

WHEN THE CURE CAUSES THE ILLNESS

Clostridium difficile is a bacterium that attacks the gastrointestinal tract of patients using antibiotics. Researchers in Colombia are currently on the trail of ways to mitigate the harsh impact of this microbe known for its strong resistance.

By José Alejandro González T.
Photos Milagro Castro

Imagine someone who turns to antibiotics to avoid a possible infection after being bitten by a cat or picking up a scissor cut, little knowing that this innocent measure will activate powerful bacteria that could cause quite profound damage within his or her organism. Although it sounds quite unlikely, this is exactly how the *Clostridium difficile* bacterium functions.

Over the last 15 years, these bacteria have caused problems and serious injuries in the GI tracts of patients who had to take antibiotics, resulting in a major epidemiological impact in certain regions of the planet. “*Clostridium difficile* is a highly interesting bacterium in genetic terms, but one barely figuring in studies from Latin American countries. A number of patients began having gastric problems when they used antibiotics, but no one knew why,” explains Dr. Juan David Ramírez, leader of this research, and director of the Microbiological Research Research Group (GIMUR) within the Universidad del Rosario’s Faculty of Natural Sciences and Mathematics.

Focusing on this issue, Dora Inés Ríos, who worked for 30 years as Professor of Microbiology at the Universidad del Rosario, carried out a literature review in 2014 alongside Prof. Ramírez, and this led to the decision to investigate this high-impact pathogen under the auspices of the University. In 2015, the GIMUR team was joined by Claudia Marina Muñoz (PhD student in Biotechnology at the National University of Colombia), who thus began her doctoral thesis. The work was to become the first study in Colombia on this microorganism, with the team identifying the infection frequency of *Clostridium difficile* nationwide, and also managing to describe factors that might be associated with the serious impact caused by certain strains.

According to Muñoz, contamination by *Clostridium difficile* can occur through contact with its spores, which are resistant to different disinfectants and can be present in hospital centers and surgical zones. Contagion generally occurs through the oral route. The bacterium’s spores inhabit the GI tract, where they remain in an inactive form and in harmony with the other microorganisms present. “This bacterium can even be found in seven percent of completely healthy individ-

THE BACTERIUM CAN BE TRANSMITTED THROUGH CONTACT WITH ITS SPORES, WHICH ARE RESISTANT TO DIFFERENT DISINFECTANTS AND CAN BE PRESENT IN HOSPITAL CENTERS AND SURGICAL ZONES.

ually healthy individuals.”



uals, who simply do not know they have it," she points out.

Once the person consumes antibiotics, the equilibrium existing between the beneficial organisms is broken, and these bacteria activate, taking on a vegetative state and causing illness, the first symptom of which is relatively benign diarrhea. "When *Clostridium* damage the intestinal barrier, they cause severe problems such as perforations of the colon, pseudomembranous colitis, and toxic megacolon. If the infection is very serious, it can even result in the patient's death," comments Muñoz.

The major problem it involves for our health systems is its powerful resistance to a great number of antibiotics, thus limiting the treatment options and medicines that could be used against it. Only a few antibiotics have been developed to specifically attack the infection caused by this bacterium. Two years ago, it was being treated with Fidaxomicin, an antibiotic that proves efficient but which is not sold in Colombia due to its high price.

A PROJECT WITH OUTCOMES

The project undertaken by the Universidad del Rosario under Ramírez's direction has enjoyed a boost thanks to the participation of co-researchers. These include Manuel Alfonso Patarroyo, professor at the School of Medicine and Health Sciences of El Rosario and director of the Molecular Biology and Immunology Working Group at the Foundation Institute of Colombian Immunology (FIDIC). The research team initially set out to find the frequency of infection in patient samples by using microbiological culture and molecular biology techniques, and by evaluating the genetic diversity of *Clostridium difficile*. They meanwhile managed to isolate the microorganism so as to compile a bacteria bank at the University, from which they began the study of other factors involving this bacterium, such as its complete genome organization and phenotypic traits. This work has allowed the researchers to move ahead with defining the causes of the disease.

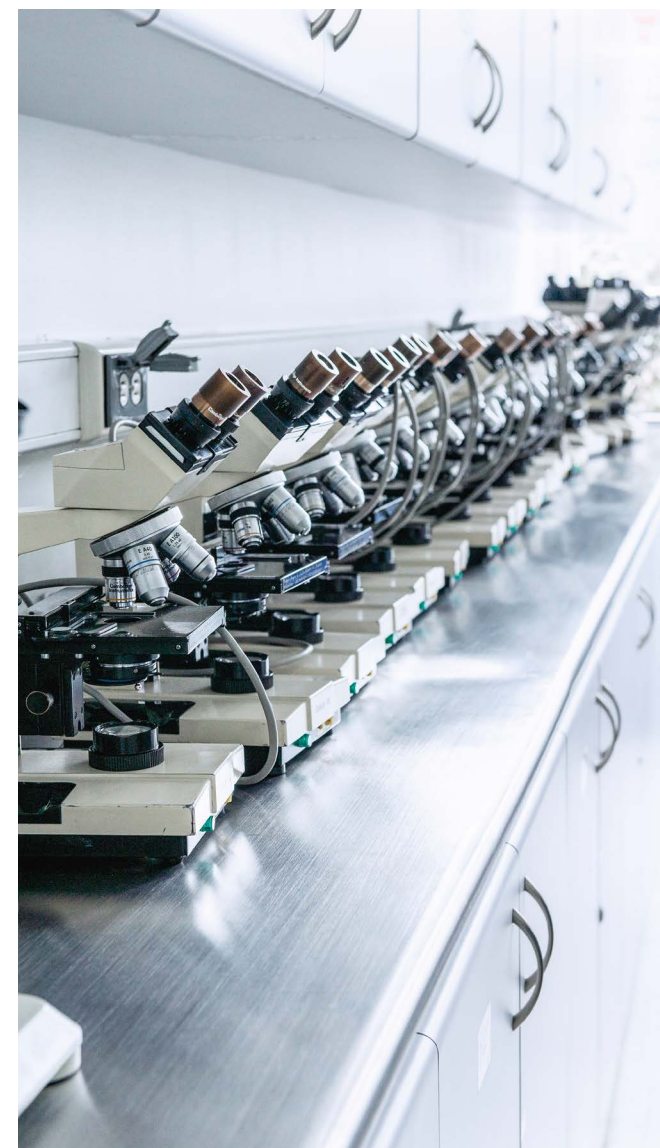
So far, the findings are the result of the analysis of 217 samples collected at both the Hospital Universitaria Mayor - Méderi, Bogotá—thanks to the participation of Drs. Juan Manuel Pardo, Claudia Birchenall, and Dario Pinilla—and the Shaio Clinic Foundation, with help from Dr. Diego F. Josa. Two groups of patients suffering from diarrhea (the main symptom caused by *Clostridium difficile*) were thought to have acquired the nosocomial infection, the Intensive Care Unit (ICU) being



the most affected, and despite belonging to the community, these were thought to be suffering from the infection. "After evaluating the frequency of infection in those analyzed, we found that 56% of the patients with diarrhea in the ICU showed positive for the bacteria," explained Muñoz.

"Besides finding it in patients who were treated in hospitals and who could have acquired the bacteria through other illnesses, a high frequency of contagion was also presenting outside medical centers. Patients were arriving at the Emergency Room with diarrhea, with an infection that was probably picked up outside this environment and which showed up positive for *Clostridium*," added Muñoz.

According to the researcher, *Clostridium difficile* is a very diverse organism that has a large number of different pop-



ulations or strains, of which some are more associated with causing infections than others. In this case, the illness begins specifically when the bacteria produce certain toxins.

In broad terms, the bacterium is able to produce two main toxins (A and B), and a third called *binary*. It should be pointed out that not all strains produce all the toxins, but that there is a range of possibilities. "Of the more than 40 combinations described for *Clostridium*, the most dangerous are those that produce all the toxins, since these are what damage the GI tract," she explains.

What has been found, therefore, is a high frequency in Colombia of *Clostridium* positives with infection-causing toxins, as well as combinations of these toxins that had not been reported in other parts of the world.

The project has now reached the stage of analyzing whole genome sequencing data using high-performance techniques in line with the most rigorous way to study microorganisms. This approach has allowed for the identification of genes associated with resistance to antibiotics, atypical organizations



Claudia Marina Muñoz, of the Universidad del Rosario's GIMUR research group, says the bacterium can be found in seven percent of perfectly healthy individuals, who simply do not know they have it.



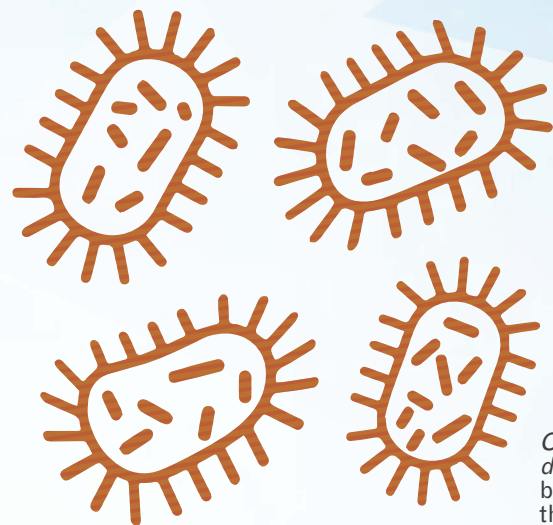
A number of patients began having gastric problems when they used antibiotics, but no one knew why," explains Dr. Juan David Ramírez, researcher at the Faculty of Natural Sciences and Mathematics, and leader of the study.



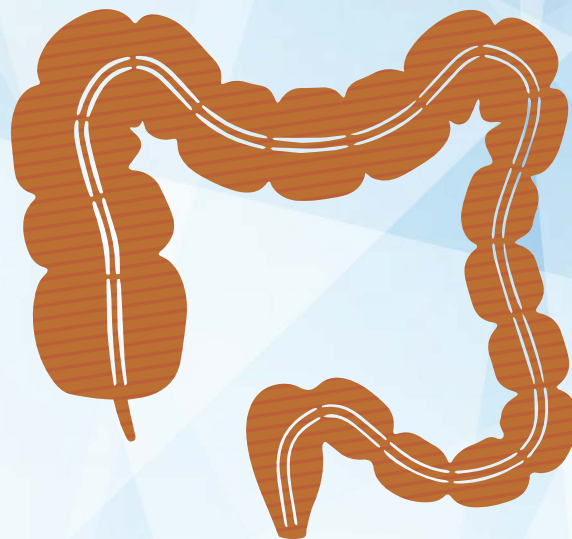
of toxins, and other virulent factors involving *Clostridium* strains circulating in Colombia.

These results enable a diagnostic tool to be created, allowing for early identification of infected Colombian patients and effective management of the illness. The idea is to integrate different strategies for its control, such as not limiting treatment to antibiotics but also reconstituting the balance in the organism, thus enabling control at a biological rather than chemical level. ■

WHEN THE CURE CAUSES THE ILLNESS



Clostridium difficile is a bacterium that attacks the GI tract of patients using antibiotics.



When *Clostridium* damage the intestinal barrier, they cause severe problems such as perforations of the colon, pseudomembranous colitis, and toxic megacolon. If the infection is very serious, it can even result in the patient's death.



Once the person consumes antibiotics, the equilibrium existing between the beneficial organisms is broken, and these bacteria activate.



Infection by *Clostridium difficile* can occur through contact with its spores, which are resistant to different disinfectants and can be present in hospital centers and surgical zones. Contagion generally occurs through the oral route.



The major problem it involves for our health systems is its powerful resistance to a great number of antibiotics, thus limiting the treatment options and medicines that could be used against it.



This bacterium can even be found in 7% of perfectly healthy individuals, who simply do not know they have it.



Nosocomial diseases

Nosocomial illnesses are those the patient acquires while receiving treatment for a medical condition in a health center and situations requiring surgical intervention. *Clostridium difficile* can be present in these environments.

According to a World Health Organization report from 2016, 1.4m people worldwide catch nosocomial infections, and this has become a patient mortality factor. According to the report, between five and ten percent of patients who enter a treatment center acquire a nosocomial infection. In Colombia, the risk of infection related to health treatment is between two and 20 times greater than in more developed countries. A report by the Bogotá Control of Bacterial Resistance Group, carried out across 36 highly-structured institutions in Colombia—29 of them in Bogotá—showed more than 320,000 people have acquired nosocomial infections in the last three years.