May 06, 2019

Microbiology

Sleeping bacteria survive antibiotic treatment and hijack the host immune system

by **Daphne Stapels**[1] | Postdoctoral Research Fellow; **Peter Hill**[1] | Postdoctoral Research Fellow

[1]: Imperial College London, London, UK

This Break was edited by Max Caine, *Editor-in-chief* - TheScienceBreaker

ABSTRACT

When we take antibiotics during a bacterial infection, most of the invading bacteria will be killed. This eases the pressure on our immune system, allowing it to clean up the few live bacteria that remain. Sometimes bacteria succeed in escaping this treatment. We are one step closer in understanding how

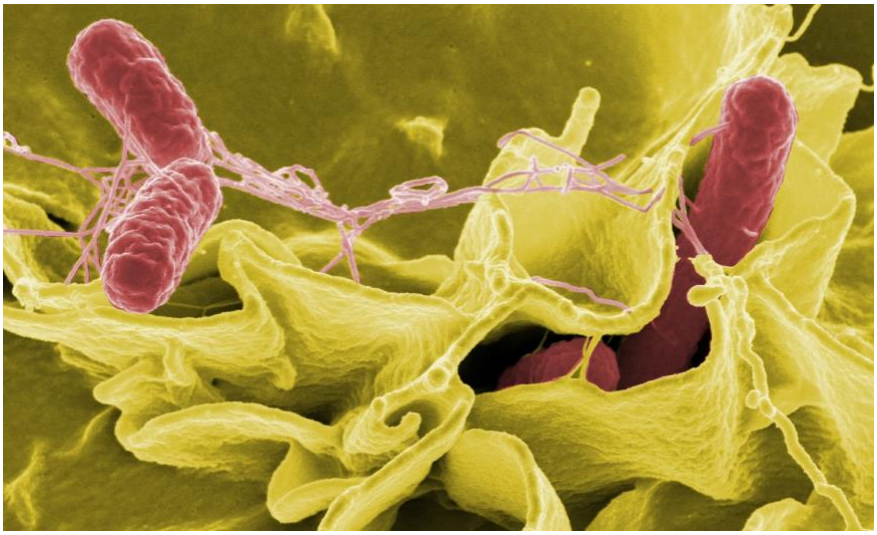

Image credits: Pixabay – CC0

Since the 1940s, it has become easier to treat bacterial infections due to the discovery of antibiotics. These drugs work by corrupting active processes in bacteria, such as the ability to make DNA or proteins. By taking antibiotics when we are infected, we kill most of the invading bacteria. This eases the pressure on our immune system, allowing it to clean up the few live bacteria that remain.

Even though antibiotic treatment is often effective in limiting bacterial diseases, many types of bacterial infections often recur, even after antibiotic treatment. Common examples are recurrent urinary tract infections, ear infections, or most famously, tuberculosis. This recurrence is thought to be caused by the reawakening of 'sleeping' bacteria, called persisters. Persisters are a small subpopulation of bacteria that, while normally sensitive to antibiotics, somehow manage to withstand killing by these drugs by entering into a transient antibiotic-tolerant state (i.e. the 'persister state').

The persister state has usually been thought of as equivalent to a 'sleeping' state for bacteria. That is, bacteria are thought to temporarily shut down their active processes that are often targets for antibiotics. So when bacteria enter into a persister state and shut down these processes, the antibiotics cannot work. Once treatment is completed, these persisters could reawaken and start a new infection episode.

With this in mind, understanding the persister state of bacteria might help us design new treatment strategies to completely eradicate an infection, including persisters.

To better understand the persister state, we used *Salmonella* as a model bacterium. *Salmonella* is best known for causing food poisoning (during infection of the gastrointestinal tract) and typhoid fever (during systemic infection). The primary niche of *Salmonella* during systemic infection is within macrophages - immune cells designed to clear bacteria and viruses. When *Salmonella* is taken up by these macrophages, many of the bacteria react by entering into the antibiotic-tolerant persister state. While antibiotics cannot kill these persisters, one might assume that at least the macrophage, specialised in killing of bacteria, could cope with these supposedly 'sleeping' persister bacteria?

Surprisingly, the answer is no. Somehow, someway, these supposedly 'sleeping' persisters survive. But how?

We hypothesised that possibly these so-called 'sleeping' persisters were not actually sleeping. What if, quietly, persisters were manipulating our immune cells to avoid being destroyed instead?

To test this, we analysed the active processes in both the persister bacteria and in the immune cells. In every living cell, a signature for all active processes going on at that time is represented by the type and amount of specific RNA molecules. We read this RNA signature from both the host immune cells and bacterial persister cells by a technique called RNA-sequencing. Our analysis showed us that macrophages that harbour the persister bacteria initially activate processes aimed at destroying the persisters.

So why are the persisters not killed?

Our analysis also showed that, rather than 'sleeping', the antibiotic-tolerant persisters are in fact very much active. Specifically, in response to the macrophages trying to kill them, the bacteria strike back, synthesizing and releasing small proteins directly inside the macrophages. These small proteins function to weaken the macrophage immune response, considerably dampening its bacteria-killing capabilities.

This is quite different from the persisters that had been discovered in 1944, surviving in laboratory culture medium, which are thought to be in a dormant 'winter sleep' state, and thus completely inactive. Our results now show that persister bacteria are not completely dormant in the human body. Rather, they weaken the defence mounted by our immune cells. This means that when antibiotic treatment is completed, these persisters might have created a much more favourable environment for their own relapse, or even for infection by other bacteria or viruses.

Despite this horrible thought, it also opens up avenues to eradicate persisters. Since persisters that do not manipulate macrophages will be killed by these immune cells, inhibiting this activity of persisters, in theory, would equip our immune cells to efficiently clear hard-to-treat infections once and for all.