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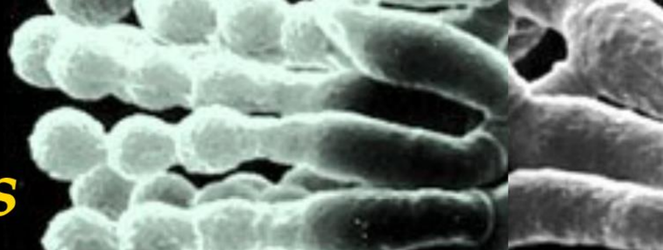
50th Anniversary Articles



Using mighty microscopes to look at microbial machinery

Rachel Harding

The features of microbes which make them harmful to human health can almost always be traced to protein molecules encoded within their genomes (genetic material). Our understanding of how these molecular machines work can be enhanced by knowing their structure; *i.e*. how are the amino acid chains of the protein arranged and organised to let the protein molecules function in the process of microbial infection? The problem is that microbes, and the protein molecules of which they are composed, are very, very small so being able to "look" at them is challenging.

To give some context to the scale of microbes and the protein molecules of which they are composed, consider this analogy. If we expanded a red blood cell to be the size of an adult human, the oxygen-carrying haemoglobin protein it contains would be about the size of a nib of a felt tip pen. In this same scenario, an *E. coli* bacterial cell would be only as large as your foot, and so is much smaller than most human cells. On this same scale, a human being would be almost as tall as the distance from the Earth's surface to the Hubble telescope – about 560 km!

Our eyes cannot see most microbes as they are too small. We can use light microscopes, but they don't give us a huge amount of detailed information. To see the finer features of microbes we can use electron microscopes. Different types of electron microscopes allow us to visualise a range of different samples, from whole organisms to individual molecules of protein. Recently, great advances have been made in the field of cryo electron microscopy (often referred to as cryoEM) and scientists are able to routinely visualise individual protein molecules and determine their precise structures to atomic detail using the information collected using these powerful microscopes.

CryoEM has been successfully used to visualise whole virus particles by many different research groups. Sevanna *et al* recently published a high-resolution structure of the entire Zika virus (**Figure 1**).

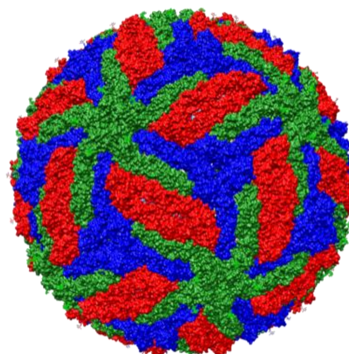


Figure 1. Zika virus particle. The different E proteins on the surface are shown in red, blue and green. From https://www.rcsb.org/structure/6CO8.

Using this method, we can see that Zika virus particles have a smooth surface composed of a total of 180 copies of both E and M envelope proteins arranged with icosahedral symmetry. With the extra information gleaned from the high resolution of the structure solved, the researchers could see that not all copies of the E protein are arranged in the same way, which is important to understand as this protein has critical roles in binding host cells in infection.

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Many microbes have complex surface structures composed of tens or hundreds of protein molecules which play roles in movement, export of material from the microbe to its host organism, or other features which can make microbes infectious or harmful to health. Uropathogenic *E. coli* bacteria cause urinary tract infections and can colonise the gut. Colonisation is dependent on the *E. coli* type 1 pilus, a long protein rod that extends out of the bacterial cells and binds to host cells. Spaulding *et al* solved the structure of this pilus protein rod using cryoEM to reveal the helical structure of repeating copies of the FimA protein molecule (**Figure 2A**). This allowed the researchers to map common variations of the pilus found in different *E. coli* strains and examine how these strain differences might influence infection.

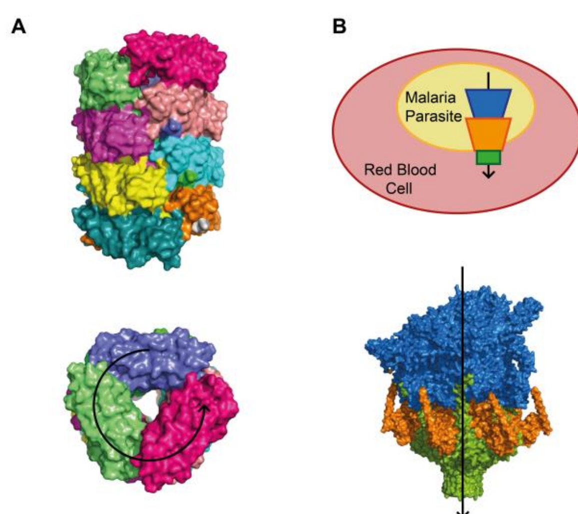

Figure 2. (**A**) 11 FimA fragment of the *E. coli* Type I pilus with side view and top view of the helical structure https://www.rcsb.org/structure/6C53. (**B**) PTEX structure which transfers protein molecules from the malaria parasite into host red blood cells. http://www.rcsb.org/structure/6E10.

The malaria parasite transports proteins from the parasite cell into host red blood cells using a system called the *Plasmodium* translocon of exported proteins (PTEX). Ho *et al* solved structures of this protein transfer apparatus by cryoEM (**Figure 2B**), capturing snapshots of the translocon at different stages of moving material from the parasite to host cells. This allowed the researchers to work out the mechanism by which PTEX might work. In turn, this might assist researchers in the future to make informed decisions on drug design to stop this structure from working with the aim of stopping malarial infection.

As cryoEM becomes an increasingly routine method for determining high-resolution protein structures from different types of microbes, there is a great opportunity for microbiologists to learn more about the molecular mechanisms which make microbes harmful to human health as well as using the structural information gathered to guide rational design of therapeutics.

REFERENCES

Sevanna *et al*. 2018. Structure 26 (9) pg 1169-1177
Spaulding *et al*. 2018. Elife, 18 (7), e31662
Ho *et al* . 2018. Nature Vol 561 pg 70–75

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Dr. Rachel Harding completed her DPhil at the Sir William Dunn School of Pathology at the University of Oxford, supervised by Professor Susan Lea, with a focus on solving structures of proteins involved in meningitis infection. Harding continued her work in structural biology at the Structural Genomics Consortium at the University of Toronto. Harding is the recipient of the Berman/Topper HD Career Development Fellowship from the Huntington's Disease Society of America Fellow, and works on structural biology targets of this disease.

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