Microbiology

Bacteria sting viral invaders

Justin Jenson & Zhijian J. Chen

The cGAS–STING signalling pathway, which has a key role in antiviral immune responses in mammals, is found to have originated as an immune-defence system that protects bacteria against viral infection. **See p.429**

Many multicellular animals respond to disease-causing agents, termed pathogens, using an evolutionarily conserved defence pathway called the cGAS–STING pathway[1,2]. Reports in the past few years of cGAS- and STING-like proteins in bacteria raised the possibility that this pathway is more evolutionarily ancient than was previously thought[3–5]. Morehouse *et al.*[6] demonstrate on page 429 that a functional cGAS–STING pathway exists in bacteria that can potently arrest bacterial growth, possibly as a means of defence against a type of virus known as a bacteriophage, or phage for short, that infects bacteria. The authors also provide evidence that the STING protein originated in bacteria before it was acquired by multicellular animals — our metazoan ancestors.

When human cells sense the presence of pathogenic DNA, this causes the cGAS–STING pathway to mount an inflammatory immune response. The enzyme cGAS (Fig. 1) is activated when it binds to DNA in the cytoplasm — the abnormal localization of DNA there can occur during viral infection[7]. Active cGAS produces a type of molecule called a cyclic dinucleotide (CDN), using the nucleotides guanosine triphosphate (GTP) and adenosine triphosphate (ATP) as starting materials to make a CDN called 2′,3′-cGAMP. This binds to STING, which is located on the membrane of an organelle called the endoplasmic reticulum. STING then activates a signalling cascade that ultimately drives the expression of a battery of antiviral genes[8,9].

CDN-based anti-phage signalling systems, known as CBASS systems, have been found in bacteria[4]. These highly diverse systems are composed of cGAS-like enzymes and downstream proteins called effectors that either kill bacteria or inhibit bacterial growth on infection by a phage, thereby stopping the phage from spreading. There are many types of CBASS effector protein[10]. Some of them contain STING-like sequences of amino-acid residues[4], raising the intriguing possibility of an intact cGAS–STING pathway in bacteria.

Using X-ray crystallography, Morehouse and colleagues showed that two different bacterial species have a protein with STING-like amino-acid sequences that is similar in structure to mouse and human STING. All of these contain a characteristic, V-shaped binding pocket that binds to CDNs. The overall structures of bacterial and mammalian STING are similar.

However, the authors noted some key differences in the composition of amino-acid residues and in the architecture of the CDN-binding pocket of bacterial STING, suggesting that its CDN-binding specificity might differ from that of mammals. The authors used these structures to guide the construction of a phylogenetic tree representing the evolutionary relationships between STING proteins from bacteria and animals. This led them to the important finding that STING probably evolved in bacteria before being acquired by an early metazoan.

cGAS-like proteins in bacteria produce many types of CDN, as well as cyclic-oligonucleotide molecules[5]. Morehouse *et al.* studied the bacterial cGAS-like protein CdnE to determine its biologically relevant CDN. They found that CdnE produces the molecule 3′,3′ c-di-GMP (also known just as c-di-GMP) *in vitro*. This discovery was surprising because 3′,3′ c-di-GMP was originally identified as a molecule that regulates the synthesis of the polymer cellulose[11], and it was subsequently discovered to have other roles, such as regulating the formation of bacterial aggregates called biofilms[12]. Constant production of 3′,3′ c-di-GMP for such other purposes would be catastrophic for bacterial growth if 3′,3′ c-di-GMP can also activate the CBASS system to cause cell death. The authors analysed the genomes of bacteria containing cGAS–STING pathways, and found that bacterial

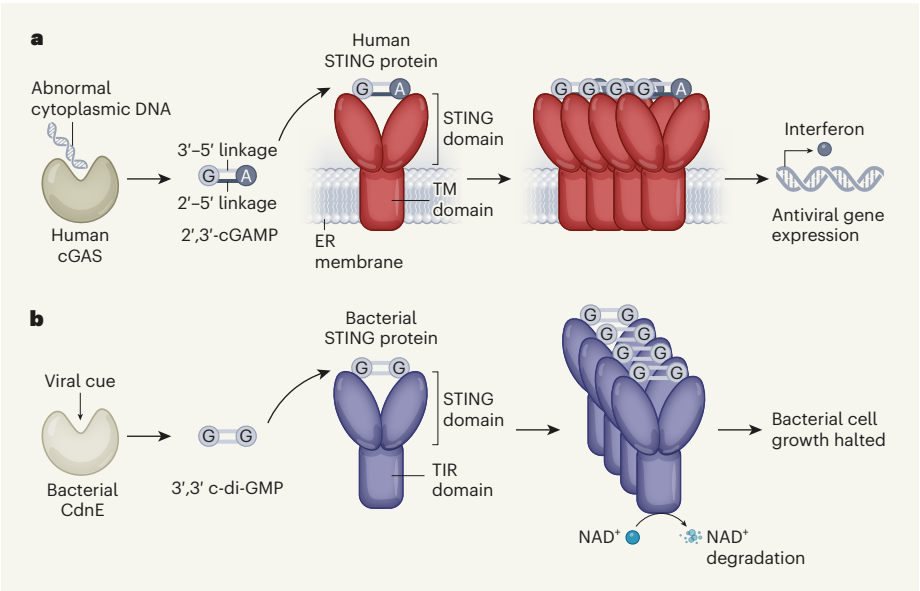

Figure 1 | A cellular defence pathway that protects bacteria and humans. a, On recognition of signs of infection, such as the abnormal presence of cytoplasmic DNA, the human enzyme cGAS produces a type of molecule termed a cyclic dinucleotide. This molecule, called 2′,3′-cGAMP, consists of the nucleotides guanosine monophosphate (G) and adenosine monophosphate (A) joined by what are known as 2′–5′ and 3′–5′ linkages, which connect nucleotide sugars and phosphate groups. When 2′,3′-cGAMP binds to the protein STING, which has its transmembrane (TM) domain located in an organelle called the endoplasmic reticulum (ER), STING proteins assemble and activate a signalling cascade. This leads to the expression of antiviral genes, including genes that encode members of the interferon family of proteins. **b**, Morehouse *et al.*[6] shed light on a related antiviral pathway in bacteria. In response to an unknown viral cue, the bacterial cGAS-like enzyme CdnE produces the cyclic dinucleotide 3′,3′ c-di-GMP, in which the nucleotides are joined only by 3′–5′ linkages. This molecule binds to a bacterial protein consisting of a STING domain and a TIR domain. The protein then assembles into long filaments, triggering the enzymatic activity of the TIR domain, which degrades the molecule NAD^+. NAD^+ depletion halts the growth of an infected cell, possibly providing a way to stop viral spread.

STING occurs almost exclusively in bacteria lacking other signalling pathways that also involve 3′,3′ c-di-GMP, thereby avoiding this potential conflict.

Human STING defends against viruses by relying on the expression of antiviral genes, such as those encoding interferon proteins, which have been identified only in vertebrates. However, bacterial CBASS systems instead fight viral infection by either arresting bacterial cell growth or inducing cell death to prevent further phage spread[10].

Morehouse *et al.* report that bacterial STING most commonly exists as a STING–TIR fusion protein that has a STING domain connected to a TIR domain, which is involved in plant and animal defence responses. The TIR domain is best known for its role in protein–protein interactions in mammalian defence pathways that are part of the innate immune response, which provides a broad defence against pathogens. Some TIR domains in plants and animals also have enzymatic activity[13,14] that degrades the molecule NAD^+, which is essential for cellular metabolism.

The authors showed that the presence of 3′,3′ c-di-GMP was sufficient to cause a bacterial STING–TIR fusion protein to assemble into long filaments that rapidly degraded NAD^+. This NAD^+ destruction halted cell growth in the bacterium *Escherichia coli*. Mutation in the CDN binding site blocked the toxicity of the system in *E. coli*, suggesting that 3′,3′ c-di-GMP controls filament formation and NAD^+ destruction mediated by the TIR domain.

STING–TIR fusion proteins are not limited to bacteria. Using a bioinformatics approach, Morehouse *et al.* identified such proteins in some invertebrates, including the Pacific oyster (*Crassostrea gigas*). Structural analysis of a STING–TIR fusion protein from *C. gigas* revealed that it binds tightly to 2′,3′-cGAMP, which is the CDN that most potently binds to and activates mammalian STING. Notably, in 2′,3′-cGAMP, the phosphodiester bonds between the nucleotides guanosine monophosphate (GMP) and adenosine monophosphate (AMP) have an asymmetric pattern of linkages (a 2′–5′ linkage between the 2′-OH group of GMP and the 5′-phosphate group of AMP, and a 3′–5′ linkage between the 3′-OH of AMP and 5′-phosphate of GMP). This arrangement is found in many multicellular animals, but not in bacteria, and suggests that the dominant ligand for STING changed after it was acquired by our animal ancestors. The reason for this change is unclear.

One unresolved question is how the bacterial cGAS–STING pathway is activated by phage infection. Morehouse *et al.* showed that, like many bacterial cGAS-like proteins[5,15], purified CdnE protein is constitutively active *in vitro*. Therefore, it is possible that the active protein is normally inhibited and is released from inhibition only on phage infection. An example of this type of system is the cGAS-like enzyme DncV in the bacterium *Vibrio cholerae*, which is inhibited by metabolites (folate-like molecules) that are presumably depleted during phage infection[16]. More research will be needed to determine whether this is how CdnE and other cGAS-like proteins are regulated, or if other regulatory mechanisms exist.

As we learn more about the diverse and complex defence systems in bacteria, it might be tempting to consider these immune systems as mirroring those of vertebrates. For example, the CRISPR–Cas system used by organisms such as bacteria can form what is akin to an immunological memory to fight specific phage reinfection. This shows echoes of our own adaptive immune systems, which can remember and respond to specific pathogens. Likewise, the bacterial CBASS systems, including the cGAS–STING pathway, provide broad protection against phage invasion, much as our own innate immune systems do. Interestingly, whereas the CRISPR–Cas system is absent in humans, and specialized immune cells called T and B cells instead do the job, the cGAS–STING pathway and its antiviral defence function are preserved from bacteria to humans.

Justin Jenson and **Zhijian J. Chen** are in the Department of Molecular Biology, University of Texas Southwestern Medical Center, Dallas, Texas 75390, USA. **Z.J.C.** is also at the Center for Inflammation Research and the Howard Hughes Medical Institute, University of Texas Southwestern Medical Center.
e-mails: justin.jenson@utsouthwestern.edu; zhijian.chen@utsouthwestern.edu

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Computer science

Brain-inspired computing becomes complete

Oliver Rhodes

Hardware modelled on the brain could revolutionize computing, but implementing algorithms on such systems is a challenge. A proposed conceptual framework could simplify implementation, accelerating research in this field. **See p.378**

The next generation of high-performance, low-power computer systems might be inspired by the brain. However, as designers move away from conventional computer technology towards brain-inspired (neuromorphic) systems, they must also move away from the established formal hierarchy that underpins conventional machines — that is, the abstract framework that broadly defines how software is processed by a digital computer and converted into operations that run on the machine's hardware. This hierarchy has helped enable the rapid growth in computer performance. On page 378, Zhang *et al.*[1] define a new hierarchy that formalizes the requirements of algorithms and their implementation on a range of neuromorphic systems, thereby laying the foundations for a structured approach to research in which algorithms and hardware for brain-inspired computers can be designed separately.

The performance of conventional digital computers has improved over the past 50 years in accordance with Moore's law, which states that technical advances will enable integrated circuits (microchips) to double their resources approximately every 18–24 months. But although such advances enable ever-more-powerful hardware, they also create challenges for system architects looking to optimize the performance of algorithms executed on these constantly changing devices.

An important feature of conventional computer design that has allowed the best performance to be obtained from new devices (chips, memory, and so on) has been the absence of a tight coupling between software and hardware