from patients with psoriatic arthritis (characterized by red, flaky skin in conjunction with joint inflammation); and 28 samples from healthy individuals.

Seventy-five percent of stool samples from patients newly diagnosed with rheumatoid arthritis carried *P. copri* compared to 21.4% of samples from healthy individuals; 11.5% from chronic, treated patients; and 37.5% from patients with psoriatic arthritis.

Rheumatoid arthritis is treated with an assortment of medications, including antibiotics, anti-inflammatory drugs like steroids, and immunosuppressive therapies that tame immune reactions. Little is understood about how these medications affect gut bacteria. This latest research offers an important clue, showing that treated patients with chronic rheumatoid arthritis carry smaller populations of *P. copri*. "It could be that certain treatments help stabilize the balance of bacteria in the gut," says Jose U. Scher, MD, director of the Microbiome Center for Rheumatology and Autoimmunity at NYU Langone Medical Center's Hospital for Joint Diseases, and an author on the new study. "Or it could be that certain gut bacteria favor inflammation."

The researchers plan to validate their results in regions beyond New York, since gut flora can vary across geographical regions, and investigate whether the gut flora can be used as a biological marker to guide treatment. "We want to know if people with certain populations of gut bacteria respond better to certain treatment than others," says Dr. Scher. Finally, they hope to study people before they develop rheumatoid arthritis to see whether overgrowth of *P. copri* is a cause or result of autoimmune attacks.

**Journal Reference:**
Discovery of HIV 'Invisibility Cloak' Reveals New Treatment Opportunities
Nov. 6, 2013 — Scientists have discovered a molecular invisibility cloak that enables HIV, the virus that causes AIDS, to hide inside cells of the body without triggering the body's natural defence systems.

Their study shows how 'uncloaking' the virus using an experimental drug triggers an immune response that stops the virus from replicating in cells grown in the laboratory.

The findings could lead to new treatments and help to improve existing therapies for HIV infection.

The innate immune system is the body's first line of defence against infection and incorporates an alarm system present in all cells of the body that detects the presence of 'foreign' material from invading bacteria and viruses. When the alarm is tripped, the infected cell begins an anti-viral programme and sends out warning signals to alert other cells that a virus is around. HIV infects vital cells of the immune system so its ability to replicate undetected without triggering this alarm system has puzzled scientists since the discovery of the virus.

The team identified two molecules inside host cells that are recruited by HIV after infection that stop the virus from reproducing its genetic material too early. The effect is to shield the virus from the alarm system and stop the innate immune system from kicking into action.

In the absence of these molecules, either by depletion from infected cells or blocking their recruitment using an experimental drug, HIV is exposed to the alarm system and an anti-virus immune response is triggered. Targeting the cloaking molecules and not the virus itself makes it much more difficult for the virus to mutate and become resistant to this treatment approach, a significant problem with standard HIV therapies.

Professor Greg Towers, a Wellcome Trust Senior Research Fellow at UCL and lead author of the study, said: "HIV is extremely adept at hiding from our body's natural defences, which is part of the reason the virus is so dangerous. Now we've identified the virus' invisibility cloak, and how to expose it, we've uncovered a weakness that could be exploited for new HIV treatments.

"There's a great deal more research needed but the potential for this approach is huge, as a possible treatment in itself but also as a complement to existing therapies. We're also interested to see whether blocking these cloaking molecules can help to boost immune responses to experimental vaccines against HIV or be used to protect against HIV transmission.

"The hope is that one day we may be able develop a treatment that helps the body to clear the virus before the infection is able to take hold."

The experimental drug used in the study is based on Cyclosporine, a drug that is widely used to prevent organ rejection in transplant patients because of its ability to dampen the immune response. Cyclosporines have been shown to block the replication of HIV and other viruses but are not suitable for treating infected patients because of their negative effects on the immune system. The team used a modified version of the drug, which blocks the effects of the two cloaking molecules without suppressing immune activity.

Dr Kevin Moses, Director of Science Funding at the Wellcome Trust said: "In 2012, 2.3 million people were newly infected with HIV. Whilst existing treatments are helping people with HIV to live longer and healthier lives, the challenge of adherence to treatment programmes means that drug resistance remains a threat and the virus continues to burden the world's poorest communities. Understanding how HIV interacts with the body's own defences might just be crucial for developing the best approaches to therapy."

Journal Reference:

Challenges to Assumptions About Bisexual Men, HIV Transmission
Nov. 6, 2013 — The number of HIV positive men who have sex with both men and women is likely no higher than the number of HIV positive heterosexual men, according to a U.S.-based analysis by University of Pittsburgh Graduate School of Public Health researchers. The finding challenges a popular assumption that bisexual men are responsible for significant HIV transmission to their female partners.

The research, which will be presented at the American Public Health Association's 141st Annual Meeting & Exposition in Boston, builds a case for federal investment in research on HIV prevalence among bisexualy behaving men.

"Some observers have exaggerated the idea of viral 'bridging'—where a bisexual man contracts HIV from another man and then transmits it to a female partner. But, at least in the U.S., the data supporting
the extent of this is quite limited," said Mackey R. Friedman, Ph.D., M.P.H., of Pitt Public Health’s Department of Infectious Diseases and Microbiology, who led the research.

Currently, the U.S. Centers for Disease Control and Prevention (CDC) does not report on HIV data specific to bisexually behaving people, though it does report data on homosexually and heterosexually behaving people, as well as injection drug users.

Dr. Friedman and his colleagues reviewed over 3,000 scientific articles to obtain data on HIV prevalence and risks among men who have sex with men only and men who have sex with men and women.

The bisexually behaving men were only 40 percent as likely to be infected with HIV as the homosexually behaving men. The researchers propose that this is because the bisexually behaving men reported lower rates of unprotected receptive anal intercourse, the biggest risk factor for HIV transmission among men in the U.S.

The analysis also estimates that there are approximately 1.2 million bisexual men in the U.S., of whom 121,800 are HIV-positive. That estimate aligns with CDC estimates for HIV infection in male heterosexuals and intravenous drug users.

Dr. Friedman, who has conducted HIV prevention and research for more than 15 years, believes that while bisexually behaving men may have a lower risk profile than homosexually behaving men, their HIV burden still warrants the development of targeted interventions.

"The HIV infection risk that bisexual men pose to their female partners has likely been overstated," said Dr. Friedman. "However, that doesn’t mean that HIV-prevention campaigns targeting bisexual men and their male and female partners aren’t needed. HIV does exist in the bisexual community, and national, bisexual-specific data collection, research, and HIV prevention and care delivery are necessary to ameliorate this population’s HIV burden."

**HIV/Aids holy water cure put to the test**

DURBAN—The Treatment Action Campaign wants the so-called 'holy water' to go under the microscope.

Rebirth Family Church’s Bishop Hamilton Nala claims he can cure HIV with water that he sells for at least R10.

Five of his followers have come forward claiming the remedy works, but activists are now calling for clinical tests.

Bishop Nala is embroiled in controversy after claiming to heal HIV/AIDS using holy water.

Activism group the Treatment Action Campaign wants Nala’s miracle remedy tested in a clinical trial.

"We are calling for a thorough investigation because we have seen people saying Bishop Nala has prayed for me and I am healed," said TAC’s Mzamo Zondi.

But the priest says the tests won’t make a difference to his followers.

“They must say medically there is no cure for Aids. I agree with that, but through prayer and through faith water and any material branded my name, as God said prophetically, I believe in that. People can be healed of Aids," said Nala.

The TAC has warned that if the so-called holy water fails the clinical trials, it will push for Nala to be arrested.

"Nala must be charged, Nala must be arrested, and anyone who is claiming to heal HIV without medication that has gone to clinical trials and has gone via the Medicines Control Council. That one must be arrested," said Zondi.

Meanwhile, the bishop has asked his followers not to get him into trouble with the law by continuing to take their ARV medication as well as his miraculous water.

November 4, 2013

**Sangamo BioSciences Presents Clinical Data From SB-728-T HIV Study Demonstrating Long-Term Immune Reconstitution and Reduction in the HIV DNA Reservoir In Subjects with Long-Term Infection**

**Data Demonstrate ZFN-dependent CCR5 Modification of Long-lived Central Memory and Stem-like T-cells**

RICHMOND, Calif., Nov. 4, 2013 /PRNewswire/ — Sangamo BioSciences, Inc. (Nasdaq: SGMO) announced today the presentation of clinical data from its program to develop a ZFP Therapeutic® for HIV/AIDS. The data demonstrate a reduction in the HIV DNA reservoir since receiving SB-728-T in seven of nine HIV-infected subjects on long-term anti-retroviral therapy (ART), despite a median duration of
HIV infection of 21 years and baseline CD4 T-cell counts prior to SB-728-T treatment of < 500 cells/µl. The data were presented at a translational medicine meeting organized by The Lancet and Cell publications and entitled, "What Will it Take to Achieve an AIDS-Free World?" which is being held in San Francisco, November 3-5, 2013.

"These data are very promising and demonstrate the important link between SB-728-T treatment and immune reconstitution of central memory and total CD4 T cells and lower HIV DNA levels in subjects at 12 months," said Rafick-Pierre Sekaly, Ph.D., Co-Director & Chief Scientific Officer, The Vaccine & Gene Therapy Institute of Florida (VGTI Florida), who presented the data and whose laboratory carried out the immunologic analyses.

ART successfully suppresses viral load in most subjects, but a significant proportion, so called immunologic non-responders (INR), do not restore normal CD4 counts above 500 cells/µl. HIV-infected subjects who were classified as INR, enrolled in Sangamo’s ongoing SB-728-902 clinical trial (Cohorts 1-3), received a single infusion of SB-728-T which resulted in a durable increase in total CD4 T-cells associated with increases in ZFN-modified CD4 central memory T-cells, specifically memory and stem cell-like cells (TSCM) which provide a long-lasting source of CCR5-modified CD4 T-cells. Notably, despite the increase in total CD4 T-cells, a long-term decrease in the peripheral blood mononuclear cell (PBMC) HIV reservoir was observed, as measured by proviral DNA using a sensitive assay technique.

"Our goal is to develop an immunologic approach to HIV that should enable the patient’s immune system to attack HIV infection at the level of both the HIV DNA viral reservoir and circulating viral load, and our data suggest that we are seeing these effects in SB-728-T treated subjects," stated Geoff Nichol, M.B., Ch.B., Sangamo’s executive vice president of research and development. "In previous clinical studies, a decline in the HIV reservoir in subjects on long-term ART has not been observed. In addition, any increase in the levels of CD4 cells in HIV-infected subjects is often associated with a concomitant increase in the size of the reservoir. In contrast, a single SB-728-T treatment of subjects on long-term ART produced a significant and durable improvement in CD4 count and, in the majority of subjects, a notable decrease in the HIV reservoir over time. In addition to data on reduction of the viral reservoir, we recently presented data at the Annual Meeting of the European Society of Gene and Cell Therapy (ESGCT) that demonstrated sustained ongoing control of viral load at or below the level of detection of HIV through 14 weeks in an SB-728-T- treated HIV-infected subject who was not on ART."

Dr. Nichol continued, "Immunologic analyses suggest that SB-728-T treatment protects long-term central memory CD4 T-cells from HIV-infection. We have also identified key immunologic markers of inflammation that correlate with the degree of engraftment and can potentially aid in the selection of subjects for which SB-728-T may be most effective. In our ongoing studies, we will continue to investigate these parameters including the threshold engraftment levels of biallelically modified T-cells and the types of HIV-reactive cells necessary to mount an immune response to the virus. We look forward to presenting the results of our SB-728-902 Cohort 5 and SB-728-1101 studies in December 2013, at the Sixth International Workshop on HIV Persistence, Reservoirs and Eradication Strategies."

Summary of Clinical Data Presented at the Lancet Meeting

SB-728-0902 Cohorts 1-3 (INR)
Subjects in this trial, so-called INR, have lower than expected total CD4 counts ( < 500 cells/µl) despite their HIV infection being well controlled by ART.

We observed that:
- Treatment of HIV subjects with a single infusion of SB-728-T leads to long-term increases in CD4 counts (up to 3 years in some subjects)
- Long-term increases in CD4 counts correlate with increased CD4 central memory, memory stem cells (TSCM)-like and increased ZFN CCR5 protected central memory T-cells
- Seven of nine subjects show decreased levels of HIV DNA at Month 12 post infusion
- An inflammatory environment pre-infusion appears to be an important predictor of CD4 reconstitution post-infusion
- Higher levels of inflammation correlate with poor immunological response and suggest a mechanism for altered survival of CM cells

Summary of Clinical Trial Design

About SB-728-902 Cohorts 1-3
The study is an open-label Phase 1 clinical trial to evaluate the safety and tolerability of single infusions of an escalating dose of an autologous (a patient’s own) CD4+ T-cell product genetically modified at the CCR5 gene by CCR5-specific ZFNs (SB-728-T). The trial enrolled nine HIV-infected subjects (three
cohorts of three subjects each) who have sub-optimal T-cell levels and no detectable viral load on long-term ART. Subjects remained on their existing antiviral therapy while receiving treatment with SB-728-T.

Updated: Nov. 5, 2013, 12:46 p.m. ET

**The HIV rebound nobody is discussing**

Is an AIDS-free generation on the horizon? Not without the help of sex workers and other marginalized "key populations," public health officials say. The world needs more prostitutes like Hawa Abdallah. At least that’s what many public health officials believe.

There’s something about the 25-year-old—the harsh angle of her eyebrows, the way her red lipstick contrasts with the dirty walls, the cold stare she directs at passing men—that says she’s in control. Abdallah has sex with as many as 20 men every day in a brothel in Dar es Salaam, Tanzania’s largest city, where 1 in 20 residents and 1 in 3 sex workers are infected with the virus that causes AIDS.

But a series of simple steps have kept Abdallah healthy until now: She knows how HIV spreads. She is tested regularly. And she’s confident enough to insist that her customers use condoms every time. Just as importantly, she’s managed to avoid the assault, arbitrary arrest and extortion that plague the profession. Health officials say that unless more prostitutes start fitting a similar profile, the world doesn’t stand a chance at beating back the virus.

It’s not just sex workers, of course. The same applies to injecting drug users, men who have sex with men, inmates and other groups in which HIV rates have moved against the global trend, and continue to rise. Which is why experts with organizations as diverse as the World Health Organization and Human Rights Watch agree that government officials must do more to support these “key populations”—no matter how illegal their activities may be—if they want to see an AIDS-free generation within their borders anytime soon.

Halting the spread of HIV among these key populations comes down to a series of calculated risks for both governments and the individuals themselves. Abdallah’s calculations started several years ago.

She never intended to spend her nights negotiating in the dark with a never-ending stream of drunks. She wanted to be a hair stylist. But the young woman quickly discovered that no matter how busy her beauty salon became, no matter how many hours she put in cutting and braiding hair, she rarely earned more than $20 per day—not nearly enough to support her two young children and a long-term boyfriend who scrapes by as a motorcycle taxi driver.

Her first night in the brothel, she made close to $60—three the amount in a fraction of the hours. Her boyfriend discovered what she was doing and tried to make her to stop. “I have refused,” she said. “It’s worth the risk.”

Among the most immediate of those risks: It’s illegal. Tanzanian law states clearly that men and women caught “loitering for the purposes of prostitution” can be locked up for three months. Male sex workers can be charged with “carnal knowledge against the order of nature” and punished with a minimum of 30 years and a maximum lifetime sentence. Officials consider both prostitution and homosexuality to be threats to the social order.

But of course, those same officials have also been preoccupied with a far more serious societal threat in recent years. When HIV began ravaging Africa, making its population the most-infected on the planet in the last decades of the 20th century, the virus struck Tanzania particularly hard. By the mid-90s, close to 1 in 10 Tanzanians were dying; a generation of children were orphaned; local economies buckled. And for years, the Tanzanian government lacked the resources to respond in force.

The situation changed to some extent in the early 2000s when the newly formed Global Fund to Fight AIDS, Tuberculosis and Malaria and the U.S. President’s Emergency Plan for AIDS Relief, among other groups, began funneling billions toward the cause. At long last, Tanzanian officials—like many of their neighbors in sub-Saharan Africa—had both bragging rights and the numbers to back them up. Treatment coverage jumped from 3.5 percent in 2005 to 55 percent in 2011. The number of health facilities providing HIV care and treatment services rose from a few dozen in 2004 to about 1,200 in 2012. Mother-to-child transmission of the virus plummeted. People started living longer.

With similar results coming in from around the world—including one from UNAIDS showing a 50 percent drop in new infections in more than 25 low- and middle-income countries over the previous decade—former Secretary of State Hillary Clinton stood before a crowd in Washington in 2012 and declared that “as we continue to drive down the number of new infections and drive up the number of people on treatment, we will get ahead of the pandemic, and an AIDS-free generation will be in sight.”
But optimism wasn’t running quite so high in the back alleys and brothels of the world—including Abdallah’s. While HIV prevalence among the general population has decreased to 5 percent in Tanzania, it’s on the upswing globally among “key populations” at the greatest risk for contracting and passing HIV, according to an August 2013 report from the Foundation for AIDS Research, or amfAR.

In fact, HIV prevalence is 22 times higher among people who inject drugs. In low- and middle-income countries, men who have sex with men and female sex workers are 19 and 13.5 times more likely, respectively, to have HIV than their peers. Many researchers have long downplayed the rates in key populations, believing they represent only a “modest share of the epidemic globally” and represent a major concern only in countries with low-level epidemics. But amfAR contends that members of these groups are critical because they are often part of “dense, high HIV-prevalence social and sexual networks” that help the disease spread rapidly from one population to another.

Consider 22-year-old Abdul Rashid. After stumbling into a community resource center several miles from Abdallah’s brothel in Dar es Salaam, Rashid—still strung out on marijuana and cocaine—was tested for HIV. The results came back quickly: Positive. And he wasn’t a bit surprised.

Rashid enjoys having sex with female prostitutes at a brothel near his drug den. But the premium he pays for sex without a condom, combined with the high price of drugs, long ago left him in such a financial bind that he started having sex with men for cash. To top it off, police also recently arrested him for selling drugs and threw him in one of the local jails—notorious places for sex among inmates. Within the last few months alone, Rashid may have spread the disease widely within all four of the primary “key populations” in Tanzania. If those individuals then have sex with their partners—who are often not a part of the high-risk groups—the virus then spills into the general population.

Sitting in a filthy orange T-shirt on the floor of the drug center, Rashid seemed too dazed to care. “I have no option to change my status, so I must say, ‘OK, if that’s the case now, I have nothing to do,’” he said.

Of course, this situation isn’t unique to Tanzania. Key populations and their sex partners account for as much as 51 percent of new infections in Nigeria, 33 percent in Kenya, 80 percent in Morocco and 47 percent in the Dominican Republic. And according to some estimates, men who have sex with men alone could make up more than half of all new infections in Asia by 2020, amfAR reports.

“Sex workers do not have a place to speak against injustices done to them ... If they go to the police, the police just become their customers for that night.”

“Unless effective strategies are put in place to mitigate the HIV burden in key populations, the global epidemic will worsen over time, preventing the world from realizing the dream of an AIDS-free generation,” the group warned in its August report.

That brings us back to Tanzania’s dilemma—similar to the one faced by most countries where prostitution is illegal. Should officials enforce the law of the land or facilitate safer testing and treatment for prostitutes in the name of public health?

When drafting their “Strategic Framework on HIV and AIDS” several years ago, the government decided to take a calculated risk and “acknowledge the vulnerability of sex workers and men who have sex with men.” The document advocated for their access to HIV prevention information and services and, surprisingly, “for decriminalization of their activities.” Despite the worries of some that it would come across as a tacit endorsement of illegal activities, the officials decided to push ahead with this approach because the potential gains in the HIV fight could be huge.

And it’s worked, to a limited extent. A report published in June by Human Rights Watch found that “a few state hospitals and some nongovernmental organizations throughout the country” have succeeded in providing friendly services to the “most at-risk populations.” Through its health agencies, the government has also supported the outreach efforts by local governments and nonprofits. Even so, this limited success is “systematically undermined” by police officers who abuse their authority and often make things worse, the report concludes.

“The Tanzanian government has committed on paper to reduce the stigma for at-risk groups, but that commitment is meaningless if the police regularly rape, assault, and arrest them,” concluded Neela Ghoshal, the author of the Human Rights Watch paper. “The government’s HIV policy can’t succeed if police are driving away the very people the public health programs most need to reach.”

A survey by the country’s National AIDS Control Program in 2010 found that a full third of sex workers in Dar es Salaam reported being beaten by their customers, but few felt comfortable reporting the crimes to police. When Human Rights Watch interviewed 66 men, women and children who were current or former prostitutes, at least 23 said the police had forced them into sex—often in exchange for release.
from custody. Some of those police officers refused to use condoms, “making the police possible conduits for transmission of HIV and other STIs.”

As a public health worker in the city of Mwanza told the group, “Sex workers do not have a place to speak against injustices done to them ... If they go to the police, the police just become their customers for that night.”

The situation is not much better at government health centers. The same Human Rights Watch report identified dozens of cases in which “health workers turned away sex workers and other key populations from health facilities, or publicly humiliated them.”

That includes a case where a drug addict went for treatment after being attacked by a mob in Dar es Salaam and was denied anesthesia while the staff stitched up his wound. “I asked for it, and the nurse said, ‘We don’t need to. We are going to sew you without. We could inject you with poison rather than with anesthesia.’” In another example, a gay man in the semi-autonomous region of Zanzibar asked to be treated for a sexually transmitted disease and was told to leave. “You already have sex with men,” a staff member told him. “Now you come here to bring us problems. Go away.”

Experiences like those create an atmosphere of fear and distrust that pushes these key populations so far underground, they become almost impossible to reach with HIV prevention messages, testing or treatment. “And that’s woeful,” said Dr. Ade Fakoya, a senior advisor to the Global Fund to Fight AIDS, Tuberculosis and Malaria. “Because it’s coverage of these very basic, essential interventions which is key to the death of the epidemic.”

Some organizations have managed to break through. As the sun sank a little lower one recent evening, some health workers partnering with the nonprofit Public Services International began setting up camp in the courtyard of Abdallah’s brothel.

She sat several feet away, watching as they erected an Army-green burlap tent and pulled out tables, pamphlets, rubber gloves and HIV test kits.

Because it was still early, and because the truck drivers and local drunks hadn’t started spilling from the nearby bars, many of the prostitutes were relaxing on the stoops outside their rooms. Some had begun drinking to brace for the hours ahead. Others played with their children.

About 150 women between the ages of 15 and 49 spend their nights in this particular brothel—a small fraction of the 7,000 plying the streets of Dar es Salaam—and most seemed to know the drill. When one emerged from behind the green flaps of the tent, another stood and entered for her check-up.

“We’re not afraid of these tests,” Abdallah said, unconvincingly. “I’ve done this before. So have they.”

To prove it, she clutched the sides of her black floral purse—embroidered with the word “Sweet”—a little more tightly and headed into the tent.

Program Manager Shahada Kinyaga watched her from a distance. “It wasn’t this easy in the beginning,” she said. “When we first approached them, they were thinking maybe we were police officers looking to exploit them or that we wanted to publicize their activity to get them into trouble. They wouldn’t let us come near.”

To change that, the PSI workers took the time to speak with a single sex worker, sharing health information and returning repeatedly. Eventually, that sex worker started sharing bits of the knowledge with her friends in the brothel—how to get free condoms, for example, and how to tell if a customer is infected with syphilis or herpes. In time, the women allowed PSI to hold workshops on building “negotiation skills” to convince their customers to wear condoms and to set up the HIV testing tent. Those diagnosed as HIV-positive began listening to the health workers about follow-up care at local clinics, and reported back about whether they were receiving the appropriate treatment. Bottom line: It’s not that the women didn’t want help—it was a matter of who to trust.

“So, more of them are doing this now,” Kinyaga said. “They are testing more regularly. They are using condoms more. Some of them don’t do it 100 percent of the time, but it’s a step.”

Inside the tent, Abdallah’s black-and-white striped pantsuit stood out starkly in the diffused light filtering through the burlap. She looked like a stylish prisoner in a war camp. She also looked worried.

These moments of dread—at waiting for test results—are part of the calculated risk Abdallah accepted from the beginning. But she tries to tell herself that if she plays the game well, if she insists that every one of her customers uses a condom every single time, if she tests regularly and receives follow-up treatment when needed, she stands a good chance of living to see her two children grow old. Within a few years, she’ll also have enough money in the bank to build a home and open her own beauty parlor. That endgame is why she’s here in the first place—why she says, “This is a good environment for me.”

Within 30 minutes, her test results are ready. Her face is stone-cold and will remain that way no matter the verdict. Abdallah knows that as long as she works in this brothel, the uncertainty will linger.
The greatest allergy myths and misconceptions, debunked
Baltimore, MD. (November 7, 2013) — From gluten allergy and hypoallergenic pets, to avoiding the flu shot because of an egg allergy, there are a lot of common myths and misconceptions about allergies. Many might be shocking due to a great deal of false information in the media and on the Internet. And some of the misconceptions can be damaging to your health if vaccinations are skipped and extreme dietary avoidances are taken.

But where did all of these misconceptions come from? According to a presentation being given at the Annual Scientific Meeting of the American College of Allergy, Asthma and Immunology (ACAAI), previously held beliefs from medical experts and public perception are partially to blame.

"Many early medical beliefs have been proven to be incorrect as research has advanced," said allergist David Stukus, MD, ACAAI member and presenter. "Unfortunately, some of these beliefs are still on the Internet, where an astonishing 72 percent of users turn to for health information."

In his presentation, Dr. Stukus outlined some of the greatest allergy myths, and explained why they are false.

1. I’m Allergic to Artificial Dyes – There is no scientific evidence to support a link between exposure to artificial coloring and allergies. Controversy exists regarding evidence for artificial coloring and behavioral changes in children, as well as dyes causing chronic urticaria and asthma.

2. I Cannot Have Vaccines Due to an Egg Allergy – Egg embryos are used to grow viruses for vaccines such as the flu, yellow fever and rabies shots. However, it’s now safe to get the flu shot, which can help prevent serious illness.

3. At-Home Blood Tests Reveal All You’re Allergic To – These tests might be able to reveal sensitization, but being sensitized to a certain allergen, like milk, doesn’t mean you’re allergic. These sort of at-home screening tests are not reliable and can often lead to misinterpretation, diagnostic confusion and unnecessary dietary elimination.

4. Highly Allergenic Foods Shouldn’t be Given to Children until 12 Months of Age – For most children, there is no evidence to support avoidance of highly allergenic foods past four to six months of age. New evidence emerging shows that early introduction of highly allergenic foods may promote tolerance.

5. I’m Allergic to Cats and Dogs, but Can Have a Hypoallergenic Breed – Unfortunately, there is no such thing as a truly hypoallergenic dog or cat. Allergens are released in saliva, sebaceous glands and perianal glands. It’s not the fur people are allergic to. It is true that some breeds are more bothersome for allergy sufferers than others.

6. I’m Allergic to Shellfish and Cannot Have Iodine Imaging – Radiologists and cardiologists often use iodinated contrast during CT scans and other procedures for better imaging. Since shellfish contain iodine, many physicians have linked a contrast reaction to a shellfish allergy. However, this is false, and a shellfish allergy has nothing to do with the reaction. In fact, iodine is not and cannot be an allergen as it found in the human body.

7. I Can’t have Bread, I’m Allergic to Gluten – You can have a gluten intolerance, but it’s extremely rare to have a true allergy. Most allergic reactions to these foods stem from wheat. Many people self-label as having gluten allergy and avoid gluten without any medical indication.

With information being widespread online via social media portals, how do you know what to believe and what not to believe?

“If you think you may have an allergy, you should see a board-certified allergist for proper evaluation, testing, diagnosis and treatment,” said Dr. Stukus. “Misdiagnosis and inappropriate treatment can be dangerous.”

Blocking the Active Site of Thiolase
Nov. 7, 2013 — Scientists at the University of Oulu, Finland, and at the Helmholtz Center Berlin (HZB) have shown the way to new directions in drug development against African sleeping sickness and other tropical parasitic infections. This was based on the structural analysis of the enzyme thiolase, which plays a central role in lipid metabolism in the parasite that causes sleeping sickness. The researchers examined the biomolecule’s structure at the MX beamline of electron storage ring, BESSY II, at the HZB.

Sleeping sicknesses—african trypanosomiasis, kala-azar, indian leishmaniasis—are infections caused by tropical parasites. Millions get sick from them each year and thousands end up dying. Anti-parasitic drugs are expensive and often have a host of unwanted side effects. In decades, there have been no new
effective therapies. Reason enough for the World Health Organization (WHO) to consider research, which can lead to the development of new anti-parasitic drugs, a top priority.

Now, Prof. Rik Wierenga and his team at Oulu University have paved the way for this type of research by shedding light on the structure of the enzyme thiolase. Thiolase figures prominently in parasitic lipid metabolism. According to Wierenga, "key is knowing the geometry of the enzyme's active site. This is the place where lipids that play a central role in parasitic metabolism attach and where chemical reactions that convert lipids into other substances take place." Which is why it's important to investigate the active site's structure and function: "It enables us to develop lipid-like substances that firmly attach to the active site and block it." The molecules that are involved represent the ideal starting points for new drug development. Studies at BESSY of the enzyme thiolase have yielded a highly detailed image of thiolase's active site. "We now have a much clearer idea of thiolase's role in all this," says Wierenga. "It would appear that the enzyme catalyzes the first step in the sterol biosynthesis pathway, which is important in a number of parasites."

"The measurements of crystalline thiolase proteins we obtained at our MX beamline has helped to unravel the active site's geometry," says HZB's own Dr. Manfred Weiss. One particular region of the protein called the HDCF loop turns out to be key. The structure, which lies deep within thiolase's interior, was previously unknown. "Understanding the HDCF loop is the ideal starting point for the development of new anti-parasitic drugs," adds Wierenga.

Journal Reference:

Autoantibodies Found in Blood Years Before Symptoms of Autoimmune Disease

Nov. 5, 2013 — Autoantibodies are present many years before symptom onset in patients with primary Sjögren syndrome, an autoimmune disease, according to a Research Letter published in the November 6 issue of JAMA.

Primary Sjögren syndrome is a disease in which immune cells attack and destroy glands that produce tears and saliva. Autoantibodies are characteristic of this syndrome and may be involved in its development. Roland Jonsson, D.M.D., Ph.D., of the University of Bergen, Norway, and colleagues measured autoantibodies before symptom onset in these patients.

All patients with primary Sjögren syndrome at Malmo University Hospital in Malmo, Sweden, have been included in a registry since 1984. Controls were randomly selected from biobanks and matched by sex, age, and date of earliest sampling (within 60 days before or after) to each case.

Of 360 cases in the registry, 44 (41 women and 3 men) provided 64 presymptomatic serum samples obtained an average of 7 years before symptom onset. In 29 cases (66 percent), autoantibodies were detected before symptom onset. All 29 cases had autoantibodies in their earliest available serum sample, as early as 18 years before symptom onset.

"To our knowledge, this is the first systematic investigation of presymptomatic autoantibodies in Sjögren syndrome. Most cases produced autoantibodies many years before clinical onset of the disease; the median [midpoint] time of 4 to 6 years is an underestimate because all seropositive cases had autoantibodies in their earliest available serum sample," the authors write.

"Autoantibody profiling may identify individuals at risk many years before disease onset. However, the significance of these presymptomatic autoantibodies for determining prognosis and treatment remains to be determined."

Journal Reference:

How Do Viruses Avoid Inhibition by Endogenous Cellular MicroRNAs?
Bryan R. Cullen

Introduction
MicroRNAs (miRNAs) are an extensive family of small regulatory RNAs that function by binding to complementary mRNAs, primarily in the 3’ untranslated region (3’UTRs), and then inhibiting their expression by reducing mRNA translation and/or stability [1]. MiRNAs are initially transcribed as long pri-miRNAs, which are sequentially processed by the RNase III enzymes Drosha, in the nucleus, and
Viruses and MicroRNAs

Upon infection of a cell, viruses encounter a wide range of miRNA species, generally more than 50 different miRNAs per cell, and these miRNAs vary greatly between tissues. For example, miR-122 is expressed at very high levels in hepatocytes, but is absent from almost all other cells, while miR-1 is primarily expressed in muscle tissue and miR-128 in neuronal cells [3]. Also, many of the more than 1000 known human miRNA species show a tissue-specific expression pattern [4]. The mechanism of avian leukosis viruses to avoid the miR-122-mediated inhibition in their normal target tissues is currently unclear. Several possible mechanisms can be proposed:

1. **Viruses block miRNA function.** This appears rare, as virus-infected cells generally contain normal levels of miRNAs, and most viruses can be inhibited by specific small interfering RNAs (siRNAs), which function indistinguishably from miRNAs in mammalian cells [5], or by insertion of target sites for endogenous cellular miRNAs into viral transcripts [6–9]. Indeed, the use of inserted cellular miRNA target sites as a way of inhibiting viral replication in tissues that express the cognate endogenous miRNA, while allowing unhindered viral replication in cells that lack this miRNA, has considerable potential in facilitating the development of novel attenuated viral vaccines or in targeting oncolytic viral vectors away from normal tissues [7–9]. Uniquely, in the case of poxviruses, it has been shown that miRNAs are degraded in infected cells [10]. In contrast, HIV-1 and influenza viruses, despite early reports to the contrary, have now been clearly shown to not block miRNA function [11, 12], and indeed, the tissue and/or species tropism of influenza virus can be readily manipulated by insertion of target sites for endogenous miRNAs [11].

2. **Viruses evolve to avoid 3′UTR targets complementary to cellular miRNAs.** Because full complementarity to the seed is generally critical for miRNA inhibition, single nucleotide mutations should block inhibition [13]. However, for viruses that can replicate in several different tissues, each expressing more than 50 distinct miRNAs, complete avoidance of all miRNAs may be very difficult to achieve. Nevertheless, especially for viruses that display a narrow tissue tropism, this mechanism seems very likely to be important.

3. **Viruses evolve very short 3′UTRs.** RISC recruitment to open reading frames (ORFs) does not effectively inhibit mRNA function, most probably because translating ribosomes sweep bound RISCs off the mRNA [12]. Therefore, miRNAs with very short 3′UTRs would be expected to be relatively refractory to miRNA-mediated inhibition. In fact, many RNA viruses express mRNAs bearing short 3′UTRs, and the short 3′UTRs that are present are often highly structured, which is predicted to also inhibit RISC binding.
RNA virus families that appear likely to use this strategy to avoid inhibition by endogenous cellular miRNAs include flaviviruses, picornaviruses, rhabdoviruses, and reoviruses.

4. **Viruses evolve structured 3′ UTRs.** Some viruses, especially retroviruses, alphaviruses, and coronaviruses, contain extensive 3′ UTRs in at least some viral mRNA species. For example, the HIV-1 mRNA that encodes Gag and Gag-Pol has a 3′ UTR that is several thousand nucleotides in length. Similarly, the coronavirus mRNA encoding the viral ORF1a and ORF1b proteins has a 3′ UTR more than 10,000 nucleotides in length. How do these very long 3′ UTRs avoid functioning as targets for multiple miRNAs? One possibility is that these 3′ UTRs have evolved high levels of RNA secondary structure, which would be predicted to globally restrict binding by miRNA-programmed RISCs [4].

Relatively little is known about the secondary structure of viral RNAs, although some data suggest that high levels of secondary structure are a common feature [23]. One viral RNA that has been examined in detail is the HIV-1 RNA genome, which also functions as the mRNA for the viral Gag and Gag-Pol proteins. This RNA has been shown to fold into an extensive secondary structure with relatively few areas that are unfolded and hence, presumably, are available for RISC binding [24]. This prediction has been validated by a comprehensive analysis of the susceptibility of the HIV-1 genome to small interfering RNAs (siRNAs), which in mammalian cells function indistinguishably from miRNAs [16]. These researchers generated over 9,000 siRNAs specific for the HIV-1 genome by sliding the siRNA target along the viral genome by one-nucleotide increments [25]. Relatively few of these siRNAs were found to inhibit HIV-1 replication and gene expression effectively, and those that did were predicted to bind to the few regions of the viral RNA genome that, using biochemical approaches, were predicted to adopt an open, unfolded conformation [24], [25].

Recently, the ability of the HIV-1 genome to bind to endogenous cellular miRNAs in relevant target cells (CD4+ T cells) or in a non-physiological target cell (HeLa cells) has been examined using a technology called photo-activatable ribonucleoside-enhanced crosslinking and immunoprecipitation (PAR-CLIP). The PAR-CLIP technique involves pulsing cells with the highly photoactivatable uridine analog 4-thiouridine and then crosslinking endogenous RNAs to bound proteins by irradiation at 365 nm [26]. Crosslinked proteins, and the bound RNAs, are recovered by immunoprecipitation of RISC using an Ago2-specific monoclonal antibody and the binding site footprinted by RNase treatment. The RISC binding sites are then comprehensively identified by deep sequencing of these small RNAs to generate sequence clusters that can be aligned to endogenous miRNA species.

Analysis of RISC binding to the HIV-1 genome indeed identified several binding sites that were occupied by RISCs programmed by endogenous cellular miRNAs and some of these could be shown, by indicator assays, to confer a modest repression of mRNA function [27]. However, perhaps the more interesting finding was that viral mRNAs, despite contributing more than 10% of the total mRNA transcriptome in HIV-1 infected cells, in fact gave rise to only approximately 0.2% of all assignable RISC binding sites, with the remaining approximately 99.8% being contributed by cellular miRNAs. That is, viral mRNAs are, at a minimum, 50-fold less likely to bind RISC than are cellular mRNAs, consistent with the idea that HIV-1-encoded mRNAs, at least, have evolved to globally avoid cellular miRNAs by adopting RNA secondary structures that preclude RISC binding.

While a more complete understanding of the interaction of viruses with cellular miRNAs must await a more detailed dissection of the effect of endogenous miRNAs on a wide range of viral species, current data suggest that viruses have likely evolved a number of strategies to avoid inhibition by these ubiquitous cellular regulatory RNAs. Whether the perturbation of these avoidance strategies has the potential to lead to the development of reagents that are useful in disease prevention, such as novel forms of attenuated viral vaccines, remains to be determined.

**An Unfolding Tragedy of Chagas Disease in North America** (long)

Peter J. Hotez mail, Eric Dumonteil, Miguel Betancourt Cravioto, María Elena Bottazzi, Roberto Tapia-Conyer, Sheba Meymandi, Unni Karunakara, Isabela Ribeiro, Rachel M. Cohen, Bernard Pecoul

In North America, Chagas disease (American trypanosomiasis caused by *Trypanosoma cruzi*) was first reported in Mexico in 1940 [1] and in the United States in Texas in 1955 [2]. However, based on ancient mummified remains discovered in the Rio Grande Valley, human *T. cruzi* infection has been present in North America since prehistoric times [3].

*T. cruzi* is a protozoan hemoflagellate that is most commonly transmitted to humans by blood-feeding triatomine bugs followed by autoinoculation [2]. Chagas disease can also be transmitted to man by non-vectorial mechanisms, namely mother-to-child-transmission [4], blood transfusion, and orally through...
food-borne transmission. When untreated in the acute stage, the disease becomes chronic and up to 30% or more of infected individuals will progress to Chagasic cardiomyopathy or megavisceral disease associated with debilitating morbidity or death. Today, Chagas disease is a leading cause of heart disease among people living in extreme poverty in the Western Hemisphere, especially in Latin America, where it is a major parasitic killer [2].

The established link between poverty and Chagas disease transmission derives largely from poor-quality housing that facilitates triatomine bug invasion, together with lack of access to adequate health care and antenatal care. Additional factors related to poverty also include lack of health education and environmental management leading to vector invasion and colonization [5]. Despite enormous strides made in Chagas vector control, through housing improvements and aggressive insecticidal spraying, and case reduction or even elimination in parts of Latin America [6], important areas of high endemicity persist, including in North America. Confirmatory data are scarce, but, according to some preliminary estimates, Mexico ranks number three, and the United States number seven, in terms of the number of infected individuals with Chagas disease in the Western Hemisphere, where 99% of the cases occur [2]. In Mexico, an earlier national seroprevalence survey reported a rate of 1.6% [7]. However, other reports have provided alternative estimates ranging between 1.0% and 5.9% (i.e., between one to six million cases nationwide) [1, 2]. In the U.S., approximately 300,000 cases are believed to be present [2], although one alternative estimate reports more than 250,000 cases in Texas alone [8], with up to one million or more cases nationwide (Figure 1) [9]. Thus, together with several thousand cases in Canada, there are between 1.5 million (lower estimate) and 7 million (highest estimate) cases of Chagas disease among the 500 million people living in North America. Clearly, there is a need for active surveillance data in order to better refine these prevalence data.
Equally troubling are suggestions that 40,000 pregnant North American women may be infected with *T. cruzi* at any given time, resulting in 2,000 congenital cases through mother-to-child transmission [4]. According to the Pan American Health Organization, congenital transmission may currently account for more than one-quarter of the world’s new Chagas disease cases [2], [6]. While North America appears to have a high disease burden resulting from *T. cruzi* infection, in actuality we know remarkably little about the epidemiology of Chagas disease in this region. No systematic effort has been undertaken to ascertain the true national prevalence of Chagas disease in Mexico, even though some isolated communities are now withering because between 30% and 50% of their populations are infected with *T. cruzi* [10]–[12]. In addition, Chagas disease is the most common cause of dilated cardiomyopathy in some areas of southern Mexico [13], and a significant percentage of mothers are infected with *T. cruzi* [14]. According to one estimate the overall economic losses from Chagas disease in Mexico may exceed $3 billion annually [8].

Similarly, in the U.S., no effort to conduct widespread surveillance testing has ever been organized in the states considered at highest risk of *T. cruzi* infection, and, while most of the human cases are presumed to have been introduced through immigration [2], it is known that triatomine vectors infected with *T. cruzi* are present throughout southern Texas and elsewhere in the U.S. [15]. Ironically, we may know more about canine Chagas disease transmission, which is widespread in Texas [16]. While seven autochthonous vector-transmitted human cases in the U.S. had been reported previously [2], the screening of blood donors initiated in 2007 has led to the identification of 16 additional cases [17], but many more may exist, especially given the pervasive ignorance among physicians and other health-care providers on how to recognize and diagnose Chagas disease and how it is transmitted [2], [18]. To underscore the lack of awareness among clinicians, the first congenital case of Chagas disease in the U.S. was reported in 2012, more than 50 years after the first documented case [19].

Alongside the gaps in surveillance is the dearth of health-care facilities offering diagnosis and treatment of Chagas disease in the U.S. Currently, we believe that Los Angeles and Houston have two of the largest clinics for diagnosing and treating the disease. Until recently, treatment of Chagas with benznidazole was thought to be safe and effective only for newborns and infants. The experience of Doctors Without Borders/Médecins Sans Frontières in Latin American countries has shown that
treatment with benznidazole is indeed possible—and safe—for children and adults alike, provided regular medical checks are performed [20]. Such activities complement similar, concurrent efforts to evaluate Chagas disease therapies [21, 22]. It is thus imperative to scale up access to benznidazole treatment while simultaneously investing in research for new diagnostics, drugs, and vaccines.

Similar to other endemic countries in Latin America where scaling up appropriate diagnosis and treatment remains a major challenge for Chagas disease patients, the need to address and reduce the burden of Chagas disease in North America is substantial: In Mexico, implementing a national program of surveillance and disease burden assessment is urgently needed, in addition to sustaining vector control efforts, providing accessible programs of case management, and implementing treatment in the most affected and impoverished areas of Mexico (i.e., Chiapas, Guerrero, Jalisco, Oaxaca, Veracruz, and the Yucatan peninsula) [1]. Without such actions, and in the absence of vector control, the Chagas disease burden could double in 25 years [8]. Moreover, in our experience, the waiting list for benznidazole (one of two drugs for the antiparasitic chemotherapy of Chagas disease) can be excessive – therefore a program to accelerate access to this essential medicine needs to be implemented. All pregnant women in Chagas disease-endemic regions of Mexico should be tested for T. cruzi infection and offered treatment after delivery and/or after completing breastfeeding. All infants with congenital Chagas disease need to be treated; a new pediatric dosage form of benznidazole developed by the Drugs for Neglected Diseases initiative (DNDi) is now available. In addition, children born during previous pregnancies should also be screened and treated once a mother is identified as infected with T. cruzi, as suggested for northern Argentina [23], while overall, new and improved algorithms are needed to address more widespread testing such as testing on women of child-bearing age that migrate from endemic to non-endemic areas.

In the U.S., increased support for federal, state, and local public health agencies is needed to implement a similar program of surveillance, disease burden assessment, and treatment programs in Texas and other southern states, and for some urban communities with large populations of immigrants from Chagas disease-endemic areas. The barriers [24] and cost-effectiveness of immigrant screening, including screening of pregnant women [25, 26], have been noted. Equally urgent is the need to assess the risks of transmission in disease-endemic areas, looking at vectors and reservoir host distributions [1], and to tease out which cases might be transmitted from within the borders of the U.S. There is an urgent need for more, and better, data on human infection in areas where infected vectors and reservoirs have been identified. Regulatory issues surrounding the use of benznidazole in the U.S. must be addressed to provide expanded treatment options. The U.S. is also faced with a health care workforce that now obtains very little, if any, formal training in medical parasitology, so that, at best, physicians are only modestly aware of how to recognize the signs and symptoms of Chagas disease, let alone identify at-risk individuals, obtain clinical testing, and begin appropriate care for patients [2, 18]. Such lack of awareness is especially acute among obstetricians caring for T. cruzi-infected expectant mothers [2, 4].

Finally, health research in Mexico needs to be prioritized based on disease burden estimates [27], while research capacity for Chagas disease across North America needs to be expanded to develop or improve current treatments. We desperately require improved drugs, as the two currently available to treat Chagas disease are of limited efficacy in some clinical situations, have undesirable adverse events, and are contraindicated in pregnancy so that we have nothing to offer pregnant women at risk of passing their T. cruzi infection onto their unborn fetuses [28]. Neither benznidazole, nor nifurtimox, is approved by the U.S. Food and Drug Administration. DNDi is currently developing a new azole drug candidate that may have a better safety profile, with clinical testing in progress. Concurrently a therapeutic Chagas vaccine is under development by a consortium of Mexican (including the Carlos Slim Health Institute) and Texan scientific institutions [29]. We also need more sensitive diagnostics that can be used at point-of-care, and biomarkers to assess therapeutic response and progression of cardiac disease in the setting of T. cruzi infection [2]. Without new tools, disease control will not be possible for Chagas disease, and the U.S. and other governments have a major role to play in ensuring innovation—from basic research to late-stage product development—is funded and carried out.

The poorest people living in the Mexico and the U.S. are silently suffering under a heavy burden of Chagas disease, with pregnant women disproportionately affected. We can save many lives with greater access to the treatments available today, while knowing the fate of tomorrow’s patients rests on increasing investments in research to develop new technologies to treat and diagnose Chagas disease, as well as improving scientific cooperation between the U.S., Canada, Mexico, and other key countries. Ultimately, scientific breakthroughs for new technologies to fight Chagas disease will be the cornerstone of efforts to control this ancient scourge in North America and beyond.
Understanding the Persistence of Plague Foci in Madagascar

Voahangy Andrianaivoarimanana, Katharina Kreppel, Nohal Elissa, Jean-Marc Duplantier, Elisabeth Carniel, Minoarisoa Rajerison,

Ronan Jambou

Abstract

Plague, a zoonosis caused by Yersinia pestis, is still found in Africa, Asia, and the Americas. Madagascar reports almost one third of the cases worldwide. Y. pestis can be encountered in three very different types of foci: urban, rural, and sylvatic. Flea vector and wild rodent host population dynamics are tightly correlated with modulation of climatic conditions, an association that could be crucial for both the maintenance of foci and human plague epidemics. The black rat Rattus rattus, the main host of Y. pestis in Madagascar, is found to exhibit high resistance to plague in endemic areas, opposing the concept of high mortality rates among rats exposed to the infection. Also, endemic fleas could play an essential role in maintenance of the foci. This review discusses recent advances in the understanding of the role of these factors as well as human behavior in the persistence of plague in Madagascar.

Introduction

Plague is a flea-borne fatal zoonosis caused by the bacillus Yersinia pestis. Primarily a disease of rodents and fleas, it has been responsible for three pandemics resulting in millions of deaths [1]. Despite advances in its control and understanding, plague is still endemic in Asia, the Americas, and Africa. It is also reemerging in countries where the disease was thought to have disappeared [2], [3]. Civil wars, urbanization, deforestation, and mining may also have an impact on the disease.

Worldwide, bubonic plague is the predominant form and is acquired after a fleabite. The bacteria multiply at the site of inoculation and disseminate via the lymphatic system to the lymph nodes. After two to six days, a painful swelling lymph node appears (the bubo), along with high fever, headache, dizziness, and prostration. Without treatment, the infection rapidly disseminates to reach the spleen, liver, and sometimes the lungs, causing a fatal septicaemia. Without treatment, lethality occurs in 40–70% of the patients. Pneumonic plague is rare but even deadlier. It may arise from a bubonic form, by haematogenous spread to the lungs, or from inhalation of aerosols during human-to-human transmission. After one to three days of latency, the onset is sudden and always fatal without early efficient treatment. Here, we review different factors that may explain how the disease is able and continue to persist in Madagascar.

Methods

The review of the literature was conducted using the online databases PubMed and HINARI. A thorough search was then undertaken in Madagascar from earlier works to recent findings, including dissertations and unpublished reports from the Ministry of Health and Institut Pasteur de Madagascar (which hosts the plague Malagasy reference center), with particular emphasis on plague dynamic. Altogether, documents cover almost 50 years of plague studies in Madagascar.

Brief Overview of Plague Epidemiology

Within the Enterobacteriaceae family, the genus Yersinia includes three human pathogenic species: Yersinia enterocolitica, Yersinia pseudotuberculosis, and Y. pestis, the causative agent of plague [1]. Although, Y. pestis and Y. pseudotuberculosis differs radically in their virulence and transmission route, they share a high genetic homology. Y. pestis diverged from Y. pseudotuberculosis within the last 20,000 years [4].

Twenty-five hundred species and subspecies of Siphonaptera are described but only 80 of these are known to be susceptible to Y. pestis [5], among which the genus Xenopsylla (especially Xenopsylla cheopis) plays a major role in pandemics. Fleas of this genus are found in all domestic and peridomestic settings where humans are at risk of infection with Y. pestis due to its high vector efficiency and broad host preference [6]. In sub-Saharan regions and in rural areas of Brazil and India, Xenopsylla brasiliensis is the predominant vector for plague [6]. Other species, like Xenopsylla astia (Indonesia, Southeast Asia) and Xenopsylla vexabilis (Pacific Islands) are also important vectors [7]. The fleas specificity to rodent hosts vary from one specific host to a broad affinity: in the northern United States, Oropsylla hirsuta parasitizes a species of prairie dogs, Cynomys ludovicianus [6], while in Zimbabwe, the four major rodent species Gerbilliscus leucogaster, Rattus rattus, Rhabdomys pumilio, and Mastomys natalensis are all hosts of X. brasiliensis [8].

The high vector efficiency of X. cheopis is reported to be related to its ability to get "blocked," which increases the transmission potential of Y. pestis. The bacterium produces biofilm required for proventricular blocking [5] of the flea leading to an increased biting rate and regurgitation of bacteria into
the wound. Partial biofilm blockage is sufficient to assure transmission, as for *Oropsylla montana* (Baker) in the United States [9].

Around 200 species of rodents and lagomorphs have been connected to the epidemiology of plague so far [6], but only few are considered significant hosts [10]. Frequency of contact between human and host varies depending on the species. *R. rattus* is a tree dwelling species nesting often in the roof of huts, whereas *Rattus norvegicus* is a ground dweller, preferably living in sewer networks of large towns. *Rattus* spp. are the major reservoir of plague in parts of Asia and Africa, especially in Madagascar [3], [11], [12]. Its population dynamics determine plague dynamics [13], [14]. Other rodents are locally involved in plague epidemiology such as the great gerbil (*Rhombomys opimus*) in Kazakhstan or the black-tailed prairie dog (*C. ludovicianus*) and the ground squirrel (*Spermophilus beecheyi*) in the United States [10].

Environmental conditions modulate seasonal transmission and global distribution of plague [15], [16]. In Asia and the United States, epidemics occur at the end of winter when rodents leave their burrows after hibernation. In other foci, seasonality in the abundance of rodents is less obvious and flea dynamics seem more important to take into account. Fleas, especially immature stages, developing in host burrows are sensitive to air temperature and humidity [17] and thus are affected by soil moisture in rodent burrows. Larvae are susceptible to desiccation [15], and their survival varies inversely with air dryness. Hot and dry days also reduce blockage in fleas [17], and low temperatures delay bacterial proliferation and early-phase transmission by *X. cheopis* [18].

**Plague in Madagascar: A Long History**

Plague arrived in the port city of Toamasina with steamboats from India in 1898 [19]. It then spread to other harbors and reached the central highlands in 1921 following the construction of the railways. It invaded the central highlands while disappearing progressively from the coasts.

From 1957 to 2001, a total of 20,900 suspected human cases were declared with an increase in the number of districts affected. Fortunately, over the years the case fatality rate decreased from 55.7% to 20.9% [20]. Still in 2004, 1,214 cases and 98 deaths were reported, but since then the incidence of human plague cases has declined continuously. However, Madagascar still accounted for 30% of human cases worldwide from 2004 to 2009 [21].

Nowadays plague is endemic in rural areas of the central highlands above 800 metres of altitude. The northern plague focus is located around the Tsaratanana Mountains (Figure 1). Additionally, plague has emerged more in the north at Ambilobe in 2011 (unpublished data) between the northern foci and Antsiranana.
From 2007 to 2011, bubonic plague accounted for 86.6% of suspected cases while pneumonic and undocumented cases accounted for 9.4% and 4%, respectively. The case fatality rate was 13% for suspected cases and 18.6% for confirmed cases. Reports of pneumonic plague cases were limited to the highlands and most often evolved from bubonic plague.

**The Bacterium-Reservoir-Flea Triad in Madagascar**

In Madagascar, all *Y. pestis* strains belong to the biovar Orientalis, which spread all over the world during the third pandemic. Isolates can be subdivided into four ribotypes (B, Q, R, and T), of which the most common is B, the original invading strain, while the three others are specific to Madagascar [22]. Antibiotic-resistant *Y. pestis* strains were also first isolated in Madagascar with one strain resistant to eight different antibiotics, including those used for plague prophylaxis and therapy [23].

Thirteen genera of Siphonaptera (four of them endemic) were described in Madagascar [24]; of those, two are involved in plague transmission: *Xenopsylla* and *Synopsyllus*. The main vector is *X. cheopis*, which parasitizes black rats living inside houses (Figure 2). The endemic genus *Synopsyllus* is composed of five species, among which *S. fonquerniei* is the most prevalent (Figure 2). It can be found in the fur or burrows of black rats living outside houses but also in open biotopes (rice fields, savannas) and in forests. This species is involved in the plague cycle above 800 metres of altitude and shows greater transmission efficiency than *X. cheopis* [25]. It also parasitizes endemic hedgehogs, rodents, and occasionally a species of lemur and insectivores.
Yet *R. rattus* remains the main plague reservoir host in Madagascar (Figure 2). Its arrival is closely linked to the history of the colonization of the island by humans [26]. The black rat is the dominant rodent species and is found everywhere: in houses, villages, fields, and also in the forests [19], [26]. Its populations can expand rapidly as it can breed inside houses all year round with an average gestation period of only 21 days and a mean litter size of 5.4 (in Madagascar). Conversely, *R. norvegicus* is limited to large towns since the 1950s, but is currently spreading on the western side of the island.

**Main Factors Impacting the Epidemiology of Plague in Madagascar**

**Rural versus Urban Foci**

In Madagascar, plague is predominantly a rural disease [21] related to agricultural activities. In the highlands, there is a hot and rainy season from October to April, followed by a cold and dry season. Harvesting occurs from February to June in dry-farming areas and in May in rice fields (in some places a second rice harvesting may occur in December). Maximum abundance of rodents in the fields is observed in July and August, followed by the maximum abundance of fleas from September to November (see [7] for more details). Villages provide three distinct habitats: houses located on top of hills, sisal hedges around livestock enclosures, and irrigated rice fields in lower areas (Figure 3A). Habitat choice and population dynamics of rodents are mainly driven by the availability of resources [14]. High plague transmission to humans has been associated with low abundance of rats and an increase in flea vectors [20]. This low number of rats is due to food shortages and an interruption of reproduction of outside rat populations during the cold season [26]. Conversely during rice harvest, an increase in reproductive rate and migration from houses to sisal hedges [14] are associated with low plague transmission to humans (Table 1). These factors are impacted by climate mediated by the availability of food and shelter.

---

Figure 2. Main vectors and rodent reservoirs in Madagascar.

Fleas involved in plague transmission in Madagascar: *Synopsyllus fonquerniei* female (1) and *Synopsyllus fonquerniei* male (3) are found on outdoor rats, whereas *Xenopsylla cheopis* female (2) and *Xenopsylla cheopis* male (4) live on indoor rats. Rat species involved in plague transmission in Madagascar: *Rattus rattus* (5) and *Rattus norvegicus* (6). doi:10.1371/journal.pntd.0002382.g002
Figure 3. Plague transmission cycle.

A) Plague cycle in the rural area of Madagascar. Rural plague foci of the highlands are organized into three habitats: houses (arrow), sisal hedges (arrowhead), and rice fields (star). The black rat, *R. rattus* (9), is the main rodent involved in transmission associated with *X. cheopis* (1) and the endemic flea *S. fonquerniei* (2). (Photo of plague foci: S. Rahelinirina). B) Plague cycle in the urban areas of Madagascar. Urban plague occurs mainly in the cities of Antananarivo (Isotry Market, left) (7) and Mahajanga (Abattoir suburb, right) (6). *R. norvegicus* (4) and *X. cheopis* (1) are involved in each focus. The Asian shrew (*S. murinus*) (5) has long been suspected to play a major role in the epidemiological cycle of plague in Mahajanga. C) Plague cycle in the forest area. A sylvatic transmission occurs in Madagascar with *R. rattus* (3) and endemic
Urban plague was mainly described in Mahajanga and Antananarivo (Figure 3B). The seaport of Mahajanga first experienced plague in 1902. A few human cases were reported between 1907 and 1928, but the town was free from plague for the next 60 years. A new outbreak occurred in 1991, followed by subsequent epidemics from 1995 to 1998 during which 1,702 suspected cases were reported [27]. In the capital Antananarivo, outbreaks of human plague were first recorded in 1921 [19]. After 58 years of silence, the disease reemerged in the city in 1979 with sporadic cases. Rodent surveillance initiated in the 1990s documented the replacement of R. rattus by R. norvegicus in the town (Table 1), favored by the construction of modern houses and sewage networks [3]. These changes were associated with a decrease in contact between humans and rat fleas due to the behavior of R. norvegicus [3]. Additionally, a lower susceptibility of R. norvegicus to plague also limited the risk of fleas leaving dead rodents in search of a new host, thus reducing human plague cases in Antananarivo [28].

### The Role of Endemic Fleas and Climate on Plague Epidemiology in Madagascar

Outside temperature may strongly affect flea abundance, thus affecting spatial and temporal distribution of the disease [29]. In Antananarivo and the surrounding highlands, plague cases are mostly reported from the warm rainy season from October to April. Conversely in Mahajanga, outbreaks of human plague occurred during the dry and cool season (from July to November). However, despite distinct plague seasons, the lowest temperature recorded in these two places during transmission is between 17° and 22°C [27], which can impact flea development. In the highlands, S. fonquerniei is exclusively found on rats caught outdoors and shows a clear seasonal cycle, thriving in the middle and at the end of the dry and cold season suggesting its role in initiating human plague epidemics [20]. This finding is supported by laboratory experiments suggesting that the development rate of flea larvae increases with temperatures below 30°C, and decreases above it. Furthermore, high temperatures with low humidity or temperatures below 9.3°C decrease the survival of the immature stages of S. fonquerniei [29]. In contrast, X. cheopis, which is mostly found on rats caught indoors, remains at relatively high abundance throughout the rainy season.

### Rats' Susceptibility to Plague in the Highlands of Madagascar

The susceptibility of rats to plague is undeniable. However, resistant R. rattus and R. norvegicus were reported in Antananarivo, which could explain the absence of epizootics and the maintenance of plague in the city [28]. Furthermore, whereas all rats in plague-free areas are sensitive to the disease, populations in plague-endemic areas are composed of sensitive and very resistant rats [28], [30]. The same was previously described for R. pumilio and M. natalensis in South Africa [31]. Yet the immune response to infection may differ even for the same species of rat within the same endemic area [32]. Dispersion of
resistant rats with their fleas could support plague dissemination [14]. Ecology seems to support selection for resistance to plague as shown by genetic structure analysis of R. rattus populations in plague foci [33]. In Mandoto (peneplain area) no difference was found [33], whereas in Betafo (mountainous area) genetic differences were observed between rats from rice field populations compared to those from houses and sisal hedges [34]. This resistance seems to be passed on to offspring as also suggested for M. natalensis [31]. A 32-base pair deletion in the chemokine receptor 5 gene (CCR5) used by HIV-1 to enter cells has been proposed to confer resistance to HIV, smallpox, and plague infections [35]. Although experimental challenges with Y. pestis in normal and CCR5-Δ32 mice did not ascertain a protective role [35], [36], a unique substitution (H184R) in a region of the CCR5 gene was found to be more prevalent in resistant animals compared to susceptible ones and is more common in rats from plague foci than from plague-free areas [37]. Other genetic markers were investigated using an AFLP genome scan approach. Twenty-two loci have been identified that may be involved in the resistant phenotype of R. rattus found in the central highlands of Madagascar. Two loci were associated with plague infection outcome in experimentally challenged rats [38].

**Diversity of Reservoir Species**

Although much less frequent and documented, sylvatic plague (Figure 3C) occurs in Malagasy primary forests where invasive R. rattus and endemic small mammals coexist and can sustain transmission through endemic fleas [25]. Human cases were reported among hunters and charcoal burners in these areas [3]. Both susceptible rodents and highly resistant insectivores live in these forests. Several endemic sylvatic small mammals such as shrews (Oryzorictinae subfamily) and tenrecs (Tenrecinae subfamily) were found infected by Y. pestis in sylvatic foci. They carry nonconventional vectors like Paractenopsyllus spp., Tsaractenus sp., and Synopsyllus estrandaei (unpublished data.). This mix of susceptible and resistant competent host species and potent vectors offers an explanation for epizootics and human plague cases. Deforestation also plays a major role in the dissemination of sylvatic plague to humans as seen in the Ikongo district after the introduction of R. rattus into this biotope. Endemic insectivores and hedgehogs in the forest were found seropositive in anti-F1 antibodies and substantiated an intense circulation of plague in this locality [39]. In the urban setting of Mahajanga, the Asian shrew, Suncus murinus, is most likely involved in plague transmission (Table 1). The abundance of X. cheopis on these shrews before the onset of human plague [3], the isolation of Y. pestis strains from their spleens [27], and their high seroprevalence after an epidemic period strongly suggests their involvement in the plague cycle. However, this hypothesis is questioned by the observation that Y. pestis strains isolated from S. murinus had different pulsotypes from those isolated from humans, rats, and fleas during the same outbreak [3].

**Plague Persistence in the Soil**

During inter-epizootic periods, Y. pestis cannot be recovered from fleas, rodents, or any other host. Persistence of the bacteria in the soil was speculated in Iran and Madagascar [19], [40], [41]. Naive rodents may thus become infected by burrowing in contaminated soil (either via inhalation or ingestion), restarting a new cycle. Although the exact mechanism remains unclear, previous studies have demonstrated the survival of Y. pestis in soil for at least 24 days under natural conditions [42]. This was previously highlighted in 1963 by inoculation of guinea pigs with soil samples collected from burrows, containing remains of Meriones vinogradovi that had been dead from plague for 7–11 months [40]. This mode of persistence could explain inter-epizootic periods. However, the virulence of Y. pestis experimentally kept for one month in soil decreased considerably [19], and it was subsequently demonstrated that dry laterite highly inactivate Y. pestis [19]. Moreover, although Y. pestis may remain viable and virulent in soil, recent studies suggested that the transmission route by exposure of susceptible mice to Y. pestis–contaminated soil seems unlikely under natural conditions. Indeed, the infectious period was short-lived and the transmission efficiency is low [43].

**Human Behavior and Plague**

Migration, poverty, and cultural practices can all have an impact on the incidence of human plague in Madagascar. A recent detailed SNP and MLVA analysis of Y. pestis strains evidenced multiple transfers of Y. pestis isolates between the highlands and Mahajanga harbor [44]. These transfers were most likely human-mediated, by transportation of goods containing infected rats or fleas by trucks or cars. In remote villages, people often prefer visiting traditional healers instead of health centers, thus delaying the implementation of an effective antibiotic treatment. Funeral ceremonies also favor the rapid spread of pneumonic plague [7], [20]. Indeed, a practice specific to Madagascar is to bury people in family burial vaults and to perform ritual corpse exhumations from time to time (Famadihana). Onsets of plague cases during these ceremonies have been observed, suggesting that handling of potentially plague-infected...
corpses may reactivate the disease. The Ministry of Health therefore recommended respecting a seven-year period between death and exhumation of a plague victim, and before any transfer of a corpse from one village to another. However, no study has been performed to determine the survival time of Y. pestis in corpses.

Poverty associated with overcrowded dwellings is another factor favoring rapid transmission and disease outbreaks in urban settings [3]. In villages, storage of crops within houses to prevent robbery attracts rats and their fleas [3, 7]. Agricultural activities, deforestation, and bushfires also promote spread of rats and dissemination of plague.

Finally, the discontinuation of plague surveillance since 2006 (due to financial shortages) has contributed to the reappearance of plague in the capital's suburbs six years after the last reported case. Two human cases were recently confirmed there outside the plague season, and Y. pestis was isolated from the spleen of R. rattus. The rat population in this area showed a higher than usual flea index, increasing the risk of Y. pestis transmission to humans and confirming that the disease is not under control, threatening the urban area of Antananarivo.

Effective plague prevention and control programs require up-to-date information on the incidence and the distribution of the disease. In Madagascar, plague surveillance (in humans and rodents) is a key priority for the Plague National Control Program (PNCP), established in 1993. The main objective of the PNCP is to reduce mortality due to plague and especially the mortality rate associated with the pneumonic form (<10% of notified cases) [7]. Surveillance is conducted by the Central Laboratory for Plague (CLP) of the Ministry of Health and the Plague Unit of the Institut Pasteur of Madagascar, which are the only facilities able to confirm plague in the country. Human surveillance is based on compulsory notification by health centers and on the biological confirmation of all suspected cases by the CLP. Y. pestis resistance to antibiotics currently used in plague treatment is also registered. The responsibility of health centers is the early detection of cases using the rapid diagnostic test at the patient’s bedside to implement i) an appropriate treatment (streptomycin relayed by sulfonamide) for all suspected cases, ii) chemoprophylaxis (sulfonamide) for the contact population, and iii) the control of fleas [7]. The community is involved in passive surveillance of plague epizootics and rodent density.

Conclusion
This review highlights the complexity of the epidemiology of plague in Madagascar and the effort made by past and present investigators to understand the reasons for the continuous presentation of human plague cases. Recent advances in various scientific fields have shown that the main host reservoir, the black rat populations of the highlands, are 1,000 times more resistant to plague than those from the coast. This is probably due to selective pressure. Adaptation of the plague bacillus to local ecological conditions may have also occurred, as suggested by the emergence and spread of new Y. pestis ribotypes in the most active foci of the highlands. The endemic flea S. fonquerniei may also play a significant role in the onset of the human plague season, whereas X. cheopis would be involved in sustaining disease transmission during several months thereafter. These various factors, along with human features, make the plague situation quite specific in Madagascar and reinforce the need for better surveillance. However, many questions still remain unanswered and represent future important challenges.

Box 1. Key Learning Points
1. Madagascar is among the top three countries that reported the most human plague cases during the past 15 years.
2. Plague occurs mainly as a rural disease, but also as an urban epidemic and a sylvatic transmission involving endemic rodents and fleas.
3. Plague is endemic in highlands above 800 metres of altitude with R. rattus as the main rodent reservoir and X. cheopis and the endemic flea S. fonquerniei as potential vectors.
4. Multidrug-resistant Yersinia pestis was first isolated in 1995 in Madagascar.
5. In less than a century, R. rattus has developed a strong resistance to the disease in endemic plague foci and this capability has a genetic basis.
6. Specific Malagasy traditions contribute to the rapid spread of pneumonic plague.

Box 2. Key Papers in the Field

Building Scenarios for Eliminating and Eradicating Onchocerciasis, Lymphatic Filariasis, and Human African Trypanosomiasis

Onchocerciasis
Onchocerciasis control relies on the control of the *Simulium* spp. vectors and the administration of ivermectin to at least 65% of the at-risk population for many years [34]. Recent evidence indicates that mass treatment with ivermectin is not only a strategy for controlling onchocerciasis as a public health problem, but that it can also interrupt transmission and eliminate the parasite in endemic foci if high treatment coverage can be maintained for a decade or more, depending on the local epidemiological situation [35]. For the purpose of the EIC, the scenario to move toward elimination of onchocerciasis is based on the current strategy of community-directed treatment with ivermectin (CDTi), with coverage extended to all areas where there is local transmission, i.e., a nodule prevalence >5% (traditionally, interventions focus on areas with a nodule prevalence >20%), and sustained mass treatment up to demonstrated elimination in the entire focus. Thereafter, periodic epidemiological and entomological surveys as well as passive surveillance need to be maintained pending global elimination (i.e., eradication). Maintaining mass treatment for the required duration is a major challenge in regions with weak governance and health system capacity.

Lymphatic filariasis (LF)
With regard to LF, spectacular results have been achieved in many settings where traditionally high prevalence and disease burden have been reduced through concerted efforts in vector control and mass drug administration, relying on ivermectin in Africa and DEC in other regions, both now usually co-administered with albendazole [36]. LF control targets all areas where local transmission of LF has been detected (e.g., though surveys). While LF control is well advanced in many countries in Asia, the Pacific, and in the Western hemisphere, implementation is much slower in Africa where a range of countries still need to update epidemiological maps and establish national programs targeting the entire at-risk population [37]. In order to be successful, elimination programs need to achieve coverage of the at-risk population in excess of 65%, and maintain it for several years, depending on the local epidemiological situation, vector fauna, and other factors (often around six years) [38]. Thereafter, regular surveys and surveillance are required in order to detect recrudescence.

The EIC scenarios take into account that in areas where onchocerciasis and LF are co-endemic (mainly in Africa), close collaboration is required between the two elimination programs as both programs rely on ivermectin distribution by community volunteers, require post-treatment surveillance, and drug distribution for either program cannot come to conclusion—and thus surveillance cannot be started—if the other program is continuing ivermectin treatment in the same area. Considering the generally much longer time horizon of onchocerciasis control programs compared to LF control programs, the duration of the former will be the decisive factor in most areas. On the other hand, it must be noted that LF has been found to reemerge after near-elimination in certain areas [39]. Last, LF elimination is furthered in areas where it is transmitted by *Anopheles* spp. mosquitoes by efforts to control malaria with long-lasting insecticidal-treated bednets (LLITNs). Other forms of vector control might also play a role even if not designed and implemented for LF control [40].

Human African Trypanosomiasis (HAT)
The scenarios for HAT control focus on eliminating the parasites causing human disease rather than the vector (tsetse flies). Hence, case detection (active and passive) and treatment are the mainstay of the scenarios, with these efforts supported by targeted interventions to reduce vector density with a view of reducing transmission (e.g., using insecticide-treated targets and cattle) [41]. Scenarios also take into account that while in *T. b. gambiense*—endemic areas eradication is conceptually feasible (none or very limited animal reservoir), in *T. b. rhodesiense*—endemic areas the mere concept of eradication is questionable as *Glossina* spp. (vector) elimination would need to be achieved due to the presence of extensive animal reservoirs, including in wild animals [42]. Thus, the scenarios for *T. b. gambiense* will aim at "eradication" while those for *T. b. rhodesiense* will aim at "elimination in humans." As a
consequence of focusing on the parasite causing human disease and neither trypanosomes in general nor the tsetse flies transmitting them, the economic benefits of elimination and eradication will chiefly result from improved public health and reduced suffering rather than increases in livestock production and improved agricultural opportunities.

**HIV may be becoming less fit as it adapts to the immune system**

Gus Cairns  
Published: 08 November 2013

HIV, at least in some parts of the world, may be developing a lower replicative capacity as it adapts to variations in the human immune system, studies in southern Africa and elsewhere suggest.

Philip Goulder of the University of Oxford told the AIDS Vaccine conference last month that competition between HIV and certain varieties of human HLA (human leukocyte antigen) genes may be contributing to a diminution in HIV virulence, a lower community viral load, and an increased proportion of ‘elite controllers’ in the population.

Goulder remarked that these changes seemed to be happening surprisingly fast, and that in some populations the introduction of antiretroviral therapy would also tend to reduce the fitness of the HIV that was still circulating.

**HLA subtype and viral fitness**

Goulder began by noting that it has been known for some time that people with certain variants of HLA tended to develop lower viral loads than average if they acquired HIV.

The HLA genes are the human version of the major histocompatibility complex (MHC), a set of proteins present in all vertebrates, which helps the body distinguish between its own tissues and foreign invaders. They sit on the surface of cells and display ‘epitopes’, short sections from protein molecules inside the cell. If the immune system does not recognise these epitopes as belonging to the self, it will mount a response and attack the foreign invader. Some HLA variants are better at displaying epitopes that stimulate a strong immune response than others.

If HLAs are too efficient, the result can be auto-immune disease and a high susceptibility to allergies. It is interesting that the HLA variant most strongly associated with a lower HIV viral load and slower disease progression, B*5701, is also associated with hypersensitivity (allergy) to the drug abacavir (Ziagen, also in Kivexa/Epzicom).

B*5701 is not the only HLA subtype associated with lower viral load. Most other varieties of the broader B*57 type are also associated with slower progression, as are B*58, B*51 and to a lesser extent B*31, 38, 42 and 44. Certain other subtypes are associated with higher viral loads and faster HIV progression.

One of Goulder’s population studies, of people with HIV in Durban, South Africa, showed that subtypes B*5701, 5702, and 5801 were associated with at least a threefold (0.5 log) lower viral load than the average for the population, and B*5703 with a fivefold reduction (0.7 log).

**HLA escape mutations**

HIV can develop resistance mutations to HLA as it does to drugs. These change the viral proteins into ones the HLA molecule cannot ‘grab hold of’ and display on the cell surface, and thus the virus becomes less visible to the immune system. However, these viral protein changes may also exact a fitness cost by causing viral replication turn over more slowly.

The first sign that this might be changing the make-up of the HIV in a population came from Japan, which has an unusually high proportion of people with the B*51 HLA variant (20% as opposed to 10% in Oxford and 1% in Durban). It also has an unusually high proportion of HIV that carries a specific, fitness-reducing HLA-resistance mutation in HIV called I135X. Three-quarters of people with HIV carry this mutation in Japan versus 40% in Oxford and 20% in Durban. Goulder wondered if this might be part of the reason Japan had maintained a particularly low HIV prevalence (less than 0.1%) despite having a developed gay scene.

The relative scarcity of HIV in Japan makes it hard to study. Instead, Goulder looked at viral loads and HLA variants in the very high-prevalence cities of Durban and Gaborone in Botswana.

The HIV epidemic is older in Botswana than it is in Durban – it reached peak prevalence in about 2000, which Durban did ten years later. The average viral load in untreated people is also lower: 15,350 copies/ml (4.2 logs) compared with 29,350 copies/ml (4.5 logs) in Durban. This is despite the fact that the average CD4 count is 50 cells/mm³ lower in Gaborone, indicating that people on average have been living with HIV for longer – which would also suggest a higher viral load.
The same survey found a higher prevalence of HIV HLA ‘escape’ mutations in Gaborone than Durban – 46% of people in Gaborone had HIV that the HLA B*57 and B*58 molecules could not display, compared to 38% in Durban.

These mutations had real effects. As we said above, in Durban, people with the B*57 and B*58 variants of HLA had lower viral loads than average. In Gaborone, they did not: their viral loads were identical to the rest of the population and in the case of the B*5702 variant, they actually had viral loads 0.3 logs higher than average. In other words, the HIV in Gaborone had evolved into a form which was essentially unaffected by more efficient HLA types. HIV in Gaborone was also less fit: in a test-tube assay, it replicated 11% more slowly than the Durban virus.

Recent drops in viral fitness – and more elite controllers
In Durban, there was evidence that a reduction in viral fitness might be happening right now. Goulder compared viral loads in female patients in the largest HIV clinic there in 2002-5 and 2012-13 (he studied people accessing antenatal care, as their viral load and treatment status was better documented). Excluding women who were on antiretroviral therapy (ART) or had been on ARV drugs for pregnancy less than three months previously, and women with CD4 counts less than 350 who might be on undocumented ART, the average viral load in women was 13,550 copies/ml (4.13 log) in 2002-5 and 5750 copies/ml (3.76 log) in 2012-13, and this difference was significant (p = 0.0013).

There also appeared to be an unusually high proportion of women in the population who were controlling HIV spontaneously. Twenty-four of the first 239 women whose viral loads were tested (10%) had a viral load under 50 copies/ml off ART. This compares with a prevalence of no more than 0.5% in most populations studied. Other explanations – false-positive HIV tests, women taking ‘black market’ or informal ART, differences in viral load assays – were ruled out, and furthermore the average CD4 count in the women with undetectable viral loads was 890 cells/mm³ – essentially normal and indicative of viral control.

Co-factors in reduced transmission and prevalence
HLA adaptation is not the only reason HIV fitness might be declining. Goulder pointed out that the fittest virus was also not necessarily the one that got transmitted most often. This is because untreated people with high viral loads tend to die quicker so have less time in which to pass on their HIV. Modelling work by the team at Imperial College, London has established that the ‘optimal’ viral load in terms of the efficiency of HIV transmission is 4.52 logs (33,000 copies/ml).

Goulder added that HLA escape mutations and higher mortality in people with high viral loads might now be joined by a third reason viral load and fitness might fall over time. In a population where ART is introduced in stages, the sickest people with the highest viral loads will get treated first, and then the next most sick, and so on. This will tend to remove the fast progressors, who have a high viral load ‘set point’, from the untreated population first.

ART will only tend to have this effect in populations where a lot of HIV is passed on in chronic infection: in populations where most people diagnosed are treated, most HIV will be passed on by the undiagnosed – who by definition will not be on ART. Nonetheless, he added, falls in HIV prevalence and rises in life expectancy in Africa and some other regions might not only be caused by behavioural changes or by putting increasing proportion of people on treatment.

“It appears there are more factors that are driving viral fitness down than we thought,” he remarked.

Reference

Did the press comply with an HIV witch-hunt in Greece?
State and private television networks went ahead and published women’s mugshots and personal details, labelling them “HIV-infected prostitutes”.
By Julie Tomlin Published 07 November 2013 13:13
When hundreds of women were rounded up in a police sweep in central Athens days before the May 2012 parliamentary elections, the move had all the hallmarks of a politically orchestrated campaign. The arrests preceded the release of figures showing a 57 per cent increase in HIV infections between 2010 and 2011, and the women were forced to take HIV tests.

Despite a lack of evidence, those who tested positive were imprisoned and charged with intentionally causing grievous bodily harm. When state and private television networks went ahead and published the
women’s mugshots and personal details, labelling them “HIV-infected prostitutes”, many saw it as evidence of a compliant press.

The mainstream media soon lost interest in the story, staying silent when most of the women were gradually released or had the charges against them reduced, but now Zoe Mavroudi, a Greek filmmaker, has directed a film on the subject – *Ruins: Chronicle of an HIV Witch-Hunt*.

When I spoke to her on Skype shortly after the Greek premiere of Ruins, she told me how she had noticed that the case was still being discussed widely on social media. “People seemed to be harking back to it to express disgust for state and police arbitrariness. I sensed that the incident had become one of the most recognisable low points of the crisis,” she said. “I wanted to create a chronicle, a kind of reference point that would help people to understand and not forget.”

Made with the support of the Unite union and Union Solidarity International, the film includes interviews with two of the women who were imprisoned, along with their mothers, as well as academics and activists.

The interviews are intercut with footage that shows journalists and commentators referring to the women as “Aids prostitutes” who “spread death”.

The women’s case has now been taken to the European Court of Human Rights.

More than 30 per cent of Greek women are unemployed, compared to 24.6 per cent of men, and more than 65 per cent of young women are out of work. They are also affected by falling standards in maternity care and cuts to services for the sick and elderly. More and more young people are returning to live in the family home.

“Greece is a very traditional society,” Mavroudi said, “and when you have the dismantling of social services . . . the burden falls on women even more than usual.

“The feminist movement in Greece has been caught off guard by the crisis,” she added.

The arrests were condemned by some female MPs and several protests were held outside one courthouse and the ministry of health in Athens.

Mavroudi hopes that next time women will be better prepared. “This case in particular was unprecedented, targeted state aggression against women,” she said.

“Without a strong and well-organised feminist movement, we cannot deal with this new reality.”

‘People Think It’s Over’

*Spared Death, Aging People With H.I.V. Struggle to Live*

Nicole Bengiveno/The New York Times

By JOHN LELAND

Published: June 1, 2013

Steve Schalchlin would be the first to tell you he lives in a time of miracles, and about how hard that can be. In 1995, as his body wasted away from AIDS, he took the limited time in front of him as a challenge: he would write songs, make amends, fill his remaining days with life. And by the end, with his digestive system shut down, his figure skeletal, he was ready to die. Then he won a lottery for a new AIDS drug that had been rushed through the approval process. Almost overnight his health began to return, and with it, another, more open-ended, challenge: life.

Multimedia

“Suddenly the future seemed like this long, empty road going toward the horizon, and I felt like, what am I gonna do with my life now?” Mr. Schalchlin, 59, said the other day, still marveling at the turn of events. “I had already accomplished all my goals that I had set for myself. And now I had this endless amount of time ahead of me, and I felt depressed.”

Mr. Schalchlin no longer worries about dying of AIDS. But he has other health problems, more often seen in people 10 or 20 years older: kidney damage, diabetes, chronic fatigue, thyroid disease, partial paralysis in one eye and general weakness that limits him throughout the day.

He is, he has learned, host to a virus that never stops working, grinding away at him, and requiring sustained, complicated treatment, each medicine bringing its own side effects. At times, he said, it becomes too much. “Sometimes I just get so tired of the fight, it just becomes exhausting,” he said. “Do I have to go another day with this, and another day, and another day? I don’t think about the future because I’m too busy worrying about the next day and the next hour and the next meal and which pill I’m supposed to take.”

Not long ago, it would have seemed unthinkable: H.I.V. is becoming a disease of the middle-aged. Nearly half of New Yorkers with H.I.V. are now 50 or older, ages many never dreamed of reaching. They
are the people who were told that they would be dead by 30, who watched their friends or lovers die, who lived to see sufferers on their death beds return to health almost overnight.

They are a diverse group, including people like Emilie Mobley, 70, a die-hard Elvis fan who gets around with a walker after a 2004 stroke, and who was once told to plan her own funeral to spare her family; or like Jan Carl Park, 63, who has had the virus since at least 1981 but has never been sick because of it. Mr. Park, director of the H.I.V. planning council at the city’s Department of Health and Mental Hygiene, lost 80 friends to the disease but moved on with his own life, getting a graduate degree and moving up the ladder at his job.

Their stories have largely moved out of the spotlight, as the disease seemed to yield to the miracle of new drugs. To many, AIDS seems like a relic of a past age, as treatable as diabetes.

Yet for people living with the virus that causes it, the experience is more layered and complex.

Now this group of almost 50,000 men and women moving through middle age is a living science experiment, entering medical and psychological territories that are largely uncharted. What are the consequences of long-term exposure to the virus, or to the medications? How do these interact with the effects of normal aging? And how, after you have braced for death, do you turn around and create a new life, often without the friends and loved ones who gave your life definition?

Interviews with a dozen members of this population elicited a mixture of wonder and anxiety. Some said they were healthier and better adjusted than they had been for decades. But for others, survival has come with consequences, both medical and social. Many said they felt forgotten by a city whose attention has turned away from H.I.V. and AIDS, and by a gay community whose activism long ago shifted to same-sex marriage. And for all of them, the journey has been filled with surprises.

“I’m not going to die,” said Osvaldo Perdomo, 52, a former Liz Claiborne executive who quit his job after developing AIDS in 2004. Though antiretroviral drugs have reduced to undetectable levels the amount of the virus in his blood, his life remains uprooted, his health sketchy. He spends long hours in bed and struggles with fatigue, depression, anxiety and memory loss. He has not dated since he got sick. And he has $40,000 in credit card debt from co-payments for his medications.

“Sometimes I feel, O.K., I’m not going to worry about what’s going to happen 10 years from now,” he said. “I’m going to worry about today. But I’m running out of resources. When I went to the drugstore last night to pick up my medication, I was crossing my fingers — if this credit card doesn’t go through, I’m going to have to borrow money from a friend. What is going to happen when I’m 65? Are you going to grow up older and alone? Who’s going to take care of you?”

Mr. Perdomo recently joined the board of GMHC, the health care and advocacy organization. “People think it’s over, you can just take a pill, there’s a cure around the corner,” he said. “It drives me crazy when people think it’s over.”

Knowledge of H.I.V. has progressed astronomically since 1980, when the first patient arrived at Bellevue Hospital with what later came to be called acquired immune deficiency syndrome, or AIDS. Paul Bellman, who was a third-year medical student at the time, can still remember trying to help with treatment. In the years since, his practice has gone from helping people die to — after the introduction of effective drugs in 1996 — helping them live.

But as his patients reached middle age, he said, they started to come to him with problems that seemed unrelated to H.I.V.

“We weren’t worried that their cholesterol was going up,” he said. “The patient was alive and gaining weight. What was developing was a profile of accelerated cardiac risk. I started noticing heart attacks in guys in their 40s and early 50s.”

What he and other doctors have learned is that H.I.V. and aging have a lot in common. Both cause inflammation and a weakening immune system. Both are associated with high levels of depression and isolation, which lead to people skipping their medication — which in turn aggravates all of the other maladies.

The cumulative effect is greater than the sum of the parts, said Dr. Antonio E. Urbina, associate medical director of St. Luke’s-Roosevelt Hospital’s Center for Comprehensive Care’s clinic in Chelsea, which sees more than 2,000 H.I.V.-positive patients over age 50 each year. He rattled off a list of common problems: high rates of cancer, heart and lung disease, kidney failure, diabetes, high blood pressure, arthritis and memory loss.

“Wherever H.I.V. hides, it isn’t quiet,” Dr. Urbina said. “It still can activate cells to spit out bad proteins called cytokines. And these cytokines drive inflammation.

“I think we were so focused in on just controlling the virus that we started to take our eye off primary care issues. We need more aggressive management, understanding that these patients are at higher risk
for heart disease, for stroke, for neurocognitive decline. We need to be aggressive, because there are things we can do now that will allow them to age healthily into their 80s and 90s.”

Almost two decades after the advent of effective treatment, the virus poses challenges for city agencies as well, said Thomas K. Duane, 58, a former state senator who believes he was infected in the early 1980s. “The city could be more prepared” for the wave of older people with H.I.V., Mr. Duane said, noting statistics that show only one-quarter to one-third of people with H.I.V. maintain treatment to suppress the level of the virus in their bodies. “It’s both a disease of poverty and not a disease of poverty,” he said. “When there’s poverty, it’s incredibly difficult for people to access the best care.”

AIDS used to present a grimly straightforward narrative. People contracted the disease, wasted to nothing and died. Or: they survived until the 1996 advent of the drug Crixivan and had remarkable recoveries. Flesh returned to withered bodies; cancerous lesions melted away.

But the story was never that simple. Many people never got sick, and others suffered unnecessarily because they lacked information or access to treatment. Poverty, mental health and discrimination have always been factors in AIDS transmission and treatment.

Now add aging to the mix. Living a few miles from each other, in Harlem and the East Bronx, Christopher Davis and Joan Warner have lived radically different lives with the virus. When Mr. Davis, 62, learned he had H.I.V. in the late 1980s and then watched a partner die, he said, “I partied harder. Took every drug in the world. I took all the money out of my 401(k) and spent it.” He quit his job, ended up homeless and dependent on the city’s H.I.V./AIDS Services Administration.

Ms. Warner, 73, took a different route. She said the doctor told her, “‘Miss Warner, if you want to stay alive’ — this was the one sentence he said — ‘you have to stop doing those drugs.’ That’s it. I’d been using and abusing drugs for 34 years. I thought, ‘I want to live.’ I have my children, I had grandchildren. But I know people who went the other way.”

Now both are coping with life with chronic disease. Both say they are healthy, then mention complaints. Mr. Davis, who is now a nonprofit financial executive, had a stroke in 2001, followed by several brain seizures; he lost most of the vision in one eye. “I have the normal problems of growing old,” he said. “My knees hurt. My hip is bad, but I still walk two or three miles a day. I think I believe in miracles.”

Ms. Warner was 51 when she went to the emergency room with acute diarrhea that was stripping pounds from her body. At four treatment centers, no one suggested an H.I.V. test. “Probably someone that old, they don’t look at risk factors,” she said. “That can be a pretty delicate situation, trying to engage older people. I guess they didn’t think about it.” Finally Ms. Warner’s daughter, who was then 22, suggested they both be tested. Her results were negative; Ms. Warner’s were not.

Her story is common, according to AIDS professionals. Many doctors hesitate to ask people their own parents’ age about their sex lives or drug use.

She described herself as “pretty healthy.” But she, too, suffers from maladies caused by inflammation, including arthritis that hampers her from walking. Her knuckles look like knotted tree roots. “Last month, the arthritis doctor drained one knee and gave me a cortisone shot in each knee. But I feel it’s beyond just the knees. The pain goes down my legs also.” Even when she lies down, her legs hurt, she said.

Surprises do not stop. Yolanda Diaz, 50, is experiencing a joy she never expected: menopause. When she first received her AIDS diagnosis in 1989, she had no interest in treatment — instead she smoked more crack and heroin, bounced in and out of prison, and let the father of her children raise them.

But age and the disease have mellowed her. Now she follows her regimen and works for an AIDS organization called Iris House, performing interventions for women with addictions. AIDS has given her life meaning and community. “Man, I’m going to be 51,” she said, lingering over the number like a fresh miracle.

“I don’t think I’m going to die from H.I.V. and AIDS. But in the long run, I am going to die from O.I.’s, from opportunistic infections. I just don’t think about it on a daily basis.”

Yet others become more isolated as they age, without support networks, religious ties or even someone to take them to a doctor when they feel ill. This isolation, doctors say, makes the disease more potent.

Nearly three-quarters of H.I.V.-positive New Yorkers ages 50 and older live alone, according to the health department; in a 2006 survey of 914 H.I.V.-positive older New Yorkers by the AIDS Community Research Initiative of America, two-thirds of the respondents were moderately or severely depressed.

Many people with H.I.V. will not go to senior centers for fear of discrimination, said Daniel Tietz, the initiative’s executive director. Others say that, as older gay men with H.I.V., they feel shut out from AIDS...
service organizations geared to younger or newly infected men, and from bars where they once felt at home.

“The amazing psychosocial programs that developed in the 1980s when there was no medical treatment are an afterthought these days,” said Perry N. Halkitis, 50, a professor of psychology and public health at New York University, who is H.I.V.-positive. “Now it’s completely biomedicalized. In the 1980s, you relied on these other therapies, which are absolutely critical as men get older.”

For an upcoming book called “The AIDS Generation,” Mr. Halkitis gathered 15 middle-aged, H.I.V.-positive gay men, many of whom had never before been in such company. “This is going to be the only group that lived through the darkest hours of the epidemic,” he said. “So there’s a particularly nuanced reality for men of my generation. You have a generation of men in need of services who really don’t have any place to get them.”

And for all of them, the virus is never dormant. Sometimes it makes its presence known, sometimes it works quietly in the background. The story of long-term exposure to H.I.V. and its medications is still spinning out new chapters.

Scott Jordan, 52, recently discovered a new wrinkle: the last treatment available to him is no longer effective.

In his Greenwich Village apartment, Mr. Jordan appeared to be a portrait of health. Married to his high school boyfriend, he is beefy, with a glowing ruddy complexion. But the muscles are from human growth hormone to prevent wasting, and the rosy cheeks are from high blood pressure.

Six months ago, when he was experiencing severe side effects from his medication, he took a brief holiday from the drugs. The side effects eased somewhat, but the amount of the virus in his blood, which had been undetectable, immediately shot up. Now the old medications will not bring it back down. Until a new drug comes along, he said, he is out of options. It was unclear, speaking with him, how confident he was that a new drug would emerge in time.

“I’ve heard people say, ‘we have these drugs now, what are you worried about?’ ” he said. “And ‘you look great.’ What it does is it invalidates our internal experience. It’s almost as if it’s not talked about. I remember a time where it was all we talked about.”

Around the apartment were notes to himself: things he wanted to do or say that he was afraid of forgetting, because his memory has declined.

“I’m a young old man,” he said. “I start to decline very rapidly in energy around 6 p.m. But I have a wonderful life. And I feel graced by God. I feel great, I guess, just physically older than my healthy contemporaries.”

**Gut Reaction: Effect of Diet, Estrogen On Gut Microbiota**

Nov. 8, 2013 — Study results from Texas A&M University and University of North Carolina School of Medicine scientists on the effect of diet complexity and estrogen hormone receptors on intestinal microbiota has been published in the September issue of Applied and Environmental Microbiology.

To date, research has shown that promoting the growth of certain beneficial intestinal microorganisms can help to improve overall health.

"In this study, we wanted to determine if steroid hormone nuclear receptors, specifically estrogen receptor beta, affect the composition of intestinal bacteria," said Dr. Joseph Sturino, lead researcher in the nutrition and food science department at Texas A&M’s College of Agriculture and Life Sciences, College Station.

"Some steroid hormones, like estradiol, and dietary phytoestrogens are known to influence the development of chronic gastrointestinal inflammation and estrogen-responsive cancers of the breast, prostate and colon," Sturino said.

Some of these effects are the result of differential and tissue-specific gene regulation by estrogen receptor beta, Sturino said. That aspect of the study was the focus of the lab work performed by Dr. Clinton Allred, also in the college's nutrition and food science department and a collaborator on the published study.

They hypothesized that some estrogenic regulatory signals are mediated, in part, by the activity of microorganisms present in the gut and that diet modification can be used to change those.

In order to investigate the effects of both receptors and diet on intestinal microorganisms, the scientists initially raised female mice on a fiber-rich diet containing plant-derived estrogenic compounds called isoflavones, comprising a complex diet. The animals were then fed an isoflavone-free diet that was
rich in highly refined sugars for two weeks, comprising a simple diet. The composition of the fecal bacteria was surveyed over the course of the study.

"As you might expect, significant differences were found between the fecal microorganisms of mice fed a biochemically complex diet containing isoflavones and those that were fed a simple diet that lacked isoflavones," he said. "Interestingly, however, we also found that the microorganisms differed between mice that expressed estrogen receptor beta and those that did not."

Distinct patterns for Lactobacillales were exclusive to and highly abundant among mice fed a complex diet containing isoflavones, Sturino explained.

"Some Lactobacillales have probiotic function when taken in adequate numbers in food or dietary supplements, so indigenous species might also act to promote gut health," he said.

In contrast, he noted, the relative diversity of Proteobacteria increased significantly following the transition to the simple, isoflavone-free diet. Proteobacteria includes a number of species commonly associated with intestinal disease, including Escherichia, the "E" in E. coli O157:H7, and salmonella.

These and other study results demonstrated that steroid receptor status and diet complexity might play important roles in microbiota maintenance, Sturino said.

"While the balance and content of microorganisms in the gut changes as we age, we are only now learning how our genetics and dietary choices affect our health by modifying the composition and activity of these microorganisms," he said.

In the long term, Sturino believes that this study will aid in the development of novel probiotics, prebiotics, nutritional strategies and pharmaceuticals to improve overall health by promoting the growth and activity of beneficial intestinal microorganisms.

**Journal Reference:**

---

**In Rare Move, Uganda Leader Publicly Tests for HIV**

Kampala, Uganda November 8, 2013 (AP)

By Rodney Muhumuza Associated Press

Uganda’s president tested for HIV in public on Friday to encourage millions of untested people to check their status, a critical step to stemming the spread of the virus in the East African country.

Public leaders rarely test for HIV in public in Uganda, despite recommendations from health workers that it would set a good example in a country that has seen HIV infection rates increasing. Uganda was once a global leader in efforts to fight AIDS.

Not all government officials at the Friday event in the capital, Kampala, joined the president in testing.

Ugandan officials have said they want to test 15 million people by the end of 2014. They acknowledge it will be hard to reach that target, the reason they want the president to be a "role model."

"Therefore, all Ugandans, test (for HIV). Find out your status and let the state and health workers manage you accordingly," said President Yoweri Museveni.

The HIV rate in Uganda stands at 7.3 percent, up from 6.4 percent in 2005, according to a 2011 survey by Uganda’s Ministry of Health. Ugandan officials who presided over its reduction from 18 percent in 1992 to 6.4 percent in 2005 say they are confounded by the increase.

Ugandans health officials say more married couples are getting infected, in part because of what campaigners have dubbed a "sexual network" in which married Ugandans maintain secret lovers. One billboard in Kampala urges couples to "put your love to the test" by testing for HIV.

Museveni and his wife are "leading by example in a bid to roll back the HIV epidemic in Uganda," the Uganda AIDS Commission, the local body tasked with fighting AIDS, said, though the first lady did not attend the event where Museveni was tested.

Experts say HIV testing is critical to preventing new infections because those who know their status are less likely to engage in risky sexual behavior. But getting people to test for HIV has proved difficult in Uganda, where rampant stigma persists and where thousands get infected each year.

The rise in new infections is stretching the ability of Uganda’s government and donors to provide HIV and AIDS treatment. More than 500,000 Ugandans need AIDS treatment, many accessing it through the U.S. President’s Emergency Plan for AIDS Relief, or PEPFAR.

If Ugandans reach their testing goal, at least 400,000 more people will likely be in need of AIDS treatment, according to Musa Bungudu, the Uganda coordinator for the United Nations’ AIDS agency.
Uganda once earned a global reputation for successfully putting in place a policy called ABC: Abstain, be faithful, or use condoms. Students of a certain generation were shown videos of the devastating toll of AIDS on the human body, and then told to postpone the first act of intercourse. But critics of Uganda’s policy to fight AIDS say the country recently has focused more on treatment rather than prevention.

Uganda’s government now has added male circumcision to the plan to fight HIV and AIDS, in response to studies showing the procedure reduces the risk among African men of getting HIV in half.

**NIH launches trial of investigational genital herpes vaccine**

Researchers have launched an early-stage clinical trial of an investigational vaccine designed to prevent genital herpes disease. The National Institute of Allergy and Infectious Diseases (NIAID), part of the National Institutes of Health, is sponsoring the Phase I trial, which is being conducted at the NIH Clinical Center in Bethesda, Md.

Genital herpes is one of the most common sexually transmitted infections in the United States. Most genital herpes cases are caused by infection with herpes simplex virus type 2 (HSV-2); however, herpes simplex virus type 1 (HSV-1) can also cause genital herpes. An estimated 776,000 people in the United States are infected with HSV-2 or HSV-1 each year. There is no vaccine to prevent genital herpes.

"Although genital herpes is treatable, it is a lifelong infection that can exact a substantial psychological and physical toll on infected individuals and places them at higher risk of acquiring HIV," said NIAID Director Anthony S. Fauci, M.D. "Furthermore, mothers with active genital herpes infection at time of delivery can transmit the virus to their newborns, which can lead to severe illness and death."

"A protective vaccine would help to reduce significantly the spread of this all-too-common sexually transmitted infection," Fauci added.

Led by principal investigator Lesia K. Dropulic, M.D., of NIAID's Laboratory of Infectious Diseases, the trial will test an investigational HSV-2 vaccine candidate, called HSV529, for safety and the ability to generate an immune system response. The investigational vaccine manufactured by Sanofi Pasteur was developed by David Knipe, Ph.D., professor of microbiology and immunobiology at Harvard Medical School, Boston.

Preclinical testing of the candidate vaccine involved a 10-year collaborative effort between Dr. Knipe and Jeffrey Cohen, M.D., chief of NIAID's Laboratory of Infectious Diseases. The experimental product is a replication-defective vaccine, meaning that scientists have removed two key proteins from the vaccine virus so that it cannot multiply to cause genital herpes.

The clinical trial is expected to enroll 60 adults ages 18 to 40, who will be divided into three groups of 20 participants each. The first group will be of people who have been previously infected with HSV-2 and HSV-1 or solely with HSV-2; the second will have individuals who had been infected with HSV-1 only; and the third will consist of those who have not been infected with HSV-1 or HSV-2. The investigational vaccine is being tested among study participants who have previously been infected with HSV to determine if it may pose any safety issues.

Within each of the three groups, researchers will randomly assign participants to receive three doses (0.5 milliliters each) of the investigational HSV529 vaccine (15 participants) or a saline-based placebo vaccine (five participants). The three vaccinations will occur at study enrollment and again one month and six months later. Participant safety will be monitored throughout the course of the trial, and researchers will follow participants for six months after they have received their last dose of vaccine. Blood samples will be used to evaluate the candidate vaccine’s ability to stimulate immune system responses to HSV-2, including production of virus-specific antibodies and T-cell responses. The study is expected to be completed by October 2016.

HSV-2 is generally transmitted through sexual contact and can spread even when the infected individual shows no symptoms. Although HSV-1 commonly infects the mouth and lips, it can also cause genital herpes. Once in the body, HSV migrates to nerve cells and remains there permanently, where it can reactivate to cause painful sores and blisters.

**Another negative result in the quest for a shorter TB treatment course**

Mara Kardas-Nelson
Published: 11 November 2013

Results from a Phase III trial that attempted to shorten the treatment duration for drug-sensitive pulmonary TB from four to six months, by using gatifloxacin instead of ethambutol, showed that the shortened treatment duration was inferior when compared to the standard treatment course. Results
from the OFLOTUB trial were presented earlier this month at the 44th World Conference on Lung Health in Paris.

A previous study, RIFAQUIN, found that it was not possible to shorten the TB treatment regimen from 6 months to 4 months by use of moxifloxacin and rifapentine twice weekly in place of rifampicin and isoniazid in the continuation phase of TB treatment.

The non-inferiority, blinded, randomised control trial was conducted in Benin, Guinea, Senegal, Kenya, and South Africa. In the control arm of the study, 919 people were given the World Health Organization-recommended-six-month course of TB treatment, whereas 917 people in the test arm received 400mg of gatifloxacin, six days a week, in place of ethambutol for two months, followed by two months of treatment with gatifloxacin, rifampicin and isoniazid.

The median age for participants in the study was 31 years, 27% were female, and 18% had HIV. All participants were adults with new TB diagnoses.

The study used a 6% non-inferiority margin, and considered patients’ “unfavourable” outcomes as a composite endpoint at two months, end of treatment, 18 months, and 24 months, with the primary outcome being at 24 months. Researchers considered participants who died, were lost to follow-up during treatment, experienced treatment failure, or experienced TB re-occurrence (a category that included relapse and re-infection), as experiencing unfavourable outcomes.

At two months and end of treatment, those in the intervention arm fared better. However, 24 months after completion of treatment, a larger proportion of people in the gatifloxacin-containing arm had unfavourable outcomes (20.9 vs 16.8%). This difference was driven by a higher frequency of tuberculosis recurrence in the gatifloxacin arm (14.6 vs 6.9% of participants were diagnosed with TB again). The intent-to-treat analysis showed a difference of 3.8% favouring the control arm, with a confidence interval that exceeded the non-inferiority margin (-0.03%, +8%). As a consequence the gatifloxacin-containing regimen was found to be inferior to standard TB treatment.

Importantly, the gatifloxacin-containing regimen was considered safe and well-tolerated: participants taking it were less likely to experience a QT prolongation in the test arm than the control arm, with a risk difference of -0.1% in the test arm (95% confidence interval -0.2, 0.1). "This is very important because almost all the [new] drugs in the pipeline have QT prolongation issues," says clinical investigator Corinna Merle, who presented the findings. "When it comes to mixing new TB drugs, you might not want to mix two drugs with QT prolongation." The risk of hyperglycemia was "almost the same in both arms", she says.

Interestingly, different countries demonstrated different outcomes: people fared better in the intervention arm in Benin, Guinea, and Kenya than those in Senegal and South Africa. Noting that countries where participants had lower BMIs when they began treatment – such as Benin and Guinea – did better on treatment than those with higher initial BMIs – such as Senegal and South Africa – Merle wonders whether the regimen’s effectiveness could be increased with better dosing. (Patients in Kenya and Senegal, however, had similar BMIs at baseline, yet Kenyan patients did better in the intervention arm than Senegalese patients.) All patients, regardless of body weight, were given 400mg a day.

A pharmacokinetic study is currently being conducted in South Africa to help determine appropriate dosing. Results are expected within the next six months. Merle also suggests that giving treatment every day of the week, instead of six days out of seven, may be important when attempting to use a shortened regimen (the team decided to treat six out of seven days as many participating countries did not have healthcare centres open on Sunday). Participants who were living with HIV also did better in the intervention arm than those who did not have HIV.

Merle says that the intricacies found in the OFLOTUB trial suggest that more research should be conducted, and that gatifloxacin shouldn't be discounted as a potential TB regimen yet. "In the control arm we had more patients lost to follow-up," Merle says. "And then more treatment failure in the control arm, but what you have more of in the [intervention arm] is re-occurrences." Merle notes that poorer outcomes in the intervention arm at 24 months may simply be because patients who had taken gatifloxacin were followed-up for longer than those on standard treatment, as they were taking two months’ less treatment. "We need to look at the dynamics in more detail to explain all of this, and why patients in some countries did better than others. We also need to look more at dosing, and at gatifloxacin treatment in HIV-positive patients."

Merle also notes that gatifloxacin is a generic drug, whereas moxifloxacin – being considered for use in a shortened TB treatment regimen as part of the REMox trial – is not. Given that both OFLOTUB and REMox are attempting to shorten TB treatment periods, Merle says "lets see [the REMox] results. If they have very good results, there’s no reason to have two types of regimens. But if they have results similar to
ours, then there could be a discussion…I really think that [gatifloxacin] should not be buried…For the moment we will wait."

Reference

**Trial of game changing new anal condom begins in Boston**
A revolutionary anal condom that simulates condom free sex is being trailed in Boston by the US National Institutes of Health in the hopes of encouraging more people to engage in safer sex
06 November 2013 | By Andrew Potts

Photo by Origami Condoms

An anal sex specific prototype of a new generation of condoms is being tested by Boston’s Fenway Institute as part of a clinical trial by the US National Institutes of Health.

Origami Condoms have developed a new kind of sexual barriers which simulate condomless sex and their anal condom hopes to be a game changer in preventing the spread of sexually transmitted infections among gay men.

Insertive condoms for women have been available in the past but the Origami Anal Condom is the first condom designed for use by a passive partner during anal sex.

The US Food and Drug Administration has never approved a condom specifically designed for use in anal sex and the National Institutes of Health wants to change that.

Origami condoms have also developed new insertive condoms for women and a penile condom for men.

The condoms are made from allergy free silicone and are designed to give greater stimulation to both partners’ genitals simultaneously during sex.

Origami hopes to conduct large scale clinical trials in 2014 to prepare for the condoms reaching the market for a 2015 target date.

For information about participating in the Fenway Institute trial contact Jake Tinsley at 617-927-6450 or email origami@fenwayhealth.org

See more at: http://www.gaystarnews.com/article/trial-game-changing-new-anal-condom-begins-boston061113#sthash.ihPJIEZU.dpuf

**The other scandal in Sri Lanka—and 80% of Commonwealth countries: endemic homophobia**

Nearly 80 per cent of Commonwealth countries are enforcing anti-gay laws perpetuated by leaders who are “wilfully turning a blind eye to homophobia on a massive scale”, a report warns today.
The study, commissioned by the Kaleidoscope Trust and compiled by Lesbian, Gay, Bisexual, Transsexual and Intersex (LGBTI) activists across the Commonwealth, calls for Britain’s former colonies to repeal anti-gay legislation, with an immediate moratorium on enforcement. Homosexual acts are a criminal offence in 41 of the 53 members of the Commonwealth, including Jamaica, Pakistan, Singapore and Malaysia.

The warning comes as leaders prepare for the Commonwealth heads of government summit that begins in Sri Lanka on Friday, where the issue of anti-gay discrimination is not on the official agenda.

“Gay rights is one of the last bastions of acceptable human rights abuses, and for Commonwealth leaders not to spend one single second discussing it – even as laws are being passed in places such as Uganda that could see gay people sentenced to death – speaks volumes for priorities,” said Kaleidoscope’s spokesman.

In a foreword to the report, Sir Shridath Ramphal, the former Secretary-General of the Commonwealth, calls for all countries to “rid ourselves of this archaic legal inheritance”, branding it “a relic of Empire”.

The report highlights a series of cases across Africa, Asia and the Americas in which anti-gay campaigners have been targeted.

One account from Caleb Orozco, executive director of the United Belize Advocacy Movement, reads:

“As the only claimant in the current constitutional challenge case, I have lost two teeth, had my family property invaded and car damaged by two masked men in the week of the supreme court hearings in May of this year.

“I have had stones thrown at me, experienced simulated gunshots, insults and physical harm on public transportation, and threats.”

Uganda, widely considered one of the worst offenders, is in the process of passing tougher anti-gay legislation, dubbed locally as the “Kill the Gays” bill. According to the report, in February 2012, the Minister for Ethics and Integrity, Simon Lokodo, was involved in a raid on a workshop for LGBTI women at which he reportedly said: “I have closed this conference because it’s illegal. We do not accept homosexuality in Uganda. So go back home.”

Singapore, the report says, has seen increasing “police surveillance and censorship of LGBTI events and activities [that] are common occurrences”.

“Across the Commonwealth LGBTI people face criminalisation and social discrimination. They face violence. They face eviction from their homes, dismissal from their jobs and estrangement from their families.” Officials insist that although such issues remain off the official agenda, talks are likely to occur behind the scenes. However it adds to concerns over Britain’s decision to attend the summit in Sri Lanka, where same-sex conduct is illegal and LGBTI activists have been threatened with detention.

Today, the Indian Prime Minister, Manmohan Singh, is expected to confirm intentions to boycott the meeting in a row over the country’s human rights record.

In March, the Queen signed a charter that marked the first time Commonwealth countries had adopted a single document promoting gay rights and gender equality. But its usefulness has come into question. “It’s effectively a talking shop designed to smooth the wheels of trade. In terms of resolving human rights issues it has been singularly ineffective,” the spokesman for the Kaleidoscope Trust said.

A Downing Street spokeswoman said last night: “The Commonwealth Charter, agreed by all Commonwealth members, explicitly states that we are opposed to all forms of discrimination, and it is important that all members live up these values. That is the message that we will be taking to the summit.”

‘Children of the devil’: victims’ voices

"I have lost two teeth, had my family property invaded and car damaged by two masked men... I have had stones thrown at me, experienced simulated gun shots, insults and physical harm on public transportation, threats that speak to, 'Caleb, you have no right to breathe!'" —Caleb Orozco, Belize

"I have had to battle against a belief that my behaviour situates me outside the 'normal'. I had been called a "child of the devil" who was at-risk, vulnerable, stigmatised and lacking... but we are challenging that prejudiced view with some success." —Joey Joleen Mataele, Tonga

"I openly live as an intersex person. Australia has a reputation for a 'macho' culture so I was proud to be elected mayor two years ago. I wish more people knew about intersex so people felt comfortable about being open about the way they were born." —Tony Briffa, Australia

"It’s been over seven months and counting since any of my family laid eyes on me. They refuse to accept me for who I am and worse off, because I advocate for what I believe in. I can never go back home now, as home has now become a memory in my past." —Charles, Zambia.
**NIH Launches Trial of Investigational Genital Herpes Vaccine**

*eNews Park Forest*, (11.08.2013)

eNews Park Forest recently reported that the National Institute of Allergy and Infectious Diseases (NIAID) has launched a Phase 1 trial of HSV529, an investigational vaccine for herpes simplex virus type 2 (HSV-2). The trial would test the safety of HSV529 and assess the vaccine’s ability to generate an immune system response. NIH estimated that 776,000 US residents acquired genital herpes each year. HSV-2 caused most genital herpes infections, but herpes simplex virus type 1 (HSV-1) also could cause genital herpes.

NIAID Director Anthony S. Fauci, MD, stated that genital herpes was a life-long infection that increased the risk of acquiring HIV. Women who had active genital herpes at the time of birth could transmit the virus to their infants, which could cause “severe illness and death.”

David Knipe, PhD, professor of microbiology and immunology at Harvard Medical School, developed the investigational vaccine, manufactured by Sanofi Pasteur. HSV529 was a replication-defective vaccine that removed two key proteins from the virus, preventing it from multiplying.

The trial plans to enroll 60 adults ages 18–40, divided into three 20-participant groups. Group 1 would consist of people coinfected with HSV-1 and HSV-2, or only with HSV-2. Group 2 would include only people with HSV-1. Group 3 would include only people never infected. Within each group, researchers randomly would assign 15 participants to receive three 0.5 milliliter doses of HSV529 and five participants to receive a saline-based placebo. Doses would take place at enrollment, one month later, and six months later. Researchers would monitor participants during the trial and for six additional months. The trial would use blood samples to evaluate the vaccine’s ability to stimulate an immune response to HSV-2. Researchers expected to complete the trial by October 2016.

Use the identifier NCT01915212 to learn more at http://www.ClinicalTrials.gov.

---

**How Zinc Starves Lethal Bacteria to Stop Infection**

Nov. 11, 2013 — Australian researchers have found that zinc can 'starve' one of the world's most deadly bacteria by preventing its uptake of an essential metal.

The finding, by infectious disease researchers at the University of Adelaide and The University of Queensland, opens the way for further work to design antibacterial agents in the fight against Streptococcus pneumoniae.

Streptococcus pneumoniae is responsible for more than one million deaths a year, killing children, the elderly and other vulnerable people by causing pneumonia, meningitis, and other serious infectious diseases.

Published today in the journal *Nature Chemical Biology*, the researchers describe how zinc "jams shut" a protein transporter in the bacteria so that it cannot take up manganese, an essential metal that Streptococcus pneumoniae needs to be able to invade and cause disease in humans.

"It's long been known that zinc plays an important role in the body's ability to protect against bacterial infection, but this is the first time anyone has been able to show how zinc actually blocks an essential pathway causing the bacteria to starve," says project leader Dr Christopher McDevitt, Research Fellow in the University of Adelaide's Research Centre for Infectious Diseases.

"This work spans fields from chemistry and biochemistry to microbiology and immunology to see, at an atomic level of detail, how this transport protein is responsible for keeping the bacteria alive by scavenging one essential metal (manganese), but at the same time also makes the bacteria vulnerable to being killed by another metal (zinc)," says Professor Bostjan Kobe, Professor of Structural Biology at The University of Queensland.

The study reveals that the bacterial transporter (PsaBCA) uses a 'spring-hammer' mechanism to bind the metals. The difference in size between the two metals, manganese and zinc, causes the transporter to bind them in different ways. The smaller size of zinc means that when it binds to the transporter, the mechanism closes too tightly around the zinc, causing an essential spring in the protein to unwind too far, jamming it shut and blocking the transporter from being able to take up manganese.

"Without manganese, these bacteria can easily be cleared by the immune system," says Dr McDevitt. "For the first time, we understand how these types of transporters function. With this new information we can start to design the next generation of antibacterial agents to target and block these essential transporters."
Molecular Interplay Explains Many Immunodeficiencies

Nov. 11, 2013 — Australian scientists have described an exquisitely balanced interplay of four molecules that trigger and govern antibody production in immune cells. As well as being an important basic science discovery, it helps explain why people with mutations in any one of the associated genes cannot fight infection effectively, and develop rare and crippling immunodeficiency disorders.

Our immune system is made of a number of different types of cells that undertake specific functions. Those that make antibodies are known as 'B cells', and they become active after infection. Once a B cell is activated, it can proliferate into thousands of clones, known as 'plasma cells', which patrol the body and secrete large amounts of antibody to destroy the invader.

Dr Lucinda Berglund and Associate Professor Stuart Tangye, from Sydney’s Garvan Institute of Medical Research, are the first to describe a specific molecular process that controls the activation and differentiation of B cells. They used human blood and tissue samples to show that the chemical messaging molecule interleukin 21 (IL-21) activates the STAT3 gene in B cells, which in turn triggers the expression of a molecule known as 'CD25', a cell surface receptor that attracts a second messaging molecule, interleukin 2 (IL-2). IL-21 and IL-2 then work co-operatively to induce plasma cell development and antibody production. Their findings are published in the international journal Blood, now online.

"The interesting and informative aspect of this finding for me is that some people have mutations in the IL-21 receptor, some have mutations in STAT3, while others have mutations in CD25, and they all have B cell defects," said Associate Professor Tangye.

"By examining B cells from people with specific genetic mutations, we revealed that both components of the IL-21 receptor are critical for B cell function—and people can have mutations in either, with equally debilitating effects. We see these effects in patients with X-linked severe combined immunodeficiency, whose impaired response to IL-21 causes severe antibody deficiency."

"Patients with mutations in the STAT3 gene develop Hyper IgE Syndrome, another rare immunodeficiency that manifests as compromised antibody production and greatly depleted immune defences.

Immunodeficiencies arising from mutations in single genes give scientists a unique opportunity to understand B cell signaling, and reveal potential targets for modulating B cell responses in immunodeficiency and autoimmunity.

The current study arose from analysing global gene expression in B cells from healthy people and people with STAT3 deficiency—which immediately highlighted genes that were poorly expressed in disease. The Tangye lab plans to investigate other genes that impact the function of B cells.

Understanding Immune System Memory—In a Roundabout Way

Nov. 11, 2013 — While the principle of immune memory has been known for decades, the exact molecular mechanisms underpinning it have remained a mystery. Australian scientists have now unraveled part of that mystery, identifying the role of a gene called STAT3, which acts as a kind of roundabout, directing chemical messenger molecules to various destinations.

An infection, or a vaccination, ‘primes’ the immune system, so that when you next encounter the same invader, your body ‘remembers’ it and quickly makes large amounts of exactly the right antibodies to quash the infection.

Once a cell is primed, traffic on the STAT3 roundabout speeds up enormously, as if the road has been upgraded and the signage much improved.

Primed immune cells, known as 'memory B cells', behave very differently from 'naïve B cells', which have never seen infection. Memory B cells act with great speed and efficiency, removing a pathogen so quickly that people frequently remain unaware they have been infected.
Patients with the rare immunodeficiency disorder, Hyper IgE Syndrome, caused by mutations in the STAT3 gene, have a ‘functional antibody deficiency’. While you can detect antibodies in their blood, those antibodies are not very good at fighting specific diseases or infections.

Through studying the blood cells of Hyper IgE patients over time, Associate Professor Stuart Tangye, Dr Elissa Deenick and Danielle Avery, from Sydney's Garvan Institute of Medical Research, have gained considerable insight into the STAT3 gene. They recently observed that naïve B cells in Hyper IgE patients barely respond to important signaling molecules, whereas their memory B cells behave in the same way as those of healthy people.

The lab members realized that naïve B cells need a very strong chemical signal indeed—targeting STAT3—to kick-start antibody production. Conversely, memory B cells only need faint signals to generate a huge antibody response. Even STAT3-compromised memory cells from Hyper IgE patients are functional. This breakthrough finding is published in the Journal of Experimental Medicine, now online.

"This study helped explain why patients who have mutations in STAT3 can't generate an effective secondary response to infection" said Associate Professor Tangye.

"STAT3 directly affects the creation of memory cells, and so while these patients have a few, they are reduced tenfold."

"When people mount a normal primary or secondary immune response, various messenger molecules known as 'cytokines' bind to receptors on the cell surface and activate STAT3."

"Many structurally different cytokines, with complementary roles in antibody production, converge at STAT3—it's literally like a roundabout, showing cytokines which route to take next within the cell."

"This study has shown us that memory cells are much more sensitive to the cytokine signals they receive. They are more robust and efficient, and the magnitude of their response is much greater than that of naïve cells."

"B cells fundamentally change their biology between the naïve state and the memory state. STAT3 appears to be the key to this molecular rewiring—because without it, memory cells cannot form properly."

"These findings explain a lot to me about how immunological memory works, and also throw more light on Hyper IgE Syndrome. They also tell us that if you want to improve antibody responses, there are certain pathways and cell types that can be targeted."

"We can see the future potential to amplify the potency of vaccines, as well as help Hyper IgE patients."

Journal Reference:
Stuart Tangye et al. Naive and memory human B cells have distinct requirements for STAT3 activation to differentiate into antibody-secreting plasma cells. Journal of Experimental Medicine, November 2013

New Cause Found for Muscle-Weakening Disease Myasthenia Gravis
Nov. 11, 2013 — An antibody to a protein critical to enabling the brain to talk to muscles has been identified as a cause of myasthenia gravis, researchers report.

The finding that an antibody to LRP4 is a cause of the most common disease affecting brain-muscle interaction helps explain why as many as 10 percent of patients have classic symptoms, like drooping eyelids and generalized muscle weakness, yet their blood provides no clue of the cause, said Dr. Lin Mei, Director of the Institute of Molecular Medicine and Genetics at the Medical College of Georgia at Georgia Regents University.

"You end up with patients who have no real diagnosis," Mei said.

The finding also shows that LRP4 is important, not only to the formation of the neuromuscular junction—where the brain and muscle talk—but also maintaining this important connection, said Mei, corresponding author of the paper in The Journal of Clinical Investigation.

Mei and his colleagues first reported antibodies to LRP4 in the blood of myasthenia gravis patients in the Archives of Neurology in 2012. For the new study, they went back to animals to determine whether the antibodies were harmless or actually caused the disease. When they gave healthy mice LRP4 antibodies, they experienced classic symptoms of the disease along with clear evidence of degradation of the neuromuscular junction.

LRP4 antibodies are the third cause identified for the autoimmune disease, which affects about 20 out of 100,000 people, primarily women under 40 and men over age 60, according to the National Institutes of Health and Myasthenia Gravis Foundation of America, Inc.

An antibody to the acetylcholine receptor is causative in about 80 percent of patients, said Dr. Michael H. Rivner, MCG neurologist and Director of the Electrodiagnostic Medicine Laboratory, who follows about 250 patients with myasthenia gravis. Acetylcholine is a chemical released by neurons
which act on receptors on the muscle to activate the muscle. More recently, it was found that **maybe 10 percent of patients have an antibody to MuSK, an enzyme that supports the clustering of these receptors on the surface of muscle cells.**

"That leaves us with only about 10 percent of patients who are double negative, which means patients lack antibodies to acetylcholine receptors and MuSK," said Rivner, a troubling scenario for physicians and patients alike. "This is pretty exciting because it is a new form of the disease," Rivner said of the LRP4 finding.

Currently, physicians like Rivner tell patients who lack antibody evidence that clinically they appear to have the disease. Identifying specific causes enables a more complete diagnosis for more patients in the short term and hopefully will lead to development of more targeted therapies with fewer side effects, Rivner said.

To learn more about the role of the LRP4 antibody, Mei now wants to know if there are defining characteristics of patients who have it, such as more severe disease or whether it’s found more commonly in a certain age or sex. He and Rivner have teamed up to develop a network of 17 centers, like GR Medical Center, where patients are treated to get these questions answered. They are currently pursuing federal funding for studies they hope will include examining blood, physical characteristics, therapies and more.

Regardless of the specific cause, disease symptoms tend to respond well to therapy, which typically includes chronic use of drugs that suppress the immune response, Rivner said. However, immunosuppressive drugs carry significant risk, including infection and cancer, he said.

Removal of the thymus, a sort of classroom where T cells, which direct the immune response, learn early in life what to attack and what to ignore, is another common therapy for myasthenia gravis. While the gland usually atrophies in adults, patients with myasthenia gravis tend to have enlarged glands. Rivner is part of an NIH-funded study to determine whether gland removal really benefits patients. Other therapies include a plasma exchange for acutely ill patient.

**Journal Reference:**
Chengyong Shen, Yisheng Lu, Bin Zhang, Dwight Figueiredo, Jonathan Bean, Jiung Jung, Haitao Wu, Arnab Barik, Dong-Min Yin, Wen-Cheng Xiong, Lin Mei. *Antibodies against low-density lipoprotein receptor–related protein 4 induce myasthenia gravis.* *Journal of Clinical Investigation*, 2013; DOI: [10.1172/JCI66039](https://doi.org/10.1172/JCI66039)

**'He’s Pretty Spunky' — Anesthesia Comes of Age During Civil War**

Nov. 11, 2013 — Anesthesia was in its infancy when the American Civil War began in 1861. The sheer number of casualties gave surgeons on both sides the opportunity to gain experience with the first two anesthetic agents developed—sulfuric ether and chloroform—according to a paper by a University of Alabama at Birmingham anesthesiologist published in the October issue of the *Scandinavian Journal of Pain.*

"As we honor the sesquicentennial, or 150th anniversary, of the Civil War, it is still widely believed that the sole anesthetic agent used was the whiskey bottle," said Maurice S. Albin, M.D., professor in the UAB Department of Anesthesiology. "But sulfuric ether was first used in 1846, and chloroform a year later."

The Mexican-American War in the late 1840s and the Crimean War in the mid-1850s saw the first battlefield use of these agents on a small scale. In the Civil War, Albin estimates that anesthesia was used at least 125,000 times by surgeons for both the North and South who, before the war, had limited or no experience with anesthetic agents. That number pales in comparison to the estimated 476,000 men wounded on both sides during the conflict and 620,000 killed, many of whom no doubt underwent a surgical procedure.

Albin’s paper reports on two wounded warriors, one a captured Union private and the other a celebrated Confederate general.

"These two case stories illustrate the profound improvement in surgical pain management made possible with anesthesia only 150 years ago," said Albin. "Most surgeons and patients today have no idea what these important improvements meant to modern medicine."

Albin, himself a World War II combat medic, tracked the story of Union Private James Winchell, a member of Berdan’s First United States Sharpshooters. Winchell was one of 3,107 Union soldiers wounded at the Battle of Gaines Mill in 1862, and one of 2,836 captured. A musket ball struck his left arm between the shoulder and elbow. His group of some 500 wounded prisoners was treated by a single surgeon, so Winchell had to wait.

Five days passed before Winchell’s shattered arm was removed, days he spent in great pain, lying under a tree shooing flies from his arm. Albin traced Winchell's story from a book on Berdan’s
Sharpshooters written by Captain C.A. Stevens, a veteran of the regiment who interviewed Winchell at a reunion in 1890. Stevens quoted Winchell's recollection of when the surgeon finally came for him. He was one of the many who did not receive anesthesia.

"I asked if he had any chloroform or quinine or whiskey," Winchell said to Stevens. "He said 'no, and I have no time to dilly-dally with you.'"

Winchell was seated in a chair, and men held his shoulders as the surgeon began to cut. After removing the bone, the surgeon suggested they pause an hour to let Winchell recover.

"I refused and told them I wanted one job of it, as I was just as ready to kick the bucket then as in one hour," Winchell told Stevens.

At this point, the surgeon and his men said of Winchell, "He's pretty spunky. Let's make a good job of it."

Amazingly, Winchell survived, was exchanged and returned home. Some 30 years later, he was still alive to tell his tale. The second individual in Albin's narrative was not as fortunate.

Lt. General Thomas "Stonewall" Jackson suffered a wound eerily similar to Winchell's. Jackson was shot in the left arm by nervous sentries from his own army following an evening reconnaissance during the Battle of Chancellorsville. He was taken to a field hospital and attended to by Hunter Holmes McGuire, a pre-eminent surgeon in the Confederate Army who determined that amputation of the arm was necessary. Chloroform was the anesthetic used, dropped on a cloth in the shape of a cone.

Jackson is recorded as saying "what an infinite blessing" as the chloroform took effect. A team of surgeons led by McGuire performed the operation to remove his arm. It took about 50 minutes, and he was under anesthesia for just over an hour, awaking shortly after. At first Jackson seemed to make a rapid recovery, but he developed pneumonia—most likely from the arduous journey from battlefield to field hospital—and died a week later.

Albin says the science of anesthesiology has come far since those days when a general got a few drops of chloroform and a private was lauded for his spunk.

"We now routinely do complicated, lengthy operations, using a variety of anesthetic agents that are best suited for each individual patient," said Albin. "Groundbreaking work done here at UAB and at other institutions has made remarkable progress in minimizing post-operative pain as well."

Battlefield medicine has also made great strides in recent years. Far-forward medical teams can provide emergency and surgical care to a wounded soldier within minutes, then transport him or her to a fully equipped theater hospital nearby. UAB is home to one of the most advanced military medical teams, the U.S. Air Force SOST-SOCCET special operations team.

Journal Reference:

Did Inefficient Cellular Machinery Evolve to Fight Viruses and Jumping Genes?

Nov. 7, 2013 — It might seem obvious that humans are elegant and sophisticated beings in comparison to lowly bacteria, but when it comes to genes, a UC San Francisco scientist wants to turn conventional wisdom about human and bacterial evolution on its head.

Far from being sleekly performing and fine-tuned athletes, the molecules guiding the activity of our genes are like sour bureaucrats that clog up the works and create unnecessary inefficiency, asserts Hiten Madhani, MD, PhD, a professor of biochemistry and biophysics at UCSF. In contrast, bacteria carry out these processes efficiently with less frustration for the gene to express itself.


Although his thinking was stimulated by his own research findings, Madhani described his Cell essay as a "just so" story, a conjecture that challenges conventional thinking, but that so far is without data to back it up. He paraphrased a source of inspiration, the renowned scientist Sydney Brenner, who won a Nobel Prize for his own studies of gene regulation. "Biology is awash in a sea of data, but it needs new theories," Madhani said.

Most scientists believe that the complexity of the molecular mechanisms that guide the expression of genes and the production of proteins within a human cell is needed to allow for flexible responses that drive the development and maintenance of multifaceted organism, Madhani said.
But he proposes that this complexity in genetic regulatory machinery did not originally evolve to allow for the development of the whole human. Instead, he suggested, complexity in gene expression might have first evolved in early eukaryotes to thwart infection by "parasitic DNA," such as retroviruses, that would otherwise invade the cell nucleus and disrupt normal genes.

In contrast to humans, bacteria control their genes and have adaptively evolved in myriad ways without complex mechanisms like those that guide human gene expression. In fact, humans, whose cells number in the many trillions, and disease-causing bacteria, which are but a single cell, have been doing battle and evolving together for ages, with multidrug-resistant bacteria perhaps being latest type of villain to emerge in this epic struggle.

Bacteria have persisted despite their simplicity. They have only one gene-bearing chromosome and lack any kind of cell nucleus. The bacterial chromosome itself lacks the modifiable, protective sheath known as chromatin. Many other details of gene expression differ between human and bacterial cells. Bacteria are known as "prokaryotes," a name that refers to the fact that they arose before cells evolved that had a nucleus—more than 3 billion years ago, according to some estimates made from fossils. Human cells have a nucleus and numerous other features that peg them as "eukaryotes."

While humans evolved from apes just a few million years ago, eukaryotes have been around since the ancestors of single-celled yeast arose, perhaps 1.5 billion years ago—with the same complex features, Madhani said.

"It might be tempting to think that the complex attributes of human gene expression evolved to drive the evolution of complex, multicellular organisms," Madhani said, "But the core elements of eukaryotic gene expression were established within the ancient unicellular progenitor of modern eukaryotes." In other words, the early eukaryotic cell already was adapting to ward off parasitic DNA, he suggested.

Madhani said his idea stems from research he published earlier this year. His research group discovered that a eukaryotic cellular machine known as SCANR plays a previously unrecognized role in thwarting corruption of the genome by parasitic DNA.

SCANR guards against DNA called jumping genes, or transposons, which long ago invaded the human genome. Transposons replicate multiple times, and insert themselves at random places within genomic DNA. When transposons insert themselves in the middle of an important gene, they may cause malfunction, disease or birth defects.

Madhani began thinking about how other mechanisms in the cell might similarly stymie certain viruses, which unlike bacterial pathogens, depend on the genetic machinery of their human hosts in order to replicate.

"Transposable elements attack from within the genome, and viruses attack from outside," Madhani said.

In addition to the chromatin that restricts access to DNA, eukaryotic cells also have embellishments to their RNA, and molecular inspectors that check to see that these eukaryotic modifications are present before protein production proceeds. The nucleus itself is gated to allow only certain molecules to get in and out. Many other eukaryotic cellular phenomena might have first evolved to defend against viruses and transposable elements, Madhani said.

**Journal Reference:**

---

**Congress opens door to allowing HIV organ donations**

By Pete Kasperowicz

The House this week will take a step toward ending a ban on organ donations from HIV-positive patients.

The House will pass S. 330, the HIV Organ Policy Equity (HOPE) Act, legislation that the Senate passed in June by unanimous consent.

The bipartisan bill would re-write language in the Organ Transplant Amendments Act of 1988, which was quickly passed by voice vote in the House and Senate in 1988. That bill, from Sen. Ted Kennedy (D-Mass.), was meant to ensure that organs from HIV patients would not be given to non-HIV patients.

However, supporters of this week's bill say the 1988 language is "medically outdated." As HIV patients live longer, many are in need of new organs, and some doctors say they would face a much shorter waiting period if organs from other HIV patients were available.

Under the bill, from Sens. Barbara Boxer (D-Calif.) and Tom Coburn (R-Okla.), the Department of Health and Human Services (HHS) would be allowed to evaluate the state of medical research into HIV
organ transplants. If the research shows the transplants can be done, HHS would be able to direct the Organ Procurement and Transplantation Network to establish procedures for these operations.

Some estimates say allowing these operations could open up another 600 organ donations each year. "This legislation offers hope for thousands of patients who are waiting for transplants by allowing scientists to research safe and effective ways to transplant these organs and save lives," Boxer said in June after the Senate passed the bill.

The House will consider the bill as early as Tuesday. It will be considered as a suspension bill, which means it will get less debate and will need a two-thirds vote for passage — suspension bills are usually non-controversial and pass easily.

**Magic Johnson Dedicates Space for World AIDS Museum in Wilton Manors**

*Sun Sentinel*, (11.07.2013) By Ariel Barkhurst

The Sun Sentinel reported about the dedication of the World AIDS Museum and Educational Center where Earvin “Magic” Johnson spoke on the 21st anniversary of his own HIV-positive announcement. In front of a crowd of approximately 100 people, Johnson touched on how the world is combatting the stigma of HIV/AIDS as well as the medical advances since his diagnosis. "When I first started treatment years ago, it was 15 pills three times a day," Johnson said. "They said it was because I'm a big guy. So my height worked against me. But now, it's pills once a day."

The museum, located in Wilton Manors, Fla., just outside of Ft. Lauderdale, is the first global space dedicated to "telling the story of HIV and AIDS," according to Museum President Steven Stagon. "I'm so glad we got this museum started," Johnson said. "We've all got to work together and that's the key."

The museum will open early next year and is approximately 3,000 square feet, although the board hopes it will expand eventually. Funding for the museum came from a $94,000 grant and 34 individual donors. Stagon believes the location is ideal because the area does not have many museums currently, yet it hosts 12 million tourists each year. "This is the epicenter for the AIDS epidemic in America right now," he said.

Board Chairperson Hugh Beswick said featured exhibits will include themes such as "AIDS chronology," where visitors walk through a multimedia timeline of the history of the disease. A dedicated room will allow people to film their personal stories, which become part of an online “virtual quilt” of multiple stories. The sole exhibit now on display is of a large red ribbon sculpture constructed from artist Ed Sparan’s 417 empty HIV medication bottles, which was worth $85,000 of medication. "That's 10 years' worth," Sparan said of the three-foot-high sculpture. "Magic is 21 years. His would be twice the size."

**HPV Rates up to Three Times Higher in American Indian Women Than US Population**

*Indian Country Today Media Network*, (11.11.2013) By Charlotte Hofer

Indian Country Today Media Network reported that Native Americans in the US Northern Plains have a higher rate of HPV-related cancer morbidity and mortality than whites in the same area and Native Americans in other regions, and are two to three times more likely to be infected with cervical cancer than white women living in the same region. Dr. Delf Schmidt-Grimminger, a scientist with Avera Research Institute in Sioux Falls, S.D., discussed the prevalence of human papillomavirus (HPV) and the benefit of the HPV vaccine and cultural-specific education among American Indian women. He spoke at a symposium on November 5 in Eagle Butte, S.D., on HPV and related cancers. Approximately 75 Cheyenne River Health Care providers, health department officials, and others attended the symposium.

Schmidt-Grimminger noted that HPV prevalence is three times higher in American Indian women than in the rest of the US population; approximately 70 of every 100 American Indian women ages 18–24 is infected. He advocated vaccinating both girls and boys as well as administering Pap tests to significantly reduce the incidence of cancer caused by this virus. Schmidt-Grimminger also commented on the need to continue sharing knowledge with other communities for additional funding to continue HPV vaccinations to save more lives.

Ann LeBeau, a Cheyenne River Sioux Tribe member working in behavioral health counseling, commented on the need for information dissemination to the community on cancer prevention. Le Beau also mentioned patients' reluctance to share personal information in their medical history with providers.

Gayle Dupris, residential aid at the Women’s Half-Way House at Cheyenne River, emphasized the need for education tailored to the community. Additional comments included the need for more outreach programs and education in the schools, and more culturally appropriate resources.
CONRAD presents new technology combining contraception, HIV and herpes simplex virus-2 prevention
Multipurpose prevention technologies featured at AAPS Annual Meeting and International Conference on Family Planning

Arlington, Va. — CONRAD Head of drug delivery, Meredith Clark, PhD, today presented preclinical data on a new intravaginal ring that provides contraception as well as HIV-1 and HSV-2 prevention at the 2013 American Association of Pharmaceutical Scientists (AAPS) Annual Meeting and Exposition in San Antonio, Texas. This multipurpose prevention technology (MPT) can remain in the vagina for up to 90 days and releases the contraceptive levonorgestrel (LNG) and tenofovir (TFV), an antiretroviral that inhibits HIV and HSV replication in susceptible cells.

The CONRAD product development team, in collaboration with Dr. Patrick Kiser at Northwestern University, performed in vitro release testing and 3-month pharmacokinetic (PK) studies of the ring in rabbits and sheep, and compared drug levels to those seen with use of tenofovir gel. The PK studies found that levels of tenofovir in the target tissue delivered from the ring are similar or higher than those obtained after TFV 1% gel application, a product that has proven to be effective in preventing HIV and HSV infections in women. In addition, release of the contraceptive agent was also consistent with previous levels tested to be efficacious in women. Stability studies will continue and lead to Phase I clinical trials in women in 2014, which will test the combination ring, as well as a tenofovir-only ring.

Tenofovir is the first microbicide proven to be efficacious in humans, with the CAPRISA 004 clinical trial showing that women using the gel before and after sex reduced their risk of HIV infection by 39-54%. CAPRISA 004 also showed the gel to be 51% effective in reducing the transmission of HSV-2, making this combination ring potentially triple protective.

"The TFV/LNG ring is the first device to be tested in women that will offer contraception as well as HIV and herpes prevention," said Dr. Clark. "And so far, tenofovir is the only microbicide that has been proven to be effective in reducing HIV infections when used topically. It's important to develop a variety of delivery mechanisms for tenofovir in order to serve different women's needs."

CONRAD’s product development director David Friend Ph.D added, "Products only work when they are used. By having a ring that can remain in the body for up to 90 days, our hope is that this ring will offer a solution to increase adherence, and therefore provide greater protection against HIV while also preventing pregnancy."

CONRAD’s deputy director of clinical research, Marianne Callahan, will also present information on MPTs later this week at the International Conference on Family Planning in Addis Ababa, Ethiopia. The CONRAD sponsored panel, "Development of Multipurpose Prevention Technologies (MPTs): Pathway from Product Development to the End Users," will discuss how the development of MPTs are tied to the regulatory approval process, the importance of acceptability research within target populations,
and the importance of taking a "systems approach" when considering feasibility of future introduction of a new technology.

In addition to the TFV/LNG intravaginal ring, CONRAD is testing the one-size-fits-most SILCS diaphragm with tenofovir gel. Used together, the diaphragm plus the gel can offer contraception plus the potential to reduce HIV and HSV-2 infections as an on-demand system providing immediate protection.

According to the World Health Organization, there are 35.3 million people living with HIV around the world and approximately 87 million unintended pregnancies occur each year. Ms. Callahan says, "An unintended pregnancy is more tangible than an invisible virus so MPTs may lead to increased product use by offering a crucial combination of protection that can have a major impact in developing countries."

**Putting the Brakes on Immunity**

Tuesday, November 12, 2013

TAU researchers discover a powerful mechanism that keeps white blood cells from going rogue

The immune system is a double-edged sword. While its primary role is to fight infections, it can also become overactive, leading to problems like allergies and autoimmune diseases.

For example, the part of the immune system responsible for resisting parasites’ acts by releasing white blood cells called eosinophil granulocytes into the blood. But elevated eosinophil levels are also responsible for allergic reactions, including most forms of asthma, gastrointestinal diseases, blood disorders, and cancers.

Now a study, led by Dr. Ariel Munitz of the Department of Clinical Microbiology and Immunology at the Sackler School of Medicine at Tel Aviv University, and conducted by graduate students Netali Baruch Morgenstern and Dana Shik, has found a mechanism that pushes eosinophils to die before they get into the blood and wreak havoc. The discovery is a breakthrough in science's understanding of the immune system and suggests powerful new treatments for eosinophilic diseases such as asthma.

"We've discovered an important and powerful pathway that works to kill eosinophils," says Dr. Munitz. "The fundamental knowledge we have gained may one day yield even bigger results and therapies."

Published online in *Nature Immunology* in November, the research was funded in part by the United States-Israel Binational Science Foundation, the Israel Science Foundation, the Israel Cancer Research Fund, and the Fritz Thyssen Stiftung. The Division of Allergy and Immunology at the Cincinnati Children's Hospital Medical Center collaborated on it.

**The body's tug-of-war**

The level of eosinophils in the blood is relatively low in healthy people, accounting for just 2 to 5 percent of white blood cells in circulation. But in eosinophilic disorders, a signalling protein called interleukin 5, or IL-5, triggers a rush of eosinophils from the bone marrow, where they are produced, and into the blood, where they are transported to various organs. IL-5 has lately been investigated as a new target for asthma medications, some of which have proven effective in clinical trials.

Analyzing the bone marrow of mice, the researchers found that the expansion of eosinophils caused by IL-5 is actually part of a broader mechanism that regulates the lifecycle of the cells. While IL-5 commands eosinophils to expand and enter the bloodstream, a cell receptor called paired immunoglobulin-like receptor A, or PIR-A, commands eosinophils to die. So eosinophils are in a constant "tug-of-war" between survival signals delivered by IL-5 and death orders delivered by PIR-A.

Although the death order by PIR-A is dominant, it is never executed. Eosinophils express another receptor, called PIR-B, which closely resembles PIR-A and inhibits its actions. In order for PIR-A to carry out its death order to the cell, PIR-B must be shut down.

"PIR-A is always inhibited by PIR-B from the very early stages of eosinophil development," says Dr. Munitz. "We had to remove the expression of PIR-B from the cells to see PIR-A's powerful effects."

**Two new approaches to nip disease in the bud**

After identifying the mechanism in cell culture systems, the researchers verified that it also operates in mice. As expected, they found that asthmatic mice without PIR-B in their bodies had very little expansion of eosinophils into their blood and lungs and therefore less asthmatic inflammation in their lungs than normal mice. Unhindered by PIR-B, PIR-A appeared to keep eosinophils from reaching harmful levels in their bodies. Because human eosinophils also express PIR-like molecules, there is good reason to believe the same mechanism works in people.
In addition to advancing knowledge of eosinophils — a basic and important cell type — the researchers’ work opens up two new avenues for treating eosinophilic disorders. Instead of lowering IL-5 levels to try to reduce eosinophil expansion, scientists can now target PIR-A to enhance its ability to kill eosinophils. Alternatively, they could weaken PIR-B so that it inhibits PIR-A less.

The researchers have preliminary evidence that PIR-B inhibits other mechanisms that drive cell death. Identifying them is the focus of their current research.

**Anthrax toxin can lurk for days in cells as a lingering threat**

The deadly toxin produced by anthrax bacteria can hide out in human cells for days, invisible both to our immune systems and to the cellular machinery responsible for destroying proteins. The findings reported in the Cell Press journal *Cell Reports* on November 14th explain why antibiotics aren’t always enough to cure anthrax infections.

"The anthrax bacteria kills people in a very short period of time, and this is in large part due to the production of the anthrax lethal toxin," said Gisou van der Goot of the École Polytechnique Fédérale de Lausanne. "This toxin disarms our immune system, but also, as very recently shown, affects our heart."

"Many years ago, we had noticed that the effect of anthrax lethal toxin was detectable for more than a week in cells that had been exposed to the toxin for less than one single hour," she added. "We wanted to understand how this was possible."

To find out just how the anthrax toxin could do its damage over time and space, in the new study the researchers examined the toxin's complex delivery route. The toxin itself has two main ingredients: the damaging lethal factor itself and a protective antigen required for cells to take up and move that killer protein.

Protective antigen helps the lethal factor enter cells by forming channels. Van der Goot and her colleagues now confirm their earlier suspicion that those channels might be capable of delivering toxin not just into cells themselves, but also into smaller sacs or vesicles within the larger cell.

Once safely inside those vesicles, the lethal factor can persist for days without degradation, the researchers show. They were surprised to find that while sheltered inside those vesicles, the toxin can also be passed on from one cell to its daughters and from one cell to another.

The findings help to explain why anthrax infection is so devastatingly deadly, but this new understanding of these bacterial weapons and their sneaky behavior does come with an upside for science.

"By studying these interactions, we can learn more than how to fight anthrax infection," van der Goot said. "We also learn a lot about how cells work."

*Cell Reports,* Abrami et al.: "Hijacking multivesicular bodies enables long-term and exosome-mediated long-distance action of anthrax toxin."

**New research reveals dengue fever mystery in 2 US cities both exposed to risk**

ASTMH Annual Meeting showcases new findings on dengue and risks of future spread in United States and abroad; One new project uses high-resolution satellite images to map potential disease hotspots

As dengue fever continues to spread from Key West north into the Florida mainland, it remains a mystery as to why this dangerous mosquito-borne illness is not yet common around Tucson, Arizona—even though outbreaks routinely occur in nearby Mexico and mosquitoes that can carry dengue are now common in the state, according to a new research presented today at the annual meeting of the American Society of Tropical Medicine and Hygiene (ASTMH).

"Key West and Tucson share a lot of risk factors. Even in arid Tucson we have a large population of mosquitoes that can carry dengue, and people here spend a lot of time outdoors, but we have yet to see evidence of locally-acquired infections," said Kacey Ernst, PhD, an infectious disease epidemiologist at the University of Arizona in Tucson.

Ernst presented preliminary findings from an ongoing study she is conducting with Mary Hayden, PhD, a scientist at the National Center for Atmospheric Research (NCAR) in Boulder, Colorado, whose work focuses on the link between human behavior and climate-related health threats.

Their survey of residents in the two cities 2,000 miles apart and with different climates turned up many similarities. More than half of the respondents in both cities reported spending at least an hour outside each day. The use of central air conditioning, thought to lower the risk of dengue, was virtually identical (55% in Key West vs. 56% for Tucson), though some of the Tucson air conditioning systems are
less dengue-friendly. They note that many people in the Tucson area use evaporative cooling systems or "swamp coolers," which increase the humidity in the home and may increase mosquito survivability, particularly in a desert environment.

"It is still a mystery as to why dengue infection has not shown up here," Ernst said. "When researchers looked at why dengue is not more common along the Texas side of the Mexico border, they cited factors limiting contact with mosquitoes, like people spending a lot of time in sealed, air conditioned buildings. Those issues are extremely important considerations, but we don't think they fully explain why Key West has dengue and Tucson doesn't."

She noted that in some ways Tucson could be seen as more vulnerable to dengue because mosquito control campaigns in the area are far less intensive than what one finds in Key West. Ernst said it's possible that there have been sporadic dengue infections in Tucson that have gone undetected or that there is something about the mosquito's life-cycle that is inhibiting its ability to transmit dengue.

Ernst and Hayden are also investigating a similar situation in Nogales in the Mexican northern border state of Sonora, where the primary dengue virus vector Aedes aegypti mosquitos are common, but no dengue appears to be present—even though social and economic conditions are similar to areas of Mexico where dengue is endemic.

Overall, the researchers want to understand more about the human and biological factors that allow dengue to move into some areas but not others. Their goal is to help prevent the disease from spreading further into the United States. A once obscure infection, dengue now threatens 40 percent of the world's population and has become endemic in Mexico.

Dengue can cause everything from mild, flu-like symptoms to an excruciating joint pain that has earned it the nickname "break-bone fever." Infections can sometimes progress to a potentially fatal form of hemorrhagic fever. There are no drugs available to treat or cure an infection. At the ASTMH meeting, scientists discussed new efforts to develop a dengue vaccine.

"I think you see researchers in the United States and around the world mounting a full-court press to try to keep dengue in check," said ASTMH President David H. Walker, MD. "One of the reasons we hold an Annual Meeting is to allow scientists who are focusing on different aspects of the disease—the biology of the virus itself, the spread of the vector, and the role of human behavior—to share their latest findings and ultimately curtail the suffering from this disease."

Researchers believe dengue was absent from the continental United States for more than a half century. But in the last nine years there have been reports of small, isolated dengue outbreaks—in 2004 and 2005 in south Texas and from 2009 to 2011 in Key West. Most recently, in August of this year, 20 cases of "locally-acquired" dengue—meaning the infections were not travel related—were confirmed in the Jensen Beach area of South Florida's Treasure Coast, evidence that the disease may be moving north.

But so far, aside from the Texas and Florida cases, there have been no recent reports of locally acquired dengue elsewhere in the continental United States.

**An Old Enemy: Aedes aegypti Mosquitoes in the United States**

The Aedes aegypti mosquito occurs in the southern United States, from Florida in the east to Arizona in the west, and was also recently discovered in California.

At the ASTMH meeting, Lars Eisen, PhD, an expert in vector-borne diseases at Colorado State University, discussed the future threat posed by Aedes aegypti and its associated pathogens for human health in the United States. He noted that the mosquito has made a steady comeback across the Americas after the discontinuation of an intense control campaign undertaken in the mid-20th century by the Pan-American Health Organization (PAHO) that eliminated the mosquito from many countries. These ultimately unsustainable control efforts did not include the United States, Eisen said, and he believes Aedes aegypti has been present continuously in the far southeastern United States for hundreds of years.

This summer, unexpected populations of Aedes aegypti were reported in Fresno and Madera counties in California's Central Valley, and in San Mateo County in the Bay Area. But Eisen cautioned that, as is being seen in Arizona, it will take more than just the presence of Aedes aegypti mosquitoes to cause a dengue outbreak.

"Things like having houses with air conditioning and screens on the windows reduce indoor biting, and basic services like regular trash collection and reliable access to piped water reduce breeding opportunities," he said. "Collectively, they can significantly limit opportunities for virus transmission."

Eisen pointed out that even when dengue does arise in the United States, these factors seem to be keeping local virus transmission at low levels. He contrasted the relatively small number of cases found in Florida and Texas to the many thousands of dengue infections regularly reported in Puerto Rico and other Caribbean islands, and in Mexico. Eisen also said that while rising temperatures caused by climate change
could bring the mosquito farther north in the United States, the many elements that influence the mosquito itself and the transmission of dengue virus argue strongly against looking solely at climate to assess disease threat.

**Spying on Mosquitoes from Space**

Eisen also noted a new effort underway using satellite imagery to help detect areas that might be at risk of dengue outbreaks.

The project involves a collaboration between Science and Technology in Atmospheric Research (STAR) and NCAR—both located in Boulder. Researchers involved in the initiative are developing, among other things, a way to search high-resolution satellite images to detect the "container" habitats preferred by *Aedes aegypti* mosquitoes, said Paul Bieringer, PhD, a scientist with NCAR's National Security Applications Program.

*Aedes aegypti* can be difficult to control because, unlike mosquitoes that spread West Nile Virus, which prefer water sources like ponds and sewers that can be easily treated with larvicides, *Aedes aegypti* seek out water-filled containers, like an empty can, bucket or old tire that collects rain water. This preference is why they are sometimes called "container breeders."

Bieringer said that satellites are now capable of providing clear images of relatively small objects on the ground, allowing researchers to employ sophisticated computer programs to scan these images and identify potential hotspots capable of harboring large populations of *Aedes aegypti* mosquitoes.

"Our goal is to develop maps that can simulate the abundance of this dengue virus carrier in a particular area, and then use these insights along with demographic and economic data, to predict where disease outbreaks are most likely to occur," he said.

**Anthrax bacteria play hide and seek**

An EPFL team discovers that, using exosomes, the lethal factor of the anthrax bacterium can travel undetected through the body for days

The bacterium responsible for anthrax develops a strategy reminiscent of the Trojan horse tale. Its pathogenic factor is able to penetrate inside a cell in such a way that it becomes completely invisible to both the immune system and medical analysis. Furthermore, it manages to exit the cell several days later, and then it continues to poison other cells.

This mechanism was discovered by researchers from EPFL, the University of California at Berkeley and the National Institute of Health in Washington. It finally explains the reason why some living organisms succumb to the disease up to two weeks after the disappearance of the last signs of bacterial presence. "This remained a mystery for more than 50 years, said Gisou van der Goot, who heads a research unit at EPFL's Global Health Institute. The bacteria would disappear after the administration of antibiotics, but the subject still died a few days later."

The researchers focused in the way the anthrax toxin was able to get inside the cell. Composed of two elements—a "protective antigen" and a "lethal factor", the toxin does not merely create a passage across
the cellular membrane. Instead, it introduces itself by endocytosis, a process by means of which the pathogen is "swallowed" by the cell.

The intoxication does not stop there. Once inside the cell, anthrax's lethal factor is sheltered by the cell's membrane, forming an "endosome", in which it can wait for several days. Then, it can either be released inside the cell, causing it to malfunction, or it can be released towards the external environment inside small vesicles – called exosomes – and get into another cell. "The immune system has no reason to react, since it only detects exosomes whose membrane is composed by the very same molecules making up the cell's endosomes," explained Gisou van der Goot.

This is the first time that scientists have been able to describe the transmission of a pathogen agent for an extended time period and throughout a long distance within the living organism. Their work has been subsidized by the Swiss National Science Foundation and the NCCR "Chemical Biology". It was published today in the Cell Reports journal. "There is still much to learn about exosomes. The results of this research will help us to better understand them" continued Gisou van der Goot.

As for the battle against anthrax, this research will lead to the development of drugs specifically targeting the lethal factor while being able to penetrate the cell's membrane.

**New Treatment Discovered to Cure MRSA Infection**

Nov. 13, 2013 — Recent work from University Distinguished Professor of Biology Kim Lewis promises to overcome one of the leading public health threats of our time. In a ground breaking study published today in the journal Nature, Lewis' team presents a novel approach to treat and eliminate methicillin resistant *staphylococcus aureus*, or MRSA, a potent bacterium whose resistance to antibiotics has kept it one step ahead of researchers. That is, until now.

The so-called "superbug" infects 1 million Americans each year. A major problem with MRSA is the development of deep-seated chronic infections such as osteomyelitis (bone infection), endocarditis (heart infection), or infections of implanted medical devices. Once established, these infections are often incurable, even when appropriate antibiotics are used.

Bacteria such as MRSA have evolved to actively resist certain antibiotics, a fact that has generated significant interest among the scientific and medical communities. But Lewis, Director of Northeastern’s Antimicrobial Discovery Center, suspected that a different adaptive function of bacteria might be the true culprit in making these infections so devastating.

The new work represents the culmination of more than a decade of research on a specialized class of cells produced by all pathogens called persisters. According to Lewis, these cells evolved to survive. "Survival is their only function," he said. "They don't do anything else."

Lewis and his research team posited that if they could kill these expert survivors, perhaps they could cure chronic infections—even those resistant to multiple antibiotics such as MRSA. Furthermore, said Brian Conlon, a postdoctoral researcher in Lewis' lab and first author on the paper, "if you can eradicate the persisters, there's less of a chance that resistance will develop at all."

Lewis, who was elected to the American Academy of Microbiology in 2011 for his scholarship in the field, has found that persisters achieve their singular goal by entering a dormant state that makes them impervious to traditional antibiotics. Since these drugs work by targeting active cellular functions, they are useless against dormant persisters, which aren’t active at all. For this reason, persisters are critical to the success of chronic infections and biofilms, because as soon as a treatment runs its course, their reawakening allows for the infection to establish itself anew.

In the recent study, which also includes contributions from assistant professor Steve Leonard of the Department of Pharmacy Practice, Lewis’ team found that a drug called ADEP effectively wakes up the dormant cells and then initiates a self-destruct mechanism. The approach completely eradicated MRSA cells in a variety of laboratory experiments and, importantly, in a mouse model of chronic MRSA infection.

Coupling ADEP with a traditional antibiotic, Conlon noted, allowed the team to completely destroy the bacterial population without leaving any survivors.

As with all other antibiotics, actively growing bacterial cells will likely develop resistance to ADEP. However, Lewis said, "cells that develop ADEP resistance become rather wimpy." That is, other traditional drugs such as rifampicin or linezolid work well against ADEP-resistant cells, providing a unique cocktail that not only kills persisters but also eliminates ADEP-resistant mutant bacteria.
Dr. Richard Novick of New York University’s Langone Medical Center and a leader in the field said the research is a “brilliant outgrowth of Kim Lewis’ pioneering work on bacterial persisters and represents a highly creative initiative in this era of diminishing antibiotic utility.”

While ADEP targets MRSA, Lewis’ team believes similar compounds will be useful for treating other infections as well as any other disease model that can only be overcome by eliminating a population of rogue cells, including cancerous tumors. They are pursuing several already.

**Journal Reference:**

### Harm reduction works: extremely low HIV incidence over almost 20 years among people who inject drugs in Australia

Michael Carter
Published: 15 November 2013

Incidence of new HIV infections among people who inject drugs in Australia is extremely low, results of a retrospective study published in the online edition of AIDS show. Investigators examined incidence among people who inject drugs who had repeat HIV tests between 1995 and 2012. The annual incidence rate remained low throughout the study period at just 0.11 per 100 person-years. The investigators attribute this “remarkable” prevention success to the early introduction of free and legal syringe and needle exchange programmes in Australia.

Globally, an estimated 3 million HIV infections involve people who inject drugs. Research involving 84 countries, conducted in 2007, showed that HIV prevalence among people who inject drugs ranged from 0.01% to 72%.

An understanding of incidence rates among people who inject drugs is important so that prevention campaigns can be planned and evaluated. Research conducted in Australia in the early 1990s found a very low HIV incidence among people who inject drugs (0.17-0.21 per 100 person-years). Investigators examined data obtained between 1995 and 2012 to see if this low incidence rate had been sustained.

The study population involved people who inject drugs who participated in the Australian Needle Syringe Program Survey. To be included in the investigators’ analysis, individuals were required to be HIV negative at baseline and to have had two or more HIV tests at least one year apart during the study period.

A total of 34,000 records were available for analysis. Approximately a quarter (26%; 8873 records) were from the 3528 individuals with repeat test results. After excluding 38 participants who were HIV positive at baseline, the study cohort comprised 3490 individuals who contributed 8763 records.

The median interval between repeat HIV tests was two years. A total of 17 repeat-testers seroconverted for HIV, yielding an incidence rate of 0.11 per 100 person-years (CI: 0.07-0.17).

The majority of incident infections (n = 12; 71%) involved gay men. Incidence was low among gay men but significantly higher among this population than other risk groups (0.83 vs 0.03 per 100 person-years; p < 0.001). No other social or demographic factors were associated with seroconversion.

"Results indicate extremely low and sustained HIV incidence over almost two decades", write the authors. “Consistent with HIV transmission patterns among the broader Australian population, the majority of HIV infections occurred among PWID [people who inject drugs] who identified as MSM [men who have sex with men].”

The authors note that Australia is acknowledged internationally as a leader in harm reduction. They believe that the early introduction of needle and syringe exchanges prevented the emergence of a large-scale HIV epidemic among people who inject drugs. Other factors possibly contributing to the low incidence include migration patterns and the self-limiting nature of HIV outbreaks among people who inject drugs.

**Reference**

### Novel microbicide gel for vagina and rectum shows potential for HIV prevention

**Research to be presented at world’s largest pharmaceutical sciences meeting**

Arlington, Va. — Researchers developed a first-of-its-kind microbicide gel formulation that shows promise for safe vaginal and rectal administration to prevent the sexual transmission of human immunodeficiency virus (HIV). This research is being presented at the 2013 American Association of
Pharmaceutical Scientists (AAPS) Annual Meeting and Exposition, the world's largest pharmaceutical sciences meeting, in San Antonio, Nov. 10-14.

There are 35.3 million people living with HIV worldwide, according to the World Health Organization, and the virus is spread most often through both vaginal and anal intercourse.

Anthony Ham, Ph.D., and the microbicide research team at ImQuest BioSciences, along with colleagues from Duke University, Magee-Womens Hospital, and University of Pittsburgh, developed the DuoGel as a task of their Integrated Preclinical and Clinical Program for Topical Microbicides grant from the National Institutes of Health. The primary goal was to create a safe and effective gel for administration of antiviral products to both the vagina and rectum, whereas current gels are only recommended for vaginal application. This **DuoGel will deliver ImQuest's antiretroviral compound IQP-0528**.

Since the environments of the vagina and rectum are dissimilar and require different conditions for safe and effective drug delivery, ex vivo toxicity, permeability, and efficacy tests were performed in both ectocervical and colorectal tissues. The DuoGel containing IQP-0528 was applied to the tissues, which were then exposed to HIV-1. The DuoGel sufficiently delivered the drug in both in vitro and ex vivo vaginal and rectal environments to prevent HIV-1 infection of these tissues.

"It is recognized that both vaginal and rectal intercourse occur during the same sexual act, so a single product that is safe for both compartments makes sense in terms of convenience, which is likely to result in higher compliance."

Currently, user compliance and acceptability are being evaluated with a placebo DuoGel. The research team is preparing the current gel for animal studies and Investigational New Drug submission, and they hope to begin phase 1 of clinical trials in early 2015. The next stage in the research is to enhance the formulation by creating a multidrug **DuoGel that also contains tenofovir**, a second antiretroviral drug.

**Scientists report human dietary supplement cures lab animals infected with human intestinal parasite**

**Preliminary success using 'probiotics' against hookworms raises hope for treating afflictions that burden 1.5 billion and cause stunting, development delays in children**

WASHINGTON, D.C. (November 15, 2013) – Laboratory animals fed a modified version of a common human dietary supplement were completely cured of intestinal worms that belong to a family of parasites that currently infect 1.5 billion people, or almost one quarter of the world’s population, according to new research presented today at the annual meeting of the American Society of Tropical Medicine and Hygiene (ASTMH).

"We need to replicate the results in other animals and also in humans, but this is an important development in our effort to find a safe, affordable and effective way to confront a major global health problem," said Raffi Aroian, PhD, principal investigator of a team of scientists at the University of California, San Diego who are seeking new treatments for a variety of parasitic worms known as "soil-transmitted helminths" or STHs.

While rarely fatal, STHs and other intestinal worms are leading contributors to disease in school-age children in low-income countries and are viewed by many experts as among the most burdensome of the world’s "neglected tropical diseases" or NTDs.

The study conducted by Aroian's team focused on hookworms, common STHs that are found in soil that has been contaminated with human feces. Hookworms can linger in the intestines for years, where they feed on blood and tissue, robbing their hosts of iron and protein and interfering with absorption of critical nutrients. They frequently cause stunting and cognitive delays in infected children. They also can have long-term effects on educational achievement and productivity.

Currently, the only drugs available to treat hookworms in humans were originally developed to combat parasites that infect farm animals. Aroian said they are only partially effective against the range of intestinal parasites that infect humans. There is also evidence of reinfections occurring rapidly after treatment and low levels of efficacy in some places.

At the ASTMH meeting, Aroian's colleague Yan Hu, PhD reported findings from a study in which hamsters were deliberately infected with hookworms. The hamsters were later divided into two groups. One group received a common strain of the bacteria *Bacillus subtilis*, which is often marketed as a "probiotic"—a dietary supplement consumed as a pill or added to food that is intended to promote digestive health. It also is the key ingredient in a popular Japanese fermented soybean dish called Natto.
The other group received the same probiotic, except the researchers modified it to express a protein derived from a closely related bacterium, Bacillus thuringiensis or Bt, which is known to be safe in humans but potentially lethal to intestinal worms.

"Five days after we administered the bacteria, we examined the animals' intestines," Hu said. "We found no worms in the animals that received the modified probiotic, while those that did not receive the modified probiotic remained infected."

Hu said the next step will be to conduct tests in different types of animals and against different types of STHs. If the probiotic continues to perform well against multiple intestinal parasites and is shown to be safe, then researchers would consider testing in humans, she said.

The research is supported by grants from the Bill & Melinda Gates Foundation and the National Institutes of Health.

"While the research has yet to move beyond tests in animals, the human health burden is so immense and the solutions so few that it's gratifying to see progress being made toward finding new treatments for intestinal worm diseases," said ASTMH President David H. Walker, MD. "It shows that new investments in neglected tropical diseases are inspiring creative solutions for the more than a billion people in need."

Aroian said the overall goal of the work is to produce a treatment for intestinal worms that is safe, effective and affordable in the world's poorest countries, where hookworms and other STHs do the most damage. "This probiotic is a food-grade bacterial product that can be easily produced in large quantities in a simple fermenter, and it can be manufactured in a form that has a long shelf-life," he said. "It could be well-suited to providing the cheap, mass treatment we need to substantially reduce the burden of this disease."

Aroian said Bt is attractive because it is a well-understood, natural substance for controlling plant pests that is believed to be safe for animals and humans. It is frequently sprayed on organic crops and is mainly lethal to insects in their larval stage. Bt also is a bacteria used in genetically engineered corn and soybean to endow the crops with resistance to plant pests.

Aroian said that while the modified probiotic under development in his lab should be safe to consume, if it proves to be an effective intervention for intestinal worms it would be marketed as a treatment, not as a dietary supplement.

**AbbVie reports 96% cure rate for interferon-free hepatitis C regimen in phase 3 study**

Keith Alcorn

Published: 18 November 2013

An interferon-free combination of three drugs plus ribavirin achieved a sustained virologic response (SVR12) in 96% of previously untreated patients with genotype 1 hepatitis C infection, AbbVie reported in a press release on Monday 18 November.

The results are the first in a sequence of six announcements of the results of phase III studies due to take place over the next few months, prior to presentation of the full results at scientific meetings in 2014.

AbbVie plans to file for registration of the combination in the United States and European Union in the second quarter of 2014, and hopes to have the first interferon-free combination for the treatment of genotype 1 hepatitis C infection available for prescription by early 2015.

The regimen consists of a fixed dose combination tablet containing the NS5A inhibitor ABT-267 and ABT-450 boosted by ritonavir, dosed once daily, and a second tablet, the non-nucleoside polymerase inhibitor ABT-333, dosed twice daily. The combination is being tested with and without ribavirin to determine whether it is possible to cure hepatitis C infection without the need for ribavirin, which can cause anaemia.

The SAPPHIRE 1 study released this week represents an easier-to-treat population of patients without cirrhosis. The study recruited 631 patients with genotype 1a or 1b hepatitis C and randomised them to receive either ABT-450 boosted by ritonavir, ABT-333 and ABT-267, or a placebo, for 12 weeks. Patients in the placebo group received the active treatment after 12 weeks.

The headline results showed that 96% of the study population achieved SVR12, and only 1.7% of participants experienced virologic relapse after completing treatment. There was little difference in virologic response by sub-genotype: 95% of genotype 1a and 98% of genotype 1b patients achieved SVR12. Treatment discontinuations due to adverse events were rare (0.6%) and the main side-effects reported by patients were nausea, headache and fatigue.
Further phase III studies will report on the efficacy of the combination in treatment-experienced patients, and also determine whether there are differences between genotypes 1a and 1b and between treatment-naïve and treatment-experienced patients in their need for ribavirin. The Turquoise study is comparing 12 and 24-week ribavirin-containing regimens in genotype 1 patients, including those with cirrhosis.

AbbVie is also conducting phase II studies of a once-daily two-drug combination, of ABT-450/r and ABT-267, and reported the first results from these studies at the Liver Meeting earlier this month.

African Commission condemns coerced sterilisation of HIV+ women

By Richard Lee | November 06th, 2013

In a landmark pronouncement for women’s rights, the African Commission on Human and Peoples’ Rights (ACHPR) has condemned the coerced sterilisation of women living with HIV as a blatant violation of their fundamental rights, which are guaranteed under the African Charter on Human and Peoples’ Rights.

Following reports of coerced and forced sterilisation of women living with HIV in numerous African countries in recent years, including South Africa, Kenya, Namibia, Swaziland, Lesotho, Tanzania, Zimbabwe and Zambia, the ACHPR’s resolution denouncing the practice has been warmly welcomed by activists and civil society groups across the continent.

“The Commission’s resolution sends a very clear message to African governments that they must take urgent measures to end the coerced sterilisation of women living with HIV in their countries,” said Nyasha Chingore, a lawyer from the Southern Africa Litigation Centre (SALC), which has long campaigned for an end to the practice. “It is time for the authorities to promote the sexual and reproductive rights of women rather than to sit idly by as they are violated.”

The Commission adopted the strongly worded Resolution on Involuntary Sterilisation and Protection of Human Rights in Access to HIV Services on November 4th during its 54th ordinary session in Banjul.

The resolution condemned all forms of stigma and discrimination in terms of access to, and provision of, health services in the context of HIV. It also made it clear that all forms of involuntary sterilisation violated women’s rights to equality and non-discrimination, dignity, liberty and security of the person, and freedom from torture, cruel, inhuman and degrading treatment, as well as the right to the highest attainable physical and mental health as enshrined in regional and international human rights instruments.

“This resolution by one of Africa’s premier human rights bodies is a major step forward, which will give women living with HIV some confidence that their rights are also protected under the African human rights system,” said Gladys Kiio, Programme Manager of the African Gender and Media Initiative Trust (GEM), which has documented the stories of women subjected to coerced sterilisation in Kenya. “It will also help to focus attention on a shameful – but still widespread – practice that most people in Africa know nothing about.”

Currently, women in Kenya and Namibia, who were subjected to coerced sterilisation, are challenging the practice in court arguing that it violated their fundamental rights. The Commission’s resolution can only strengthen their legal attempts to bring an end to the involuntary sterilisation of women living with HIV and other marginalised women, including women with disabilities.

The Lancet/Cell Conference Asks: What Will it Take to Achieve an AIDS-free World?

Published on Friday, 15 November 2013 00:00

Written by Matt Sharp

"What will it take to achieve and AIDS-free world?” was the question on everyone’s mind at a small meeting of the same name held November 3-5 at the St. Francis Hotel in San Francisco. A Cell press release stated that the goal of the meeting was to "bridge the gap between researchers and clinicians in a joint effort to identify what needs to be done before an AIDS-free world can go from dream to reality."

The translational medicine conference, organized and sponsored by The Lancet and Cell medical journals, brought together over 200 leading HIV research and clinical practice experts from 6 continents.

Richard Horton, editor-in-chief of The Lancet, opened the meeting with a compelling question about the possibility of a cure, asking whether it would cause complacency. He pointed out that the best we can ultimately hope for is "low endemic levels of HIV." He added that besides the research questions, there are enormous political, economic, and structural forces that will press the questions about maintaining
funding and neglecting key populations and geographies. In the end, these issues became the underlying theme of the meeting, where there was otherwise little new data.

"It's much too soon for a victory lap," claimed Anthony Fauci, director of the National Institute of Allergy and Infectious Diseases, in his opening review of scientific advances, but he spoke of a shift towards translation and implementation of recent findings.

In terms of prevention, Fauci said that we have not yet reached the goal of a 33% reduction in HIV infections and have much work to do, especially in the areas of expansion of testing and male circumcision. He spoke of the fact that HIV vaccine research has seen years of disappointment, but this past October there were many papers published on new vaccine discoveries.

With regard to the 35.3 million people currently living with HIV, Fauci discussed optimization of treatment and the need for more potent, less toxic, and longer acting drugs for antiretroviral therapy (ART). As far as a cure for HIV, he stated that researchers must extrapolate from what has been learned with the various cure cases thus far, but concurred that the most practical approach would be to move forward with efforts to achieve "sustained remission" after early treatment, as seen in the VISCONTI Cohort and the Mississippi baby cure case. Finally, Fauci reiterated that a cure would need to be safe, administered with no tertiary care needed, and most definitely scalable.

The meeting's first session consisted mostly of basic science review talks on targeting HIV reservoirs and other cure-related research. Robert Siliciano of Johns Hopkins spoke about the recent research published in CELL that found that the size of the latent reservoir is 60 times larger than previously thought.

Romas Gelezuninas, lead researcher at Gilead Sciences working on eradication therapies, described new work on a specific neutralizing antibody known as PGT121; with the Gates Foundation he will be moving forward in studying this approach in monkeys. Gilead has other work ongoing on several approaches for reservoir reduction such as TRL-7 agonists and the use of combination therapies, which almost everyone regards as the way forward for any cure strategy.

Warner Greene from the Gladstone Institutes in San Francisco spoke about his studies that for the first time mechanistically link 2 HIV pathogenic signatures commonly mentioned in eradication work, inflammation and CD4 T-cell depletion. According to Greene, such discoveries could lead to "an affordable bridge therapy for 16 million people who currently need but do not have access to ART, a potential solution for those on ART who are developing aging-related diseases a decade or more before the non-infected population, and a potential clearing of the latent reservoir that could contribute to a cure for HIV/AIDS."

HIV vaccines were a recurrent thread throughout the meeting, with a session dedicated to "Vaccines and Harnessing the Immune Response." Despite already having moved the goalpost for a successful vaccine forward 10 years, it will likely be moved yet another 10 years into the future. While there have been many setbacks and disappointments, a lot has also been learned. The central discussion concerned recent advances in broadly neutralizing antibody research by Gary Nabel, Dennis Burton, and Michel Nussenzweig. The trajectory seems to be looking at the kinds of antibodies people make in response to long term HIV infection. David Baltimore from the California Institute of Technology stated in his talk, "We need a vaccine, but we still don't have an open road in front of us."

Any discussion of ending AIDS must address prevention, and indeed the conference included a session on "Antiretrovirals for Primary and Secondary HIV Prevention." An examination of ending AIDS through global access to antiretrovirals was the focus of a talk by Wafaa El-Sadr from Columbia University. She spoke of the need for a roadmap for ART scale-up in diverse settings, emphasizing that "it is critical to achieving optimal outcomes for both people living with HIV and for prevention."

The "AIDS-free generation" sound-bite has been perceived as insulting by many people currently living with HIV. The organizers adeptly addressed the issue with a session on co-morbidities and aging, opened by long-time AIDS researcher Jay Levy from the University of California at San Francisco (UCSF), dedicated to the issues of people who have been living and surviving with HIV/AIDS for decades.

"By definition AIDS is a syndrome of complications associated with advanced immunodeficiency," stated Steve Deeks, also from UCSF. "If we can get everyone on [antiretroviral] therapy, then AIDS will essentially be gone." He stressed that understanding the links between HIV, ART, and aging will be critical to healthy and thriving older age in people with HIV. Community members allowed entry to the conference as press stressed the necessity of research into mental health issues, especially for long-term survivors.

In a dinner keynote address, the "Berlin Patient," Timothy Brown, spoke to conference delegates about his experiences as the first known person to be cured of HIV. Paula Cannon, a stem cell researcher
from the University of Southern California, said of his talk, "I have heard Tim speak several times now, but the simple power of his message remains just as strong as ever. We are lucky to have such a sincere, humble, and generous individual as an ambassador for this effort."

The meeting ended on a somewhat positive note with a panel discussion by several local and global leaders in the quest for an end to AIDS. Nobel Laureate Francoise Barre-Sinoussi spoke about the ongoing work of the International AIDS Society and the growing interest in finding a cure. "I'm here as a witness of 30 years of science and translational research," she exclaimed. "We need to continue the effort of working together if we want to make progress toward a cure."

David Evans, a community advisory board member for the Delaney AIDS Research Enterprise and Director of Research Advocacy at Project Inform, said of the conference, "A case was well made for the potential of cure research and what might be able to be achieved with HIV treatment as prevention. What was lacking, however, was both the science and advocacy on the role that people living with HIV can play as equal partners in ending the epidemic."

To that end, an interesting discussion will continue on the complexities and challenges the global community will need to address before an end to AIDS is ever achieved.

Source

Gemma Ahaibwe | November 15, 2013 4:06pm

**Halting and Reversing the Spread of HIV/AIDS in Uganda: President Museveni Publically Tests for HIV**

On November 8, Ugandan President Yoweri Museveni and his wife publically tested themselves for HIV/AIDS to encourage all Ugandans to take steps to curb the spread of disease, which currently affects 7.3 percent of the population. Although Uganda has been successful in increasing HIV/AIDS education and also reducing the HIV/AIDS prevalence rate from an average of 18 percent during the early 1990s to approximately 6.4 percent by 2004/05, (UNAIDS 2011 figures), challenges remain. In fact, the HIV/AIDS prevalence rate stagnated at an average of 6.0 percent in 2004, but this trend reversed and the rate rose to close to 7.3 percent in 2011. With less than two and a half years left until the 2015 Millennium Development Goals (MDG) deadline, most countries—Uganda included—are still striving to meet this and other MDG goals and targets.

Approximately 130,000 new HIV infections are registered in Uganda annually according to estimations from the Ugandan Ministry of Health. Worse still, as noted above, the infection rate has been on the rise. This trend poses a challenge for Uganda in achieving MDG 6A on halting and reversing the spread of HIV/AIDS by 2015.

The increase in the HIV/AIDS prevalence has been blamed on the complacency of Ugandans—especially regarding sexual behavior—with the availability of anti-retroviral therapies (ARTs). Uganda has also experienced a slow uptake of proven prevention strategies like safe male circumcision (SMC). Despite the compelling evidence that SMC reduces the risk of heterosexually acquired HIV infection in men by approximately 60 percent, its uptake in Uganda has been slow, with only 26 percent of males aged 15-49 circumcised. Circumcision has largely been practiced on religious and cultural grounds and is not typically used as an HIV prevention strategy.

In addition, Uganda has not yet achieved its universal coverage of HIV/AIDS testing target by 2015. Knowledge of one’s status is an effective strategy for behavioral change and a critical link for obtaining care, treatment and other HIV/AIDS support services. Indeed, the recent move by Uganda’s president to publicly test for HIV underscores the importance of testing as a first step towards curbing the spread of the HIV virus.

Expanding coverage and uptake of HIV/AIDS testing and SMC could boost Uganda’s fight against HIV/AIDS according to recent research undertaken by the Economic and Policy Research Centre under the Global Development Network—Strengthening Institutions to Improve Public Expenditure Accountability Project.

In that report, my colleague, Ibrahim Kasirye and I find that scaling up safe male circumcision to 66 percent of uncircumcised adult males by 2020 would avert 121,278 adult HIV infections and would lead to total net cost savings of almost $790 million. Additionally, scaling up voluntary counseling and testing to reach 100 percent of “never tested” adults in Uganda by 2020 would avert 113,813 adult HIV infections and yield a total net cost savings of almost $734 million. The net cost savings are defined as the lifetime antiretroviral therapy costs multiplied by the annual number of infections averted, less the cumulative net...
costs of implementing the scaled-up programs. In less technical terms, cost savings are savings due to changes in policy strategy that reduce associated policy expenses, in this case provision of antiretroviral therapy and other therapies after infection.

For both policies, the cost savings are greater per infection averted than the values of the cost per HIV infection. This means that both policies can be feasibly scaled up. Thus to reverse the increasing prevalence and new infection trends, Uganda should expand coverage and uptake of a mix of different HIV/AIDS prevention methods and interventions as we move towards achieving the HIV/AIDS related MDG.

**Two Steps Obama Can Take to Defeat AIDS**

By Desmond Tutu  
November 18, 2013

We are making historic progress against HIV/AIDS: The global rate of new HIV infections has leveled, and the number of annual AIDS deaths has decreased by nearly a third since 2005. Antiretroviral drugs are driving these gains by stopping progression of the disease and, we now know, preventing the spread of HIV infections.

Yet AIDS remains the leading cause of death in sub-Saharan Africa, where poverty limits access to lifesaving treatments and 25 million people are living with HIV—representing 70 percent of cases worldwide. President Barack Obama should be commended for uniting the world behind the goal of creating an AIDS-free generation. I share his passion and believe we can achieve this in the next decade—but only if we accelerate the provision of antiretrovirals to the poorest and most vulnerable people.

The opportunity has never been clearer. New data published in the New England Journal of Medicine project that early treatment with antiretrovirals in South Africa, my home country, would prove very cost-effective over a lifetime (costing $590 per life-year saved) and generate both public health and economic benefits. The World Health Organization now recommends early and preventive treatment with antiretrovirals, including administration to children and uninfected partners of people living with the disease. The WHO estimates that this could save an additional 3 million lives and prevent at least as many new HIV infections through 2025.

When Obama and I met in South Africa in June, I reminded him that, given his deep familial roots in the continent, his success is our success—his failure, our failure. With that in mind, there are two decisions Obama can make before the end of this year to fulfill the promise of an AIDS-free generation.

The first is to commit to doubling the number of people receiving antiretroviral treatment through the President’s Emergency Plan for AIDS Relief. Through PEPFAR, the U.S. government has already made treatment available to 6 million people living with HIV—an increase from only 50,000 when Congress authorized the program in 2003. Millions of lives have been saved, and at least 1 million babies have been born HIV-free thanks to the program.

Analysis in PEPFAR’s own blueprint for creating an AIDS-free generation has demonstrated that rapid scale-up of antiretrovirals is among the most effective ways to interrupt HIV transmission at the community level in hard-hit countries like Kenya and Uganda. Because PEPFAR has helped build the capacity in many African countries to massively scale up access to antiretrovirals, it should be possible—with proper funding and sustained political commitment—to treat 12 million people by the time Obama leaves office in 2017. This swift attack would deal a devastating blow to the virus where it is spreading most aggressively.

The second decision Obama could make concerns America’s role in expanding access to antiretrovirals through the Global Fund to Fight AIDS, Tuberculosis and Malaria. In just over a decade, the Global Fund has provided antiretrovirals to 5.3 million people, including 2.1 million pregnant women living with HIV. It is aiming to raise $15 billion through its third “replenishment” cycle—the centerpiece of which is a pledging conference next month in Washington, D.C.—to support its work over the next three years.

As the conference host, the United States is leading a diplomatic effort to urge other donors to increase their contributions to the Global Fund. The implicit promise from the United States, based on previous contributions, to give $1 for every $2 contributed by other donors in the lead-up to the December conference has already produced encouraging results. Among other recent pledges, the United Kingdom has announced it will double its already sizable commitment, and several Scandinavian countries have agreed to increase their contributions by 25 percent.
The donor meeting next month is not the end of this important fundraising drive, so I hope the president will agree to provide up to one-third of the total Global Fund’s donations—up to $5 billion—over the next three years. A matching grant from the United States would encourage those countries that have not yet pledged, or that make unambitious commitments, to do more—or risk leaving U.S. dollars on the table. Bold commitments from Australia, Canada, Germany, Japan and the European Commission, backed by a one-third U.S. match, could quickly turn the tide against AIDS. We can defeat this disease and create an AIDS-free generation over the next decade if we remain focused and driven.

Desmond Tutu is archbishop emeritus of Cape Town and honorary chairman of endgame, a global campaign to defeat AIDS, tuberculosis and malaria.

Canada: Nova Scotia court acquits young man with undetectable viral load of aggravated sexual assault for HIV non-disclosure despite no condom use

November 19, 2013

IN THE PROVINCIAL COURT OF NOVA SCOTIA
Citation: [R. v. J.T.C.], 2013 NSPC 105

Date: November 8, 2013
Docket: 2608453, 2608454
Registry: Halifax

Her Majesty the Queen
v.
J.T.C., a young person

DECISION

by Cecile Kazatchkine, Senior Policy Analyst, Canadian HIV/AIDS Legal Network

On November 8 2013, the Provincial Court of Nova Scotia in Canada released a very encouraging decision in a case of HIV non-disclosure. A young man with an undetectable viral load who had not disclosed his HIV positive status to his sexual partner before engaging in unprotected sex was acquitted of aggravated sexual assault.

The couple had engaged in vaginal sex on three occasions. Twice, they used a condom. On the third occasion, however, it was found that they had unprotected vaginal sex without ejaculation. At no time, did the young man disclose his HIV status. In fact, the judge found that he had actively concealed that he was HIV positive to his sexual partner who had inquired about rumours that he had AIDS.

In 2012, the Supreme Court of Canada ruled in R. v. Mabior and R. v. D.C., that a person living with HIV has a legal duty to disclose his or her HIV positive status to a sexual partner where there is a “realistic possibility of HIV transmission.” The Supreme Court was clear that where a condom is used and the HIV positive partner has a low viral load, there is no “realistic possibility of HIV transmission” and thus, no duty to disclose under the criminal law. These decisions were understood to mean that a person living with HIV must disclose his or her HIV positive status before having vaginal sex unless he or she uses a condom and has a low viral load.

None the less, the Provincial Court of Nova Scotia acquitted the young man, despite the factual finding that he had engaged in unprotected sex. The Court described two different routes to its conclusion.

The first route relates to the analysis of the consent given by the complainant. In Canada, one element that the prosecution must prove in a non-disclosure prosecution is that the complainant would not have consented to sex if he or she had known about his or her partner HIV positive status. At trial, the complainant testified that had she known that the accused was HIV positive she would not have had...
unprotected sex with him. But she also said that had she known that his risk of transmitting HIV was virtually non-existent, she would have consented.

As described by Justice Campbell, that the risk of transmission was infinitesimally small was the “true state of affairs” based on the evidence before the Court. Indeed, the unchallenged medical expert called by the defence testified that he did not believe that there was any risk of transmission in this case. He further concluded that “in an act of sexual intercourse someone with an undetectable viral load such as [the accused] had a one in one million chance of transmitting the virus. That might be as high as one in 500 000 (…)” and described the risk as “very close to zero.”

According to the Court, the complainant’s statement that had she known the extremely low degree of risk she would have consented to unprotected sex with the accused is part of the context that needs to be taken into account when determining whether the consent was vitiated or not. As summarised by Justice Campbell:

[...] to ignore [the complainant]’s acknowledgement that with full knowledge of the facts she would have had unprotected sex with [the accused] would amount to a strange privileging of half-truth, deception and misconception over truth. The truth is that she would have had unprotected sex with him had she known the facts. My conclusion is that her consent was not vitiated by the deception.

The second route relates to the realistic possibility of transmission. The Court found that that element had not been met either. This conclusion is at odds with the predominant interpretation of Mabior and D.C. — that unprotected sex, even with an undetectable viral load, would necessarily be considered as representing a “realistic possibility of transmission.”

In a recent decision, the Ontario Court of Appeal had ruled that there was no need for the Crown to bring medical evidence of “a realistic possibility of transmission” in each case. The Court of Appeal ruled that proving unprotected sex would be sufficient to establish “a realistic possibility of transmission” and that evidence of the accused’s exact viral load at the time and the associated degree of risk of HIV transmission would be irrelevant in such circumstances. (There was no medical evidence on the risks of transmission before the Ontario Court of Appeal or evidence of the accused’s viral load.)

The Provincial Court of Nova Scotia, however, did not accept that the Supreme Court of Canada or the Ontario Court of Appeal decisions had definitely closed the doors to different findings with respect to whether “a realistic possibility of HIV transmission” existed based on the medical evidence before the judge in a particular case. Concerned about the potential for discrimination against people living with HIV in the absence of any risk, the Provincial Court of Nova Scotia stated that the Supreme Court decisions “can and should be interpreted in a way that in not incompatible with an approach that respects both the scientific evidence in each case and the fact finding role of trial courts.” According to the Court, “[t]he Supreme Court did not intend (…) to impose evidentiary findings on trial courts that are incompatible with the evidence actually before those courts.”

In the case at bar, the medical evidence called by the defence was clear: the risk of transmission was approaching zero. The Court was careful to specify the risk determination was a finding of fact (versus a finding of law), specific to the case, and ruled that the legal conclusion arising from that fact was that, even in the absence of a condom, the legal test of a “realistic possibility of transmission” was not met.

This decision is an encouraging development in the law on HIV non-disclosure in Canada. While trial court decisions have limited precedential authority in the Canadian legal system, this decision remains important as it demonstrates that Mabior and D.C — which have been strongly criticised for being at odds with the science and previous case law — need not prevent science from prevailing over prejudice. Medical evidence can and should play a critical role in cases of HIV non-disclosure, exposure and transmission, something both defence lawyers and medical experts in HIV will need to be very mindful of.

**New findings could help target the bacteria that cause Lyme disease and syphilis**

The bacterial pathogens that cause Lyme disease and syphilis are highly invasive. These pathogens, or spirochetes, can invade the central nervous system and, in the case of syphilis, enter the placenta, causing disease in the unborn child. In the November 19 issue of the Biophysical Journal, a Cell Press publication, researchers provide new insights into how these spirochetes penetrate tissue barriers. The findings might be used to develop new treatment strategies to help affected patients or even prevent infections.

"We are one of the few groups that are trying to understand the physical interactions with the environment that make spirochetes such successful pathogens," says senior author Dr. Charles Wolgemuth of the University of Arizona in Tucson. "We've previously understood very little about how
these bacteria move through and into our organs, tissues, and central nervous system, but our work sheds light on these processes and could form the basis for novel therapeutics that target the bacterium’s ability to invade.

Dr. Wolgemuth and his team, in collaboration with Dr. Justin Radolf at the University of Connecticut Health Center, found that the swimming speeds of the bacteria decrease with increases in the viscosity of their external environment, even though their motors—called flagella—are entirely intracellular. The team then used mathematical modeling to determine how these flagellar motors propel the undulating bacteria forward through viscous fluids. Finally, they fit their simulated data to their experimental data to reveal how external forces affect the movement of the Lyme disease and syphilis spirochetes.

The researchers also showed that both types of spirochetes (syphilis's *Treponema pallidum* and Lyme disease's *Borrelia burgdorferi*) respond to changes in viscosity in a similar manner and can be explained by the same biophysical model. "Since the syphilis bacterium cannot be cultured in the lab, our results show that data derived from studying the Lyme disease bacterium is highly informative about the syphilis bacterium and can be used as a 'surrogate' for it," says Dr. Wolgemuth. *Biophysical Journal* Castellano et al.: “Viscous Dynamics of Lyme Disease and Syphilis Spirochetes Reveal Flagellar Torque and Drag”

**New method to diagnose sepsis is faster, cheaper**
WASHINGTON, DC – November 15, 2013 – A new method could cut hours off the time it takes to diagnose blood infections while also eliminating the need for complicated manual processing and expensive equipment, according to a report to be published in mBio®, the online open-access journal of the American Society for Microbiology, on November 19. The method combines a selective lysis step in which blood cells in the sample are destroyed, a centrifugation step to collect any bacteria or fungi in the sample, and a fluorescence step that analyzes the particular fingerprint of any pathogens present in the sample. Tests show the method correctly identifies the species of bacteria or fungi in 96.5 % of positive blood culture samples, crucial information for doctors to provide the appropriate drugs for their patients.

"The primary benefit of getting a rapid identification is making sure the patient is on the right [antibiotic] therapy and to quickly make any needed adjustments to the initial therapy," says co-author John Walsh of bioMérieux, Inc. in Durham, North Carolina. Patients with bloodstream infections are usually in very serious condition, says Walsh, and faster identification of the organism causing the infection can help get patients on the most effective antibiotics faster and save lives. Proper diagnosis is also important from the perspective of antibiotic stewardship: using more appropriate, targeted antibiotics reduces the risk of contributing to the spread of resistance to broad-spectrum antibiotics.

Walsh says the current standard approach to diagnosing bloodstream infections, Gram staining and overnight sub-culture followed by phenotypic ID tests, have limitations that can prevent timely treatment. Gram staining provides early, low level information about the type of microorganism present, but it sometimes takes hours to deliver a result, and technicians can make mistakes in the process that provide misleading results. Other more specific identification methods are also available for diagnosis, but they can take at least a day or two to produce results and many require expensive equipment.

In the technique developed by Walsh et al., a sample of positive blood culture is treated with lysis buffer to "pop" the blood cells, then transferred to a specialized optical tube. The tube is centrifuged, which drives bacteria or fungi, which are denser than the solution, down through a liquid density cushion to form a pellet at the bottom of the tube.

Then comes the intrinsic fluorescence spectroscopy (IFS): the microbial pellet is irradiated with light ranging from the deep ultraviolet to infrared, which excites certain organic molecules in the microorganisms, including tryptophan, NADH, FAD, porphyrins, and others, and causes them to fluoresce in a characteristic way depending on the identity of the microbe. The exact pattern of fluorescence is compared with a database of 37 of the most common known pathogens to identify the organism present.

"We're using intrinsic fluorescence to identify the microorganisms. It's an innate property of most living organisms. Because it's intrinsic, no reagents are needed for the identification step," which removes many of the opportunities for mistakes and lowers test costs, says Walsh.

Testing in a controlled laboratory study shows the method can correctly identify the species in 96.5% of all test samples, and in the 2.7% of samples for which no species identity was provided, the system was able to correctly identify 67% to the family level, which is often enough information to select an effective therapy. Among over a thousand samples tested, the method never gave an incorrect result as to the family level or the Gram type.

Walsh says the research and development team in Durham is actively working on automating the system with robotics to make it a fully hands-off process. Blood cultures grow in their own time, often producing a positive result at an inconvenient time of the day for clinical labs, he points out, so automation could speed up diagnosis significantly.

"Our vision is to have a system that will automatically identify the blood culture isolate within 15 minutes of the culture being called positive," says Walsh. If a culture is positive at 2 AM, he says, automating this method could make it possible to identify the organism by 2:15 AM and send an electronic report to a patient's physician. They hope to begin testing and evaluating the feasibility of an automated form of the system in a clinical environment within months.

Rapid ID Procedure: An overall schematic of the simple three-step process (lyse-spin-read) is given in Figures 1a-c. Briefly, a 2.0 mL sample of warm (35-37°C) positive broth is removed from the test blood culture bottle and added to 1.0 mL of warm (35-37°C) selective lysis buffer (0.45% w/v Brij-O10 in 0.3M CAPS, pH 11.7), contained in a 15 mL screw capped polypropylene tube. The mixture was vortexed for 5 seconds at maximum speed and then placed in a 35-37°C waterbath for 60 seconds. After an additional 1-2 second vortex, 1.0 mL of the lysate was removed and layered onto a single 5/16 inch diameter polypropylene ball (CIC Ball Co.) floating on the surface of 0.5 mL of a solution of 24% w/v cesium chloride + 0.005% w/v Pluronic F-108 + 10 mM HEPES, pH 7.4 contained within an optical micro-
centrifuge tube. The polypropylene ball was used to control the layering process and create an undisturbed interface. The tube was sealed with a screw-cap and centrifuged for 2 minutes at 10,000 rpm at 20-25°C in a microcentrifuge with A-8-11 swing-out rotor.

3-D Imaging Technique Utilizes Famous Mathematician’s Theory
Nov. 19, 2013
UT Dallas computer scientists have developed a technique to make 3-D images that finds practical applications of a theory created by a famous mathematician.

This technique uses anisotropic triangles – triangles with sides that vary in length depending on their direction – to create 3-D “mesh” computer graphics that more accurately approximate the shapes of the original objects, and in a shorter amount of time than current techniques. These types of images are used in movies, video games and computer modeling of various phenomena, such as the flow of water or air across the Earth, the deformation and wrinkles of clothes on the human body, or in mechanical and other types of engineering designs. Researchers hope this technique will also lead to greater accuracy in models of human organs to more effectively treat human diseases, such as cancer.

“Anisotropic mesh can provide better simulation results for certain types of problems, for example, in fluid dynamics,” said Dr. Xiaohu Guo, associate professor of computer science in the Erik Jonsson School of Engineering and Computer Science whose team created the technique.

The technique finds a practical application of the Nash embedding theorem, which was named after mathematician John Forbes Nash Jr., subject of the film A Beautiful Mind.

“The underlying mathematics we used to solve this problem are rigorous and beautiful,” Guo said. “Finding a way to use the theory in a practical application will have a great impact in the field.”
Guo and his team found that replacing isotropic triangles (part 1) with anisotropic triangles (part 2) in the particle-based method of creating images resulted in smoother representations of objects.

The computer graphics field represents shapes in the virtual world through triangle mesh. Traditionally, it is believed that isotropic triangles – where each side of the triangle has the same length regardless of direction – are the best representation of shapes. However, the aggregate of these uniform triangles can create edges or bumps that are not on the original objects. Because triangle sides can differ in anisotropic images, creating images with this technique would allow the user flexibility to more accurately represent object edges or folds.

Guo and his team found that replacing isotropic triangles with anisotropic triangles in the particle-based method of creating images resulted in smoother representations of objects. Depending on the curvature of the objects, the technique can generate the image up to 125 times faster than common approaches. For example, 155 seconds to create a circular image with Guo’s approach, versus more than 19,500 seconds for a common approach to generate an image of similar quality.

Objects using anisotropic triangles are of a more accurate quality, and most noticeable to the human eye when it comes to wrinkles and movement of clothes on human representatives.

The next step of this research is moving from representing the surface of 3-D objects to representing 3-D volume.

"If we are going to create accurate representations of human organs, we need to account for the movement of cells below the organ’s surface," Guo said.

The research was presented at the Association for Computing Machinery SIGGRAPH conference earlier this year.

Study finds youth prefer and benefit more from rapid point-of-care HIV testing

TORONTO, Nov. 19, 2013 – Youth prefer, accept and receive HIV results more often when offered rapid finger prick or saliva swab tests rather than traditional blood tests according to a study by researchers at St. Michael's Hospital.

More than 50 per cent of youths who took part in 14 North American studies preferred the rapid point-of-care tests because they are less invasive and provide faster results, said family physician Dr. Suzanne Turner.

Her literature review of studies published between 1990 and 2013 on HIV, POC testing and youth, appear in the Journal of Adolescent Health today.

Rapid point-of-care HIV tests are done by finger prick or saliva swab, with results available in minutes. Traditional HIV testing requires a blood test that is sent to a lab for analysis.

"Traditional testing provides results within one to two weeks," said Dr. Turner. "But many high-risk youth are transient or homeless and won’t come back for a follow-up, so having the rapid POC testing available means these individuals can know their status within the same appointment."

Multiple studies within the review also found that participants who chose this option were more likely to receive their results within the follow-up period, at 91.3 per cent, compared with those who chose traditional testing, with 46.7 per cent.

In Canada, youth between the ages of 15 and 29 account for 26.5 per cent of all positive HIV test reports. Another 83 per cent of adolescents in a Toronto study reported they've never visited a health care provider for any sexual health-related reason.

"As a physician, I have a lot of anxiety about youth who don’t know or don’t receive their HIV status," said Dr. Turner, who is also an addictions physician. "Results within the same appointment provide the opportunity to address high-risk behaviours with youth if they’re negative and identify next steps, prevention measures and treatment for those who are positive."

Fewer than 32 per cent of youth are tested for HIV and a reported 60 per cent of HIV positive youth didn’t know their status, American data shows.

Rapid POC HIV testing is only offered at about 50 locations in community clinics in Ontario – despite the positive findings and potential benefits. Expanding the test into primary and emergency departments could increase acceptance rates, with between 83 and 93 per cent of youth accepting rapid HIV POC testing in an emergency department setting, the review found.

"Not being offered HIV testing directly was also identified as a barrier with youth," said Dr. Turner. "Point-of-contact HIV testing must become routine and be offered in all patient environments, from emergency departments to community programs."
Acceptance rates for HIV testing increase when POC testing is offered directly and when offered alongside traditional testing, the study showed.

"In an era of patient-centred care, reducing wait-times for results and offering less invasive HIV testing options should be considered to improve patient acceptance and reduce failure-to-notify, especially with such high positive HIV rates in youth," said Dr. Turner.

The specific POC test preference among youth was observed across a diverse range of racial, gender, sexual orientation, cultural, geographic and socio-economic populations in a variety of health care settings.

Dr. Turner states that more research must be done on youth, HIV and POC testing to determine specific trends in particular populations and create youth-targeted testing programs.

**Staphylococcus aureus bacteria turns immune system against itself**

Around 20 percent of all humans are persistently colonized with *Staphylococcus aureus* bacteria, a leading cause of skin infections and one of the major sources of hospital-acquired infections, including the antibiotic-resistant strain MRSA.

University of Chicago scientists have recently discovered one of the keys to the immense success of *S. aureus*—the ability to hijack a primary human immune defense mechanism and use it to destroy white blood cells. The study was published Nov 15 in *Science*.

"These bacteria have endowed themselves with weapons to not only anticipate every immune defense, but turn these immune defenses against the host as well," said Olaf Schneewind, MD, PhD, professor and chair of the Department of Microbiology at the University of Chicago and senior author of the paper.

One of the first lines of defense in the human immune response are neutrophils, a type of white blood cell that ensnares invaders in neutrophil extracellular traps (NETs), a web-like structure of DNA and proteins. Captured bacteria are then destroyed by amoeba-like white blood cells known as macrophages. However, *S. aureus* infection sites are often marked by an absence of macrophages, indicating the bacteria somehow defend themselves against the immune system.

To reveal how these bacteria circumvent the human immune response, Schneewind and his team screened a series of *S. aureus* possessing mutations that shut down genes thought to play a role in infection. They looked to see how these mutated bacteria behaved in live tissue, and identified two strains that were unable to avoid macrophage attack. **When these mutations—to the staphylococcal nuclease (nuc) and adenosine synthase A (adsA) genes respectively—were reversed, infection sites were free of macrophages again.**

Looking for a mechanism of action, the researchers grew *S. aureus* in a laboratory dish alongside neutrophils and macrophages. The white blood cells were healthy in this environment and could clear bacteria. But the addition of a chemical to stimulate NET formation triggered macrophage death. Realizing that a toxic product was being generated by *S. aureus* in response to NETs, the team used high performance liquid chromatography and mass spectrometry techniques to isolate the molecule.

They **discovered that *S. aureus* were converting NETs into 2'-deoxyadenosine (dAdo), a molecule which is toxic to macrophages.** This effectively turned NETs into a weapon against the immune system.

"Sooner or later almost every human gets some form of *S. aureus* infection. Our work describes for the first time the mechanism that these bacteria use to exclude macrophages from infected sites," Schneewind said. "Coupled with previously known mechanisms that suppress the adaptive immune response, the success of these organisms is almost guaranteed."

*S. aureus* bacteria are found on the skin or in the respiratory tracts of colonized humans and commonly cause skin infections in the form of abscesses or boils. Normally not dangerous, severe issues arise when the bacteria enter the bloodstream, where they can cause diseases such as sepsis and meningitis. Antibiotic-resistant strains, such as methicillin-resistant *S. aureus* (MRSA), are difficult to treat and have plagued healthcare systems around the world.

Schneewind and his team hope to leverage their findings toward therapies against *S. aureus* infections. But both genes and the dAdo molecule are closely related to important human physiological mechanisms, and Schneewind believes targeting these in bacteria, without harming human function, could be difficult.

"In theory you could build inhibitors of these bacterial enzymes or remove them," Schneewind said. "But these are untested waters and the pursuit of such goal requires a lot more study."
Bacteria Recycle Broken DNA: Modern Bacteria Can Add DNA from Creatures Long-Dead to Its Own

Nov. 18, 2013 — From a bacteria’s perspective the environment is one big DNA waste yard. Researchers have now shown that bacteria can take up small as well as large pieces of old DNA from this scrapheap and include it in their own genome. This discovery may have major consequences – both in connection with resistance to antibiotics in hospitals and in our perception of the evolution of life itself.

Our surroundings contain large amounts of strongly fragmented and damaged DNA, which is being degraded. Some of it may be thousands of years old. Laboratory experiments with microbes and various kinds of DNA have shown that bacteria take up very short and damaged DNA from the environment and passively integrate it in their own genome. Furthermore this mechanism has also been shown to work with a modern bacteria’s uptake of 43,000 years old mammoth DNA. The results are published now in the scientific journal Proceedings of the National Academy of Sciences (PNAS). The discovery of this second-hand use of old or fragmented DNA may have major future consequences.

Postdoc Søren Overballe-Petersen from the Centre for GeoGenetics at the Natural History Museum of Denmark is first author on the paper and he says about the findings: "It is well-known that bacteria can take up long intact pieces of DNA but so far the assumption has been that short DNA fragments were biologically inactive. Now we have shown that this assumption was wrong. As long as you have just a tiny amount of DNA left over there is a possibility that bacteria can re-use the DNA. One consequence of this is in hospitals that have persistent problems with antibiotic resistance. In some cases they will have to start considering how to eliminate DNA remnants. So far focus has been on killing living pathogen bacteria but this is no longer enough in the cases where other bacteria afterwards can use the DNA fragments which contain the antibiotic resistance."

The research group’s results reveal that the large reservoir of fragments and damaged DNA in the surroundings preserve the potential to change the bacteria’s genomes even after thousands of years. This is the first time a process has been described which allows cells to acquire genetic sequences from a long gone past. We call this phenomenon Anachronistic Evolution – or Second-hand Evolution. Professor Eske Willerslev from the Centre for GeoGenetics at the Natural History Museum of Denmark is the leader of the project. He says: "That DNA from dead organisms drives the evolution of living cells is in contradiction with common belief of what drives the evolution of life itself."

Furthermore old DNA is not limited to only returning microbes to earlier states. Damaged DNA can also create new combinations of already functional sequences. You can compare it to a bunch of bacteria which poke around a trash pile looking for fragments they can use. Occasionally they hit some ‘second-hand gold’, which they can use right away. At other times they run the risk of cutting themselves up. It goes both ways. This discovery has a number of consequences partially because there is a potential risk for people when pathogen bacteria or multi-resistant bacteria exchange small fragments of ‘dangerous’ DNA e.g. at hospitals, in biological waste and in waste water.

In the grand perspective the bacteria’s uptake of short DNA represents a fundamental evolutionary process that only needs a growing cell consuming DNA pieces. A process that possibly is a kind of original type of gene-transfer or DNA-sharing between bacteria. The results show how genetic evolution can happen in jerks in small units. The meaning of this is great for our understanding of how microorganisms have exchanged genes through the history of life. The new results also support the theories about gene-transfer as a decisive factor in life’s early evolution.

Soren Overballe-Petersen explains: "This is one of the most exciting perspectives of our discovery. Computer simulations have shown that even early bacteria on Earth had the ability to share DNA – but it was hard to see how it could happen. Now we suggest how the first bacteria exchanged DNA. It is not even a mechanism developed to this specific purpose but rather as a common process, which is a consequence of living and dying."

Journal Reference:
More Than Skin Deep: New Layer to the Body's Fight Against Infection
Nov. 18, 2013 — The layers of skin that form the first line of defense in the body's fight against infection have revealed an unanticipated secret.

The single cell type that was thought to be behind the skin's immune defense has been found to have a doppelganger, with researchers from the Walter and Eliza Hall Institute showing the cells, despite appearing identical, are actually two different types.

Institute scientists Dr Michael Chopin, Dr Stephen Nutt and colleagues from the institute's Molecular Immunology division have been investigating Langerhans cells, the immune cells that provide the first line of defense against attacks through the skin.

Until recently, scientists believed that, because they looked identical, all Langerhans cells were also genetically identical and had the same function. However Dr Nutt said the research team, with collaborators from the National Institutes of Health, US, have shown this is not the case.

"Langerhans cells are produced and found in the skin and are quite unique among immune cells because they do not have a definite lifespan, they can last for a lifetime," Dr Nutt said. "They are only replaced when necessary, such as when the skin is damaged by a burn or a cut. When that happens, new Langerhans cells have to be produced by the bone marrow. These cells look the same, so it was always thought that they were genetically the same and their function was the same. We have shown that this isn't the case."

This surprise finding, published today in the *Journal of Experimental Medicine*, could have repercussions for developing and refining therapies for skin infections and skin cancers.

Although Langerhans cells were discovered nearly 150 years ago, Dr Chopin said there were still a lot of gaps in our knowledge about how they develop and their role in responding to foreign invaders. Dr Chopin said the research team was initially trying to understand the role of Langerhans cells. "Not everything that makes contact with the skin is harmful, so it is important the immune system doesn’t overreact," he said. "We were trying to find out whether Langerhans cells were there to activate an immune response to invaders, or to suppress the immune system to prevent it from overreacting."

"While designing the experiment, we found that the genes that define the Langerhans cells that are produced in the skin were different to those of Langerhans cells that came from bone marrow. In essence we now know that there are two different types of Langerhans cells where we thought there was one. We now need to find out if they behave differently as well."

Dr Nutt said the research could explain why some promising new drugs have not had the desired effect in the clinic. "Some clinical trials of drugs that were designed to help boost Langerhans cells in response to infections have not responded as the researchers expected," Dr Nutt said. "Our finding may help explain why these drugs didn't work outside the laboratory and our current research may provide guidance in developing therapeutics to treat skin infections or skin cancer."

*Journal Reference:*

Fruit Bat Population Covering Central Africa Carries Two Deadly Viruses
Nov. 19, 2013 — The study, conducted jointly by the University of Cambridge and the Zoological Society of London's Institute of Zoology and published today in the journal *Nature Communications*, found that the "gregarious" bats span over 4,500 km of central Africa (around the distance from California to New York). The researchers also discovered that thirty-four per cent of the bats had been infected with Lagos bat virus, a disease similar to rabies, and 42 per cent had been infected with henipaviruses.

The African straw-coloured fruit bat (Eidolon helvum), which can live in roosts of over one million and often congregates near cities, was previously known to be a reservoir for these viruses, but it was not known to what extent.

For the study, the researchers tested over 2,000 bats in 12 different countries across Africa, measuring DNA from blood and tissue samples. They discovered that the bats were largely genetically similar, meaning that they travelled and mated across the continent without any evidence of population subgroups or specific migratory patterns—the largest example of this freely mixing population structure ever found in mammals. The species’ homogeneity and extensive movement means that the two viruses can be spread easily.

Professor James Wood, the study's senior author from the University of Cambridge's Department of Veterinary Medicine, said: "We now not only know how widespread viral infections are in this bat population, but we also know much more about its population structure. This new information indicates
that the unique population of freely mixing bats across the entire continent facilitates the spread of the viruses. This has important implications for the monitoring of these viruses in order to prevent its spread to other animals, including humans."

Fruit bats are often hunted for meat, a process which can result in a spill-over of these pathogens from animals to humans. Henipaviruses can also be spread through contact with urine and faeces. While no instances of either disease have been reported in humans in Africa, the viruses have previously been detected in pigs in Ghana. Henipaviruses have caused fatal disease in humans, pigs and horses in SE Asia and Australia.

Although potential human infection raises public health concerns, the study’s lead author, Dr Alison Peel, cautions restraint. She said: "Sometimes, a knee-jerk response can be to try and remove bats from urban areas via culling or dispersal. However, there is evidence to suggest that actions such as this can stress the bats and lead to a greater risk of spill-over. The most appropriate response is ongoing studies and public awareness to avoid handling bats, and to wash the wound thoroughly if you are bitten by a bat."

Journal Reference:

Bacteria Use Lethal Cytotoxins to Evade Antibiotic Treatment

The figure shows the bacterial 70S ribosome with the cleavage point for the cytotoxin VapC20 marked with red. To the right is transfer RNA, which is cleaved by a similar mechanism in the pathogenic organism Shigella flexneri. Behind this is an RNA gel showing the actual cleavage reaction in the ribosome. (Credit: Ditlev E. Brodersen)
Nov. 18, 2013 — In spite of the fact that the first antibiotics were discovered almost a century ago, infectious diseases such as tuberculosis, encephalitis and meningitis are still serious diseases for humans in the twenty-first century. The World Health Organization (WHO) estimates that there are more than 8 million new cases of tuberculosis per year on a global scale, and that more than 300,000 of these are due to multidrug-resistant strains that are not only difficult to treat, but are also emerging rapidly in regions such as Eastern Europe.

Bacterial tolerance is not just due to resistance, but also to the formation of persistent cells that have gone into a dormant state where they are no longer sensitive to antibiotics. On the molecular level, this process is controlled by a number of advanced cytotoxins produced by the bacteria themselves in order to survive. In *Mycobacterium tuberculosis*—the organism that causes tuberculosis—there are no fewer than 88 such toxins, all of which presumably help the organism to survive.

In a new article in the journal *Nature Communications*, an international team of researchers with the participation of the Department of Molecular Biology and Genetics, Aarhus University, has revealed the mechanism behind one of these toxins—VapC20. It turns out that when the toxin is activated, it destroys the tuberculosis bacteria’s own protein ‘factory’ (the ribosome) by cleavage. The bacteria are thereby unable to produce proteins in the short term, and thus avoid the effect of antibiotics that also often attack the ribosome.

When treatment with antibiotics is completed, the pathogenic bacteria ‘wake up’ and are ready to synthesise new ribosomes. Surprisingly, it appears that the location in the ribosome that is cleaved by VapC20 is the same place that is destroyed by the strong cytotoxins α-sarcin and ricin, which are found in plants such as castor beans and are twice as venomous as cobra snake poison.

Further analysis of the cleavage point in the ribosome also shows that the mechanism is presumably general for a number of the many toxins, and the new knowledge could therefore be used in future to develop new ways of treating pathogenic bacteria by impairing their ability to use such cytotoxins.

**Journal Reference:**

### Control Malaria by Segmenting Sleeping Arrangements

Nov. 18, 2013 — Better malaria control might come from segregating household sleeping arrangements, according to a new study co-authored by a University of Guelph professor.

The researchers found malaria eradication related more to household size than to a country’s wealth or temperature. Guelph economics professor Ross McKitrick and two Finnish professors, Larry and Lena Huldén, found that when average household size drops below four persons, malaria extermination is much more likely.

Malaria is transmitted by infected mosquitoes. The research team examined data on malaria insect vectors, as well as demographic, sociological and environmental factors for 232 countries. Malaria is still prevalent in 106 countries.

"When we controlled for all the variables, the factor that had the most explanatory power on malaria control was household size," said McKitrick.

"Malaria-bearing mosquitoes mainly feed at night, and tend to return to the same location for blood meals. The more people who sleep in one area, the greater the likelihood of an infected mosquito spreading the parasite to a new, uninfected victim."

Malaria infects red blood cells and can cause anemia, nausea, fever and, in some cases, death. Each year, 225 million people are infected and 800,000 die, mostly children.

"It is a common misconception that malaria is a tropical disease, and with 90 per cent of malaria deaths taking place in Africa, it is easy to see why people believe this," said McKitrick.

"But historically, malaria has occurred in all climate zones including the Arctic, and was endemic in North America and Europe a hundred years ago. In many cases, the disease disappeared even in countries that made no efforts to fight it, while others that tried to eradicate it failed. We found declining average household size key to explaining this pattern."

The researchers looked at factors such as gross domestic product per capita, urbanization and slums, latitude, mean temperature, forest coverage, national DDT use, household size and even religion.

Countries with a significant Muslim population generally had large households but did a better job of eradicating malaria, with the researchers speculating it may be because of their segregated sleeping arrangements. Males and females generally sleep in separate areas.
As household size continues to decline, McKitrick said malaria should gradually disappear. But countries need not wait for that to happen.

In Vanuatu—with an average 5.6 people per household—providing bed nets and effective drug distribution and surveillance since 1996 has effectively wiped out malaria.

"The key factor is segmenting sleeping quarters and greater use of bed nets in those countries where malaria is still prevalent," he said.

"Individual bed nets can emulate a household with several bedrooms, making it difficult for the mosquitoes to transmit the parasite to other household members."

Journal Reference:

**Ritonavir the cause of the interaction between telaprevir and ritonavir-boosted atazanavir**

**Atazanavir dosing without ritonavir may be feasible during telaprevir treatment**

Michael Carter
Published: 25 November 2013

Ritonavir is the cause of the detrimental bi-directional interaction between telaprevir and ritonavir-boosted atazanavir, according to Spanish research published in the online edition of Clinical Infectious Diseases.

The research involved 14 people with HIV and hepatitis C virus (HCV) co-infection who were taking simultaneous HCV therapy based on telaprevir (Incivo or Incivek) and an HIV treatment combination including ritonavir (Norvir)-boosted atazanavir (Reyzataz). Plasma concentrations of telaprevir and atazanavir were monitored when patients were taking the ritonavir booster and again after the withdrawal of this drug. Marked increases in plasma levels of both telaprevir and atazanavir were observed after the cessation of ritonavir.

Telaprevir and boceprevir (Victrelis) are HCV protease inhibitors. The standard of care for people with HCV genotype-1 infection now includes treatment with one of these drugs in combination with pegylated interferon and ribavirin.

Telaprevir is metabolised through the liver using the P450/CYP3A4 pathway. HIV protease inhibitors are metabolised in a similar way, meaning there is a potential drug-drug interaction between telaprevir and this class of antiretrovirals. Indeed, significant decreases in telaprevir and ritonavir-boosted darunavir (Prezista) and fosamprenavir (Telzir) have been observed when these drugs are co-administered.

Currently the only HIV protease inhibitor recommended for co-administration with telaprevir is ritonavir-boosted atazanavir.

Atazanavir is only a weak inhibitor of CYP3A. This led investigators to hypothesise that ritonavir was the main driving force behind the interaction between telaprevir and ritonavir-boosted atazanavir. They designed an open-label study to see if this was the case.

Their study population consisted of 14 people with HIV and HCV co-infection taking simultaneous HCV and HIV therapy that included telaprevir (1125mg twice daily) and ritonavir-boosted atazanavir (100/300mg twice daily).

The investigators first monitored plasma levels of telaprevir and atazanavir when the participants in the study were taking the ritonavir booster.

The patients then discontinued ritonavir therapy, switching to unboosted atazanavir at a dose of 200mg twice daily. The pharmacokinetic monitoring was then repeated.

Withdrawal of ritonavir therapy had a marked impact on plasma concentrations of both telaprevir and atazanavir.

The telaprevir area under concentration-time curve (AUC0-12) level increased by 19%, with maximum concentrations of the drug (Cmax) increasing by 19% and minimum concentrations (Cmin) by 12%. The half-life of the drug was unchanged (12 vs 11 hours).

The atazanavir AUC0-12 increased by 39%, Cmax by 19% and Cmin by 48%. However, the half-life of atazanavir was reduced from 22.6 to 16.4 hours.

"RTV [ritonavir] is responsible for the detrimental interactions that occur between TVR [telaprevir] and ATVr [atazanavir/ritonavir] when administered together, likely by influencing either the absorption phase of first-pass metabolism of TVR", write the authors.
They believe their research is of clinical significance, noting that plasma concentrations of telaprevir are known to be associated with treatment outcomes. Similarly, higher telaprevir C_{min} concentrations have been associated with an increased risk of anaemia. "TVR would be a good candidate for therapeutic drug monitoring if its best therapeutic range were known," suggest the authors.

They conclude, “the coadministration of TVR and unboosted ATZ gives rise to increased exposure to both drugs compared with their coadministration with RTV.”

Reference

Urgent need to address "resurgent" gay global epidemic, says English public health chief
Gus Cairns
Published: 23 November 2013
There is an increasing and potentially catastrophic HIV and sexual health epidemic in gay men and men who have sex with men (MSM) in every part of the world, Professor Kevin Fenton, the National Director of Health and Wellbeing at Public Health England, told the BHIVA autumn conference last week.

Despite having an increasing number of tools to prevent HIV, Professor Fenton added, HIV prevalence in MSM is increasing almost everywhere and incidence (the proportion who acquire HIV every year) is stubbornly refusing to change.

Professor Fenton said that optimising HIV testing programmes for MSM, particularly in countries where they faced criminalisation and discrimination, was key to controlling the epidemic, but that this would be a “challenge”.

HIV in gay men and MSM globally
Gay men everywhere have higher rates of HIV than in the general population, Fenton said. It is estimated that the HIV rate in MSM is eight times that of the general population in low-income countries and 23 times the general-population rate in high-income countries. The most reliable prevalence figures suggest that, regionally, Latin America and the Caribbean have especially high rates, but in no country of the world is HIV prevalence lower in MSM than in the population as a whole. Even in South Africa, the country with more people living with HIV than any other, HIV is twice as common in MSM as it is in other people.

In the developed world, the US has, by some way, the highest rate of HIV new diagnoses in MSM, though the UK has possibly the highest current rate in Europe. However, in terms of the rate of increase of new diagnoses, it is central-European countries such as Poland, the Czech Republic and Hungary, historically with very low prevalence, that are seeing the fastest-growing European epidemics in MSM.

In the lower-income world, reliable estimates of incidence are harder to come by, but over the last few years studies have found huge increases in HIV diagnoses in MSM in countries ranging from Thailand and China through Kenya to Nicaragua and Peru. The highest-ever annual HIV incidence recorded in a study comes from a group of MSM in Kenya, where a quarter of those originally HIV negative had HIV a year later. Unfortunately, African countries – and some in other areas of the world – initially denying they had MSM at all or any HIV problem in them, are now responding to the realisation that many people with HIV are MSM by introducing repressive measures such as increased criminalisation.

HIV was especially high, Fenton said, partly for biological reasons. It is now estimated that anal sex is 18 times better at transmitting HIV than vaginal sex. The chance of HIV being caught from having receptive anal sex once with a partner with a detectable HIV viral load is about 1.4% or one in 71 encounters; but because people have sex together more than once, the per-partner likelihood of catching HIV from a sero-different partner is, in gay men, about 40%.

Another factor in gay men’s higher HIV prevalence, Fenton added, was that because gay men have more partners and higher changeover rates, their sexual networks are more closely connected: 25% of gay men diagnosed with HIV were members of a cluster that had HIV viruses that were genetically identical, suggesting rapid transmission within the network, compared with 5% of heterosexual people.

Gay men do not just have more HIV for biological reasons, however, noted Fenton. The rates of smoking (at 27 to 66%, according to area), recreational drug use, lifetime depression (about 40%) and lifetime severe anxiety (at 20%) are all roughly double in gay men what they are in the general population. Underpinning these are higher rates of traumatic experience such as child sexual abuse (CSA) and intimate partner violence (IPV).
In several studies that looked at rates of depression, drug use, CSA and IPV in gay men, and related them to HIV prevalence and high risk sex, gay men with three or four of these conditions were twice as likely to have HIV and three times as likely to have had recent high-risk sex than men with none of them.

Socioeconomic disadvantage and racism also magnified the effects of HIV in MSM: this was one of the reasons why black gay men in the US are three times more likely to have HIV than white gay men.

**Preventing HIV in gay men and MSM**

How can we start to reduce the burden of HIV in gay men? Fenton commented that survey after survey showed that MSM, especially in parts of the world where they are criminalised and/or especially stigmatised, were reluctant to ‘come out’ to healthcare providers; often justifiably so, as there are many cases of their being refused HIV care and treatment if they do.

He called for healthcare workers to have training in the diversity of MSM and their health issues; culturally competent care is a basic human right, he added, especially as healthcare workers may be able to help MSM in their coming-out process because of their unique social role.

We have more HIV prevention options than we used to, and others based on antiretroviral therapy – such as pre-exposure prophylaxis (PrEP) – were in development, he said. HIV testing was now not just a gateway to treatment if MSM tested positive but could also be a gateway to a more tailored approach, and access to, behavioural and biomedical interventions for HIV. HIV testing frequency needed to increase in gay men, and options such as testing in routine care, at home and within social networks needed to be considered.

Gay men also needed to be helped to develop communication skills and more respectful attitudes towards their own health and towards other MSM. “We need to increase healthy, responsible and respectful sexual behaviours and relationships,” Fenton said, and to look at ways to help gay men have better sex with less harm.

Health workers needed training to provide “supportive, non-judgmental care”, he added, and the “sometimes systematic” exclusion of MSM from HIV prevention, services and research had to be combated.

Policies for enhancing HIV prevention in MSM did exist, Fenton concluded. As well as addressing the social and structural epidemic drivers, they included “ensuring effective and culturally competent combination prevention and treatment approaches”.

Fenton K *The resurgent global HIV epidemic in men who have sex with men (MSM).* Plenary lecture, BHIVA Autumn Conference, 2013.

**Identifying targets of autoantibodies**

Patients with the autoimmune disease systemic lupus erythematosus (SLE) produce autoantibodies that target can cause damage to multiple organ systems. The host factors that are targeted by autoantibodies produced by SLE patients are not fully understood.

In this issue of the *Journal of Clinical Investigation*, Jordan Price and colleagues at Stanford University developed a microarray to identify cytokines, chemokines, and other circulating proteins as potential targets of the autoantibodies produced by SLE patients. The authors identified several autoantibody targets, and determined that SLE patients with high levels of autoantibodies directed against the B cell activating factor (BAFF) had more severe SLE-associated symptoms.

In an accompanying commentary, Maureen Su of the University of North Carolina and Stephanie Sarantopoulos of Duke University discuss how identification of autoantibody targets produced by patients with autoimmune disorders will be informative for diagnosis and therapeutic strategy development.

**Reference**

Fenton K *The resurgent global HIV epidemic in men who have sex with men (MSM).* Plenary lecture, BHIVA Autumn Conference, 2013.

**Identifying targets of autoantibodies**

Patients with the autoimmune disease systemic lupus erythematosus (SLE) produce autoantibodies that target can cause damage to multiple organ systems. The host factors that are targeted by autoantibodies produced by SLE patients are not fully understood.

In this issue of the *Journal of Clinical Investigation*, Jordan Price and colleagues at Stanford University developed a microarray to identify cytokines, chemokines, and other circulating proteins as potential targets of the autoantibodies produced by SLE patients. The authors identified several autoantibody targets, and determined that SLE patients with high levels of autoantibodies directed against the B cell activating factor (BAFF) had more severe SLE-associated symptoms.

In an accompanying commentary, Maureen Su of the University of North Carolina and Stephanie Sarantopoulos of Duke University discuss how identification of autoantibody targets produced by patients with autoimmune disorders will be informative for diagnosis and therapeutic strategy development.

**TITLE:** Protein microarray analysis reveals BAFF-binding autoantibodies in systemic lupus erythematosus

**View this article at:** [http://www.jci.org/articles/view/70231?key=58cb6df2455064a28a39](http://www.jci.org/articles/view/70231?key=58cb6df2455064a28a39)
Controlling our circadian rhythms

Most people have experienced the effects of circadian-rhythm disruption, after traveling across time zones or adjusting to a new schedule. To have any hope of modulating our biological "clocks," to combat jet lag or cope with alternating shifts, we need to first understand the physiology at play. A new study in The Journal of General Physiology helps explain some of the biophysical processes underlying regulation of circadian rhythms.

BK ("Big Potassium") channels, which are activated during nerve impulses and can reduce neuronal excitability, affect a variety of physiological functions. One of the channel's most intriguing roles is to regulate the frequency of nerve impulses conducted by the SCN, a structure located in the brain that acts as a master clock to synchronize circadian rhythms throughout the body. During the nighttime, SCN neurons are less active than during the day, consistent with the increase in SCN BK channel abundance that occurs at night. During the day, however, BK channels have little effect on neuron excitability in the SCN, even though one might expect the increase in neuronal firing rate to increase BK activation.

In a new study, Andrea Meredith and colleagues from the University of Maryland School of Medicine show that, in addition to the reduction in overall number of BK channels present in the SCN during the day, decreased channel activity might also depend on an increase in the prevalence of a channel variant containing a four-amino acid chain (SRKR). The researchers demonstrated that stimuli comparable to spontaneous daytime firing elicited a diminished response from the SRKR variant compared with a different BK variant.

The findings provide new evidence about how BK channels modulate SCN activity, solving one more piece of the puzzle of complexities surrounding circadian rhythm regulation.


Bonding together to fight HIV

(BOSTON, MA) A collaborative team led by a Northeastern University professor may have altered the way we look at drug development for HIV by uncovering some unusual properties of a human protein called APOBEC3G (A3G).

APOBEC3G

It is well known that in response to virus infection, the body makes specific antibodies to counteract the infection. However, we are also born with another way to fight infection, namely through the action of defense proteins that are always present in our system. These proteins provide the first line of defense against invading pathogens. For example, we are all potentially protected against HIV because we have an antiviral protein called A3G. However, HIV has evolved a strategy to circumvent the activity of this protein by tricking our cells into destroying our own A3G proteins. This is where Prof. Williams's research comes into play.

A Multi-Functional Protein

A3G moves along a DNA strand as part of its function as an enzyme, and when it reaches a particular one of the four bases in DNA, it chemically alters the DNA, causing HIV to mutate. This was originally thought to be the only way A3G blocks HIV infection. However, some researchers found that even when A3G could not chemically alter the DNA, it still inhibited HIV. To explain this, Prof. Williams's collaborator Dr. Judith Levin from NIH, together with postdoctoral fellow Dr. Yasumasa Iwatani, proposed that A3G forms a roadblock that prevents the virus from making a DNA copy of its genome, thereby stopping HIV replication. This would require A3G to be more slow-acting, yet because the protein normally has to move fast to perform its chemical function, there seemed to be an apparent contradiction in the experimental results.

Professor Williams' research resolves this paradox and shows that the A3G protein does not always have the rapid movement needed for chemical function. Instead, its activity changes over time. **First,**
A3G is a really fast protein,” said Williams. “Then, gradually over time, it becomes a slow protein and remains bound to the DNA, blocking replication.”

**Challenging Popular Opinion**

Many researchers doubted that a protein could have both enzyme and roadblock functions. An enzyme is designed to act rapidly, so the idea of the A3G protein starting off fast, and then gradually slowing down seemed physically impossible. Professor Williams' collaborator Dr. Ioulia Rouzina from the University of Minnesota came up with the novel idea that when A3G proteins group together, they become slower over time. To test the idea, the Williams lab used an instrument called optical tweezers that allowed them to stretch single DNA molecules with A3G proteins bound. By measuring the change in DNA length over time as the proteins came on and off the DNA, they could show that the rates at which A3G bound to DNA became slower over time.

How does this happen? It was already known that A3G proteins bind to each other and form a multi-protein complex. "Once the complex is formed, the A3G proteins are no longer able to move rapidly along the DNA strand as needed for chemical modification of the DNA," said Williams. "This suggests that slow binding can also block HIV replication.”

**Impact On Hiv Research**

The A3G protein has at least two mechanisms by which it can block HIV replication. We have known for over 10 years that A3G can, in principle, provide protection from HIV. However, finding a drug that can counter the anti-A3G activity of the virus has been elusive. This new work has the potential to develop alternative approaches to HIV therapy and development of drugs that can enhance the roadblock activity of A3G. This provides an alternate pathway for drug development that has not previously been pursued.

---

**Two Y Genes Can Replace the Entire Y Chromosome for Assisted Reproduction in Mice**

Nov. 21, 2013 — The Y chromosome is a symbol of maleness, present only in males and encoding genes important for male reproduction. But live mouse offspring can be generated with assisted reproduction using germ cells from males with the Y chromosome contribution limited to only two genes: the testis determinant factor *Sry* and the spermatogonial proliferation factor *Eif2s3y*.

"Does this mean that the Y chromosome (or most of it) is no longer needed? Yes, given our current technological advances in assisted reproductive technologies," said Monika A. Ward, Associate Professor at the Institute for Biogenesis Research, John A. Burns School of Medicine, University of Hawai‘i. At the same time, however, she also emphasized the importance of the Y chromosome for normal, unassisted fertilization and other aspects of male reproduction.

In a new manuscript scheduled for online publication in the journal *Science* on November 21, 2013, Ward and her UH colleagues describe their effort to identify the minimum Y chromosome contribution required to generate a healthy first generation mouse, capable of reproducing a second generation on its own without further technological intervention.

For this study, Ward and her colleagues used transgenic male mice with only two Y genes, *Sry* and *Eif2s3y*. The mice were considered infertile because they had meiotic and postmeiotic arrests—that is, the germ cells that should have normally developed into sperm did not fully mature in these mice—but researchers were able to find few usable cells. Yasuhiro Yamauchi, a post-doctoral scholar on Ward's team, harvested these immature spermatids and used a technique called round spermatid injection (ROSI) to successfully fertilize oocytes in the laboratory. When the developed embryos were transferred to female mouse surrogate mothers, live offspring were obtained.

Because the overall efficiency of ROSI with two Y genes was lower than with regular, fertile mice, the researchers then looked to see whether the addition of other Y genes could improve it. They increased the live offspring rate by about two-fold when *Sry* was replaced with the sex reversal factor *Sxr*, which encodes three additional Y genes. These results demonstrated that *Sxr* encodes a gene or genes that enhance the progression of spermatogenesis.

The study's findings are relevant but not directly translatable to human male infertility cases. In the era of assisted reproduction technologies, it is now possible to bypass several steps of normal human fertilization using immotile, non-viable, or immature sperm. At present, ROSI is still considered experimental due to concerns regarding the safety of injecting immature germ cells and other technical difficulties. The researchers hope that the success of ROSI in mouse studies may serve to support this approach as a viable option for overcoming infertility in men in the future.
As for the human Y chromosome, the researchers agree that it's not on its way to oblivion. Its genetic information is important for developing mature sperm and for its function in normal fertilization. The same is true for mice.

"Most of the mouse Y chromosome genes are necessary for normal fertilization," Ward said. "However, when it comes to assisted reproduction, our mouse study proves that the Y chromosome contribution can be brought to a bare minimum. It may be possible to eliminate the mouse Y chromosome altogether if appropriate replacements are made for those two genes."

**Journal Reference:**

### Study Identifies Protein Essential for Immune Recognition, Response to Viral Infection

**Nov. 24, 2013** — A Massachusetts General Hospital (MGH)-led research team has identified an immune cell protein that is critical to setting off the body's initial response against viral infection. The report that will be published in an upcoming issue of *Nature Immunology* and is receiving early online release describes finding that a protein called GEF-H1 is essential to the ability of macrophages — major contributors to the innate immune system — to respond to viral infections like influenza.

"The detection of viral genetic material inside an infected cell is critical to initiating the responses that signal the immune system to fight an infection and prevent its spread throughout the body," says Hans-Christian Reinecker, MD, of the Center for the Study of Inflammatory Bowel Disease in the MGH Gastrointestinal Unit, senior author of the report. "Our findings indicate that GEF-H1 may control immune responses against a wide variety of RNA and DNA viruses that pose a threat to human health."

The body's first line of defense against infection, the innate immune system rapidly responds to invading pathogens by mobilizing white blood cells, chemical factors called cytokines and antimicrobial peptides. When viruses invade cells, they often move towards the nucleus in order to replicate and sometimes to integrate their own genetic material into that of the host cell, traveling along structures called microtubules that cells use for internal protein transport. But how microtubule-based movement of viral components contributes to induction of the immune response has been unknown.

GEF-H1 is known to bind to microtubules, and previous research indicated that it has a role in immune recognition of bacteria. A series of experiments by Reinecker's team found that GEF-H1 is expressed in macrophages — key components of the innate immune system — and activated in response to viral RNA and that it controls the expression of beta interferon and other cytokines. Mice in which expression of GEF-H1 was knocked out were unable to mount an effective immune response to influenza A and to encephalomyocarditis, a virus that causes several types of infection in animals.

"The sensing of intracellular viral nucleic acids for induction of interferons is so important that many viruses, including influenza A, have evolved specific strategies to interfere with activation of the interferon defense system," says Reinecker, an associate professor of Medicine at Harvard Medical School. "We are hopeful that this discovery will allow the development of new strategies to curtail viral mechanisms that impede the immune responses to infections that are often associated with high mortality rates."

**Journal Reference:**
Hao-Sen Chiang, Yun Zhao, Joo-Hye Song, Song Liu, Ninghai Wang, Cox Terhorst, Arlene H Sharpe, Megha Basavappa, Kate L Jeffrey, Hans-Christian Reinecker. *GEF-H1 controls microtubule-dependent sensing of nucleic acids for antiviral host defenses.* *Nature Immunology*, 2013; DOI: [10.1038/ni.2766](https://doi.org/10.1038/ni.2766)

### Mysterious Virus Could Be Signal of Weak Immune System

**Nov. 21, 2013** — More than 260,000 Americans are alive today thanks to transplant operations that have replaced their failing kidneys, hearts, lungs or livers with healthy organs donated by volunteers or accident victims.

But treatment doesn't end with surgery. Transplant recipients follow strict drug regimens designed to suppress their immune systems just enough to prevent rejection of the donated organ, but not so much as to leave them prone to infection.

Until now, maintaining this delicate balance has been something of a medical guessing game. But in a study to be published Nov. 21 in *Cell*, Stanford University scientists report the discovery of what may be a barometer of immune system strength: a little-known virus that proliferates as the medications suppress the immune system.
The work was led by senior author Stephen Quake, PhD, the Lee Otterson Professor in the School of Engineering and professor of bioengineering and of applied physics.

Quake and a team of collaborators, including transplant specialists from the Stanford School of Medicine, isolated specific DNA fragments from the blood of 96 heart and lung transplant patients for the study.

These fragments came from the blood plasma; red and white cells were extracted, leaving behind proteins and free-floating DNA. The free-floating DNA was from dead cells—the biological version of flotsam and jetsam.

Quake is a pioneer in genomics and was one of the first scientists to use genome sequencing to identify and quantify this free-floating DNA for diagnostic purposes.

One notable application of this technique relates to pregnancy. By studying fragments of cell-free DNA from maternal plasma, Quake has developed tests that can ascertain whether a woman is carrying a fetus with an extra copy of chromosome 21, which causes Down’s syndrome. Additionally, cell-free DNA analysis of fragments from cancerous cells has been used to assess disease progression.

"Cell-free DNA provides an amazing window into human health, and the applications are multiplying well beyond its traditional areas of cancer and prenatal diagnostics," Quake said.

In the new study, Quake’s team used these techniques to study the immune systems of transplant patients. They knew that these patients would begin taking powerful immunosuppressant drugs immediately after surgery. They reasoned that these drugs would affect the microbiome, which is the sum total of all the bacteria, viruses and fungi that inhabit the body.

Although we may not notice these microorganisms unless they cause illness, they help us digest food, excrete waste, make our feet itch or just float around with no discernable effect. In fact, the nonhuman cells that make up the microbiome "outnumber human cells in our bodies at least 10 to one," said lead author Iwijn De Vlaminck, PhD, a postdoctoral scholar in Quake’s lab.

The goal of the experiment was, essentially, to use cell-free DNA analysis to perform a census of the microbiome. The hypothesis was that a systematic analysis of the nature and number of these microorganisms would reveal something about the interplay between the immune system and these nonhuman guests.

"Initially we didn't know what to expect, and we didn't know how the immunosuppressive medications would affect the numbers," De Vlaminck said.

The scientists collected plasma samples from each patient. From each sample, they extracted fragments of nonhuman DNA. They used genetic sequencing techniques to compare the fragments to a library of genetic sequences previously compiled by scientists all around the world. In this way, the team created a snapshot of the various types of bacteria, viruses and fungi in each patient’s microbiome.

The scientists wanted to understand how the microbiome in each patient responded to immunosuppressant medications over time. So they took these plasma samples from each patient about seven times over a two-year period. Each time, they ran a genomic analysis of nonhuman DNA fragments in the plasma. This allowed them to chart the composition of each patient’s microbiome, in a statistical version of time-lapse photography.

The scientists also studied the entire data set of 656 snapshots of all 96 patients as they looked for differences or similarities among patients.

Through this process, one stunning and unmistakable finding emerged: a mysterious micro-organism known as the anellovirus exploded into prominence as the immunosuppressive drugs kicked in, going from very low levels immediately after surgery to dominating the microbiome over time.

"It looks like the anellovirus takes advantage of the lack of immune system surveillance," De Vlaminck said.

Why or how is unknown. In fact, scientists know very little about the anellovirus. Since it was first identified in 1997, it has been found in human subjects whenever scientists have looked for its genetic fingerprints. But this common bug has not yet been identified as the cause of any disease.

But the Stanford scientists did find previous studies involving patients infected with HIV in which levels of anellovirus rose as those unfortunate patients progressed toward AIDS and the full-blown collapse of their immune systems.

These two data points—the increased prevalence of anellovirus measured in the plasma of transplant patients in the Stanford study, and the prior findings from AIDS research—provided strong hints that rising levels of anellovirus indicated a weak immune system.

But that still left a question: Was there a correlation between anellovirus levels and organ rejection?
Working with transplant specialists at the School of Medicine, the scientists looked at their data from a different angle and found out that the answer was yes. Twenty patients suffered moderate or severe episodes of organ rejection during the two years of the experiment. In these 20 patients, levels of anellovirus were "significantly lower ... at almost every point in time," the authors write, adding that "the lower viral load observed for rejecting patients is thus indicative of a higher level of immunocompetence."

Put another way, lower levels of anellovirus suggest a stronger immune system and an elevated risk of organ rejection, while higher levels of anellovirus suggest a weaker immune system with a corresponding shift in risk toward vulnerability to infection.

"We are often walking this fine line between infection and rejection, and we don't have good tools to measure immune system function for individual patients," said co-author Kiran Khush, MD, assistant professor of medicine and a cardiovascular transplant specialist at Stanford. Instead, transplant physicians prescribe medicines according to formulas derived from factors such as time after transplant, age and weight, even though it is likely that different patients, fitting these profiles, will respond to the medications in different ways.

"We end up practicing reactive medicine," Khush said, adding, "A reliable indicator of immune system health is one of the holy grails in transplantation."

Co-senior author Hannah Valantine, MD, professor of cardiovascular medicine and senior associate dean for diversity and leadership at the School of Medicine, described the apparent linkage between anellovirus population and immune system strength as "one of the most exciting observations in my 30-year journey" of transplant research.

"These findings suggest an effective tool to individualize the monitoring and, ultimately, the treatment of rejection," she said. "In the future, this could allow us to safely lower the doses of immunosuppressive drugs patients receive, thereby avoiding devastating side effects."

**Journal Reference:**

---

**Intestinal Bacteria Influence Food Transit Through the Gut**

Nov. 21, 2013 — Food transit through the small intestine affects the body's absorption of nutrients and, consequently, our health. The discovery that food transit time is regulated by a hormone indicates new ways to increase the intestinal absorption of nutrients, and thus potentially treat malnutrition.

One of the tasks of the gut microbiota is to break down essential nutrients from our diet to provide a usable energy source in the colon.

Researchers at the Sahlgrenska Academy, University of Gothenburg, have now shown that lack of energy in the colon leads to increased release of a hormone primarily associated with appetite control and insulin secretion, GLP-1.

Importantly, they also showed that the released GLP-1 regulates how quickly food passes through the small intestine. These findings may open up new possibilities to treat malnutrition and malnutrition-related diseases.

"Food transit through the small intestine is a complex balancing act, in which the gut lining must be given time to absorb nutrients but without allowing pathogenic bacteria sufficient time to colonize the small intestine. We have discovered that food transit through the small intestine is regulated by a specific hormone called GLP-1, which is linked to our glucose metabolism and appetite," says Anita Wichmann, postdoctoral researcher at the Sahlgrenska Academy and the study's lead author.

The study, published in the journal *Cell Host & Microbe*, was led by Professor Fredrik Bäckhed, who heads an internationally recognized research group that investigates the links between the gut microbiota and regulation of the body's metabolism.

"We are continuously discovering new functions that are regulated by the gut microbiota, which highlight its incredibly important function for health and development of diseases," he says.

**Journal Reference:**
Health Group Retracts Claim That 'Self-Inflicted' H.I.V. Is Common in Greece

By Danny Hakim
Published: November 26, 2013
LONDON — The news spread like wildfire after the World Health Organization reported that about half of new H.I.V. cases in Greece were “self-inflicted” as a way to get state benefit payments.

Social media erupted on Monday. There were headlines everywhere from The Daily Mail to the Drudge Report to Al Jazeera. The conservative American commentator Rush Limbaugh weighed in, saying the story shows “what the welfare state does to people.”

But on Tuesday morning, the World Health Organization and the group that produced the report conceded that the H.I.V. claim was not true.

“There is no evidence of people in Greece or anywhere else in Europe deliberately infecting themselves,” said Martin Donoghoe, a spokesman for the health organization.

So what happened? It was an editing error, the group said. It apologized.

The claim was part of a single sentence on page 112 of the organization’s European region report, which was first published in September and more broadly publicized by the agency in late October. And it was startling.

“H.I.V. rates and heroin use have risen significantly, with about half of new H.I.V. infections being self-inflicted to enable people to receive benefits of €700 per month and faster admission on to drug substitution programs,” the report said.

The report was produced by the Institute of Health Equity at University College London and overseen by Sir Michael Marmot, an epidemiologist. (Its full title was “Review of social determinants and the health divide in the W.H.O. European Region: final report.”)

In response to questions from The New York Times, a spokeswoman for the Institute of Health Equity said Monday that the claim came from a Lancet study produced by researchers from the University of Cambridge, the London School of Economics, the London School of Hygiene and Tropical Medicine and the University of California, San Francisco.

But that 2011 Lancet study said that only “a few” such cases had been found, and it cited yet another report, by researchers in Greece. That report, however, said only that it was a “well-founded suspicion that some problem users are intentionally infected with HIV, because of the benefit they are entitled to.”

On Tuesday morning, the Institute of Health Equity spokeswoman told The Times that the W.H.O. report should have said “about half of infections are due to needle injection, some of which is deliberate self-infection.”

However, it was not even clear that there was evidence to say that much. There was no apparent documentation of any such cases in the various reports.

By Tuesday afternoon, the W.H.O. had its own correction: “The sentence should read: ‘half of the new H.I.V. cases are self-injecting and out of them few are deliberately infecting the virus.’” Later in the day, it recalled that statement, however, and said, “There is no evidence suggesting that deliberate self-infection with HIV goes beyond a few anecdotal cases.”

The Greek H.I.V. claim did not receive much attention when it first came out, because it was buried in a lengthy report, though it was included in an article in late October on the website of the magazine New Scientist. It gained traction quickly on Monday on Twitter, and was reposted by European journalists and politicians.


Not all accepted it at face value, however. Media Matters for America, a nonprofit organization that pillories right-leaning news outlets, wrote that “the W.H.O. report is incorrect” in a posting on Monday, after studying the citation.

What is not in dispute is that Greece has had a sharp rise in cases of H.I.V., the virus that causes AIDS, amid its economic crisis.

A report from the European Center for Disease Prevention and Control last year said that, because of a breakdown of preventive services, “the number of new cases reported among people who inject drugs exceeded the number of new cases reported among men who have sex with men” in the first eight months of 2012.
“The current economic turmoil will continue to have adverse effects on H.I.V. prevention not only in Greece, but also in other parts of Europe,” the report said.

A study focused on Athens published in the scientific journal PLOS One said the increase in H.I.V. infections among intravenous drug users jumped annually to 260 in 2011 from between 10 and 20. And that jumped to 522 in 2012, the report said. It noted that it had been previously established that “economic and sociopolitical transitions have helped unleash H.I.V. epidemics in some countries.”

On Tuesday, the World Health Organization said the recent rise of H.I.V. in Greece was “largely driven by infections among people who inject drugs.”

Researchers, the agency added, were working “to fully understand the underlying reasons and recommend appropriate measures” to help those infected.

Researchers have a nose for how probiotics could affect hay fever
A study has shown that a daily probiotic drink changed how cells lining the nasal passages of hay fever sufferers reacted to a single out-of-season challenge. However, it did not lead to significant changes in hay fever symptoms, although this challenge test may not have accurately represented natural allergen exposure.

Our immune system must distinguish between “friends” that can be beneficial to our health and "foes" that can have harmful effects. There is now a growing body of evidence that the gut microbiota, the trillions of bacteria that live in our gut, influences that recognition. When it fails an immune response occurs. This is the case with hay fever, or seasonal allergic rhinitis, when the immune system reacts to pollen or fungal spores.

Previously, a research team at the Institute of Food Research (IFR) found that taking a drink containing the probiotic bacterium Lactobacillus casei changed how our immune system responds to grass pollen, measured through changes in molecules produced by the immune system.

A new study, published in the journal PLOS ONE, shows for the first time how these probiotics can interact with cells in our gut to produce systematic changes in cells lining our nasal cavity.

Funded by Yakult and the Biotechnology and Biological Sciences Research Council (BBSRC), clinicians and scientists at IFR and the University of East Anglia (UEA) on the Norwich Research Park gave 60 hay fever sufferers daily drinks for 16 weeks, outside of the hay fever season. One group was given a drink containing Lactobacillus casei Shirota, and the other group received very similar drinks without the probiotic. The study was double-blinded and placebo controlled, so neither the volunteers nor the scientists knew which group was receiving the probiotic. Samples were taken from the volunteers’ nasal cavities and blood, both before and after being challenged with pollen to trigger their allergy. This was then repeated at the end of the 16-week intervention. Clinical measurements of the symptoms of hay fever were also recorded.

Volunteers who received the probiotic drink saw changes in allergic inflammation in their nasal lining, as well as changes in their blood, that are associated with immune responses. This is strong evidence of how the gut microbiota can influence cells of the gut lining, and have a systematic influence on our bodies and distant cells, such as those lining our nasal passages. But despite this, the probiotic had no detectable effect on the symptoms of hay fever.

Hay fever is a complicated condition to assess, and mimic in a clinical setting. The researchers used a single allergy challenge, applied to the volunteers’ nasal passage, to provide a standard, reproducible test to help ensure all the subsequent results are comparable. In the real world hay fever is usually triggered by longer term exposure to the allergen, variable in strength and timing over a period of days or weeks. The IFR researchers are now exploring the possibility of carrying out a seasonal study to investigate whether the changes in the nasal mucosa seen in this single challenge study relate to changes in hay fever symptoms triggered by a more realistic natural exposure to pollen.

Reference: Oral Delivery of a Probiotic Induced Changes at the Nasal Mucosa of Seasonal Allergic Rhinitis Subjects after Local Allergen Challenge: A Randomised Clinical Trial, PLOS ONE DOI: 10.1371/journal.pone.0078650

Lessons Learned About Influenza A, Four Years After Epidemic
Nov. 22, 2013 — With autumn upon us, and a new flu strain lying in wait, the presentation of a study analysing the management of the 2009 influenza A crisis could not, if anything, be better timed. The paediatrician Eider Oñate has just read her thesis in which she analyses how the H1N1 virus affected the paediatric population in Gipuzkoa during that period: what was done well, what was done badly, and whether the health services allowed themselves to be led by the public alarm rather than by exclusively clinical criteria when deciding hospital admissions. Dr Oñate has analysed the information gathered
during the pandemic to address all these questions, and responses to them could be of use when tackling a similar situation in the future.

"On the basis of the theoretical assumptions the alarm of the health authorities was justified, but fortunately the pandemic did not turn out to be as serious as expected," recalls the paediatrician Eider Oñate from her place of work in the Paediatric Intensive Care Unit at the Hospital Universitario Donostia-San Sebastian. She knows what she's talking about. She was in the firing line during the 2009 influenza A crisis. "What would have been inexcusable would have been the failure to adopt the measures that were taken, and that such caution should have turned out to be responsible for many avoidable victims." Oñate sought to quantify the impact of the pandemic on the paediatric population of Gipuzkoa, during as well as in the periods prior to and following the appearance and spread of the H1N1 virus. She has gathered the results into her thesis "Clinical and epidemiological features of the children hospitalised owing to infection by the influenza virus during the 2009 pandemic in Gipuzkoa: comparison with the pre- and post-pandemic period." The research has confirmed that in this sector of the population (one of the a priori risk groups), the impact of this influenza subtype was similar compared with that of seasonal flu, even though older children and young adults were the ones mostly affected.

The World Health Organisation (WHO) announced the "ongoing pandemic" in June 2009 and a set of protocols were activated in hospitals; as a result, all the cases of children suspected of being infected by influenza A were required to be studied in depth. The gathering of information that under normal circumstances is not obtained offered the chance to study, for the first time, in an organised health system and with the resources for viral diagnosis, the impact of the new virus on the population and on health care and its clinical and epidemiological behaviour. It was also a scenario that had not been repeated since 1968, the year of the previous flu pandemic caused by the appearance of a new strain of the influenza A virus.

**The consequences of public alarm**

The conclusion drawn from the analysis of the data indicates that influenza A caused mostly mild symptoms, even in patients belonging to risk groups, and mortality, as in non-pandemic flu, was low and lower than expected. Despite that, the study has also confirmed that, compared with the data prior to and following the pandemic, there was a substantial increase in the number of consultations handled by the emergency departments and an increase in the number of hospital admissions. "The public alarm had a lot to do with that," admits Oñate. "It not only changed the attitude of the population, but also the routine clinical practice of the paediatricians." So, when an asthmatic child with influenza A for example was involved, they acted differently without basing themselves so much on clinical training. Usually a child with asthma and common flu without any breathing problems is sent home, but during the pandemic the most usual thing was to have him/her admitted to hospital as a precaution.

Aside from the controversy on whether the actual severity of the crisis was distorted by factors outside the health sphere, Oñate stresses that the experience acquired after the first pandemic in the 21st century by the A (H1N1) pandemic influenza virus will make it possible to set up new diagnostic and therapeutic strategies over the coming years. Firstly, one should not allow oneself to be ruled by the alarm. At the same time, it would be necessary in the light of the new experiences to review and assess the strategies affecting the use of antiviral drugs, including the famous Oseltamivir, of proven effectiveness in specific situations. On a practical level it would be necessary to have available methods for early diagnosis that are sufficiently sensitive and reliable for detecting the flu and which would obviate the need for aggressive tests.

History has shown that sooner or later there will be another pandemic. "Nobody knows when it's going to happen again, but we are better prepared now," says the paediatrician, but she issues a warning. "Yes, we could have one as virulent as that of 1918. In these situations you have to prevent contagion and stop it spreading." The paediatrician cites Asia as the possible source where it could emerge, and highlights the importance of the world surveillance network to detect the appearance of new strains sufficiently in advance. "One should not be afraid of flu, but it is important that the people for whom vaccination is recommended be vaccinated," she concludes.

**Emergence of the Middle East Respiratory Syndrome Coronavirus**

Christopher M. Coleman, Matthew B. Frieman
Published: Sep 05, 2013; DOI: 10.1371/journal.ppat.100359
Introduction

It began routinely enough. A patient with severe respiratory disease at the Dr. Soliman Fakeeh Hospital in Jeddah, Saudi Arabia was getting worse and no one knew why. A sample of sputum was sent to Dr. Ali Mohamed Zaki to identify the culprit, as he had identified these diseases many times before. However, this time would be different. The sample showed no positive hits on any of the virus assays he normally used. He contacted Dr. Ron Fouchier, at Erasmus Medical College in Rotterdam, Netherlands, to see if he could be of help. Dr. Zaki’s initial idea was that the virus was a paramyxovirus, and Dr Fouchier had recently published a Pan-paramyxovirus polymerase chain reaction (PCR) assay [1]. In Dr. Fouchier’s lab, the virus was identified as a novel coronavirus, one that had never been seen before.

This novel coronavirus, now called Middle East respiratory syndrome coronavirus (MERS-CoV), has been identified in several countries across the Middle East and Europe, with primary infections found in Saudi Arabia, Qatar, Jordan, and The United Arab Emirates (UAE) (http://www.who.int/csr/disease/coronavirus_infections/en/). Infections in the United Kingdom, Tunisia, France, Italy, and Germany have been imported by travel from the Middle East. The Italian cluster is believed to be from a patient traveling to Jordan and back, and the French cluster originated from a patient traveling to the UAE. The largest cluster of cases, 23 in total, is in Saudi Arabia. As of July 25, 2013, there are 90 confirmed infections, of which 45 have resulted in death, resulting in a 50% case fatality rate. MERS-CoV has been sequenced from nine infected individuals, and its genome sequence places it in the same sub-family (Group 2) as SARS coronavirus (SARS-CoV), but in a new lineage (called Group 2c) (sequences reported in [2]–[4] and at http://www.hpa.org.uk/webc/HPAwebFile/HP Aweb_C/1317138176202; http://www.ncbi.nlm.nih.gov/nuccore/KC776174) (http://www.virology- bonn.de/index.php?id=46).

What Is the Name of This Novel Coronavirus?

The initial name of this novel coronavirus was hCoV-EMC, which stood for human coronavirus–Erasmus Medical College, where the first isolate was sequenced [3]. An additional isolate, provisionally named human coronavirus England 1, was isolated from a patient in London, UK, who had been flown from Qatar to London on July 22, 2012. A report from the Coronavirus Study Group of the International Committee on Taxonomy of Viruses (ICTV) has proposed naming this virus Middle East respiratory syndrome coronavirus (MERS-CoV) [5]. MERS-CoV is provisional until ratified by the ICTV.

Where Did MERS-CoV Come From? Is There a Natural Reservoir?

The closest phylogenetic neighbors to MERS-CoV are putative bat coronaviruses in China (BtCoV-HKU4 and BtCoV-HKU5) [4], the Netherlands (BtCoV/VM314/2008) [2], and a recently discovered isolate from South Africa [6]. All four of these bat coronaviruses have been sequenced only from bat samples and have never been isolated as live viruses from either bats or the environment. The natural reservoir of MERS-CoV has not been identified, although its similarity to these other four viruses suggests that it is of bat origin. Importantly, SARS-CoV emerged from bats as well [7]. Anecdotal evidence suggests that MERS-CoV may have been transmitted to humans via livestock (camels or goats); however, there is no scientific data yet to support this theory.

Does MERS-CoV Share Any Features with SARS-CoV beyond the Zoonotic Origin?

Given the similarities in emergence and apparent zoonotic origins between MERS-CoV and SARS-CoV, initial experiments on MERS-CoV focused on direct comparison with the known molecular biology of SARS-CoV. Infection experiments in cell culture showed that MERS-CoV does not use the SARS-CoV receptor, angiotensin converting enzyme 2 (ACE2), for entry, and that MERS-CoV has a much broader host range than the epidemic isolate of SARS-CoV [8]–[14]. The genome structure of MERS-CoV is similar to other coronaviruses, with the 5′ two-thirds of the genome encoding the non-structural proteins (NSPs) required for viral genome replication, the remaining 3′ third of the genome encoding the structural genes that make up the virion (spike, envelope, membrane, and nucleocapsid proteins), and four accessory genes interspersed within the structural gene region [2]. One additional similarity between MERS-CoV and SARS-CoV is their abilities to inhibit a robust type I interferon (IFN) response in infected cells. However, MERS-CoV has been shown to be much more sensitive to exogenous type I IFN treatment compared to SARS-CoV, which may be important for pathogenesis [8], [9], [11], [14], [15]. Several SARS-CoV-encoded proteins have demonstrated innate immune signaling antagonism [16], and MERS-CoV encodes several IFN antagonists as well (Matthews et al, submitted, Muller et al, submitted).

What Is the Receptor for MERS-CoV and What Cells Does It Infect?

MERS-CoV has been shown to infect a range of human, primate, porcine, and bat cell lines [11]. Ex-vivo infections of human lungs and human airway epithelial cell cultures identified type II alveolar cells and non-ciliated lung epithelial cells (Clara cells) as the targets of infection, rather than the ACE2-expressing
ciliated epithelial cells that SARS-CoV targets [9, 15]. Interestingly, in at least one case, endothelial cells were infected as well [15], showing a distinct difference between the biology of SARS-CoV and MERS-CoV, as SARS-CoV specifically infects ciliated epithelial cells in the lung [17, 18]. The receptor for MERS-CoV was recently identified as dipeptidyl peptidase 4 (DDP4) by mass spectrometry analysis of Huh7 cell protein bound to the MERS-CoV Spike protein in vitro [10]. Transfection and localization experiments demonstrated that DPP4 is indeed the receptor for MERS-CoV and is necessary for infection of a non-permissive cell line [10]. DPP4 has many diverse functions in glucose homeostasis, T-cell activation, neurotransmitter function, and modulation of cardiac signaling [19]. ACE2 does not require enzymatic function in order to act as a receptor for SARS-CoV entry, but the enzymatic function of ACE2 has been linked to severity of the disease following SARS-CoV infection [20]. Similarly, inhibition of the enzymatic function of DPP4 did not affect virus entry in vitro; however, the role of DPP4 enzymatic activity has not been investigated in vivo [10].

What Is the Host Response to MERS-CoV?

Transcriptional analysis of MERS-CoV infected cells has identified several pathways specifically modulated during infection [9]. MERS-CoV is shown to modulate the innate immune response, antigen presentation, mitogen-activated protein kinase (MAPK), and apoptosis pathways. Inhibition of the MAPK pathway showed reduction in viral replication in culture, pointing to potential therapeutics. Importantly, several studies show that MERS-CoV, similar to SARS-CoV, does not induce an early type 1 IFN response, suggesting that MERS-CoV may encode proteins that inhibit sensing of the viral RNA during infection [8, 11, 14, 15]. The modulation of these pathways may explain the increased lethality of MERS-CoV.

Is There a Small Animal Model of MERS-CoV for the Study of Pathogenesis?

There is currently no small animal model for MERS-CoV. Rhesus macaques infected with MERS-CoV display pneumonia, reduced appetite, significant lung pathology, and inflammatory infiltrates [21]. However, MERS-CoV does not replicate in BALB/c, C57B/6, 129SvEv, or STAT1 knockout mice on the 129SvEv background (Coleman et al, submitted). Interestingly, mouse DPP4 is highly similar to the human DPP4, varying at only 62 positions out of 767 amino acids residues total (92% similarity). However, the differences tend to be on surface-exposed residues which, therefore, could affect binding of viral spike protein to mouse DPP4. Future structural and functional interaction experiments are needed to identify if the mouse DPP4 interacts differently with MERS-CoV spike, as compared to human DPP4, and if the known mutations allowing for this binding could be used for the development of a mouse model of MERS-CoV.

Are There Approved Treatments or Vaccinations for MERS?

There is no current treatment or vaccination available for MERS-CoV, but, with the continuation of the outbreak, identification of therapeutics is a top priority. Several manuscripts have demonstrated that a variety of therapeutics inhibit MERS-CoV replication in cell culture [9, 15]. None have been tested in vivo, in part due to the lack of a small animal model, as described above. One promising avenue is to use the knowledge of SARS-CoV and compare it to MERS-CoV. IFNα was shown in multiple models to protect against SARS-CoV-induced disease. MERS-CoV is also sensitive to IFNβ treatment in vitro [15]. Ribavirin, a known inhibitor of RNA viruses, has also been demonstrated to inhibit MERS-CoV replication, and together they can inhibit MERS-CoV at nanomolar levels [22]. Other inhibitors were shown to affect specific pathways, specifically the MAPK pathway. The MAPK inhibitor SB203580 was shown to inhibit MERS-CoV replication in VerE6 cells [9]. Additional therapeutics and vaccinations are in development, with a focus on FDA compounds already in use.

Conclusion and Questions

Many unanswered questions remain on this newly identified virus:

1. What is the environmental reservoir of MERS-CoV? Is it transmitted from bats to camels, goats, or cats? Is the virus linked to date palm harvesting? How did it spread to people from the environment?
2. Are there associated comorbidities that predispose someone to contracting MERS? With the age of infected patients skewed toward older males, is there a genetic link to infection? Are the patients generally immunosuppressed?
3. What is the seroprevalence of MERS-CoV in the general population? Has MERS-CoV been circulating for many years between animals and people and only now mutated enough to be able to cause disease in people? Or is this a new spillover event that has not been seen by humans until now?
4. Why doesn’t MERS-CoV grow in mouse cells or cause disease in mice? Is it because the viral spike protein doesn’t bind mouse DPP4 at all, is it because there are other host factors necessary for entry and replication in mouse cells, or is it due to location and amounts of receptor expression?
5. How do the MERS-CoV proteins contribute to disease? Are there any specific functions of the proteins that allow for enhanced pathogenesis?
6. Since this virus is similar to bat coronaviruses identified in China, Africa, and Europe, why haven’t other bat coronaviruses spilled over into people, causing serious disease (with the exception of SARS-CoV[7] and, potentially, hCoV-229E[23])? What is it about MERS-CoV and the conditions in the Middle East that have contributed to viral infection and the high mortality rate?

With the spread of MERS-CoV through the Middle East, one thing is certain at this point: The emergence of the novel SARS coronavirus in 2003 from a zoonotic source in China and its spread around the world is not an isolated incident of coronavirus spread. Continued spillover events will occur from animals to humans in the future. The sooner we understand these current microbial threats, the more people we can save from infection and possible death. If we can identify these microbes in our environment before they infect us, we can better protect ourselves against future infections.

**Disseminated Histoplasmosis in HIV-Infected Patients in South America: A Neglected Killer Continues on Its Rampage**

Mathieu Nacher, et al
Published: Nov 21, 2013; DOI: 10.1371/journal.pntd.0002319

HIV/AIDS is not a neglected disease. Histoplasmosis is not considered a neglected disease in North America. However, in South America, it should be. It often affects neglected populations and represents a lethal blind spot of the HIV/AIDS data collection systems. Counts of new AIDS cases and AIDS-related deaths are useful to follow the epidemic; however, they overlook the exact cause of death. In the context of the South American pathogen ecology, the systemic mycosis due to *Histoplasma capsulatum* var. *capsulatum* is probably on the top of the list of AIDS-defining illnesses and AIDS-related deaths[1], yet it is mostly undiagnosed and is not even on the diagnostic algorithm used by a significant proportion of clinicians facing a febrile, severely immunodepressed patient in the region.

**The Invisible Burden**

Studies performed in the 1950s and 1960s on the histoplasmin skin test positivity in South America showed positivity rates around 30% from Trinidad and Tobago in the North, to Uruguay and Argentina in the South. The pathogen is there[2]. Despite this, expertise and awareness of this disease is limited to mycologists and some clinical teams scattered throughout the South American continent[2–16]. But those with expertise are the exception rather than the rule. Imported cases in Europe occurring in HIV-infected residents or travellers from South America, notably in France, Spain, and Italy, are starting to be recognized, but often late in the course of the disease because clinicians are not familiar with this “endemic” disease[17–19].

For too long, the absence of a simple, reliable, and affordable diagnostic test has made it difficult to determine the burden of this disease in HIV-infected patients in much of South America. The gold standard for diagnosis relies, so far, on the culture of fluid and tissue samples[20]. This requires invasive investigations by clinicians (bone marrow aspirates; biopsies of the liver, lymph node, and intestine; etc.). From the lab perspective, direct examination may accelerate diagnosis, but culture may take weeks and require a BSL 2 laboratory. Detection of specific antigens in serum or urine samples is not available for diagnosis in most of South America, and galactomannan detection (cross reactivity during histoplasmosis) is not being used as a standard of care. Contact with clinicians from various countries suggests that, although severe histoplasmosis often kills in a few days, most clinicians are not very aggressive in their investigations. Moreover, presumptive antifungal therapy is rare. Biopsies are usually immersed in formalin by surgeons rather than sent for culture. Most often, clinicians do not take proper samples for mycological diagnosis, creating a vicious cycle that diminishes the capacity of mycological laboratories and perpetuates underfunding and the absence of diagnosis and, thus, the “nonexistence” of the very disease that is killing numerous patients.

In French Guiana, there has been a mycology laboratory since 1997 with a BSL 3 laboratory. The virtuous cycle between laboratory and clinicians has been fruitful: clinicians are well informed about histoplasmosis and are quite aggressive in looking for the infection, while the mycology laboratory has the capacity to appropriately process and identify *Histoplasma* in clinical specimens[21]. Although published
maps [2] show no histoplasmosis in most of French Guiana, the recent figures that have emerged are striking. With 1.5 cases per 100 patient-years, histoplasmosis is the most common cause of AIDS-defining illness. Interestingly, despite awareness of this disease and availability of liposomal amphotericin B, it has also been the leading cause of AIDS-related death for decades. A recent 2-year study of all HIV patients admitted in Saint Laurent du Maroni hospital showed that 41% of admitted patients with CD4 counts below 200 had disseminated histoplasmosis, and 85% of admitted patients with CD4 counts below 50 and isolated fever had disseminated histoplasmosis. This is a clear message for physicians when admitting a severely immunodepressed HIV patient in the region: “Don’t miss histoplasmosis!”

No Data = No Existence. Meanwhile, Patients Continue Dying from a Treatable Disease...
The high prevalence of HIV–Histoplasma coinfections on the South American continent is not a trivial problem. The scarcity of the published research on this topic reflects the tragic fact that this problem is evolving under the radar of health care systems and is truly a neglected disease. Generations of young doctors will learn to look for tuberculosis, pneumocystosis, and bacterial pneumonia when confronted with a febrile patient with respiratory signs, but not for histoplasmosis. Similarly, important clinical clues, such as cytopenia (ascribed to bone marrow involvement with or without haemophagocytic syndrome) and liver enzyme abnormalities will often not lead to the suspicion of disseminated histoplasmosis. A big danger is that a smear negative, treatment resistant, “tuberculosis-like” syndrome may often be labelled drug-resistant tuberculosis, when it was never tuberculosis in the first place. It is thus of paramount importance to fill this knowledge gap and revise the diagnostic and therapeutic algorithms in the region. The “Know your epidemic, know your response” UNAIDS slogan should also be applied to histoplasmosis.

We Need Research and We Need to Act Now
The HIV/AIDS epidemic is still active in countries in the Amazon basin. Guyana, Suriname, French Guiana, and the Brazilian state of Amapa all have HIV prevalence rates over 1% of the population. Although the AIDS incidence has steadily declined in the southern states of Brazil, the situation in the northern (Amazonas, Roraima, and Amapa) and the northeastern states of Brazil is still concerning. A very coarse calculation based on 600,000 HIV patients and an annual histoplasmosis incidence rate of 1.5% would estimate the annual number of cases to be in the thousands. Unfortunately, histoplasmosis thus still has a future in HIV patients. Although there is a need for epidemiologic research to measure the true burden of disease in various regions of South America, actions can, and should, be taken to diagnose and treat patients now. We need to develop standard mycological practices in the area that emphasize early and aggressive clinical diagnosis, and we need to develop new rapid diagnostic tools and advocate for affordable treatment. New affordable diagnostic assays (CDC, Immy) are presently being tested in Brazil, French Guiana, Suriname, and Colombia. We hope they will allow us to improve our knowledge of local epidemiologies and reduce patient mortality through early diagnosis and increased awareness. Although amphotericin B is available, it has potential significant renal side effects. Liposomal amphotericin B is the treatment of choice of the most severe cases of HIV-associated histoplasmosis, but its cost exceeds 800 US dollars per day. Gilead Sciences, Inc. has committed to the procurement of HIV drugs at affordable prices. Extending this policy to the problem of treating HIV-associated histoplasmosis, a neglected disease, would be an important step in improving the health of HIV/AIDS patients in the region. In the near future, DNDi (Drugs for Neglected Diseases initiative) should provide a low-cost, heat-stable alternative to liposomal amphotericin B that could be valuable for histoplasmosis treatment. Our focus has been HIV-associated disseminated histoplasmosis; however, it should be emphasized that the problem of missing the diagnosis of histoplasmosis in South America also extends to some immunocompetent patients or patients with causes of immunodepression other than HIV [22].

We need tests; we need treatments; but first of all, we physicians need to integrate our South American epidemiology in our diagnostic algorithms. Looking for malaria in febrile patients in malaria-endemic areas is automatic; looking for histoplasmosis in febrile, immunosuppressed, HIV-infected patients in South America, Central America, and perhaps way beyond [23], [24], is not. The sooner we do it, the better for our patients.

The Leprosy Bacillus, circa 1873
A scientist’s desperate attempts to prove that Mycobacterium leprae causes leprosy landed him on trial, but his insights into the disease’s pathology were eventually vindicated.

By Kate Yandell | October 1, 2013
Gerhard Armauer Hansen observed *Mycobacterium leprae* for the first time in infected nodules excised from leprosy patients. Barely distinct, rod-shaped bacteria (purple) became apparent under Hansen’s microscope. However, it took German bacteriologist Albert Neisser’s stain for the bacterium, developed after visiting Hansen in 1879, to make *M. leprae* clearly visible. Pictured are illustrations of *M. leprae*-infected cells from a testicle, taken from Hansen’s 1895 book *Leprosy: In its Clinical and Pathological Aspects*. WELLCOME LIBRARY, LONDON

Norwegian physician Gerhard Armauer Hansen first saw rod-shaped microbes in samples harvested from leprosy patients in 1873. Seven years later, Hansen, who worked in the leprosy hospital in the coastal town of Bergen, was on trial for attempting to infect a patient with bacteria without permission, using a cataract knife to inoculate a woman's eye with material from leprosy lesions.

Hansen resorted to such an extreme measure because he was having trouble proving his conviction that the microbes caused leprosy—which results in peripheral nerve damage and skin lesions—and that the disease was infectious. He had tried in vain to infect rabbits and to cultivate the microbe in vitro—evidence considered necessary to prove contagiousness. "Leprosy was afterwards called the least contagious of contagious diseases," says Tony Gould, author of *A Disease Apart: Leprosy in the Modern World*, which might explain why Hansen had struggled to come up with the necessary proof.

Hansen’s unfortunate patient, a 33-year-old woman named Kari Nielsdatter, already had tuberculoid leprosy, one form of the disease, but Hansen hoped to infect her with a second form, called lepromatous leprosy. The infection did not take hold, but Hansen was punished for conducting the experiment. He was stripped of his position at the leprosy hospital but allowed to keep his position as Norway’s chief medical officer for leprosy, which he used to push through measures that kept leprosy patients in partial isolation.

Despite his misdeeds, Hansen was later honored as the discoverer of *Mycobacterium leprae*, which was officially accepted as the cause of the disease at the first International Leprosy Conference, held in Berlin in 1897. Today, leprosy is often called Hansen’s disease.

Some of the early skepticism about the contagiousness of the disease came from Daniel Cornelius Danielssen, Hansen’s mentor and a preeminent leprosy expert of the day. Danielssen was convinced that leprosy could not be transmissible and instead thought it ran in families, or arose from poor living conditions. He had even inoculated himself and others with material from leprosy patients without causing illness, which bolstered his conviction.

While Hansen’s assertion that leprosy is infectious was ultimately vindicated, “there appears to be a very strong genetic predisposition to leprosy,” according to Richard Truman, acting chief of the laboratory research branch at the National Hansen’s Disease Program in Baton Rouge, Louisiana. Only up to 5 percent of people are susceptible to leprosy, and susceptibility appears to run in families, but is additionally enhanced by malnutrition and conditions that compromise the immune system.

In the end, then, perhaps Hansen and Danielssen were both partly right: Hansen’s mysterious rods cause leprosy, but only in those with the poor luck to be genetically and environmentally susceptible.
Secret Botulism Paper Published
The discovery of a new form of the deadly botulinum toxin gets published, but its sequence is kept under wraps until an antidote is developed.
By Bob Grant | October 18, 2013
In a publishing first, the sequence of a newly discovered protein is not divulged in papers announcing the finding. Researchers at the California Department of Public Health in Sacramento discovered the protein, a new type of the extremely dangerous botulinum toxin, lurking in the feces of a child who displayed the symptoms of botulism. They published their findings in two reports on the website of The Journal of Infectious Diseases, but absent from either paper was the DNA sequence of the protein, the eighth form of botulinum toxin recovered from the bacterium Clostridium botulinum. The move represents the first time that a DNA sequence has been omitted from such a paper. “Because no antitoxins as yet have been developed to counteract the novel C. Botulinum toxin,” wrote editors at The Journal of Infectious Diseases, “the authors had detailed consultations with representatives from numerous appropriate US government agencies.”

These agencies, which included the Centers for Disease Control and Prevention and the Department of Homeland Security, approved publication of the papers so long as the gene sequence that codes for the new protein was left out. According to New Scientist, the sequence will be published as soon as antibodies are identified that effectively combat the toxin, which appears to be part of a whole new branch on the protein’s family tree.

Fighting Viruses with RNAi
The long-debated issue of whether mammals can use RNA interference as an antiviral defense mechanism is finally put to rest.
By Ruth Williams | October 10, 2013
Plants, fungi, and invertebrates use RNA interference (RNAi) to fend off invading viruses. Mammals, on the other hand, are known to contain RNAi machinery, but researchers have never been able to prove that they use the molecular obstruction strategy for fighting viruses. Two papers published in Science today (October 10) provide the long-elusive evidence that indeed they do.

“There was controversy in the field as to whether or not RNAi ever acts as an antiviral response in mammals, and I think what is clear from these papers is that it does, in at least some laboratory conditions,” said Christopher Sullivan, a professor of molecular genetics and microbiology at the University of Texas in Austin, who was not involved in the work. “Now the argument will shift to when or if this matters in the wild,” he said.

When a single-stranded RNA virus infects a cell, its first actions are to hi-jack the protein production machinery and make essential viral proteins such as replicase. This enzyme makes a complimentary strand to the viruses’ genetic material, which is then used as a template to generate multiple copies of the virus. With each round of replication the viral genome thus temporarily becomes double-stranded. And it is this unusual two-ply RNA molecule that triggers the host to initiate RNAi—an ancient mechanism that chops the double-stranded RNA into 21- to 23-nucleotide-long fragments called siRNAs, which target and destroy the virus’s single-stranded genome.

Over the course of evolution, however, the same machinery has been adopted for processing non-coding RNAs of the host into microRNAs that target and suppress host messenger RNAs. That is, in addition to its role in virus combat, RNAi machinery is used as a gene-regulatory mechanism.

Since RNAi’s virus-fighting ability was first discovered in plants and insects, scientists have been hunting for its equivalent in mammals. But despite repeated attempts they could only find evidence for RNAi’s gene-regulatory role.

“A lot of people have tried—ramming their heads against the wall time and time again—to discover this [in mammals] . . . and didn’t find it,” said Sullivan. In fact the results were “strikingly negative,” he said. However, added Eric Miska, a professor of molecular genetics at the Gurdon Institute in Cambridge, UK, who also was not involved in the work, “lack of evidence is not evidence for the lack of this pathway.”

There are a number of reasons why an antiviral RNAi pathway may have been elusive in mammalian cells, explained Olivier Voinnet, a professor at the Swiss Federal Institute of Technology in Zurich who led one of the new studies. First, mammals have evolved an alternative virus-combating innate immune response, involving proteins called interferons. Second, many viruses carry RNAi-suppressing proteins, which may have hidden the process. Third, researchers may simply have been looking in the wrong cells.
Multipotent cells, such as embryonic stem cells, do not produce or respond to interferon—possibly because the interferon response often leads to cell death, and multipotent stem cells, which are essential for replenishing tissues, might be considered more valuable than their differentiated counterparts, said Voinnet. Whatever the reason, Voinnet thought that these interferon-less cells might expose a virus-fighting RNAi process hidden in other cell types. Sure enough, infection of mouse embryonic stem cells with encephalomyocarditis virus led to the production of siRNAs characteristic of RNAi. Furthermore, infection of embryonic stem cells with Nodamura virus that lacked its B2 protein—a suppressor of RNAi—also gave rise to siRNAs.

“For the first time we’ve seen real siRNAs being produced in mammalian cells,” said Voinnet excitedly. “All we’d seen before were microRNAs.”

Why had no one thought to look in stem cells previously? “The majority of our cells are not pluripotent and the majority of cells that first come in contact with viruses, then, are not pluripotent,” said Sullivan. Thus people had assumed stem cells were not an important site of virus-fighting action. Additionally, in plants and insects, RNAi is clearly an antiviral response in differentiated cells, he said.

It might not be only pluripotent cells where antiviral RNAi is active, however. Shou-wei Ding, a professor and plant pathologist at the University of California, Riverside, who led the second study, found that deleting the B2 gene in Nodavirus was sufficient to produce siRNAs in baby hamster kidney cells and newborn mice infected with the virus.

These reports of infection-induced siRNA production provide the first proof that antiviral RNAi is possible in mammalian cells and live mice. How commonly the mechanism is used by mammals, however, remains to be seen.

“[The work] challenges the field to now . . . look for [natural] examples where this really matters,” Miska said.


A New Antibiotic?

Scientists show that peptide-conjugated phosphorodiamidate morpholino oligomers can effectively silence essential bacterial genes.

By Tracy Vence | October 16, 2013

A new type of antibiotic based on DNA/RNA analogs that silence the expression of specific bacterial genes is reported in The Journal of Infectious Diseases today (October 16) by researchers from Oregon State University (OSU), the University of Texas Southwestern Medical Center, and the Corvallis-based firm Sarepta Therapeutics. Called peptide-conjugated phosphorodiamidate morpholino oligomers (PPMOs), the compounds were effective at reducing the viability of Acinetobacter lwoffii and A. baumannii both in vitro and in infected mice.

“With infections from drug-resistant pathogens rising rapidly, there is an urgent need to come up with new approaches such as the use of PPMOs to spur antibiotic development,” study coauthor David Greenberg from UT Southwestern noted in a statement. “There is a lot of promise in developing new antibiotics that target specific pathogens as opposed to so-called broad-spectrum antibiotics that target whole classes of bacteria.”

Fierce Drug Delivery noted that a PPMO is “essentially a synthetic form of microRNA,” such as those that have already been used to silence specific disease-causing genes. While further research is required before PPMOs move to clinical trials, the researchers said their approach shows promise for precisely targeting specific pathogens. “The mechanism that PPMOs use to kill bacteria is revolutionary,” OSU’s Bruce Geller, coauthor on the study, said in a statement. “They can be synthesized to target almost any gene, and in that way avoid the development of antibiotic resistance and the negative impacts sometimes associated with broad-spectrum antibiotics.”
Drug Widens Immunity to Flu

An immune suppressive drug can unexpectedly help immunized mice fight off many strains of flu.

By Ed Yong | October 20, 2013

The immunosuppressant drug rapamycin paradoxically helped to protect mice against a diverse range of influenza viruses after the animals were vaccinated against just one flu strain.

Rachael Keating from St. Jude Children’s Research Hospital said it seems that rapamycin steers immune cells away from producing antibodies that strongly target a particular flu strain, in favor of those that block a wide variety of strains. Her results, published today (20 October) in Nature Immunology, could help in the long-running race to develop a universal flu vaccine.

There are many subtypes and strains of influenza, which evolve at great speed and often hybridize into entirely new strains. Current flu vaccines cannot protect against all of these strains, which forces scientists to try and predict those most likely to cause problems in the coming year. This imperfect system often leaves people unprotected against unexpected strains, let alone emerging ones that have the potential to cause pandemics. This has driven an intense search for a universal vaccine, and the new study could further help researchers working toward that goal.

In 2009, a different group unexpectedly showed that rapamycin boosted the immune response against a rodent virus in mice by increasing the quantity and quality of CD8 T-cells, which destroy infected cells. Inspired by this discovery, Keating showed that the drug provides the same benefits against flu.

She and her colleagues gave mice a dose of rapamycin while immunizing them against a strain of seasonal H3N2 flu, finding that the rodents could also ward off lethal doses of other strains, including H1N1, H5N1, and the emerging H7N9.

To the team’s surprise, CD8+ T cells were not responsible for rapamycin’s positive effects. Instead, the drug seemed to work by affecting B cells, which make antibodies, and CD4 T cells, which help in this process.

Over the course of a flu infection, B cells typically change the class of antibodies that they produce. They also fine-tune those antibodies into a narrower set, which bind very strongly to variable regions of flu viruses—regions that are usually specific to different strains. This produces an immune response that efficiently deals with a single strain, but is ill-equipped to protect against others.

“Rapamycin prevents that narrowing,” explained senior author Maureen McGargill. In treated mice, the B cells produced a more diverse repertoire of antibodies, which targeted different parts of the incoming viruses, including regions that are conserved across many strains. This provided protection against flu viruses regardless of strain.

Such cross-reactive antibodies bind relatively weakly to their targets and, under normal circumstances, would probably get outcompeted by antibodies with a narrower focus but higher affinity. “For whatever reason, antibodies to the conserved regions are very rare,” said McGargill. That’s why humans do not naturally become immune to all flu strains after just encountering one.

Rapamycin blocks mTOR, a receptor protein that affects the production of B-cells and the switch between antibody classes. How exactly this leads to a wider array of antibodies is unclear, and the team is now working to unpick the details.

“It’s interesting that it is possible to skew the response towards more broadly cross-reactive antibodies, in mice, in a particular situation,” said Sarah Gilbert, an immunologist from the University of Oxford who was not involved in the work. “[But] it is very difficult to understand how the findings in this paper relate to vaccinating humans.”

The problem is that current flu vaccines use inactivated flu viruses, while McGargill’s team used live viruses. The researchers said that their viruses—which were either inactivated at body temperature or injected into parts of the body where they cannot reproduce—effectively acted like a vaccine. But “if they
Genetic Roots of the Ashkenazi Jews
Most Ashkenazi Jews, traditionally believed to have descended from the ancient tribes of Israel, may in fact be maternally descended from prehistoric Europeans.

By Kate Yandell | October 8, 2013

The majority of Ashkenazi Jews are descended from prehistoric European women, according to study published today (October 8) in Nature Communications. While the Jewish religion began in the Near East, and the Ashkenazi Jews were believed to have origins in the early indigenous tribes of this region, new evidence from mitochondrial DNA, which is passed on exclusively from mother to child, suggests that female ancestors of most modern Ashkenazi Jews converted to Judaism in the north Mediterranean around 2,000 years ago and later in west and central Europe.

The new findings contradict previous assertions that Ashkenazi mitochondrial lineages originated in the Near East, or from mass conversions to Judaism in the Khazar kingdom, an empire in the north Caucasus region between Europe and Asia lasting from the 7th century to the 11th century whose leaders adopted Judaism. “We found that most of the maternal lineages don’t trace to the north Caucasus, which would be a proxy for the Khazarians, or to the Near East, but most of them emanate from Europe,” said coauthor Martin Richards, an archaeogeneticist at the University of Huddersfield in the U.K.

Richards and colleagues’ story “seems reasonable,” said Harry Ostrer, a human geneticist at Albert Einstein College of Medicine of Yeshiva University in New York City who was not involved in the study. “It certainly fits with what we understand about Jewish history.”

The Ashkenazi Jews make up the majority of Jews today and most recently have ancestry in central or Eastern Europe. Previous work has demonstrated that just four mitochondrial types, pass down from four mothers, account for 40 percent of variation in Ashkenazi Jews’ mitochondrial DNA, and some researchers have published evidence of Near Eastern origins for these Ashkenazi mitochondrial types.

To further investigate the matrilineal lines of the Ashkenazi Jews, Richards and colleagues looked at mitochondrial genome sequences in living Jews and non-Jews from the Near East, Europe, and the Caucasus. Based on the results, the team concluded that, in contrast to the evidence for many Ashkenazi males, whose Y chromosomal DNA suggests a likely origin in the Near East, the female lineage of Ashkenazi Jews have substantial ancestry in Europe. Specifically, the researchers found that the four main Ashkenazi founder mitochondrial types were nested within European mitochondrial lineages, not Near Eastern ones, and an analysis of more minor haplogroups indicated that an additional 40 percent of mitochondrial variation found in Ashkenazi Jews’ mitochondrial DNA was likely of European origin. The remaining variants appeared to be from the Near East or are of uncertain origin, and there was no evidence for Ashkenazi Jewish origins in the Khazar kingdom, according to the authors.

Historical evidence indicates that Jewish communities began to spread into Europe during classical antiquity and migrated north during the first millennium CE, arriving in the Rhineland by the 12th century. Local European women could have begun to join the Jewish population around 2,000 years ago or earlier, Richards and colleagues suggest, and the Ashkenazis may have continued to recruit additional women as they headed north.

But some scientists question these conclusions. “While it is clear that Ashkenazi maternal ancestry includes both Levantine [Near Eastern] and European origins—the assignment of several of the major Ashkenazi lineages to pre-historic European origin in the current study is incorrect in our view,” physician-geneticists Doron Behar and Karl Skorecki of the Rambam Healthcare Campus in Israel, whose previous work indicated a Near Eastern origins to many Ashkenazi mitochondrial types, wrote in an e-mail to The Scientist. They argue that the mitochondrial DNA data used in the new study did not represent the full spectrum of mitochondrial diversity.

R. Keating et al., “The kinase mTOR modulates the antibody response to provide cross-protective immunity to lethal infection with influenza virus,” Nature Immunology, doi:10.1038/ni.2741, 2013.
mitochondrial lineages of the Ashkenazis but disagreed that it rules out a Khazarian contribution. “Jews and non-Jews residing in the regions of Khazaria are underrepresented, which biases the results toward Europe as we have seen in many other studies,” he said in an e-mail to The Scientist. Elhaik recently concluded from autosomal DNA that European Jews did, in fact, have a Khazarian background.

David Goldstein, a geneticist and director of the Center for Human Genome Variation at the Duke University School of Medicine, said that the questions of whether there was a Khazar contribution to the Ashkenazi Jews’ lineage, or exactly what percentage of mitochondrial variants emanate from Europe, cannot be answered with certainty using present genetic and geographical data. Even if a set of variants are present in a specific region today, that doesn’t mean that the region always had that set of variants. Some variants could have been lost due to drift, or perhaps migration altered the balance of variants present in the population.

“These analyses really do not have any formal statistical inference about evolutionary history in them,” Goldstein wrote in an e-mail to The Scientist. “They are based on direct interpretations of where one finds different [mitochondrial DNA] types today. And so the analyses are largely impressionistic.”

Nevertheless, Goldstein noted that the new study “does offer better resolution of the [mitochondrial DNA] than earlier ones, and so the suggested interpretation could well be right.”


Brain-Washing During Sleep
Rest clears out interstitial clutter in the mouse brain.
By Kerry Grens | October 18, 2013
The restorative power of sleep may originate outside of our neurons, in the interstitial space between cells. During sleep, this space expands by 60 percent, perhaps to more effectively clear away toxins, researchers report this week in Science. “Sleep changes the cellular structure of the brain. It appears to be a completely different state,” lead author Maiken Nedergaard, the co-director of the Center for Translational Neuromedicine at the University of Rochester Medical Center in New York, said in a press release.

Nedergaard and her colleagues showed that the mouse glymphatic system—which exchanges cleansing cerebrospinal fluid for protein-laden interstitial fluid in the brain—enlarges during sleep or anesthesia, and shrinks during awake periods. As an example of sleep’s cleaning power, the group demonstrated that β-amyloid in the interstitial space disappeared faster while animals were sleeping.

Sleep researchers say the finding makes sense. It “fits with a long-standing view that sleep is for recovery—that something is paid back or cleaned out,” David Dinges of the University of Pennsylvania told Science Now. “It’s not surprising, our whole physiology is changing during sleep,” Raphaelle Winsky-Sommerer, a lecturer in sleep at Surrey University, told the BBC. “The novelty is the role of the interstitial space, but I think it’s an added piece of the puzzle, not the whole mechanism.”

Nedergaard told the BBC that the brain has limited resources, and can’t devote energy to both the functions of awake states and house cleaning. “You can think of it like having a house party,” she said. “You can either entertain the guests or clean up the house, but you can’t really do both at the same time.”

Lnc-ing p53 and epigenetic silencing via a novel noncoding RNA
Posted by Biome on 16th October 2013
It is now well-established that noncoding regions of the genome have a vital role to play in gene expression. Genome-wide approaches have led to the identification of an abundance of noncoding RNAs, from microRNAs to long non-coding RNAs (lncRNAs). LncRNAs were initially discovered in the 1980’s but the extent of their transcription was only realized over twenty years later with the advent of improved high-throughput sequencing technologies. Subsequent research has revealed the breadth of cellular processes they are involved in. Maite Huarte from the University of Navarra, Spain and colleagues probe how the novel
In your study you describe a novel lncRNA, Pint, that you studied in mouse cells. What were the main findings of these experiments? What did you uncover about the function of Pint?

We have identified Pint, a ubiquitously expressed lincRNA (long intergenic noncoding RNA) that is finely controlled by the tumor suppressor p53. We found that in mouse cells, Pint regulates the expression of genes of the TNF-β, MAPK and p53 pathways, promoting proliferation and survival. We therefore decided to investigate the mechanism of gene regulation by Pint, and found that it has an effect on the epigenome: Pint is a nuclear lincRNA that interacts with Polycomb Repressive Complex 2 (PRC2), an important epigenetic regulator, and is required for PRC2 targeting of specific genes for their repression. In fact, we observed that Pint functional activity is highly dependent on PRC2 expression.

How did you identify human PINT?

After finding Pint in the mouse genome, we wondered if an equivalent lincRNA exists in human cells. Indeed, we identified a human transcript that is produced from the human syntenic region. Despite relatively low sequence homology, human PINT presents suggestive analogies with the mouse lincRNA (Pint): PINT is similarly regulated by p53, and its expression correlates with the same cellular pathways as the mouse ortholog, including the p53 pathway. Interestingly, PINT is under-expressed in colon cancer, while its enforced expression inhibits the proliferation of tumor cells. These data are consistent with a possible role of PINT as a tumor suppressor lncRNA.

What do we know about lncRNAs relationship with the p53 pathway? How can this knowledge be useful to us?

We know that p53 controls the expression of many lncRNAs, and we are only starting to understand the biological meaning of this. Our work is revealing that these p53-regulated lncRNAs can effectively fine-tune the p53 response. Our findings help understand how lncRNAs contribute to gene networks in general and, in the particular case of p53, have important implications: lncRNAs have an impact in the way other p53 regulated lncRNAs are expressed, and in the particular case of p53, have important implications: lncRNAs have an impact in the way other p53 regulated lncRNAs are expressed, and, in the particular case of p53, have important implications: lncRNAs have an impact in the way other p53 regulated lncRNAs are expressed, and, in the particular case of p53, have important implications: lncRNAs have an impact in the way other p53 regulated lncRNAs are expressed. Additionally, although the possibility is still distant, therapeutic expression of PINT may carry fewer negative effects than that of protein coding genes, given that it regulates specific facets of p53 and PRC2.
What are your plans for future studies?
There are still many unresolved questions that we continue to investigate. First of all, regarding the mechanism of gene repression: How can Pint-PRC2 specifically recognize and target specific genes? Are there other proteins/RNAs involved in this repressive complex? Another aspect that interests us is the role of human PINT as a tumor suppressor: How does PINT loss affect the biology of a tumor? Is the mechanism conserved between mouse and human? These are some of the questions that we would like to answer with our future work.

Measuring Latent HIV
The latent HIV provirus reservoir persisting in CD4+ T cells is potentially 60 times larger than previously believed, researchers show.
By Abby Olena | October 24, 2013
Most HIV treatments depend on the activation of all CD4+ memory T cells that harbor the latent reservoir (LR), which is composed of inactive HIV proviruses integrated into the genomes of these resting cells. Knowing the size of the LR is essential for measuring the success of antiviral therapies. Two common ways to test for provirus in a patient with HIV are the PCR-based method, in which integrated HIV DNA is amplified and quantified using PCR and the viral outgrowth assay (VOA). For the VOA, a patient’s CD4+ T cells are collected and then activated in vitro, so that they produce virus from the LR and release viral proteins that can then be quantified. PCR detects much more provirus than the VOA, and in the past, it was assumed that not all of the provirus that is detected by PCR is transcription-competent. Now, researchers from Johns Hopkins University School of Medicine in Baltimore, Maryland, have characterized the LR more completely than ever before, and show that its size is perhaps 60 times larger than predicted by the VOA. Their work was published today (October 24) in Cell.

“I think it’s an excellent paper and a very provocative finding,” said HIV researcher Jonathan Karn, a professor of molecular biology at Case Western Reserve University, who was not involved in the work. The evidence “does indicate that the VOA probably underestimates the latent reservoir,” added Karn, which is “really quite an important observation.” Knowing the LR’s true size is essential for clinicians who must decide when a patient can discontinue antiretroviral therapy, he noted.

The VOA “is a very sensitive assay to detect all possible viruses released after maximum T cell activation, so people have believed that this scale of the latent reservoir is what we need to eradicate, which is around one in one million resting CD4+ T cells,” said first author Ya-Chi Ho, who is a postdoctoral fellow at Johns Hopkins. The research team thus began as it would have approached a VOA: by activating patients’ CD4+ T cells. Then they used PCR to amplify proviruses not induced by activation, and sequenced them directly.

The researchers found that 88 percent of proviruses had obvious defects that would make them replication-incompetent, such as deletions or start codon mutations, which was consistent with previous observations. However, almost 12 percent had no obvious sequence defects, yet were not induced during the first round of T cell activation. The scientists examined the genomic location and methylation status of these proviruses. They found that most of the noninduced proviruses were in active transcription units and were not CpG-methylated. They also showed that more rounds of stimulation could induce some of the proviruses to produce active virus. Based on these results, the researchers proposed that provirus induction is stochastic, and that the size of the LR is likely around 60 fold higher than previously predicted by VOA.

“I think everybody in the field had a sense that there was something like this to explain the discrepancy between the VOA and the PCR-based assay,” said Fabio Romero, an assistant professor at the Institute of Human Virology at the University of Maryland School of Medicine in Baltimore, who did not participate in the study. “What was really striking, at least to me, was that there is a high frequency of cells that do not produce virus when they are cultured in the VOA, and yet they contain replication-
competent HIV sequences,” he continued. “That is scary,” Romerio said, because latent HIV must be induced in vivo in order for anti-latency drugs to be successful.

The paper confirms that “there are obviously difficulties with every ex vivo assay,” said David Margolis, a professor of medicine at the University of North Carolina, Chapel Hill, who studies HIV latency but was not part of the work. “None of the data is really surprising, but it’s really important that it’s definitively shown,” he added, because “some people keep calling measures of HIV DNA the HIV reservoir. That’s just not true.” It’s a measure of persistent infection, but most of what you’re measuring is not a reservoir anymore, as demonstrated by this work.”

A larger HIV reservoir may sound daunting, but experts agreed that although the paper showed caution is required when interpreting all assays, it is better to understand the full size of the LR than to be in the dark.

“I think this paper is part of a very exciting time, where we’re starting to see that there is a way forward from where we are,” said Margolis. “I think we just have to be very rigorous and continue to do careful, but difficult studies like this one that will ensure important progress.”

The field will need to grapple with how to proceed with activation and treatment in vivo, based on new understanding that latent, replication-competent HIV may not be activated, said Karn. But “I am very optimistic that we can get this right,” he added.


HIV Structural Studies Undermine Prior Work

New research on the structure of the surface protein the virus uses to infiltrate human cells clashes with an earlier paper’s findings, causing some scientists to call for a retraction.

By Bob Grant | November 4, 2013

Scientists have inched closer to the goal of understanding the structure of the HIV trimer, an envelope glycoprotein that the virus uses to establish contact with the human cells it will infect, according to new research published last week. The new findings clash with previously published results, prompting calls for the earlier paper to be retracted.

Three late-October papers—one published in Nature Structural & Molecular Biology by researchers at the National Cancer Institute and two Science papers written by a collaborative group from The Scripps Research Institute in La Jolla, California, and Weill Cornell Medical College in New York City—arrive at similar structures for the elusive protein, which could be a valuable vaccine target. This latest understanding of the protein, which holds that a spike-like structure is surrounded by three helices that help swivel the spike into position to hook on to a target human cell, contravene a previous model that posited the presence of a cavity at the center of the spike. According to some structural biologists, that earlier work—published in a May 2013 issue of PNAS by Youdong Mao of the Dana-Farber Cancer Institute in Boston, Massachusetts, and colleagues—should now be retracted. “I give no weight to the previous paper,” Marin van Heel of Leiden University in the Netherlands told Nature.

Since researchers began questioning Mao’s results in June, he and his colleagues have failed to provide raw data or detailed methods to their critics. “They are throwing up a lot of smoke screens, not releasing the data, and not retracting the paper,” van Heel continued. “The next step is due with these new structures.”

The recent findings add to an expanding body of work surrounding the solving of structures related to HIV’s ability to infect human cells. Earlier this year, researchers in China produced a 3-D portrait of CCR5 a protein on the surface of white blood cells that is instrumental in HIV’s incursion into host cells.

The new papers describing the structure of the HIV trimer represent independent corroborations for the structure, and Mao told Nature he welcomes the fresh data, but noted that they do not invalidate his results. “A full understanding of the [glycoprotein] structure likely will require scientist to capture all different ‘snapshots’ under different contexts and ‘connect’ them together,” he wrote in an e-mail to Nature.
Gut Microbes May Impact Autoimmunity

Researchers show that the prevalence of one genus of bacteria correlates with the onset of rheumatoid arthritis.

By Abby Olena | November 6, 2013

The more scientists learn about the gut microbiome, the more roles it seems to play. New evidence from researchers at the New York University (NYU) School of Medicine, the Memorial Sloan-Kettering Cancer Center in New York, and the Harvard School of Public Health in Cambridge, Massachusetts, shows a correlation between onset of rheumatoid arthritis (RA) with the prevalence of a certain microbe—Prevotella copri. The work was published this week (November 5) in eLife.

“It’s been suspected for years and years . . . that the development of autoimmune diseases like arthritis is dependent on the gut microbiota,” Diane Mathis, a professor of microbiology and immunobiology at Harvard Medical School in Boston, Massachusetts, who was not involved in the work, told ScienceNOW. “It’s a very striking finding,” she added.

The researchers sequenced bacterial genes in 114 fecal samples from patients who had recently been diagnosed with RA, patients who had been treated for RA, patients with a different type of autoimmune arthritis (psoriatic), and healthy controls. They found P. copri in 75 percent of the samples from patients who had just been diagnosed with RA, but only in 21 percent of samples from healthy controls, 38 percent of samples from patients with psoriatic arthritis, and in less than 12 percent of samples from patients who had been treated for RA. Then the researchers compared P. copri DNA from several of the samples from newly diagnosed RA patients and controls and found that P. copri strains from recent-onset RA patients had fewer genes to metabolize purines and vitamins. The team also inoculated mice with P. copri and showed that the bacteria not only colonized their guts, but also seemed to make the rodents more susceptible to inflammation.

“That they were able to associate one bacterium with one pathology is remarkable,” immunologist Yasmine Belkaid of National Institute of Allergy and Infectious Diseases in Bethesda, Maryland, who did not participate in the work, told ScienceNOW.

“At this stage, however, we cannot conclude that there is a causal link between the abundance of P. copri and the onset of rheumatoid arthritis,” coauthor Dan Littman, a professor of immunology at NYU Langone Medical Center, said in a statement. “We are developing new tools that will hopefully allow us to ask if this is indeed the case.”

Thwarting Persistence

Researchers show that activating an endogenous protease can eliminate bacterial persisters.

By Abby Olena | November 13, 2013

Bacterial antibiotic resistance is widespread and presents ongoing challenges in treating infections. A less widely known—but equally challenging—problem is antibiotic tolerance, where bacteria called persisters are not sensitive to antibiotics because the drugs’ targets are inactive in these dormant cells. Kim Lewis’s group at Northeastern University in Boston, Massachusetts, had previously shown that persisters give biofilms their drug-tolerant properties. Now, Lewis and his colleagues have eradicated a biofilm infection with a derivative of the drug acyldepsipeptide called ADEP4, and showed that it activates a protease in persister cells, causing them to self-digest. Their work was published in Nature today (November 13).

“From a treatment perspective or a translational research perspective, [this study is] probably one of the most profoundly important advances that I’ve seen in the field for more than a decade,” said Garth Ehrlich, who is a professor of microbiology and immunology at the Drexel University College of Medicine in Pennsylvania, and was not involved in the work. “This is really transformative,” he continued. “The fact that they were actually able to eradicate a biofilm infection in deep tissue is essentially unheard of.”

Based on previous work in actively dividing cells, Lewis’s team predicted that ADEP4 would activate ClpP—a protease that typically identifies and degrades misfolded proteins—in dormant persister, too. The researchers showed that in a stationary population of methicillin-resistant Staphylococcus aureus (MRSA), treatment with ADEP4 caused widespread, non-specific degradation of proteins, suggesting that activation of ClpP led to the cells’ self-digestion. ADEP4 alone did not kill all bacterial cells, which the authors suggested is likely due to mutations in ClpP that render those cells resistant to the compound. But ClpP mutants were susceptible to other antibiotics, so combinations of ciprofloxacin or rifampicin with ADEP4 led to a decrease in S. aureus to below the level of detection in vitro, in both biofilm and stationary populations.
The ultimate test of ADEP4’s efficacy came when the scientists moved into a mouse model of chronic *S. aureus* biofilm infection. They showed that ADEP4 and rifampicin together eliminated the biofilm from muscle tissue of the mice to the limit of detection. “It’s a terrific weapon to kill cells,” said Lewis, and “it does especially good job killing those that conventional antibiotics do not.”

The paper is an “important advance in therapeutics against persistent infections,” said Daniel Wozniak, a professor of microbiology and microbial infection and immunity at Ohio State University, who did not participate in the work. “The concept of exploiting or rewiring a pathway in a bacteria, which is normally meant for normal homeostasis [and] normal cellular biology . . . is a new therapeutic twist,” he added.

“The beauty of this system is it seems to have a mechanism for going directly after the persister population—they basically chew themselves up from within,” explained microbiologist Lynn Hancock, an associate professor of molecular biosciences at Kansas State University, who was not involved in the work. Both hyperactivating a housekeeping gene and using ADEP4 in combination with other antibiotics represents “a really novel way of thinking about treating infection,” he continued.

Work still remains before this treatment reaches the clinic. Lewis explained that ADEP4 seems to work well for gram-positive pathogens like *S. aureus*, but is too large to penetrate the outer membrane of gram-negative bacteria, like *Pseudomonas aeruginosa*, which is often responsible for persistent biofilm infections in the lungs of cystic fibrosis patients. Lewis’s group is currently collaborating with a team of chemists to develop and identify other small molecules that could activate proteases in different bacteria and to produce ADEP4 variants with higher solubility and better activity. “There’s room to produce better analogs,” he said.

“An even harder test” of the treatment, and one that Ehrlich would really like to see, “would be to use an animal model [with] an implantable device and see if their therapy can eradicate a biofilm” on an implant, like a screw or an artificial joint, he said. “If they can eradicate that, that’s the holy grail.”

Wozniak pointed out that looking at other antibiotics besides rifampicin in combination with ADEP4 in vivo would be helpful for translating the findings. “In reality, clinicians have few choices to treat tolerant *Staph aureus* infections,” he said, “so it’d be nice to know how broadly applicable this will be.” He also would like to know how effective ADEP4 is for treating polymicrobial infections.

Despite the remaining challenges, experts believe the team’s findings will still have an impact. Gram-positive pathogens “are a worthy target to go after because those organisms are the leading cause of hospital-associated infection, and they’re often tolerant and resistant to a variety of antibiotics,” Hancock said. “Coming up with new ways and strategies to treat them is . . . a cool thing.”

“Over the past 15 years or so, we’ve established that most of these chronic infections are biofilms,” said Ehrlich. “For the first time, with this paper, we’re getting close to developing treatments. It certainly looks hopeful.”


**HIV’s Stealth Revealed**

**HIV-1 evades the immune system with a protein shield, which can be lifted.**

By Ed Yong | November 21, 2013

HIV-1, the virus that causes AIDS, is renowned for its ability to escape the immune system. A new study shows that its sneaky talents depend on the capsid protein that makes up the virus’s outer coat.

Xavier Lahaye and Takeshi Satoh from the Institut Curie in Paris showed that HIV-1 uses its capsid to cloak its DNA from dendritic cells—sentries that detect incoming threats and mobilize the immune system. The researchers also managed to lift this immunity cloak by mutating the capsid.

“By playing with the capsid, we made an HIV-1 that does not replicate but can stimulate an immune response,” said Nicolas Manel, who led the study. “We could imagine modifying the virus and using it as a vaccine.” The results are published today in *Immunity*.
“It’s a very nice piece of work,” said Greg Towers from University College London, who was not involved in the study but recently published similar results. “I think we’re all on the same page, and there’s a paradigm shift in our understanding of how HIV interacts with the immune system. We used to think that the capsid came off the virus when it entered the cell, and its job was done. It turns out that it also protects the virus’s DNA from being seen.”

Dendritic cells can recognize viruses that infect them with a range of sensor molecules, and they instruct other parts of the immune system to target these threats. “They are the key orchestrators of the immune response,” said Manel. “They’re first to detect pathogens and direct what happens after that.”

Back in 2010, Manel’s team showed that dendritic cells can detect HIV-1 under some circumstances, although this is atypical because the virus does not usually infect the cells efficiently. By contrast, a related virus called HIV-2 does fully infect dendritic cells and triggers a strong immune response. This partly explains why HIV-1 can evade the immune system and cause AIDS, while HIV-2 does not.

The team wanted to find out how the dendritic cells were detecting the viruses, and which parts they were sensing. They began by tweaking HIV-2’s capsid, and found that changing a single amino acid produced a virus that cannot infect dendritic cells but can still be detected by them. This implied that detection takes place at an early part of the virus’s life cycle, before it has a chance to make copies of itself.

Once HIV-1 and HIV-2 enter cells, they copy their RNA genome into a DNA molecule that is integrated into the genetic material of their host. The team found that this DNA is what the dendritic cells detect. “You need synthesis of DNA for detection, but you don’t need the steps after,” said Manel.

The team also identified the sensor molecule—a protein called cGAS, which was discovered by investigators at University of Texas Southwestern Medical Center in Dallas this year. When cGAS detects viral DNA in dendritic cells, it triggers a chain of molecular signals that marshals an immune response.

That explains HIV-2, but how does HIV-1 evade detection? The team showed that the capsid somehow masks the viral DNA, preventing cGAS from sensing it. This allows the DNA to integrate into the host’s genome, be copied, and produce new virus particles.

The team managed to peel back this invisibility cloak by mutating the capsid, leaving HIV-1’s DNA exposed and visible to the cGAS sensors. This suggested that similar modified viruses may, after much development, form the basis of a vaccine. This modification happens naturally to an extent—people who are infected with HIV-1 live longer if they had previously been infected with HIV-2, and are less likely to proceed to develop AIDS.

Alternatively, “we can think about small chemical compounds that stimulate this pathway and mimic the immune response,” said Manel.

“The study highlights the importance of studying HIV-2 [to increase] our understanding of underlying mechanisms important for HIV-1 pathogenesis,” added Joakim Esbjörnsson from Lund University in an e-mail.


New, more aggressive HIV strain found in West Africa
By Agence France-Presse
Thursday, November 28, 2013 18:55 EST
A new and more aggressive strain of HIV discovered in West Africa causes significantly faster progression to AIDS, researchers at Sweden’s Lund University said Thursday.

The new strain of the virus that causes AIDS, called A3/02, is a fusion of the two most common HIV strains in Guinea-Bissau. It has so far only been found in West Africa.

“Individuals who are infected with the new recombinant form develop AIDS within five years, and that’s about two to two-and-a-half years faster than one of the parent (strains),” said Angelica Palm, one of the scientists responsible for the study based on a long-term follow-up of HIV-positive people in Guinea-Bissau.

Recombinant virus strains originate when a person is infected by two different strains, whose DNA fuse to create a new form.

“There have been some studies that indicate that whenever there is a so-called recombinant, it seems to be more competent or aggressive than the parental strains,” said Palm of the study published in the Journal of Infectious Diseases.

The strain was first discovered by the Swedish team in Guinea-Bissau in 2011.

According to researchers, the speed with which A3/02 leads to people falling ill from AIDS does not impact on the effectiveness of medication on infected individuals.
“The good news is that as far as we know the medicines that are available today are equally functional on all different subtypes of variants,” Palm said.

The study warns that such recombinants may be spreading fast, especially in regions with high levels of immigration, such as Europe or the United States.

“It is highly likely that there are a large number of circulating recombinants of which we know little or nothing,” said Patrik Medstrand, professor of clinical virology at Lund University.

Some 35.3 million people around the world are living with HIV, which destroys the immune system and has caused more than 25 million deaths since AIDS first emerged in the early 1980s, according to the World Health Organisation.

Existing treatments help infected people live longer, healthier lives by delaying and subduing symptoms, but do not cure AIDS. Many people in poor communities do not have access to the life-giving drugs, and there is no vaccine.

**Micronutrient supplements delay HIV disease progression for patients with higher CD4 cell counts**

Michael Carter  
Published: 28 November 2013

Long-term micronutrient supplementation delays HIV disease progression in patients with higher CD4 cell counts, results of a study published in the *Journal of the American Medical Association* shows. The research was conducted in Botswana and involved antiretroviral-naïve patients with CD4 cell counts above 350 cells/mm$^3$. **Supplementation with vitamins B, C and E plus selenium significantly slowed the rate of HIV disease progression.**

“A single supplement providing the combination of multivitamins with B vitamins, vitamins C and E, and selenium, as compared with placebo, administered early in HIV disease, reduced the risk of reaching a CD4 cell count of 250 cell/mm$^3$ or less in two years,” write the authors. “The benefit was also evident with an earlier end point of a CD4 cell count of 350 cells/mm$^3$ or less, which is the current standard for starting ART [antiretroviral therapy] in Botswana.”

Vitamins B, C and E support immunity and selenium has been shown to reduce HIV replication in test tube studies.

Earlier research showed that micronutrient supplements slow the rate of HIV disease progression in patients with advanced HIV disease. Supplementation with selenium alone **has also shown benefits in a randomised trial.** An international team of investigators wanted to see if supplementation was also beneficial for patients with higher CD4 cell counts, and in particular, whether it might delay the need for antiretroviral treatment in this group of people with HIV.

They designed a double-blind, placebo-controlled study involving 878 antiretroviral-naïve patients in Botswana. All were infected with HIV-1 subtype C, had a CD4 cell count above 350 cells/mm$^3$, were of normal body weight and were asymptomatic.

The patients were randomised into four study arms:
- Placebo.
- Multivitamins (B, D and E) alone (thiamin, 20 mg; riboflavin, 20 mg; niacin, 100 mg; vitamin B$_6$, 25 mg; vitamin B$_{12}$, 50 μg; folic acid, 800 μg; vitamin C, 500 mg; and vitamin E, 30 mg).
- Selenium alone (200 μg per day)
- Multivitamins plus selenium.

Micronutrients were taken daily.

The study was conducted between 2004 and 2009 and follow-up lasted 24 months. The main outcome was progression to a CD4 cell count below 250 cells/mm$^3$. Secondary outcomes were progression to a combined outcome of a CD4 cell count below 250 cells/mm$^3$, an AIDS-events or AIDS-related death, or a CD4 cell count below 350 cells/mm$^3$.

Patients had a median CD4 cell count of 420 cells/mm$^3$ at baseline and a third had a count above 500 cells/mm$^3$. Median baseline viral load was approximately 15,000 copies/ml.

Outcomes clearly showed the benefits of supplementation.

Compared to patients in the placebo arm, individuals who received therapy with multivitamins alone or multivitamins plus selenium were significantly less likely to experience a fall in their CD4 cell count to below 250 cells/mm$^3$ ($p = 0.04$ and $p = 0.02$, respectively). Selenium supplementation alone did not reduce the risk of the CD4 cell count falling below this threshold.
For secondary outcomes, only supplementation with multivitamins and selenium reduced the risk of progressing to the composite outcome of CD4 cell count below 250 cells/mm$^3$, AIDS-events or AIDS-related death ($p = 0.04$). Similarly, only combined therapy with the multivitamins and selenium were shown to reduce the risk of progression to a CD4 cell count below 350 cells/mm$^3$ ($p = 0.04$).

Multivariate analysis controlling for age, sex, and baseline characteristics including CD4 cell count and viral load confirmed that supplementation with multivitamins plus selenium reduced the risk of CD4 cell count falling to below 250 cells/mm$^3$ ($p = 0.01$) and also the risk of progressing to secondary outcomes ($p = 0.03$). Neither multivitamins alone nor selenium alone reduced the risk of disease progression.

**Supplementation had no effect on viral load.**

No adverse events were related to any of the study medication, and 90% of patients had high levels of medication adherence (at least 96% of doses taken).

“These results…demonstrate the effectiveness and safety of multivitamins and selenium when administered together in a single supplement in sowing HIV disease progression in the early stage of disease prior to ART,” comment the investigators. “The evidence from our study presented herein supports the provision of low-cost supplementation with multivitamins combined with selenium for HIV-infected individuals in early stages of the disease who are ART-naïve to prolong adequate immune response and prevent AIDS-defining conditions.”

**Reference**

Baum MK et al. *Effect of micronutrient supplementation on disease progression in asymptomatic, antiretroviral-naïve, HIV-infected adults in Botswana: a randomized clinical trial.* JAMA 310: 2154-63, 2013. (Full text article freely available).

**Sweden divided over criminalising HIV unprotected sex**

By Erik Fau (AFP) – 19 hours ago

Stockholm — When Lina Afvander got her HIV diagnosis, it came with a set of prescriptions and a disclosure obligation, which legally requires HIV-positive people in Sweden to reveal their status before having sex.

"My strategy is that I don't expose myself to this situation that often, I just don't have that much casual sex," the 35-year-old Swede said.

"I wait until I can trust the other person, until I know it's someone who can handle this."

Quite a bitter pill to swallow in itself, an HIV diagnosis in Sweden also brings with it the risk of criminal prosecution if the legal responsibility, intended to prevent the spread of the virus, is not met. But Sweden's use of criminal law in HIV cases, one of the most stringent in the world, is now being challenged as experts argue that people on effective anti-retroviral therapy have an extremely low risk of transmitting the disease.

The debate around HIV criminalisation has been highlighted by the run-up to World AIDS Day on Sunday.

In Sweden a failure to comply with the HIV disclosure obligation, followed by unprotected sex, can result in charges of attempted aggravated assault and a prison sentence, even if the virus is not transmitted.

Thus people with HIV cannot legally have unprotected sex in any circumstance—even if they reveal their HIV status and have the full consent of their uninfected partner, since a person cannot consent to an assault.

Around 6,500 people are HIV-positive currently in Sweden and 90 percent of them are on an effective anti-retroviral therapy.

According to the 2010 Global Criminalisation Scan Report, published by the Global Network of People Living with HIV, Sweden has the highest rate of HIV-related convictions in the world, with 6.12 sentences per 1,000 people living with the virus, 60 times more than France and 24 times more than the United States.

But last October things began to change.

A court of appeal cited a report from the National Centre for Communicable Diseases Control (SMI) to acquit an HIV-positive man sentenced to one year in prison for having unprotected sex with four women who never contracted the virus.

"Patients who are on effective anti-retroviral therapy are in essence non-infectious when it comes to sexual contact," said Jan Albert, professor of infectious disease control at Karolinska Institutet and one of the authors of the report.
The report argued that prosecution was wrong as the risk of transmitting the virus is extremely low. It debunked the myth that HIV-positive people who are aware of their condition and take medication are the ones spreading the disease.

"If you have protected sex with an HIV-positive untreated person, the risks of transmission are higher than if you have unprotected sex with an HIV-positive person on effective therapy," said Marielle Nakunzi, a lawyer at the Swedish Association for Sexuality Education, commenting on the report, which needs further research to be conclusive.

"The evidence is still not 100 percent proved in all settings," Albert said, referring for example to cases where the presence of other sexually transmitted diseases like chlamydia or gonorrhoea can increase the risks of HIV transmission.

Sweden, which is often praised in international forums for its gender equality policies, does not comply with the recommendations of the UNAIDS programme, which advocates that "non-disclosure of HIV-positive status and HIV exposure should not be criminalised."

While Swedish rights organisations have criticised the law, the legal status of HIV may mirror general beliefs that are widespread among Swedes.

According to the latest SMI report which assesses attitudes towards HIV since 1987, 40 percent of Swedes in 2011 believed HIV-positive people should avoid having sex altogether.

"These figures are insane," Nakunzi said.

"We have this tendency to think: if you have HIV, you have to tell me or the police will come and get you."

Matthew Weait, professor of law and policy at Birbeck College and an expert on the impact of law on HIV prevention, also thinks the regulation has to do with Swedish mentality.

"Swedes report that they are more likely to trust a stranger than distrust him or her, so if there is a perceived violation of trust, the response will be more extreme," he said.

"And if one couples this with the high levels of trust which Swedes have in their legal system, it is perhaps unsurprising that they will readily turn to the law for a solution."

The country's two main political parties have yet to take a clear stand on the matter.

Swedish organisations campaigning for a change in HIV criminalisation, such as Hiv-Sverige or the Swedish Association for Sexuality Education, see the acquittal in October as a promising breakthrough.

For Afvander, directly affected by the law, disclosure should be left as a moral obligation.

"I think there is a lot you should tell before you have sex, but it should not be legislated," she said.

"If I were the other one, I'd like to know, so I guess I have a moral disclosure obligation."

**How Will AIDS Be Eradicated?**

**In the U.S. and around the world, there are obstacles to prevention and treatment. How will they be overcome?**

**Every Nation Needs Affordable Drugs**

Michel Sidibé is the executive director of U.N. AIDS.

**Updated** November 28, 2013, 7:19 PM

As World AIDS Day approaches and we gather to remember friends and family lost to AIDS, we can also look with hope to the future and a generation free from H.I.V.

It is only because of the great strides of recent years that we can even talk of the end of the AIDS epidemic. New H.I.V. infections have fallen by 33 percent, and new infections among children have fallen by 52 percent in the last decade. AIDS-related deaths have fallen by nearly 30 percent since the peak in 2005. And nearly 10 million people in low- and middle-income countries now have access to antiretroviral therapy.

Antiretroviral therapy reduces the risk of transmission to below 5 percent. Everyone in the world needs access to these treatments.

We need to be determined in achieving universal access to H.I.V. services to be able to realize the full potential of the tools we have available. Research is showing that antiretroviral therapy not only keeps people living with H.I.V. alive and well, but also has important preventive benefits – reducing the risk of H.I.V. transmission to below 5 percent.
By pursuing innovative preventive strategies and in making the full range of lifesaving H.I.V. services available to everyone in need – including people most affected by the epidemic such as sex workers, men who have sex with men, and people who inject drugs – we will start to realize the end of AIDS.

To get there, we need to create a greater demand for testing so that everyone knows their H.I.V. status. Treatment programs need to be enhanced. And all efforts must be made to reduce the price of medicines so that universal access is achievable for all nations.

Although incredible progress has been made in reducing prices, especially for the first-line medicines that are used in initial treatment, many countries still struggle to procure medicines at affordable prices. Second- and third-line medicines, which are used when patients develop resistance to the initial treatments, still have prohibitive prices for the great majority of patients who need them in the developing world.

This is why it is important to manage intellectual property rights with public health considerations in mind, allowing developing countries to establish their own capacities to produce high-quality medicines and, where it is not feasible, to increase their negotiating power with pharmaceutical companies.

No one should become sick or die because they lack access to medicines or services. The response to H.I.V. is about human rights and human dignity. On the journey to the end of AIDS, we cannot allow anyone to be left behind.

Researchers Block Replication of AIDS Virus
Nov. 28, 2013 — A multidisciplinary team of scientists from Spanish universities and research centres, among which is the University of Valencia, has managed to design small synthetic molecules capable of joining to the genetic material of the AIDS virus and blocking its replication.

This achievement has been obtained for the first time in the world by a group of researcher led by José Gallego from Universidad Católica de Valencia "San Vicente Mártir." The University of Valencia, the Príncipe Felipe Research Centre, and the Instituto de Salud Carlos III have participated. The work has been recently published by Angewandte Chemie International Edition.

The newly designed synthetic molecules inhibit the output of genetic material of the virus from the infected cell nucleus to the cytoplasm, thus the virus replication is blocked and avoids the infection of other cells.

The genetic material of the AIDS virus, or HIV-1, is formed by ribonucleic acid (RNA), and encodes several proteins that allow it to penetrate the human cells and reproduce within them. The new virus inhibitors, called terphenyls, developed by this group of scientists, were designed by computer to reproduce the interactions of one of the proteins encoded by the virus, the viral protein Rev.

In this way, the terphenyls join Rev’s receptor in the viral RNA, preventing the interaction between the protein and its RNA receptor. This interaction is necessary for the virus genetic material to leave the infected cell nucleus and, thus, it is essential for the survival of HIV-1. The fact that the terphenyls block the virus genetic material output of the cell prevents the infection of other cells.

This discovery is the result of a close collaboration between three research groups throughout several years. Thus, the scientists of the Universitat Católica de Valencia were in charge of the computational design and verified experimentally that the terphenyls were capable of joining the Rev receptor in the viral RNA and inhibit the interaction between this RNA and the protein.

For its part, the molecules were synthesised in professor Santos Fustero's organic Chemistry laboratory in the Príncipe Felipe Research Centre and the University of Valencia. Also, through experiments with cells infected by the virus, the group of José Alcamí in the Instituto de Salud Carlos III demonstrated that the inhibitors block the replication of the HIV-1 and inhibit the function of the Rev protein, confirming this way the validity of the models generated by computer.

Traditionally, pharmaceutical companies have focused on the development of medicines that act on target proteins, as the approach to the receptors made out of RNA is considerably complex.

Although several natural antibiotics act at the bacterial ribosomal RNA level, up to now designing by computer a new synthetic chemical entity capable of joining RNA target and have a relevant pharmacological effect was not possible. The terphenyl structures identified in this research could open new ways to approach other therapeutic targets formed by nucleic acids.

On the other hand, the infection by HIV affected 34 million people worldwide in 2010, according to the World Health Organisation (WHO). The emergence of resistance to the current antiretroviral therapies and the lack of an effective vaccine highlight the necessity of identifying the new medicines that
act on other virus targets. Rev protein constitutes one of this alternative targets, but so far they it has not been possible to develop antiviral agents based in their inhibition.

The results of this research have been the objectives of a patent application, and the three laboratories involved in the research keep their collaboration with the objective of improving the pharmacological properties of new Rev inhibitors.

**Journal Reference:**


---

**Mother-To-Child HIV Transmission in Gipuzkoa Reduced Significantly Over Last 25 Years**

Nov. 29, 2013 — Miren Apilánez, researcher in the Department of pediatrics of the UPV/EHU-University of the Basque Country, has studied the evolution that took place between 1984 and 2011 in pediatric HIV infection in Gipuzkoa. The development of methods to diagnose the disease coupled with increasingly more effective treatments have made it possible to reduce mother-to-child transmission (vertical transmission) from 23.9% to 2.4%, thus virtually eradicating infection in children.

Vertical transmission occurs between mothers infected with HIV and their offspring. This infection can take place at three different moments or phases: during pregnancy, during birth or during breastfeeding, "but in actual fact, the most critical moment is the birth because the child comes into contact with the mother's blood or vaginal secretions," explains the researcher. "Infection is also possible during pregnancy, but it is less likely because the placenta acts as a barrier." As regards breastfeeding, "the moment it became known that it was a way of transmitting the infection, it was contraindicated in developed countries," explains Apilánez.

In the course of the research, Apilánez studied 239 children of HIV-infected mothers and born between 1984 and 2010 in Gipuzkoa, and their mothers. 30 children were infected by the virus, and 209 seroreverted during the first months of life.

**Four periods, four scenarios**

Throughout this time there is proof of an evolution in various aspects relating to the infection in Gipuzkoa: the transmission rate itself, the implementing of diagnostic and therapeutic methods in mothers and children, or the channel through which mothers acquire the HIV infection.

So Apilánez has established four periods in the course of time, defined mainly by the implementing of diagnostic measures and therapies. The first period was the one between 1984, when the first child was diagnosed, until March 1994, and Apilánez defines it as "the period of few resources, characterized by the absence of effective therapies."

In March 1994 the results of the ACTG076 protocol were published worldwide showing that the administering of the first antiretroviral drug known as AZT during pregnancy and birth reduced vertical transmission considerably. The protocol stated that it had to be administered during pregnancy in order to improve the immunovirological situation of the mother, who would reach birth with an undetectable viral load and, therefore, with a minimal risk of passing the infection onto the child. The treatment is completed with intrapartum therapy as well as therapy for the newborn during the first 45 days. After starting this treatment in pregnant women infected with HIV in Gipuzkoa (second period), it was seen that within three years transmission fell from 25% to 8%. "However, children continued to become infected because there were women who did not know, while they were pregnant, that they had been infected, and therefore did not receive any treatment," pointed out the researcher.

In view of these results it was in June 1997 that the HIV early detection protocol was set up for pregnant women in Gipuzkoa (third period); in other words, when it was established that all pregnant women should undergo the HIV test to rule out the infection. "This was crucial, because many women who were unaware that they were infected were detected, although the entire population was not reached," says Apilánez. In this period, too, the combined therapy or highly active antiretroviral therapy (HAART) also began to be used. This therapy is characterized by the combining of various drugs that attack the virus at different moments in the infection process. That way the treatment is much more effective.

The fourth and final period began in March 2000. It was characterized by considerable work to spread the guidelines in early detection and treatment of HIV infection in pregnancy through the Professional Associations of Doctors and Nurses in Gipuzkoa; it was finally possible to detect all the pregnant women infected and therefore achieve 100% therapeutic coverage.
The prevention strategies in the vertical transmission of HIV have enabled the transmission rate to be cut from 23.9% in the first period down to 2.4% in the last one. Highly active antiretroviral therapy during pregnancy has emerged as the most significant protection factor to tackle the transmission of HIV infection between mothers and children; that way, the mother-to-be reaches the birth with a correct immunovirological situation, in particular with an undetectable viral load and therefore a minimal risk of transmitting the infection to the child.

However, the researcher insists that these measures need to be carried out 100% in order to be effective and recalls that even in this way there are still a number of not very well-known factors that hamper the total eradication of the mother-to-child transmission of the infection.