Crack patterns of drying dense bacterial suspensions†

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Drying of bacterial suspensions is frequently encountered in a plethora of natural and engineering processes. However, the evaporation-driven mechanical instabilities of dense consolidating bacterial suspensions have not been explored heretofore. Here, we report the formation of two different crack patterns of drying suspensions of Escherichia coli (E. coli) with distinct motile behaviors. Circular cracks are observed for wild-type E. coli with active run-and-tumble swimming, whereas spiral-like cracks form for immotile bacteria. Using the elastic fracture mechanics and poroelastic theory, we show that the formation of the circular cracks is determined by the tensile nature of the radial drying stress and facilitated by the locally ordered structure of bacteria due to their collective swimming. Our study demonstrates the direct link between the swimming behaviors of individual bacteria and the mechanical instabilities and pattern formation of drying bacterial films at macroscopic scales. The results provide new insights into the dynamics of active matter in a drying process and supply important information for understanding a wide range of biological processes associated with drying bacterial suspensions.

1 Introduction

Active matter is nonequilibrium systems consisting of autonomous units that convert local internal or ambient free energy into mechanical motions. A large number of biological and physical systems including suspensions of self-propelled cytoskeleton1,2, swarms of bacteria3–5 and clusters of synthetic active colloids6–8 can be categorized as active matter, which exhibit fascinating statistical and mechanical properties that draw tremendous attention in recent years from both the fundamental and engineering perspectives9–11. While extensive studies have been conducted in understanding the emergent collective dynamics of active matter in fluid states12,13, it is still unclear whether and how local activity affects the macroscopic mechanical properties of consolidating active matter during a drying process.

Drying of bacterial suspensions as a premier model of active matter plays a crucial role in many biological, environmental and industrial processes and influences diverse phenomena ranging from biofilm formation14, spreading of disease15 and food hygiene16 to interbacterial competition for survival17, coating and self-assembly18–20. The active swimming of bacteria can profoundly modify the complex interplays between solid, liquid and vapor phases as a suspension passes from a fluid to a solid state during drying, giving rise to the unusual growth dynamics and morphologies of “coffee rings” in dried deposits18–22. However, these existing studies are all limited to the dilute limit of bacterial suspensions, where bacteria are deposited near the edge of drying drops. The mechanical instabilities of thick consolidating bacterial films formed by drying dense bacterial suspensions remain elusive, despite that such instabilities and the resulting crack patterns have been extensively investigated in counterpart passive systems23–32.

Here, we explored the role of bacterial swimming in the mechanical instabilities and the resulting crack patterns in the dense consolidating bacterial suspensions. We used a wild-type strain of Escherichia coli (E. coli) as our model bacteria, which display the classic run-and-tumble swimming in water (swimmer)33. As a control, we also examined a mutant strain of E. coli that show only tumbling (tumbler). While spiral-like cracks were found in the dried deposits of tumblers and dead swimmers, we observed circular cracks in the dried deposits of wild-type swimmers. Using the elastic fracture mechanics and the poroelastic theory, we showed that the circular cracks form due to the tensile nature of the radial drying stress, which is further promoted by the locally ordered structure induced by the collective swimming of wild-type E. coli. In contrast, the spiral-like cracks arise from the dynamic interplay between cracking and delamination of bacterial films. Our study unambiguously demonstrates the effect of bacterial swimming on the mechanical instabilities of consolidating bacterial films and illustrates the unique features of active matter in the infamously complicated drying process. Our results are also helpful for deciphering different crack patterns in the dried bacterial films frequently encountered in natural and engineering applications.

2 Experiment

We used two different E. coli strains with distinct swimming behaviors in our experiments, i.e., a wild-type strain with active
swimming (BW25113) and a mutant strain of tumblers (RP1616). The two strains share a similar body geometry, which has the average length of 3 µm and the average width of 0.8 µm (Fig. 1a). The culturing protocols of the two strains are detailed elsewhere. In addition, we have also studied the drying behaviors of suspensions of dead swimmers. The dead bacteria were obtained from suspensions of active wild-type bacteria sitting in sealed micro-centrifuge tubes for at least four days. The bacteria were confirmed to be immotile from direct optical microscopy. To avoid any potential complication due to the change of the quality of the buffer over the long waiting, we washed the dead bacteria with 0.9% saline solution for our experiments.

In a typical experiment, we prepared a suspension of bacteria with an initial volume fraction \(\phi_i\) of 10-20%. A glass slide cleaned with DI water and dried by a blowgun was used as the substrate, which was hydrophilic with a water contact angle \(\approx 25^\circ\). A drop of the suspension with an initial volume \(V_i\) of 2-3 µL was gently deposited onto the substrate for drying. A bright-field inverted microscope was used to image the drying process at a frame rate of 1-20 fps. All experiments were performed at the room temperature of 20 ± 2 °C with uncontrolled relative humidity of RH = 30 ± 4%. We used a scanning electron microscope (SEM) to image the microstructures of dried bacterial deposits, and an optical non-contact profilometer to measure the temporal evolution of the height profile of drying bacterial films.

### 3 Results and discussions

#### 3.1 Circular cracks of wild-type swimming bacteria

We observed circular cracks in the consolidating suspensions of wild-type bacteria with active swimming (Figs. 1b and c). Figures 2a-f show the snapshots of different stages during the formation of circular cracks by drying a drop of wild-type swimmer suspension with \(V_i = 2.5 \, \mu L\) and \(\phi_i = 13\%\). After the drop was deposited on the substrate (Fig. 2a), evaporation initially occurred predominantly near the pinned contact line of the drop, driving the formation of a compaction front where the concentration of bacteria increased drastically from \(\phi_i\) in the bulk fluid to that close to the random close packing in the consolidating film \(\phi\). The compaction front displayed an approximately constant length \(L\) (Fig. 2g), and continuously moved toward the center of the drop over a time interval of 414 s (Fig. 2b and Video S1 in ESI†). After the passing of the compaction front, the bacterial film was wet and gel-like and continued to undergo evaporation along the top surface of the film, leading to the accumulation of stress. Once the critical material strength was reached around 427 s, cracks were first initiated near the edge of the film to release the excess stress\(^3\) (Figs. 2c and 2h), which then propagated along a circular path (Figs. 2d-2e) over a short interval of about 73 s in stick-slip motion and eventually formed the circular cracks (Fig. 2f). Accompanying the crack propagation, the film also delaminated radially toward the drop’s center as indicated by the interference fringes shown in Figs. 2i-2j. The extent of film delamination in the final dried deposit can be visualized by the height profile of the deposit in Fig. 1c, for instance.

To interpret the circular cracks of wild-type swimmers, we calculate the tensile stress distribution in the consolidating bacterial film. Figure 3a illustrates the geometry of the consolidating film in a cylindrical coordinate. Drying stress was accumulated in the film behind the compaction front in response to the continuously decreasing local pore pressure \(P_{\text{pore}}\) over time. As evaporation proceeded, the film became flat and thin and evaporation predominantly occurred at the top surface of the film in the late stage of drying. Therefore, \(P_{\text{pore}}\) satisfies an one-dimensional diffusion equation with \(z = 0\) defined at the top surface of the film\(^5\):

\[
\frac{\partial P_{\text{pore}}}{\partial t} = \frac{\kappa E}{\eta} \frac{\partial^2 P_{\text{pore}}}{\partial z^2},
\]

where \(\eta \approx 10^{-3} \, \text{Pa} \cdot \text{s}\) is the dynamic viscosity of water and \(E\) is the Young’s modulus of the dehydrated film on glass substrate. We estimated \(E \approx 100 \, \text{MPa}\) based on our direct measurement using atomic force microscopy, and this value is consistent with the modulus of an isolated dehydrated bacterium. Although the Young’s moduli of isolated bacteria and bacterial films are of the same order of magnitude at the microscopic scales, the lat-
ter is generally larger which depends on substrate stiffness, humidity, bacterial type and even the model. Here, $\kappa$ is the permeability of the film, which is given by the Carman–Kozetsky relation $\kappa = \frac{1}{15} \left(1 - \phi\right)^2 \phi a^2 \sim 10^{-15}$ m$^2$, $\phi \approx 0.7$ is the packing fraction of bacteria (Appendix C) and $a \approx 1$ µm is the characteristic size of bacteria. The initial condition is $P_{pore}(z, 0) = P_{atm}$, where $P_{atm} = 10^5$ Pa is the atmospheric pressure. The boundary conditions are $\partial P_{pore}/\partial z|_{z=0} = -\eta V_E/\kappa$, where $V_E$ is the steady surface evaporation rate. The evaporation rate is expressed as $V_E = \frac{D_w}{R} \frac{n_{wat}}{n_w} A(\theta)(1 - RH)^{42}$, in which $D_w = 25 \times 10^{-6}$ m$^2$ s$^{-1}$ is the diffusion coefficient of water into air at the room temperature, $R = 2$ mm is the drop radius pinned at the contact line, $n_{wat} = 0.02$ kg/m$^3$ is the water density in the vapor at the air-water interface, $n_w = 1 \times 10^3$ kg/m$^3$ is the water density in the bulk liquid, $A(\theta) \approx 1.3$ for a contact angle $\theta \approx 25^\circ$, and the relative humidity $RH \approx 30\%$. Therefore, $V_E \approx 5 \times 10^{-7}$ m/s. Solving Eq. 1, we have $P_{pore}$ given by $^{43}$:

$$P_{pore}(z, t) = P_{atm} - P'(z, t),$$

where

$$P'(z, t) = \frac{2\eta V_E}{\kappa} \left[ \frac{\sqrt{\pi}}{\sqrt{2}} \right] e^{\frac{-z^2}{2\sigma^2}} \text{erfc} \left( \frac{z}{\sqrt{4\pi\sigma^2}} \right)$$

gives the deviation of the pore pressure from the atmospheric pressure. Here, $c = E\kappa/\eta$ is a constant.

During consolidation, a film is constricted by the substrate, thus the out-of-plane strain $\varepsilon_{zz}$ is significantly larger than the in-plane strain $\varepsilon_{rr} + \varepsilon_{\theta\theta}$ driven by shrinkage. Figure 3b shows the temporal variation of the height profile $h(r)$ along a diameter of a consolidating film from $10$ min to $19$ min, during which a circular crack was generated as indicated by the sharp drop of $h$ at $r \approx 1.4$ mm. The film thickness $h$ decreases significantly during this period, whereas the in-plane contraction is negligible, suggesting that $\varepsilon_{zz} \gg \varepsilon_{rr} + \varepsilon_{\theta\theta}$. Furthermore, since the top surface of the film is traction-free, the deformation of the thin film is predominantly driven by the in-plane stress, suggesting $\sigma_{zz} \ll \sigma_{rr} + \sigma_{\theta\theta}$ $^{36,42}$.

The in-plane stress components $\sigma_{rr}$ and $\sigma_{\theta\theta}$ responsible for crack formation are correlated by the equilibrium condition of the stress field in the cylindrical coordinate:

$$\frac{\partial \sigma_{rr}}{\partial r} + \frac{1}{r} (\sigma_{rr} - \sigma_{\theta\theta}) = 0.$$

Using the Biot constitutive relation of homogeneous and isotropic solids $^{35}$ under the approximations $\varepsilon_{zz} \gg \varepsilon_{rr} + \varepsilon_{\theta\theta}$ and $\sigma_{zz} \ll \sigma_{rr} + \sigma_{\theta\theta}$, we have (Eq. A3 in Appendix A):

$$\sigma_{rr} + \sigma_{\theta\theta} = \frac{2P^* (1 - 2\nu)}{1 - \nu},$$

where $\nu$ is Poisson’s ratio of the bacterial film. Given the boundary condition $\sigma_{rr}|_{R} = P^*$, $\sigma_{rr}$ and $\sigma_{\theta\theta}$ can be analytically solved with Eqs. 4 and 5 (Eqs. A6 and A7 in Appendix A):

$$\sigma_{rr} = \langle P^* \rangle \frac{r^2 (2\nu - 1) - R^2 \nu}{r^2 (\nu - 1)},$$

$$\sigma_{\theta\theta} = \langle P^* \rangle \frac{r^2 (2\nu - 1) + R^2 \nu}{r^2 (\nu - 1)},$$

where $\langle P^* \rangle$ the film-thickness-averaged liquid pressure (Eq. A5 in Appendix A). We take the Poisson’s ratio of the film $\nu \approx 0.4$ $^{38,44}$, $R \approx 2$ mm and $h \approx 20$ µm (the thickness of the film at which the crack is generated, see Fig. 3b), so that Eqs. 6 and 7 can be used to predict the spatiotemporal stress distribution.

Figure 3c shows $\sigma_{rr}(r, t)$ and $\sigma_{\theta\theta}(r, t)$ at a fixed location $r = 0.75 R$ where the cracks were generated in the experiment. Note that $\sigma_{rr}$ is tensile ($\sigma_{rr} > 0$) and reaches $\sigma_c \approx 10^5$ Pa when $t > 200$ s, suggesting the formation of the cracks in the circumferential direction at large times. Here, $\sigma_c$ is the critical stress for cracking, which can be estimated based on the the critical film thickness at which cracks form (Fig. B1 in Appendix B). In comparison, $\sigma_{\theta\theta}$ is compressive ($\sigma_{\theta\theta} < 0$), precluding the formation of cracks in the radial direction that are commonly observed by drying drops of colloidal suspensions $^{42,45}$ according to the principles of fracture mechanics.

The above calculation is generic and applies to any consolidating thin films satisfying the strain and stress conditions $\varepsilon_{zz} \gg \varepsilon_{rr} + \varepsilon_{\theta\theta}$ and $\sigma_{zz} \ll \sigma_{rr} + \sigma_{\theta\theta}$, independent of the swimming behavior of bacteria. The active swimming of bacteria further modifies the microscopic structure of the consolidating bacterial films, which further facilitates the formation of circular cracks. Figure 4a shows the two-dimensional (2D) flow field of a suspension of wild-type bacteria near the compaction front during drying, which exhibits the characteristic swimming vortices driven by the collective swimming of bacteria $^{4,5}$. Such a coherent dynamic structure was preserved throughout the drying course and
eventually led to the locally ordered configuration of bacteria in the dried deposit as indicated by the red arrows in Fig. 4b. The circular crack marked by the yellow arrows propagated through this locally ordered structure, reminiscent of the crack formation in the consolidating drops of passive colloidal rods during which circular cracks were generated along the order direction of the deposited colloidal rods near the contact line\textsuperscript{27,46}. The preferred propagation of cracks along the ordered structure can be understood by the strain energy release rate $G \equiv -\partial \mathcal{U}_s/\partial A$. Here, $\mathcal{U}_s$ is the strain energy available to release and $A$ is the surface area of crack. For a given $\mathcal{U}_s$, a crack path along the order direction creates a smaller $A$ and, therefore, a larger $G$. Thus, the crack path along the order direction is more energetically favorable.

Finally, Figs. 2i-j show that along with the propagation of the circular cracks, the film also delaminated toward the center of the drying drop. Owing to the strong humidity gradient across the film thickness during drying\textsuperscript{47}, the in-plane tensile stress $\sigma_{rr}$ is localized near the top surface of the film and gives rise to a stress gradient across the film as illustrated in Fig. 6. Such a stress gradient creates a bending moment $M$, which promotes the film delamination once the accumulated stress is beyond the critical stress for delamination\textsuperscript{43,47,48}. To gain more insight into the mechanical instabilities of cracking and delamination in the consolidating films, we estimated the critical stress for cracking $\sigma_c$ and the critical stress for delamination $\sigma_D$ (in Appendix B), and we found $\sigma_c$ and $\sigma_D$ are of the same order of magnitude $\sim 10^5$ Pa. Hence, immediately following the formation of the circular cracks, the film delaminated from the new crack surface toward the center of the drying drop. Figure 1c shows the film delamination triggered by the gradient of $\sigma_{rr}$, which provides further evidence supporting the mechanism underlying the mechanical instabilities of the consolidating swimmer films.

### 3.2 Spiral-like cracks of immotile bacteria

To verify directly the role of active swimming in the formation of circular cracks, we examined the crack patterns of mutant tumblers as well as dead wild-type swimmers. Neither of the two types of bacteria showed the collective swimming in drying drops. In contrast to the circular cracks of wild-type bacteria with active swimming, we observed spiral-like cracks in the dried deposits of these two types of immotile bacteria as shown in Fig. 5.

Figures 7a-f show the snapshots of different stages during the formation of the spiral-like cracks of a tumbler drop with $\phi = 14\%$ and $V_f = 2.5 \mu$L. The compaction front of the drying tumbler drop was significantly wider than that of the wild-type swimmer drop (Fig. 2) and increased in width over time (comparing Videos S1 and S3 in ESI\textsuperscript{†}). As the accumulated stress reached the material strength, hairpin-shaped cracks facing the center of the drying drop first appeared (boxed regions in Figs. 7c and d), which simultaneously initiated film delamination between the two arms of the hairpin (interference fringes in Figs. 7h and i). Upon the creation of new surfaces by cracking, the film subsequently delaminated perpendicularly to the arms of the hairpins and propagated along the circumferential direction (Fig. 7h). The delamination front along the circumferential direction displayed an arc-shaped profile, which triggered the formation of a spiral-like crack at the delamination front (Figs. 7e and j). The crack in turn created a new surface for further delamination. This cycle of crack and delamination repeated with time until the cracks initiated by different hairpins met, which ultimately gave rise to the spiral-like crack pattern shown in Fig. 7f. Thus, the spiral-like cracks of immotile bacteria stem from the dynamic interplay between film cracking and delamination.

Due to the absence of the collective swimming, the tumblers...
Circular cracks were observed in the consolidating films of wild-type driven consolidating bacterial suspensions, a model active matter. We investigated the mechanical instabilities of dense evaporation-manipulating the swimming behaviors of microorganisms. The consolidating films of microorganisms can be tuned by manipulating the swimming behaviors of microorganisms. More broadly, our experiments demonstrate that the crack patterns in the consolidating films of wild-type tumbler and dead wild-type swimmers suggest that the locally ordered structure driven by the collective swimming of wild-type bacteria plays a key role in the formation of circular cracks. More ordered structure driven by the collective swimming of wild-type tumblers and dead wild-type swimmers suggest that the locally ordered structure driven by the collective swimming of wild-type bacteria plays a key role in the formation of circular cracks. More.

4 Conclusions

We investigated the mechanical instabilities of dense evaporation-driven consolidating bacterial suspensions, a model active matter. Circular cracks were observed in the consolidating films of wild-type swimming E. coli, which were followed by film delamination along the radial direction toward the center of the drying drop. Using the framework of elastic fracture mechanics and poroelastic theory, we showed that the circular cracks are determined by the tensile nature of the radial drying stress. The formation of the circular cracks is further promoted by the locally ordered structure induced by the collective swimming of bacteria. The tensile radial stress in combination with the humidity gradient across the film thickness leads to the delamination of the consolidating films upon the creation of the free surface by cracking. Moreover, we also observed spiral-like cracks in the dried deposits of immotile bacteria. Such an intriguing pattern likely arises due to the complex interplay between cracking, delamination, film geometry and stress anisotropy during drying. The detailed mechanism of this complicated process is an open question for future research. Taken together, our results elucidate the critical role of the microscopic bacterial activity on the macroscopic mechanical instabilities and pattern formation of consolidating bacterial films. Our study also provides insights into diverse biological, environmental and industrial processes associated with drying bacterial suspensions, such as spreading of pathogens, biofilm formation, painting and coating of biological fluids.

Conflicts of interest

There are no conflicts to declare.

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Appendix A Derivation of the in-plane stresses $\sigma_{rr}$ and $\sigma_{\theta\theta}$

For linearly poroelastic solids, the stress and strain are related by the Biot constitutive equation:

$$
\varepsilon_{ij} = \frac{1 + \nu}{E} \sigma_{ij} - \frac{\nu}{E} \sigma_{kk} \delta_{ij} + \frac{\alpha}{3K} \left( P_{\text{pore}} - P_{\text{atm}} \right) \delta_{ij}
$$

$$
= \frac{1 + \nu}{E} \sigma_{ij} - \frac{\nu}{E} \sigma_{kk} \delta_{ij} + \frac{\alpha P^*}{3K} \delta_{ij},
$$

(A1)

where $\alpha \approx 1$ is the Biot-Willis coefficient, $\nu$ and $K = E/(1 - 2\nu)$ are Poisson’s ratio and bulk modulus of the film, respectively. Here, $E$ is the Young’s modulus of the film. $P^* = P^*(r,z)$ is given by Eq. 3 in the main text. $(i,j)$ take the values of $(r,\theta,z)$ in a cylindrical coordinate system. Accordingly, $\sigma_{zz} = \sigma_{rr} + \sigma_{\theta\theta} + \sigma_{zz}$.

Summing up the three principle strains according to Eq. A1 under the approximations $\varepsilon_{zz} \approx \varepsilon_r + \varepsilon_{\theta\theta}$ and $\sigma_{zz} \approx \sigma_{rr} + \sigma_{\theta\theta}$ explained in the main text leads to:

$$
\varepsilon_{zz} = \frac{1}{3K} \left( \sigma_{rr} + \sigma_{\theta\theta} \right) - \frac{P^*}{K}.
$$

(A2)

In combination with $\varepsilon_{zz} = -\frac{\nu}{E} \left( \sigma_{rr} + \sigma_{\theta\theta} \right) - \frac{P^*}{K}$ from Eq. A1, we have:

$$
\sigma_{\theta\theta} + \sigma_{rr} = \frac{2P^*}{3K(1+\nu)} = \frac{2P^*(1-2\nu)}{1-\nu}.
$$

(A3)
Plugging Eq. A3 into Eq. 4 in the main text leads to:

\[
\frac{r}{h} \frac{\partial \sigma_{rr}}{\partial r} + 2\sigma_{rr} = 2P^* \left(2 + \frac{1}{\nu - 1}\right). \tag{A4}
\]

Since \(P^*\) shows small variations across the film thickness, we calculate the film-thickness-averaged liquid pressure to remove the weak \(z\) dependence:

\[
\langle P^* \rangle = \frac{1}{h} \int_0^h \left[ 2\eta V_k \frac{E}{\kappa} \frac{\kappa^2 - \rho^2}{\kappa} - \eta V_k \frac{r}{\kappa} \text{erfc} \left( \frac{r}{2\sqrt{h/k}} \right) \right] dz
\]

\[
= \frac{V_k E t}{h} \left[ 1 + \frac{he^{-\rho^2/c^2}}{\sqrt{\pi} c} - \frac{h^2 + 2ct}{2ct} \text{erfc} \left( \frac{h}{2\sqrt{ct}} \right) \right], \tag{A5}
\]

where \(c = E\kappa/\eta\).

The stress boundary condition at \(r = R\) is \(\sigma \cdot n |_{r=R} = \sigma_{rr} |_{r=R} = \rho_{atm} - \rho_{pore} = P^*\). Applying the condition and replacing \(\langle P^* \rangle\) (Eq. A5), Eq. A4 can be solved:

\[
\sigma_{rr} = \frac{\langle P^* \rangle}{h} \left( \frac{1}{2} (2\nu - 1) - \frac{R^2\nu}{h^2} \right). \tag{A6}
\]

Plugging Eq. A6 into Eq. 4 in the main text yields:

\[
\sigma_{\theta\theta} = \frac{\langle P^* \rangle}{h} \left( \frac{1}{2} (2\nu - 1) + \frac{R^2\nu}{h^2} \right). \tag{A7}
\]

**Appendix B  Critical stress for cracking \(\sigma_c\) and delamination \(\sigma_d\)**

An evaporation-driven consolidating film is prone to crack to release the excess stress once the accumulated stress is beyond the critical stress \(\sigma_c\). The value of \(\sigma_c\) can be determined by measuring the corresponding critical film thickness \(h_c\) as

\[
\frac{\sigma_{at}}{2\gamma} = 0.1877 \left( \frac{2a}{h_c} \right)^{2/3} \left( \frac{GM_0\phi a}{2\gamma} \right)^{1/3}, \tag{B1}
\]

where \(G = \frac{E_b}{2(1+\nu_b)}\) is the shear modulus of bacteria, \(E_b\) and \(\nu_b\) are the Young's modulus and Poisson's ratio of the dehydrated bacteria, \(a\) is the characteristic radius, \(\gamma\) is the surface tension, \(M_0\) is the coordination number, \(\phi\) is the 3D packing fraction. To measure \(h_c\), we prepared bacterial drops with \(V_i = 2.5\ \mu\text{L}\) and various \(\phi_i\), and deposited them on glass substrates for drying to obtain dried deposits of different thicknesses.

Figure B1 shows the measurement of the height profiles along the diameters of two dried deposits of swimmer drops, in which cracks were initiated in the dried deposit with the larger film thickness, but not in the deposit with the smaller film thickness. The critical film thickness for cracking of swimmer films, \(h_c\), should lie between these two thicknesses. We simply took the average of the maximum values of the blue and black curves as indicated by the dashed lines in Fig. B1, which gives the estimate \(h_c \approx 13\ \mu\text{m}\). Plugging the typical values of \(E_b = 300\ \text{MPa}\) \cite{37}, \(\nu_b = 0.237\ \alpha = 1\ \mu\text{m}, \gamma = 72\ \text{mN/m}, M_0 = 6, \text{ and } \phi \approx 0.7\) (see Appendix C) into Eq. B1, we have the critical stress for swimmer films \(\sigma_c \approx 10^5\ \text{Pa}\).

The critical stress for film delamination \(\sigma_d\) can be estimated...
where $\Gamma_d$ is the adhesion strength of the film, $E$ and $h_d$ are the Young’s modulus and critical delamination thickness of the film, respectively. Although it is challenging to directly measure the adhesion between an evaporation-driven consolidating bacteria film and a glass substrate, the adhesion strength for the majority of biofilms is on the order of magnitude of 5 mJ/m² [44,53–57]. Here, the Young’s modulus of the dehydrated bacteria films can be estimated as $E \sim 100$ MPa. In our experiments, the film delamination and cracking develop nearly simultaneously. Therefore, we simply take $h_d \approx h_c \approx 13 \mu m$. Plugging the values of $\Gamma_d$, $E$ and $h_d$ into Eq. B2 yields $\sigma_d \sim 10^5$ Pa for swimmer films, which is of the same order of magnitude as $\sigma_c$.

### Appendix C Estimate of the 3D packing fraction of bacteria in dried deposits

To estimate the 3D packing fraction of bacteria in a dried deposit, we first calculate the relation between the 3D packing fraction $\phi$ and the 2D area fraction $\phi_{2d}$. Assuming a cylindrical shape of a bacterial body with a length of $l_b$ and radius of $r_b$. The volume of bacterial body is $V_0 = \pi r_b^2 l_b$ and the cross-sectional area of the body along its major axis is $A_0 = 2\pi r_b l_b$. Consider bacteria within a horizontal layer of length $K$ and width $W$ parallel to the substrate. We assume that all the bacteria are confined within the layer with their major axes aligned parallel to the substrate, a configuration agreeing reasonably well with our SEM images (Fig. 4b). The number of bacteria within the layer is $N$. The 2D area fraction within the layer is given by $\phi_{2d} = \frac{N_{2d}}{K} = \frac{2\pi r_b l_b}{W K}$, whereas the 3D volume fraction is $\phi = \frac{N_{3d}}{K W^2} = \frac{\pi r_b}{K W}$ . Thus, $\phi = \frac{\pi}{4} \phi_{2d}$. We experimentally measured $\phi_{2d}$ for wild-type bacteria based on the SEM images (Fig. 4b), which gives $\phi_{2d} \approx 0.96$. Consequently, $\phi \approx 0.75$. We note that the experimental $\phi$ is slightly larger than the theoretical maximum values of 3D random packing fraction of rigid cylinders ($\approx 0.7$) [58–60], which is likely due to the locally ordered structure of bacteria and the approximation taken in our estimate.

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