

Active motility and wetting cooperatively regulate liquid-liquid phase separation

Dixi Yang^{1*}, Anheng Wang^{2,3*}, Chunming Wang^{2,3†}, Hajime Tanaka^{4‡}, and Jiaxing Yuan^{1§}

¹ Hong Kong University of Science and Technology (Guangzhou), ² Institute of Chinese Medical Sciences, University of Macau ^{*} equal contribution, [†] corresponding author
³ Faculty of Health Sciences, University of Macau, ⁴ The University of Tokyo [‡] senior collaborator, [§] corresponding author

Introduction

Liquid-liquid phase separation (LLPS) in aqueous two-phase systems (ATPS) is well understood for passive systems, but how active matter interacts with and regulates this process remains largely unknown. Here we study motile *Pseudomonas aeruginosa* in a dextran-PEG ATPS and show that bacterial motility, combined with amphiphilic wetting, generates a series of nonequilibrium morphologies—including self-spinning droplets, colloidal chains, and capillary-like clusters. Activity also produces a striking dual kinetic effect: it suppresses coarsening at high DEX fraction via hydrodynamic repulsion, yet promotes capillary clustering at low DEX fraction. These findings reveal a robust coupling between active stresses, wetting, and LLPS, offering a new route to control phase separation using living active agents.

Methods

Experiments: We added motile *P. aeruginosa* were to a DEX-PEG ATPS. Contact-angle measurements confirm amphiphilic wetting. And use spinning-disk confocal microscopy to image 3D structures.

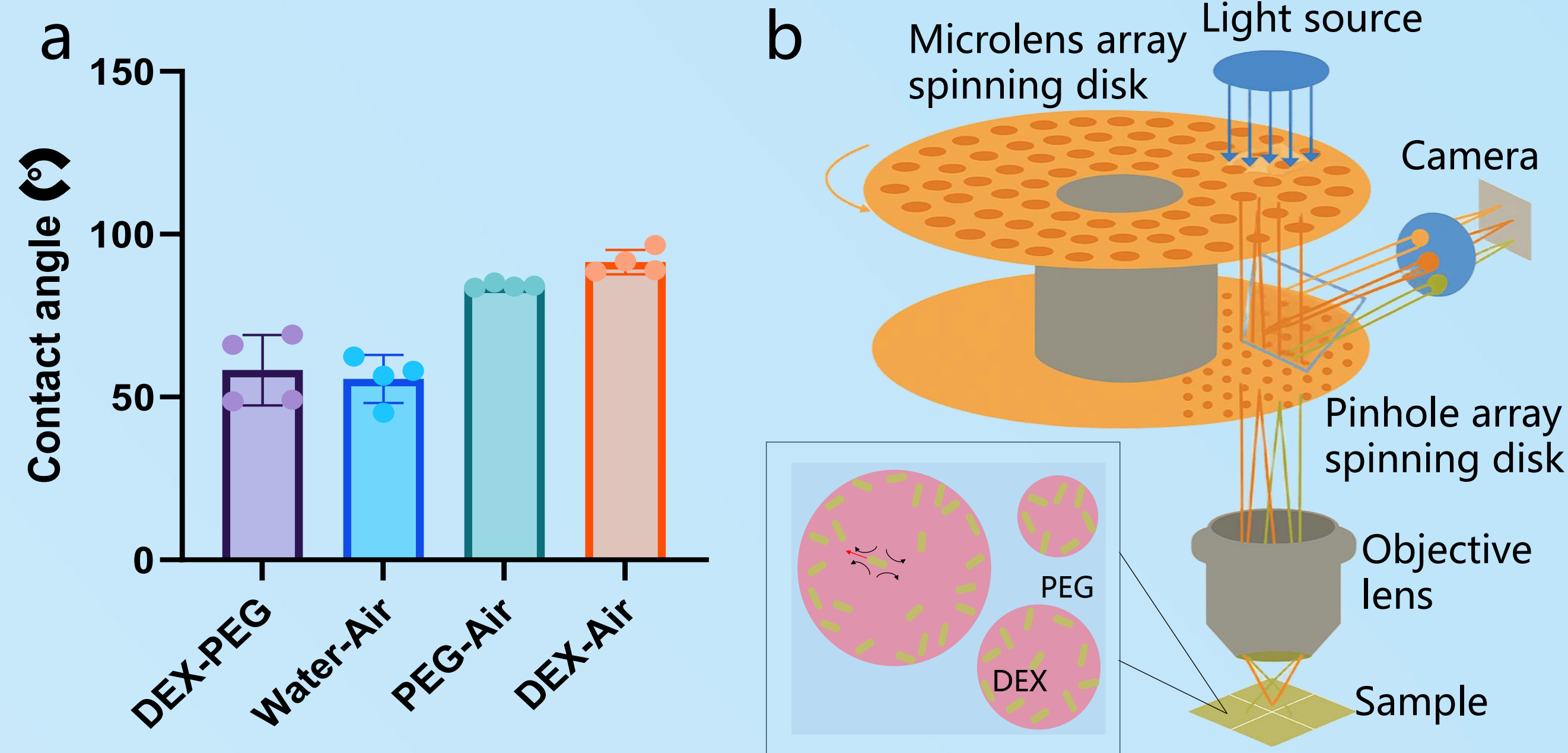


FIG. 1. Interfacial wettability and confocal visualization. (a) Contact angles characterizing the wettability of *P. aeruginosa* across four interfaces: the DEX-PEG-bacteria interface and the water/PEG/DEX-air-bacteria interfaces. (b) Schematic of the spinning-disk confocal microscopy setup used to image the DEX-PEG system containing *P. aeruginosa*.

Results

I. Morphology transition controlled by DEX fraction

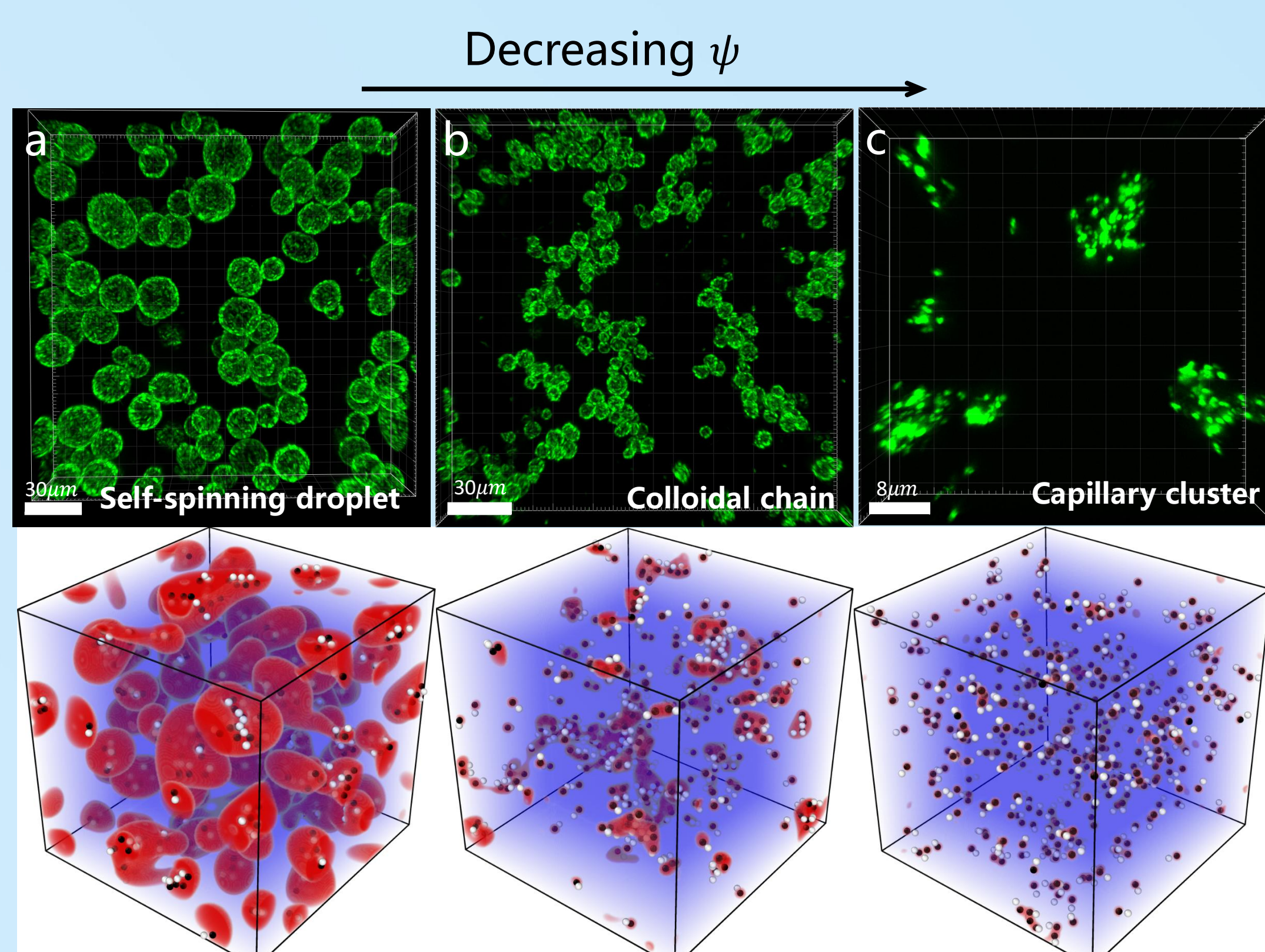


FIG. 2. Morphologies of phase-separated domains in an ATPS with a small amount of *P. aeruginosa*, showing a transition upon decreasing the DEX volume fraction ϕ_{dex} . (a) Self-spinning droplets. (b) Elongated, interconnected droplet chains. (c) Branched, capillary-like bacterial clusters. Upper panels: experimental images of green fluorescent protein (GFP)-tagged bacteria. Lower panels: corresponding simulation snapshots (black: head; white: tail; red: DEX phase; blue: PEG phase).

II. Activity suppresses coarsening at high ϕ_{dex}

Passive (non-motile) systems exhibit normal coarsening: droplets merge and grow. Active systems remain composed of small droplets.

Hydrodynamic flows generated by motile bacteria repel neighboring droplets, preventing coalescence. Simulations reproduce slowed domain growth and flow-mediated repulsion.

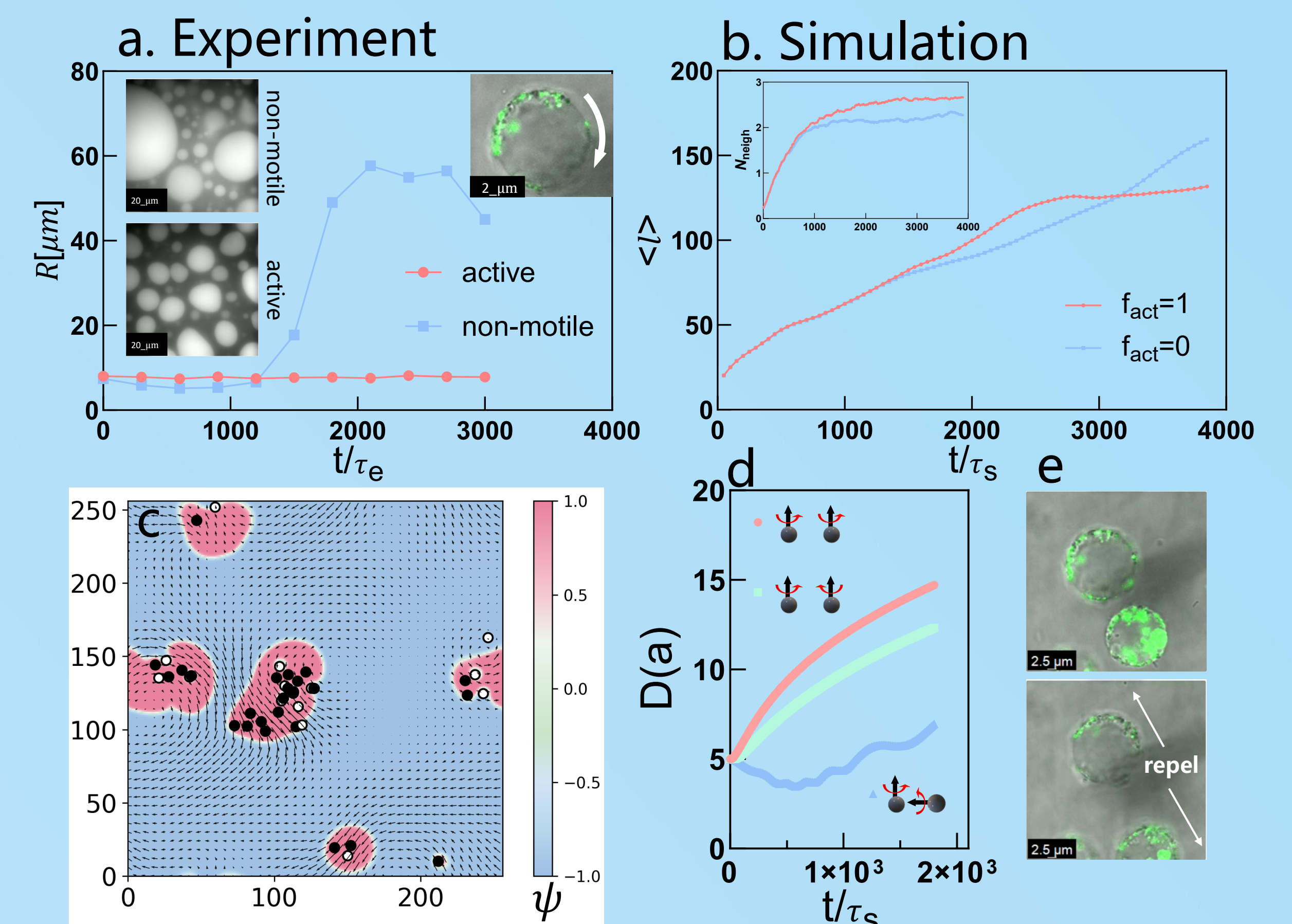


FIG. 3. Dynamic behavior and mechanism of coarsening in self-spinning droplets. (a) Experimental time evolution of the average droplet radius R . Inset: visual comparison of R for active and non-motile systems (left), and a magnified confocal image of a self-spinning droplet (right). (b) Simulated evolution of the domain size, showing suppressed coarsening in the active case. Inset: time evolution of the average neighbor number N_{neigh} . (c) Heat map of the composition field ψ with the velocity field v , demonstrating spontaneous droplet rotation. (d) Simulated center-to-center distance D between two rotating spheres for various rotation orientations, showing effective hydrodynamic repulsion. (e) Experimental time-lapse images showing two self-spinning droplets separating over time without coalescing.

III. Activity enhances capillary clustering at low ϕ_{dex}

At low DEX fraction, activity acts oppositely: DEX-rich clusters become larger and more coordinated. Active swimmers promote interfacial aggregation and increase local coordination numbers. Simulations match experimental trends, demonstrating enhanced capillary-cluster growth under active conditions.

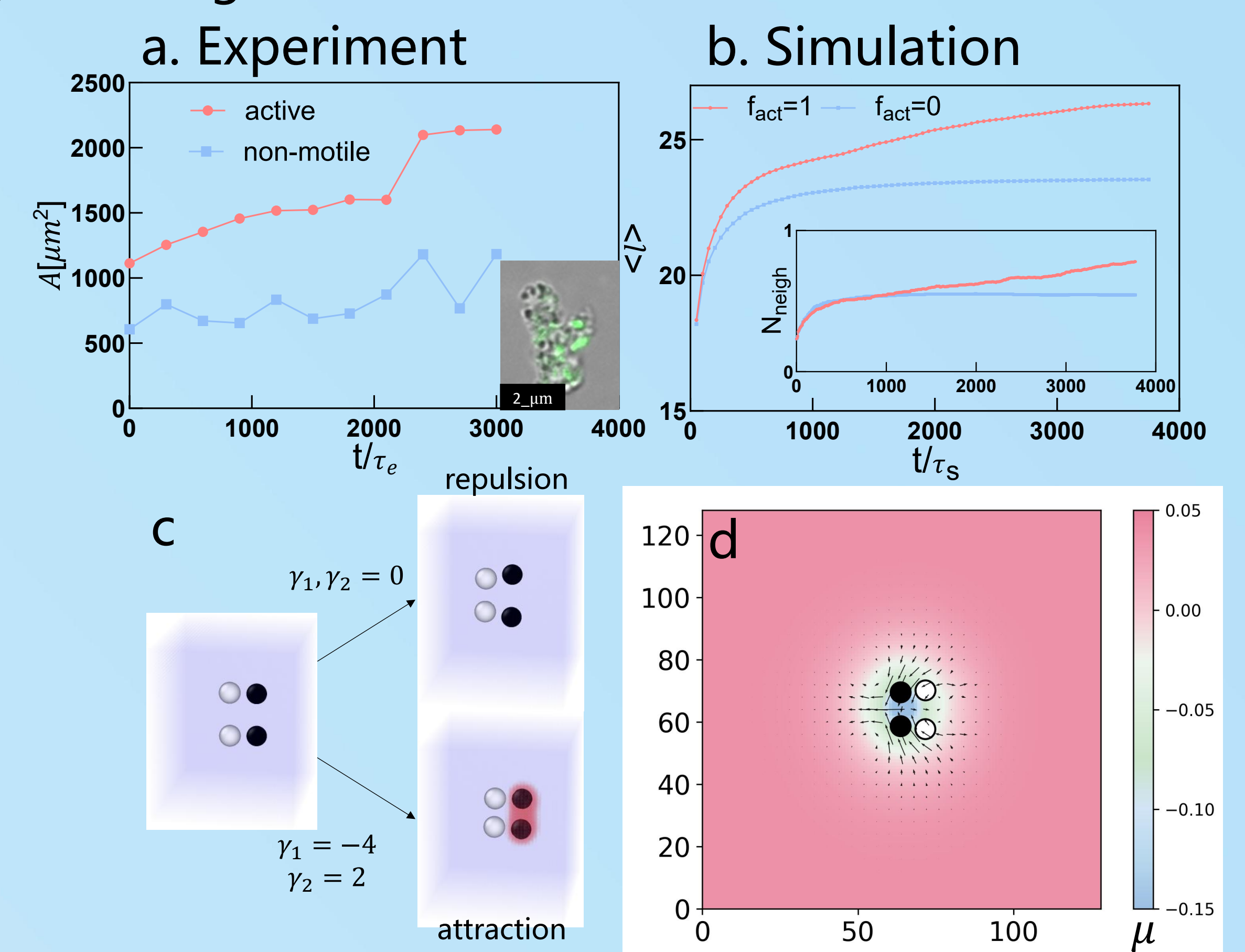


FIG. 4. Dynamic behavior and mechanism of branched bacterial clusters. (a) Experimental time evolution of the DEX-phase cluster area A , showing accelerated phase-separation kinetics under activity. Inset: magnified confocal image of a branched bacterial cluster. (b) Simulated evolution of the ψ -based domain size $\langle \ell \rangle$. Inset: time evolution of the average neighbor number N_{neigh} . (c) Two-bacterium simulation showing that attractive interactions arise only when wetting affinity is present; without affinity, no attraction is observed. (d) Heat map of the chemical potential μ overlaid with the velocity field v , revealing solvent fluxes directed toward the inter-bacterial region that forms a capillary bridge.

Conclusion

Activity exerts a dual influence on phase separation.

At high DEX fraction, active flows repel droplets, suppressing coarsening. At low DEX fraction, the same activity promotes capillary aggregation and accelerates cluster growth. This duality reveals a general mechanism by which motility, wetting, and LLPS are strongly coupled, enabling active control of phase-separation morphology and kinetics using living agents.