combination with the phenomenon of nuclear magnetic resonance (NMR). Brief (a few milliseconds) application of a second magnetic field that oscillates at the characteristic NMR frequency of the experimental set-up rotates the nuclear spins so that they are no longer aligned with the static field. Subsequently, both the average spin orientation and the anisotropic γ-ray distribution will precess (rotate) about an axis parallel to the static field, and the number of γ-rays detected by an appropriately oriented detector will oscillate with time (Fig. 1b). This effect can be used to infer the nuclear-spin dynamics — an approach that has proved fruitful in nuclear-physics studies[3]. The degree of nuclear polarization that is needed to produce sufficiently anisotropic γ-ray emission in these studies is more than 10,000 times greater than can be achieved by 'magnetizing' the sample in the field of a typical NMR magnet[2]. Such hyperpolarization can instead be obtained by using methods such as spin-exchange optical pumping[4], in which electron polarization produced by laser irradiation is transferred to the nuclei in collisions.

Zheng and collaborators adapted apparatus that they had previously used[5] for conventional, low-field (0.7 millitesla) MRI of hyperpolarized xenon-129 (^{129}Xe). They used optical pumping to produce 60% of the maximum possible polarization in a sample of about 70 picomoles of metastable ^{131}Xe. The nuclei were contained in a glass cell shaped like the Chinese character for the word 'middle' (see Figure 1b of the paper[1]). The authors then manipulated the nuclear-spin orientation using an additional magnetic field that oscillated at the NMR frequency of 960 hertz. Finally, they measured the time-varying flux of γ-rays using detectors aligned parallel and perpendicular to the 0.7-mT static field.

Using this arrangement, Zheng *et al.* demonstrated a new way to measure NMR spectra — they used γ-ray detection to infer changes in spin orientation that occurred while the oscillating magnetic field was applied to the sample. More importantly, by applying magnetic-field gradients across the sample, the authors generated a 2D image that shows the shape of the glass cell (see Figure 1a of the paper[1]). Their imaging technique is based on a truly inventive idea. In conventional MRI[6], a mathematical operation known as a Fourier transform is used to relate the shape of a sample to its constituent 'spatial frequencies'. The authors realized that, when magnetic-field gradients are used to produce a sinusoidal spatial variation of the angle made by the nuclear-spin orientation to the direction of the static field, the γ-ray flux at the detectors is directly related to the strength of specific spatial-frequency contributions to the sample's shape. An image of the sample was consequently formed by taking multiple measurements and then summing the different contributions.

The authors' imaging method therefore shares many of the favourable characteristics of MRI, particularly in terms of spatial resolution, controllable contrast and spectral sensitivity. But the technique can also produce images from samples that contain only trace amounts of radioactive nuclei — if the glass cell had been filled with water, rather than metastable ^{131}Xe, an image could have been made using conventional proton MRI, but about 50 billion times more nuclei would have been participating in the imaging process.

Zheng and colleagues' approach is a fantastic idea, but there are many hurdles to negotiate along the path from their reported proof-of-concept experiments to practical applications in the clinic. Given that the authors' image took about 60 hours to acquire, the first essential step will be to increase the speed of image acquisition. Immediate gains might be reasonably straightforward to obtain by improving the efficiency of data collection. For instance, the sample was repolarized by optical pumping — a time-consuming process — after each spatial-frequency contribution was measured. This meant that only 6% of the acquisition time was spent measuring the γ-ray flux.

But even if information about all spatial frequencies could be obtained after a single hyperpolarization step, the 200-minute acquisition time needed to count an adequate number of γ-rays would still be unfeasibly long for *in vivo* studies. Further modifications that increase the γ-ray flux will therefore be required. The authors suggest that this might be accomplished by using other radioactive noble gases, such as metastable forms of ^{127}Xe or krypton-79, which have shorter half-lives than metastable ^{131}Xe.

A further problem for *in vivo* application is that the lifetime of nuclear polarization for nuclei with spins greater than one-half is generally much less than one second in the liquid state[7]. Therefore, the high level of polarization required for anisotropic γ-ray emission will be difficult to maintain over the time needed for tracer transit from a site of injection to the organ of interest. Delivering the tracer in gas bubbles or solid nanoparticles could potentially overcome this problem, but would probably reduce the possibilities for tracer targeting. These approaches, and others that involve more-complex manipulation of nuclear polarization using ideas from NMR, are definitely worth exploring in the quest to bring the full power of the authors' technique to bear in diagnostic imaging. ■

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MICROBIOLOGY

The bacterial cell wall takes centre stage

An unexpected function has been assigned to part of the molecular machinery that synthesizes the bacterial cell wall — a dramatic shift in our understanding that may have major implications for antibiotic development. SEE ARTICLE P.634

KEVIN D. YOUNG

Shakespeare's comedies are full of mistaken identities — a woman is thought to be a man, lovers pursue egregiously misidentified objects of their affections, a duke pretends to be priest — and hilarity ensues. Although this works well in comedy, it's not so humorous in science. A paper on page 634 by Meeske *et al.*[1] raises the curtain on an analogous plot twist as the authors shatter a deeply held assumption about how bacterial cell walls are built, a conclusion that is also supported by the work of Cho *et al.*[2] in a paper published in *Nature Microbiology*.

Previously, only class A penicillin-binding proteins (aPBPs) were known to polymerize the molecule peptidoglycan[3], the backbone of most bacterial cell walls. Now, Meeske *et al.* and Cho *et al.* report that this enzymatic step can be performed by an unrelated protein, RodA, which had been thought to have a completely different role. The discovery overturns long-held ideas about a fundamental biological process and exposes a different target towards which antibiotic development might be directed.

Our view of cell-wall synthesis had seemed

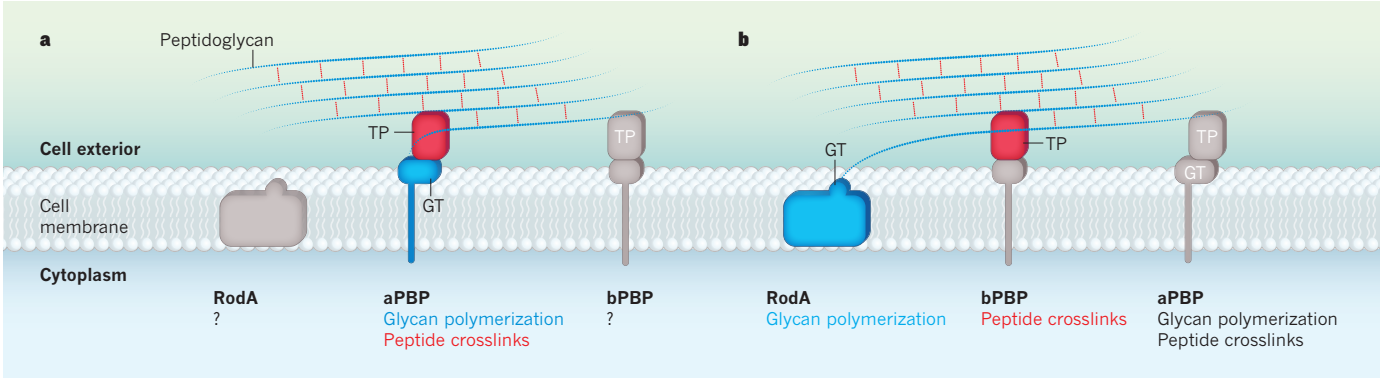

Figure 1 | Models of peptidoglycan synthesis in bacterial cell walls. **a**, In the conventional model, a dissaccharide (not shown) is flipped across the cell membrane from the cytoplasmic side to the cell exterior, and delivered to a class A penicillin-binding protein (aPBP), which contains a glycosyltransferase enzyme domain (GT, blue oval) and a transpeptidase enzyme domain (TP, red oval). The glycosyltransferase domain polymerizes the disaccharide substrates into long glycan chains (blue lines), and the transpeptidase domain links these chains through short peptides (red lines) to create the peptidoglycan cell wall. The RodA protein and a class B penicillin-binding protein (bPBP) have unknown roles in this model. **b**, In the model proposed by Meeske *et al.*[1] and Cho *et al.*[2], substrate is delivered to RodA (blue rectangle), the newly recognized glycosyltransferase that catalyses chain formation. This chain is crosslinked to existing strands by the transpeptidase domain of bPBP. In this scheme, the aPBPs also polymerize and crosslink glycan chains, but their relationship to the RodA or bPBP complex is undefined.

fairly straightforward (Fig. 1a). Two carbohydrate molecules are linked to one another to form a disaccharide, which is then 'flipped' from inside the cell across the cell membrane to the cell exterior. There, the glycosyltransferase domain of an aPBP adds this disaccharide to a growing polysaccharide chain, and a transpeptidase enzyme binds this new chain to older ones by creating peptide crosslinks[4]. The result is a large, interconnected, net-like mesh known as a peptidoglycan that wraps around the cell and acts like an external skeleton. Breach this wall, and most bacteria will burst like overinflated balloons, which is one reason that cell-wall synthesis has been studied so intensively as a potential target for antibiotics.

In the established view, only aPBPs have glycosyltransferase polymerizing activity, and for decades no other such enzymes were thought to be involved. The major upheaval precipitated by Meeske *et al.* and Cho *et al.* is caused by their finding that RodA can perform this glycosyltransferase step, perhaps even displacing aPBPs from a central role in the process (Fig. 1b).

Although this is something of a shock, we really should have been prepared. Whispers (or shouts) that something was amiss were heard as long as 13 years ago, with reports that two bacteria, *Bacillus subtilis*[5] and *Enterococcus faecalis*[6], can survive in the absence of all known aPBPs. This led to predictions that an unidentified protein — possibly RodA (ref. 5) — might have glycosyltransferase activity. But RodA represents an entirely different family of proteins (the SEDS family) and, in its absence, bacterial cells that are usually rod-like in shape[7] round up and grow as spheres. What RodA actually does has engendered vigorous debate[8]. An old but recently resurrected proposal is that RodA and other SEDS proteins flip disaccharides across the membrane to deliver these substrates to the aPBPs[8,9].

Thus, the idea that RodA and other SEDS proteins might function as peptidoglycan polymerases was not just unexpected, it was all but actively disregarded. That it has taken so long to confirm the identity of this new class of polymerase is a somewhat dubious tribute to our ability to ignore data that run counter to preconceptions.

In defence of the field, measuring this kind of glycosyltransferase activity in living cells was mostly out of reach until the advent of super-resolution microscopy about five years ago. Meeske and colleagues monitored peptidoglycan synthesis by visualizing the microscopic motion of Mbl, a protein that is part of the RodA machinery that elongates the rod-shaped cells of *B. subtilis*. Cho and colleagues used a similar approach by monitoring MreB, a protein in the RodA complex of *Escherichia coli*. In both species, these complexes constantly circle the cells, inserting new peptidoglycan as they go. However, Meeske and colleagues confirmed that adding a β-lactam antibiotic inhibited cell-wall synthesis and stopped this motion, indicating that peptidoglycan polymerization is the motor that drives the RodA machine.

Surprisingly, though, both groups found that the RodA apparatus continued to move when aPBPs were absent or inhibited; this indicated that another synthase enzyme must be operating. Furthermore, Cho and colleagues observed that motion stopped when RodA was removed or inactivated. Meeske *et al.* found that overexpression of RodA restored rapid growth to a mutant that lacked aPBPs, and a partially purified cell extract containing RodA exhibited glycosyltransferase activity. All of these results argue that RodA can polymerize peptidoglycan.

Why is this transformative? First, we now know that aPBPs are not essential for peptidoglycan synthesis, although they help a lot. For example, *B. subtilis* strains that lack aPBPs produce aberrant clumps of peptidoglycan and have abortive and oddly placed cell-wall invaginations, and such mutants grow much more slowly than normal[5]. Thus, aPBPs might still be required for robust growth and division in most bacterial cells. Second, as both Meeske *et al.* and Cho *et al.* infer, other members of the SEDS protein family might also have glycosyltransferase activity. For example, the SEDS protein FtsW, which is associated with cell division, might drive peptidoglycan polymerization during this stage of the bacterial cell cycle. In other circumstances, SEDS proteins might be co-opted for different purposes, and some bacteria might exist with no aPBPs at all. Third, because RodA is impervious to compounds that normally inhibit aPBPs, there might be undiscovered antibiotics that target the newly recognized glycosyltransferase abilities of SEDS proteins.

As always with new discoveries, there are questions and concerns. For example, if the SEDS proteins are glycosyltransferase enzymes, then why are there any aPBPs at all? Why has evolution produced (and combined) these two disparate agents? Might RodA flip its own peptidoglycan precursors across the membrane? And how does RodA interact with aPBPs in cells that require these latter enzymes?

Finally, and perversely, I worry that we might exchange one case of mistaken identity for another. Both groups propose that all SEDS proteins have this enzymatic activity, a conclusion drawn from the abilities of a single member, RodA, and the fact that two possible catalytic residues are evolutionarily conserved among the RodA and FtsW proteins of other organisms. Such inductive reasoning certainly opens other lines of inquiry, but the danger is that focusing attention too strongly in one direction might blind researchers to alternative possibilities, recreating our earlier error.

In the end, Shakespeare resolves his characters' mistakes, unites the lovers and allows all to live (mostly) happily ever after. But, for microbiologists, this is only Act II, and the stage may yet be set for even more interesting drama. ■

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FOOD SECURITY

A collaboration worth its weight in grain

Interventions to improve crop yields in rural China through collaboration between researchers and farmers illustrate how the goal of increasing global food production can be approached locally. SEE LETTER P.671

LEAH H. SAMBERG

A defining challenge of our time is to feed an increasingly populated, urban and affluent planet while minimizing the loss of diverse and crucial ecosystems. The focus of attempts to increase global food production has therefore turned to closing yield gaps — increasing productivity of existing farmland to reach a crop's potential regionally attainable yield[1,2]. On page 671, Zhang *et al.*[3] tackle this challenge at the ground level, working closely with small-scale farmers in rural China to increase crop yields in practical and locally appropriate ways. Their work raises the question: what exactly does it mean to talk about yield gaps on a local scale?

On a global scale, yield gaps are viewed as a function of a relatively small set of factors. One study[4] estimated that climate, fertilizer application and water-irrigation techniques explain 60–80% of the variation in yield for major crops, and that closing gaps to meet attainable yields would increase global production of key crops by 45–70%. At the local scale, on-farm productivity depends on these factors, as well as on management practices such as sowing date and planting density, and on socio-economic aspects such as labour availability and market access. On the ground, the conversation about yield gaps leaves the conceptual realm of productivity in optimal conditions, and turns to the adoption of agricultural technology and the design of interventions to remove constraints on productivity. This is the scale at which Zhang and colleagues offer their contribution: the results of a project designed to improve actual yields on actual farms.

FUSUO ZHANG

Figure 1 | Collaborating to improve farming skills. Zhang *et al.*[3] report on a five-year project involving local direct interactions between agricultural researchers and farmers in Quzhou County, China, that resulted in better shared knowledge about farm-management approaches and improved crop yields.

Zhang *et al.* provide a detailed description of a technology-transfer programme known as the Science and Technology Backyard in Quzhou County, China. Over a five-year period, the project team assessed crop-management techniques and measured maize (corn) and wheat yields at a local research station and in farmers' fields. By assuming that the productivity achieved at the research station represents attainable yields, they identified management practices associated with lower yields on farms, including choice of crop variety, planting density and timing, and management of soil tillage and water-irrigation infrastructure. Project staff worked with farmers (Fig. 1) to redesign cultivation recommendations to meet local needs, and to implement techniques designed to surmount yield constraints.

As reported by Zhang and colleagues, these efforts led to impressive results. Within the five-year period, the research team estimated that combined maize and wheat yields in the fields of lead farmers — skilled farmers who worked closely with the researchers — increased from 67.9% to 97.0% of attainable levels, with an increase from 62.8% to 79.6% of attainable yields countywide.

When viewed through the lens of closing yield gaps, this is a remarkable achievement. But questions arise about whether that is the most useful way to frame this work, and how this local-scale technology-transfer project is linked to global food-production goals. Is this story really about yield gaps, and should it be?

It is unclear whether the data reported in this paper can be described accurately as yield gaps. Zhang *et al.* compare yields on an annual basis or averaged over a few years, and these data naturally display high levels of variability between years, fields and crops. Such patterns are probably a function of spatial and temporal variability in weather, pest and disease burdens, soils and topography, in addition to management factors. In general, yield-gap studies assess yields across many years, and ideally include crop-simulation models to account for multiple sources of variability[2,4–6].

It is also worth considering whether a focus on the yield of maize and wheat is the best metric of success. Zhang *et al.* attribute farmers' management 'deficiencies' to factors such