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Taming tuberculosis — again

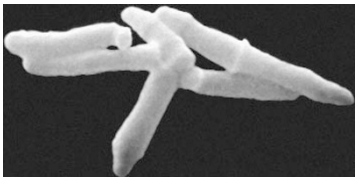

Fig. 1 *Mycobacterium tuberculosis*, the causal agent of the disease tuberculosis. Image courtesy of B. Jacobs, Albert Einstein College of Medicine.

Tuberculosis kills an estimated three million people per year, and its prevalence is increasing at an alarming rate around the world — despite the availability of effective anti-tubercular drugs such as isoniazid and rifampin. Even more frightening is the fact that *Mycobacterium tuberculosis* strains resistant to multiple drugs have been observed in infected individuals, making treatment extremely difficult. In addition, the bacille Calmette-Guérin (BCG) vaccine, which is widely used in developing countries to immunize infants against tuberculosis, is only partially effective. Thus, there is a clear need to develop additional drugs and vaccines to combat tuberculosis. On page 141 of this issue of *Nature Structural Biology* (and discussed in the News and Views on page 94), Ronning *et al.*[1] present the X-ray crystal structure of the *M. tuberculosis* antigen 85c protein, which is highly immunogenic (making it a possible vaccine candidate) and was recently shown to play a major role in a late step of cell wall synthesis[2] (making it a novel potential drug target).

History of tuberculosis

Tuberculosis — known as phthisis (to waste) to ancient Greeks, and as consumption (to consume) to the Western world in the 1700–1800s — has been a threat to humans since antiquity. It has been suggested that *M. tuberculosis* and related bacteria, which can also be responsible for this disease, began to take up residence in the human body ~10,000 years ago, transferred from cattle that had been domesticated. References to tuberculosis-like symptoms and characteristics can be found throughout early writings, such as those of Hippocrates, and tuberculosis is well known for claiming the lives of many famous individuals, including the Bronte family of writers, John Keats, and Henry David Thoreau. In the 1800s, common treatments for tuberculosis included induction of lung collapse (which helped because the bacteria require a high oxygen environment to survive) and isolation of patients in institutions called sanitoria, which were located away from cities where the air was fresh. In 1882 Robert Koch discovered the bacteria that cause tuberculosis, and the era of modern medical treatment began in 1944, when the antibiotic streptomycin was first administered to a person with tuberculosis.

Tuberculosis infection

Approximately one third of the world's population is currently infected with *M. tuberculosis* (see Fig. 1 for an image of these bacteria). However, only ~10% of infected individuals, who are otherwise healthy, will manifest an active infection — characterized initially by coughing, fever, weight loss, night sweats, and shortness of breath. Contrary to popular thought, *M. tuberculosis* is not just a pulmonary disease: infections can occur in many organs, including the brain, kidneys, and heart, but active infection usually does begin in the lungs. 'Droplet nuclei' containing *M. tuberculosis* can be coughed out by person with an active infection and may remain suspended in the air for several hours, thus making tuberculosis a contagious disease.

In most cases, inhaled bacteria are rapidly destroyed by the immune system. However, sometimes *M. tuberculosis* bacteria remain dormant in the lungs, inside macrophages lodged in calcified structures called tubercles, from which the name of the disease derives and which are scar-like structures that result from the body's attempt to isolate the area of infection. Such

dormant bacteria can persist in the body for years, held off by the immune system, and may emerge as an active infection under times of stress or immune compromise. It is not surprising, therefore, that tuberculosis is a major problem in the care and treatment of HIV-infected individuals.

Resurgence of tuberculosis and treatment

Without treatment, the death rate from active tuberculosis is ~40–60%, but that can be reduced to ~10% with proper use of antibiotics. During the 1940s to early 1980s, antibiotic treatment facilitated a decline in the number of tuberculosis cases in countries such as the United States. However, the number of cases in the US began to increase again in the mid 1980s, and tuberculosis rates are also rising around the world. A number of interrelated factors have contributed to the resurgence in the US, including: (i) the HIV/AIDS epidemic (ii) increased immigration from countries with many cases of tuberculosis (iii) increased poverty and homelessness (iv) decline in the health care infrastructure, and (v) poor compliance with treatment regimens.

The current treatment for infection usually involves a combination of drugs: pyrazinamide and ethambutol (or streptomycin), for the first two months of treatment, and isoniazid and rifampin, for at least six months. Unfortunately, the long time course required for effective treatment, the unpleasant side effects of the drugs, and the foibles of human nature often lead to poor compliance, one of the major factors contributing to rising infection rates. Thus, the World Health Organization has been pushing for universal adherence to a process known as directly observed therapy (DOT)[3], through which health care workers counsel patients, monitor their progress, and ensure sure that the swallowing of each dose of medication is observed by a health care worker or volunteer. Obviously, this is a time consuming and expensive enterprise, but one that appears necessary to halt the spread of the disease and the emergence of multi-drug resistant strains.

Research for new therapies and vaccines

The extraordinary measures (such as DOT) that are required to ensure effective treatment of tuberculosis patients, given the limited available therapies and preventive measures, underscore the need for both new drugs and new vaccines. However, the process of developing these new weapons against tuberculosis may be long and difficult — in part because *M. tuberculosis* is not the most tractable research system. A number of factors make *M. tuberculosis* nearly as difficult to study in the lab as it is to treat. For example, like other mycobacteria, it has an extremely resilient cell wall that is comprised of unusual lipids. This coat allows the bacteria to survive inside macrophages, which normally destroy phagocytosed pathogens, and it also apparently renders the bacteria resistant to many drugs. The same coat facilitates clumping of the bacteria, making them difficult to work with and to count. In addition, *M. tuberculosis* bacteria divide only once in a 24 h period under the best of conditions. Thus, a visible colony emerges only after about a month (instead of after ~8 h like an *E. coli* colony). *M. tuberculosis* is also a highly contagious pathogen, and thus labs must be well equipped to handle working with such organisms.

Despite the difficulty of working with the system, *M. tuberculosis* appears ripe for structural and functional genomics initiatives — ones that would not only contribute new folds to the data base, but also identify potential drug and vaccine targets. The complete genome of *M. tuberculosis* was recently sequenced[4], and ~16% was estimated to encode novel proteins that may have mycobacterial specific functions. Moreover, there is clearly much work to be done on the structures of *M. tuberculosis* proteins: in a search of the Protein Data Bank for the word tuberculosis, only 25 structures were obtained as of this printing, whereas in a search for typhimurium (*Salmonella typhimurium*, another pathogen that is a common cause of food poisoning) 123 structures were listed. The antigen 85c story — the latest installment of which is presented on page 141 — will hopefully, in the coming years, show how such basic structural research can aid in the development of an effective treatment for tuberculosis.

1. Ronning, D. *et al. Nature Struct. Biol.* **7**, 141–146 (2000).
2. Belisle, J.T. *et al. Science* **276**, 1420–1422 (1997).
3. World Health Organization tuberculosis site. http://www.who.int/gtb/
4. Cole, S.T. *et al. Nature* **393**, 537–544 (1998)