J. Indian Inst. Sci.
A Multidisciplinary Reviews Journal
ISSN: 0970-4140 Coden-JIISAD

REVIEW ARTICLE

Swarming in Bacteria: A Tale of Plasticity in Motility Behavior

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Abstract | One of the most fascinating sights in nature is to witness certain insects, birds, and fish move together in a very coordinated and precise fashion for food search, to avoid predation and for migration. The collective movement is called swarming. In 1885, Gustav Hauser, a German pathologist discovered collective movement in a bacterium he later named *Proteus mirabilis* (Armbruster and Mobley in, Nat Rev Microbiol 30: 186–194, 2013). It was not until 1972 when this mode of bacterial movement was characterized and classified by Henrichsen (Bacteriol Rev 36: 478–503, 1972). Several bacteria are now known to exhibit swarming. Here we describe the how and why of swarming with a focus on plasticity.

1 What is Swarming?

Swarming in bacteria can be defined as the locomotion of a population of flagellated bacteria on a semi-solid surface. To understand swarming, it is important to understand other modes of movements that most bacteria employ (Fig. 1). In liquid growth media, also called the planktonic phase, a bacterium can swim using the rotation of its **flagella**. When it comes to solid surfaces, bacteria can use a wide range of modes for motility. These include sliding which is a passive spreading of cells due to the push from the dividing cells; gliding—the use of focal adhesion complexes to attach to the surface to move—and twitching—**pilus** retraction as a means of pulling itself forward. These modes of movement (except sliding) are single-cell motilities and this is where swarming stands apart. Swarming is a mode of motility on semi-solid surfaces where the cells make use of flagella and by far it is the fastest mode of motility bacteria use on a surface[2]. The striking feature of the swarming motility is that unlike the other modes, swarming is a **quorum-sensing** dependent collective movement of cells in many if not all swarming bacteria. A list of swarming bacteria with different features is presented in Table 1.

2 Swarm Patterns and Plasticity

Swarming bacteria are recognizable by the pattern they produce during swarming as shown for a few in Fig. 2. *Proteus mirabilis* has unique swarming behavior. Unlike other species that have a single initiation into the swarming phase, *P. mirabilis* proceeds through iterative swarming and consolidation (**dedifferentiation**) steps to create a bulls-eye pattern of colony[3] (Fig. 2). *Pseudomonas aeruginosa* forms a swarm composed of tendrils or dendrites. *Paenibacillus dendritiformis* forms a curly branched pattern. *Escherichia coli* and *Rhizobium etli* show no pattern. *Bacillus subtilis* forms a featureless mat.

The **plasticity** of swarm patterns has been investigated at the phenomenological and molecular levels in *P. aeruginosa*. This bacterium does not swarm on nutrient-rich media such as brain heart infusion agar or Luria–Bertani agar, but it can swarm easily on minimal media or peptone growth media (Fig. 4); however, the number of dendrites and area coverage are dependent on media used[4]. Tendrils in *P. aeruginosa* swarms can sense other tendrils and change the direction of movement[5]. Recent studies also show that tendrils of a *P. aeruginosa* swarm can avoid non-biological and inert obstacle reflecting the plasticity of swarming[6]. Bacteria taken from non-swarming (nutrient-rich) media can easily form dendrites when introduced to minimal medium (unpublished observation from Varsha Singh) suggesting adaptability of the bacteria. It is believed that a trace element present in rich media prevents swarming. For example, iron limitation induces swarming in *P. aeruginosa* while an excess of iron

Flagellum: A lash-like locomotory appendage extending from the cell membrane. It is associated with a motor protein for rotation, driven by a proton or sodium ion pump.

Pilus (plural pili): A hair-like appendage on the bacterial cell surface to facilitate adhesion, infection, and conjugation.

Quorum sensing: A mechanism by which bacterial populations regulate gene expression or behavior depending upon cell density in the population.

Dedifferentiation: Reversal of cell type to its previous form.

Plasticity (or phenotypic plasticity): "The ability of an individual organism to alter its phenotype (observable traits of an organism) in response to changes in environmental conditions"[73].

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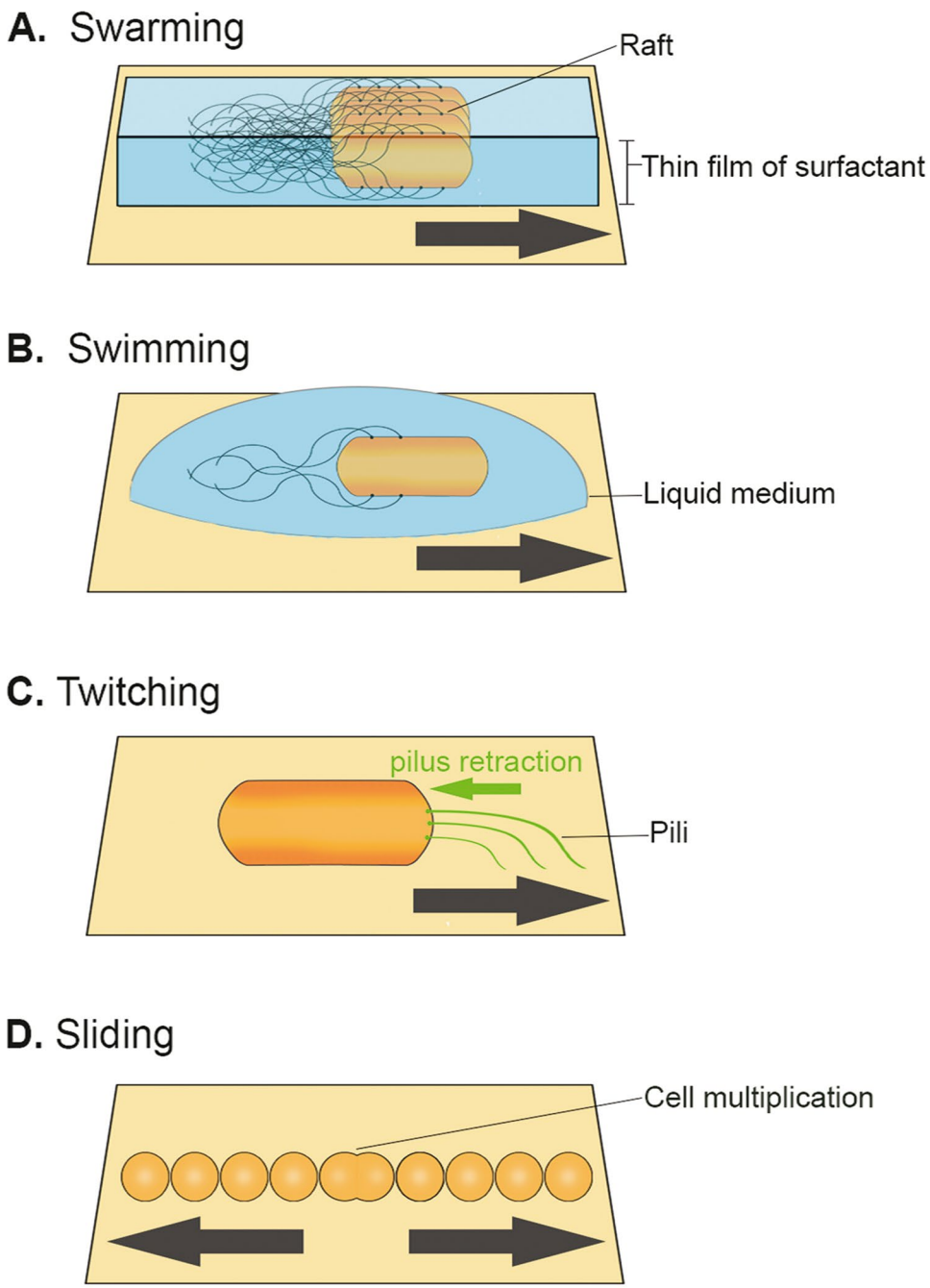

Figure 1: Modes of locomotion in bacteria.

can suppress swarming[7]. Phosphate limitation can also induce swarming[8]. There is evidence to suggest that the number of flagella at the pole of *P. aeruginosa* rod-shaped cells determines the pattern of swarming. Mutation in *fleN*, a regulator of flagella number, can generate 2–5 flagella per cell and causes loss of dendrites in a *P. aeruginosa* swarm. While non-flagellated mutants are non-swarmers, multi flagellated *fleN* mutants have a swarm pattern resembling that of *E. coli* and are classified as hyperswarmers[9]. The ratio of non-flagellated and flagellated cells can determine swarm patterns[10]. Studies in *P. aeruginosa* indicate that both environmental and genetic perturbations can influence swarming motility in this bacterium making it a plastic behavior. As discussed later, plasticity allows the bacterium to swarm over antibiotics and to spread when the host is compromised.

Table 1: Some of the swarming bacteria with their flagella type, quorum sensing requirement, and surfactant.

Name	Flagella type in swarming	Quorum sensing during swarming	Surfactant
Proteus spp.	Peritrichous[41]	Yes[19]	capsular polysaccharide
Salmonella spp.	Peritrichous[42]	?	LPS (?)[27]
Bacillus subtilis	Peritrichous[43]	Yes[19]	Surfactin25
Escherichia coli	Peritrichous[42]	Yes[19]	LPS (?)[27]
Pseudomonas aeruginosa	Lophotrichous[16]	Yes[19]	Rhamnolipid, HAA[16, 26]
Serratia marscescens	Peritrichous[20]	Yes[19]	Serrawettin[24]
Vibrio spp.	Peritrichous[44]	Yes[19]	?
Burkholderia dolosa	Polar/Peritrichous[45]	Yes[23]	Rhamnolipid[23]
Yersinia enterocolitica	Peritrichous[46]	Yes[47]	?
Aeromonas spp.	Peritrichous[44]	?	?
Azospirillum spp.	Peritrichous[44]	Yes[48]	?
Clostridium tetani	Peritrichous[49]	?	?
Chromobacterium spp.	Peritrichous[50]	?	?
Rhizobium leguminosarum	Subpolar[51]	Yes[22]	Long-chain N-acyl homoserine lactones[22]
Myxobacteria	No flagella[52]	?	

3 Substratum and Swarming

The bacterial cells undergo **differentiation** for swarming upon a semi-solid surface. The solid nature of the surface is crucial for this motility. In the laboratory, swarming is observed on a bacterial growth medium solidified with a moderate concentration of agar, though a few species can swarm on a higher concentration of agar (up to 3%)[11]. Swarmer bacteria can be classified into two groups on the basis of the percentage of agar it can swarm on. Robust swarmers can swarm across hard agar surfaces of up to 3% agar while temperate swarmers can swarm only on softer agar surface (0.5–0.8% agar). *Proteus mirabilis* and *Vibrio* spp. are robust swarmers while *Escherichia coli, Salmonella* spp., *Bacillus* spp., and *Pseudomonas* spp. are temperate swarmers[12]. On a single swarming surface, different bacteria can produce different patterns of the swarm (Fig. 2). It is also dependent on media used[4] (Fig. 4). This suggests that swarming pattern is rather plastic.

How do bacteria sense the substratum/surface to switch their physiology and morphology to become swarm proficient? In *V. parahaemolyticus*, the role of flagella in sensing appears to be important. When cells encounter increased viscosity, the flagella rotation is restricted and there is a decrease in sodium-ion flow into flagellar motor proteins. The reduction of flagellar rotation triggers the induction of lateral flagella. Hence flagella serve a dual role of motility appendage and **dynamometer**[13]–[15]. Interestingly, in *P. aeruginosa*, type IV pili are also required for swarming[16]. Type IV pili are involved in surface sensing[17] and they may perform the same function on the swarm surface or they facilitate locomotion.

4 Swarm Lag

When cells are transferred from the planktonic phase to a soft agar surface, there is a period of growth due to expansion (sliding) where the cells multiply and grow as a circular colony without a recognizable swarm pattern. This period is called the swarm lag. The duration of swarm lag depends on the species and the nutrient media used[4]. It is believed that the swarm lag is used to produce wetting agents or surfactants which facilitate the movement of a population of cells since the surface tension on the agar surfaces is not suitable for the movement of the cells. In many cases, the **surfactant** production is dependent on reaching a certain cell density and quorum sensing[18]. Swarm lag can be reduced with an increase in cell density transferred to swarm agar plate[18]. This suggests that a specific threshold of cells is required to initiate swarming motility. This could be in turn due to insufficient wetting agent or surfactant production, which is under the regulation of quorum

Differentiation: A process by which a cell changes from one type to another with a defined function.

Dynamometer: A device used to measure force, torque or power.

Surfactant: Molecules which lower the surface tension between a liquid and another phase.

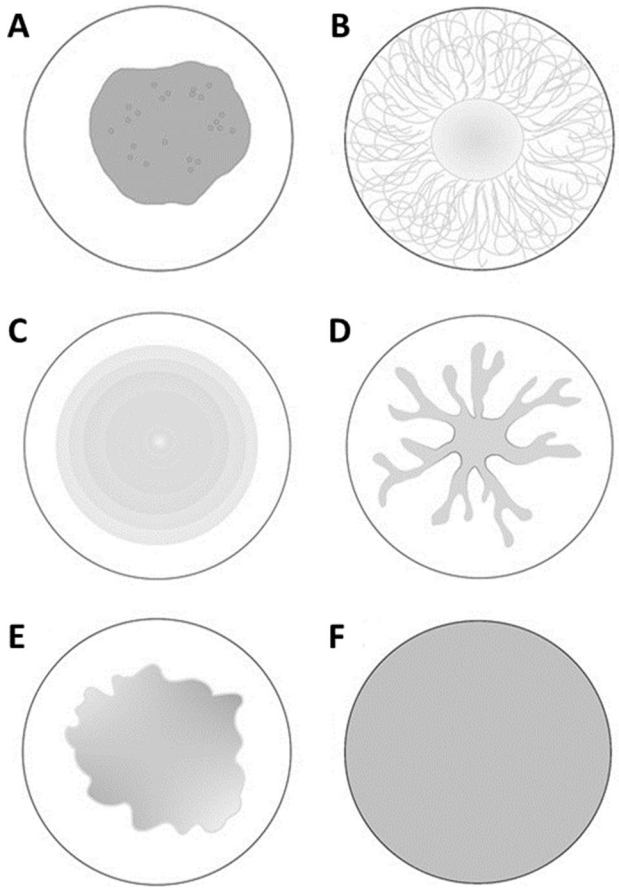

Figure 2: Examples of swarm patterns seen in nature. Swarm patterns of **a** *Escherichia coli*, **b** *Paenibacillus dendritiformis*, **c** *Proteus mirabilis*, **d** *Pseudomonas aeruginosa*, **e** *Rhizobium etli*, **f** *Bacillus subtilis*.

sensing[19–23]. Hence, the bacterial cells collectively secrete surfactant molecules to modify the surface properties. Providing purified surfactant to a freshly spotted *B. subtilis* significantly reduced their swarm lag[18], this could mean that there is a critical amount of surfactant, not cell density per se, required to initiate swarming. An increase in cell density could allow the population to reach the critical surfactant amount quickly.

The surfactant molecules, mostly glycolipids or lipopeptides, vary from species to species. These include surfactin produced by *B. subtilis;* serrawettin by *Serratia* spp.; long-chain *N*-acyl homoserine lactones by *Rhizobium* spp., capsular polysaccharide by *Proteus mirabilis*; rhamnolipid and its precursor HAA (3-(3-hydroxyalkanoyloxy) alkanoic acid) by *Pseudomonas aeruginosa*[16, 18, 24–26]. LPS-O-antigen is believed to be used by *Salmonella* spp. and *E. coli*[27]. In many species, surfactant production is under the regulation of quorum sensing[19–23]. In *P. mirabilis*, RsbA a sensory protein is involved in sensing the environment to initiate swarming motility[3]. In *Serratia*, an autoinducer identified as *N*-butanoyl-L-homoserine lactone (BHL) is involved in quorum sensing. In *P. aeruginosa* also quorum sensing is a core requirement to initiate swarming and surfactant production[28]. In all, swarm lag is used by different bacteria to generate surfactant using a number of strategies, most relying on reaching a high cell density.

5 Heterogeneity in Swarming Population: Cell Elongation

One notable difference in swarming and swimming cells is that the former is longer. In *B. subtilis,* the cell aspect ratio of 4.9, of **wild type** cells, supports effective swarming when compared with mutants of either lower or higher aspect ratios[29]. Cells can become elongated due to inhibition of cell division[30]. The proliferation of cells happens in swarmer cells of *V. parahaemolyticus* in a length-dependent fashion. The shorter cells divide in the middle while longer cells divide at a

Wild type: A phenotype and genotype (genetic makeup) t found in nature and used as a reference in research.

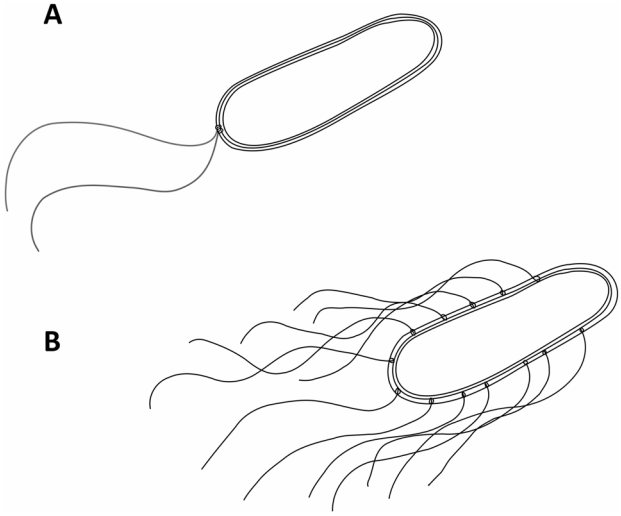

Figure 3: Types of flagella. **a** Lophotrichous, **b** Peritrichous.

distance away from the middle ensuring that there are enough elongated cells after the divisions[31]. This ensures that elongated cells have more area to accommodate lateral flagella and adequate cell-to-cell contact to increase swarming efficiency[32]. In *P. mirabilis*, the length of individual cells increased with an increase in agar concentration and with enhanced population migration[33]. Swarming proficiency associated with cell elongation comes with a trade-off, the elongated cells have a higher susceptibility to osmotic pressure and antibiotics due to changes in thickness and composition of peptidoglycan cell walls[34]. In general, cell elongation is more common in robust swarmers than in temperate swarmers[12].

6 Heterogeneity in Swarming Population: Modification of Flagella

As motility is a requirement of swarming, flagella being the major motility appendage in bacteria undergoes several modifications during swarming. Swarm proficient cells have additional **peritrichous** flagella in *Aeromonas caviae*[35] or a second polar flagellum (**lophotrichous**) in *P. aeruginosa*[16, 36] (Fig. 3). The latter can also produce a specialized set of stator motors associated with flagella[36] that enhance the propeller function of the flagellum. These modifications help swarmer cells to overcome surface friction and might also help in hydration[37]. The morphological changes in bacteria upon exposure to swarming agar is important for their differentiation into swarm proficient cells. When differentiated cells are directly transferred onto a new surface, they swarm without any lag[18]. These cells are able to revert to swimmers when exposed to liquid media[38].

In many bacteria, regulators of flagella are upregulated upon exposure to a solid surface and overexpression of such a regulator can reduce the swarm lag. In *E. coli,* the presence of additional flagella makes cells swarm proficient without a lag[39]. The experimental evolution of *P. aeruginosa* produces hyperswarmer populations with a mutation in a flagella synthesis regulator called fleN[9]. Many bacteria produce additional flagella during swarm lag[13, 18, 40].

7 Chemotaxis and Swarming

Chemotaxis is the movement of organisms to or away from chemicals. Swimming is chemotaxis-driven motility. The cells swim towards nutrients and away from harmful substances. Chemotaxis is an essential component of swarming in certain species. In *Myxococcus xanthus*, the rate of swarm expansion increases in presence of prey[53]. In *Vibrio parahaemolyticus* and *Serratia marcescens*, iron limitation induces swarming and the mutation in iron acquisition genes affects swarming[54, 55]. Swarming is induced when certain amino-acids like glutamate, aspartate, histidine, or proline are provided as the sole nitrogen source in the agar media[16]. In *E. coli*, chemotaxis systems sensing serine, aspartate and maltose are required for swarming, while the identity of **chemoeffectors** remains unknown[56]. Many other species also show a similar requirement of chemotaxis

Peritrichous: Arrangement of flagella on all sides of a rod-shaped cell, usually along the length of the cell (Fig. 3).

Lophotrichous: Multiple flagella arise from a single point of the cell, usually at the polar end (Fig. 3).

Chemoeffector: An attractant or repellent molecule to which the chemotaxis system responds.

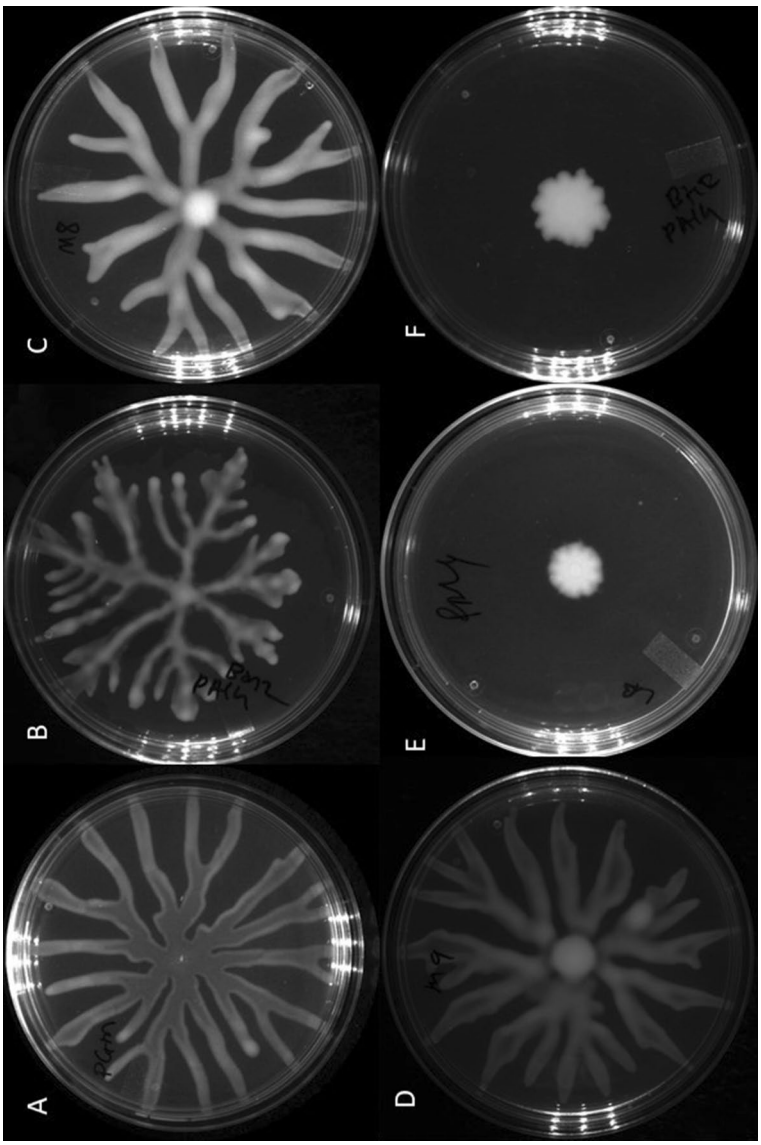

Figure 4: Media dependent plasticity in swarming in *Pseudomonas aeruginosa* PA14. A 24 h swarm on **a** Peptone growth medium (PGM)-0.6% agar, **b** BM2 minimal medium-0.6% agar, **c** M8 minimal medium-0.6% agar, **d** M9 minimal medium-0.6% agar, **e** Luria–Bertani (LB)-0.6% agar and **f** Brain heart Infusion (BHI)-0.6% agar.

systems[18, 57]. Some exceptions to this rule exist in *P. mirabilis* and *Rhodospirillum centenum* where swarming appears to be unaffected by mutations in chemotaxis systems[58, 59].

8 Collective Movement in a Swarm

In a swarming population, peritrichous flagella on individual cells bundle together and rotate in a counter-clockwise direction to push the fluid behind. The cells swarm in a thin film of surfactant with a height close to the cell width on the agar surface[60] (Fig. 1). Frequently, a cell can undergo a tumble when a single flagellum rotates in the opposite direction opening the flagellar bundle and changing the orientation of the cell. This can play a role in short-range interactions[61].

Bacteria in a moving swarm also orient their flagella to form a packet of cells called a **raft**, with the cells having the same direction and motility (Fig. 1). In the swarm interior, each cell tries to swim straight but collides with other cells. These collisions keep the population in a consistent direction[62] leading to the formation of rafts. Cells move in and out of rafts continuously. Raft formations promote swarming as the cells isolated from these rafts becomes non-motile but regain motility when returned to the raft[2, 63]. This evidence again supports the fact that swarming is indeed a collective movement. In *B. subtilis*, extracellular proteolytic activity contributes to swarming. Though the exact reason for this is not known, it is speculated that proteolytic cleavage of certain cell surface proteins enhances cell-to-cell interactions and raft formation[64].

Microscopy studies on *E. coli* show that at the edge of swarm there are cells that are jammed and stalled[32, 62, 65]. These non-motile cells continue to rotate their flagella to pump fluid to the virgin agar ahead[32]. This layer is followed by highly motile cells in the interior which sometimes dart through the layer of jammed and stalled cells only to become a part of it. When stalled cells in the lagging layers encounter moving cells, they reverse their direction by switching the direction of flagellar rotation (without changing the cell orientation), join the motile layer and regain their forward motility[32, 62]. Modeling based on experimental observations in *E. coli* suggests that the flow of wetting fluid around the swarm in a clockwise direction could be used for long-range communication. This constitutive flow is speculated to be achieved by the rotation of flagella sticking outward from the swarm[60].

The movement of the cells in a population of *B. subtilis* and *S. marcescens* follows super-diffusion consistent with Levy walks[66]. It is defined as a movement composed of longer displacements between shorter localized movements. This is likely to be facilitated by the collective flow of the population, rather than a single cell-based mechanism. This movement is widely used to describe foraging mechanisms in other systems[66]. Bacteria could also use swarming as a foraging mechanism along with other goals.

Is swarming a surface phenomenon? Can swarming happen in the planktonic or liquid phase of bacterial growth? These questions often initiate discussions on the physics of coordinated motion in liquid and on dilution effects in the liquid phase. Swimming motility of bacteria in the liquid phase as well as swarming motility on surface have shared features such as the use of flagella. Unlike swarming, swimming does not depend on quorum or on surfactant. However, bacteria can produce quorum signals as well as some surfactants in liquid phase making swarming a possibility in the liquid phase of growth. However, a number of things might make swarming hard to achieve in liquid phase: (i) surface sensing might be necessary to achieve threshold level of surfactant, (ii) surfactant can readily diffuse or dilute away in liquid phase and (iii) random collision required for raft formation is less likely to happen in three-dimensional liquid phase. Molecular and system-level understanding of swarming in different bacteria such as the effect of surface sensing on flagella, raft formation, and surfactant production will enhance our understanding.

Raft: An arrangement of bacterial cells along their length.

9 Division of Labor in a Swarm

In a swarm, the population at the center of the colony have more **vegetative cells** compared to elongated, multi-flagellated cells at the growing edges. This is also reflected in the level of gene expression[67]. Cells at the center express genes involved in oxidative and copper stress responses, while those at the edges show upregulation of genes involved in energy metabolism including components of electron transport chain and ATP production. This along with the observations of **biofilm**-like biomass at the center of *P. aeruginosa* swarm suggests that cells at the edges take up the role of expanding into new surfaces, while the vegetative cells establish new settlements[67]. Another study suggests that shorter cells (presumably undifferentiated) of *V. parahaemolyticus* are released from flooded swarm colonies into a liquid environment, but the longer cells remain stuck to the surface. Thus, swarm colonies can

Vegetative cells: Refers to the cells which have not differentiated to swarming state or which has dedifferentiated from a swarming state.

Biofilm: A group of cells adhering to each other using secreted extracellular matrix to form a functional community. It can evade the immune system of hosts and share nutrients.

serve as a source of cells for colonisations elsewhere[68].

10 Advantages of Swarming

Proteomic and lipidomic studies comparing swimmer and swarmer populations of *Paenibacillus polymyxa* show upregulation of genetic regulators involved in signaling, antibiotic resistance and surfactant production in swarming populations[69]. Swarming *P. aeruginosa* shows resistance to as many as 13 different antibiotics[70]. This resistance is believed not to be an intrinsic ability of the cells, but an effect of multicellularity and rapid movement. Upon encountering an antibiotic patch, the swarming population continues to swarm such that a small fraction of the population dies acting as a shield for the rest of the population[11]. **Virulence** and invasion capabilities of *P. mirabilis* have also been attributed to its swarming behaviour[71]. Swarming *P. aeruginosa* also shows upregulation of virulence-related genes[72]. Effect of swarming on antibiotic resistance and virulence has therapeutic implications in the field of medicine.

Virulence: Ability of a microbe to infect and harm a host organism.

11 Future Perspectives

A wide range of bacterial species engage in swarming motility. An increase in speed, collective antibiotic resistance, and virulence-associated with swarming suggests that swarming motility might have evolved in bacteria to impart survival advantage in a competitive environment. Specific chemotactic cues for swarming and external factors that influence this behavior are of clinical and medical importance. Bacterium specific pattern of swarming suggests that continued investigation will lead to the discovery of additional environmental factors, flagella modulation, and surfactant production strategies. Patterns of swarming also present an interesting problem for understanding and modeling of fluid flow, resource allocation and optimal utilization of space by bacteria on a swarming surface.

Publisher's Note

Acknowledgement

We thank M.S. Akhil and Sandeep Xavier for illustrations. We thank S. Joge and M.A. Kollaran for images of *P. aeruginosa* swarm.

Received: 7 May 2020 Accepted: 9 May 2020
Published online: 28 May 2020

References

1. Armbruster CE, Mobley HLT (2013) Merging mythology and morphology: the multifaceted lifestyle of *Proteus mirabilis*. Nat Rev Microbiol 30:186–194
2. Henrichsen J (1972) Bacterial surface translocation: a survey and a classification. Bacteriol Re. 36:478–503
3. Belas R, Schneider R, Melch M (1998) Characterization of Proteus mirabilis precocious swarming mutants: identification of rsbA, encoding a regulator of swarming behavior. J Bacteriol 180:6126–6139
4. Kollaran AM et al (2019) Context-specific requirement of forty-four two-component loci in *Pseudomonas aeruginosa* swarming. iScience 13:305–317
5. Caiazza NC, Shanks RMQ, O'Toole GA (2005) Rhamnolipids modulate swarming motility patterns of *Pseudomonas aeruginosa*. J Bacteriol 187:7351–7361
6. Kotian B, Abdulla A, Hithysini K, Harkar S, Joge S, Mishra A, Singh V, Varma MM (2020) Active modulation of surfactant-driven flow instabilitiesby swarming bacteria. Phys Rev E 101(1):012407
7. Boyle KE, Van Ditmarsch D, Deforet M, Xavier JB (2015) Integration of metabolic and quorum sensing signals governing the decision to cooperate in a bacterial social trait. PLoS Comput Biol 11:e1004279
8. Bains M, Fernández L, Hancock REW (2012) Phosphate starvation promotes swarming motility and cytotoxicity of *Pseudomonas aeruginosa*. Appl Environ Microbiol 78:6762–6768
9. van Ditmarsch D et al (2013) Convergent evolution of hyperswarming leads to impaired biofilm formation in pathogenic bacteria. Cell Rep. 4:697–708
10. Boyle KE et al (2017) Metabolism and the evolution of social behavior. Mol Biol Evol 34:2367–2379
11. Butler MT, Wang Q, Harshey RM (2010) Cell density and mobility protect swarming bacteria against antibiotics. Proc Natl Acad Sci USA 107:3776–3781
12. Partridge JD, Harshey RM (2013) Swarming: flexible roaming plans. J Bacteriol 195:909–918
13. Belas R, Simon M, Silverman M (1986) Regulation of lateral flagella gene transcription in *Vibrio parahaemolyticus*. J Bacteriol 167:210–218
14. McCarter L, Hilmen M, Silverman M (1988) Flagellar dynamometer controls swarmer cell differentiation of *V. parahaemolyticus*. Cell 54:345–351
15. Kawagishi I, Imagawa M, Irnae Y, McCarter L, Homma M (1996) The sodium-driven polar flagellar motor of marine Vibrio as the mechanosensor that regulates lateral flagellar expression. Mol Microbiol 20:693–699
16. Köler T, Curty LK, Barja F, Van Delden C, Pechére J-C (2000) Swarming of *Pseudomonas aeruginosa* is dependent on cell-to-cell signaling and requires flagella and pili. J Bacteriol 182:5990–5996

17. Maier B, Wong GCL (2015) How bacteria use type IV pili machinery on surfaces. Trends Microbiol 23:775–788
18. Kearns DB, Losick R (2003) Swarming motility in undomesticated *Bacillus subtilis*. Mol Microbiol 49:581–590
19. Daniels R, Vanderleyden J, Michiels J (2004) Quorum sensing and swarming migration in bacteria. FEMS Microbiol Rev 28:261–289
20. Eberl L, Molin S, Givskov M (1999) Surface motility of *Serratia liquefaciens* MG1. J Bacteriol 181:1703–1712
21. Ochsner UA, Koch AK, Fiechter A, Reiser J (1994) Isolation and characterization of a regulatory gene affecting rhamnolipid biosurfactant synthesis in *Pseudomonas aeruginosa*. J Bacteriol 176:2044–2054
22. Daniels R et al (2006) Quorum signal molecules as biosurfactants affecting swarming in *Rhizobium etli*. Proc Natl Acad Sci USA. 103:14965–14970
23. Nickzad A, Lépine F, Déziel E (2015) Quorum sensing controls swarming motility of burkholderia glumae through regulation of rhamnolipids. PLoS One 10:e0128509
24. Hara-hotta H, Yano I (1992) A novel extracellular cyclic lipopeptide which promotes flagellum-dependent and -independent spreading growth of Serratia marcescens. J Bacteriol 174:1769–1776
25. Arima K, Kakinuma A, Tamura G (1968) Surfactin, a crystalline peptidelipid surfactant produced by *Bacillus subtilis*: Isolation, Characterization and its inhibition of fibrin clot. Appl Environ Microbiol 31:488–494
26. Déziel E, Lépine F, Milot S, Villemur R (2003) rhlA is required for the production of a novel biosurfactant promoting swarming motility in *Pseudomonas aeruginosa*: 3-(3-hydroxyalkanoyloxy)alkanoic acids (HAAs), the precursors of rhamnolipids. Microbiology 149:2005–2013
27. Toguchi A, Siano M, Burkart M, Harshey RM (2000) Genetics of swarming motility in *Salmonella enterica Serovar typhimurium*: critical role for lipopolysaccharide. J Bacteriol 182:6308–6321
28. Yeung ATY et al (2009) Swarming of *Pseudomonas aeruginosa* is controlled by a broad spectrum of transcriptional regulators, including MetR. J Bacteriol 191:5592–5602
29. Ilkanaiv B, Kearns DB, Ariel G, Beer A (2017) Effect of cell aspect ratio on swarming bacteria. Phys Rev Lett 118:1–5
30. Howery KE, Clemmer KM, Emrah S (2015) Regulation of the min cell division inhibition complex by the Rcs phosphorelay *in Proteus mirabilis*. J Bacteriol 197:2499–2507
31. Muraleedharan S, Freitas C, Mann P, Glatter T, Ringgaard S (2018) A cell length-dependent transition in MinD-dynamics promotes a switch in division-site placement and preservation of proliferating elongated *Vibrio parahaemolyticus* swarmer cells. Mol Microbiol 109:365–384
32. Turner L, Zhang R, Darnton NC, Berg HC (2010) Visualization of flagella during bacterial swarming. J Bacteriol 192:3259–3267
33. Little K, Austerman J, Zheng J, Gibbs KA (2019) Cell shape and population migration are distinct steps of proteus mirabilis swarming that are decoupled on high-percentage agar. J Bacteriol 201:1–15
34. Auer GK et al (2019) Bacterial swarming reduces *Proteus mirabilis* and *Vibrio parahaemolyticus* cell stiffness and increases β-Lactam susceptibility. MBio 10:e00210–19
35. Kirov SM et al (2002) Lateral flagella and swarming motility in Aeromonas species. J Bacteriol 184:547–555
36. Kuchma SL et al (2015) Cyclic Di-GMP-mediated repression of swarming motility by *Pseudomonas aeruginosa* PA14 requires the MotAB stator. J Bacteriol 197:420–430
37. Partridge JD, Harshey RM (2013) More than motility: *Salmonella flagella* contribute to overriding friction and facilitating colony hydration during swarming. J Bacteriol 195:919–929
38. Alberti L, Harshey RM (1990) Differentiation of Serratia marcescens 274 into swimmer and swarmer cells. J Bacteriol 172:4322–4328
39. Kearns DB, Losick R (2005) Cell population heterogeneity during growth of *Bacillus subtilis*. Genes Dev 19:3083–3094
40. Hoeniger JFM (1964) Cellular changes accompanying the swarming of *Proteus mirabilis*. Can J Microbiol 10:1–9
41. Tuson HH, Copeland MF, Carey S, Sacotte R, Weibel B (2013) Flagellum density regulates *Proteus mirabilis* swarmer cell motility in viscous environments. J Bacteriol 195:368–377
42. Manson MD, Armitage JP, Hoch JA, Macnab RM (1998) MINIREVIEW bacterial locomotion and signal transduction. J Bacteriol 180:1009–1022
43. Guttenplan SB, Shaw S, Kearns DB (2013) The cell biology of peritrichous flagella in *Bacillus subtili*. Mol Microbiol 87:211–229
44. Mccarter L (2004) Dual flagellar systems enable motility under different circumstances. J Mol Microbiol Biotechnol 7:18–29
45. Roux D et al (2018) A putative lateral flagella of the cystic fibrosis pathogen *Burkholderia dolosa* regulates swimming motility and host cytokine production. PLoS One 13:e0189810
46. Young GM, Smith MJ, Minnich SA, Miller VL (1999) The *Yersinia enterocolitica* motility master regulatory operon, flhDC, is required for flagellin production, swimming motility, and swarming motility. J Bacteriol 181:2823–2833
47. Atkinson S, Chang C-Y, Sockett RE, Cámara M, Williams P (2006) Quorum sensing in *Yersinia enterocolitica* controls swimming and swarming motility. J Bacteriol 188:1451–1461

48. Fukami J et al (2017) Revealing strategies of quorum sensing in *Azospirillum brasilense* strains Ab - V5 and Ab - V6. Arch Microbiol. https://doi.org/10.1007/s00203-017-1422-x
49. Hoeniger JF, Tauschel HD (1974) Sequence of structural changes in cultures of *Clostridium tetani* grown on a solid medium. J Med Microbiol 7:425–432
50. Sneath PHA (1956) The change from polar to peritrichous flagellation in *Chromobacterium spp*. J Gen Microbiol 15:99–105
51. Tambalo DD, Yost CK, Hynes MF (2010) Characterization of swarming motility in *Rhizobium leguminosarum* bv. viciae. FEMS Microbiol Lett 307:165–174
52. Kaiser D, Warrick H (2011) *Myxococcus xanthus* swarms are driven by growth and regulated by a pacemaker. J Bacteriol 193:5898–5904
53. Berleman JE, Kirby JR (2009) Deciphering the hunting strategy of a bacterial wolfpack. FEMS Microbiol Rev 23:1–7
54. Lin CS et al (2016) An iron detection system determines bacterial swarming initiation and biofilm formation. Sci Rep 6:1–13
55. McCarter L, Silverman M (1989) Iron regulation of swarmer cell differentiation of Vibrio parahaemolyticus. J Bacteriol 171:731–736
56. Arshey RAMH (1998) The chemotaxis system, but not chemotaxis, is essential for swarming motility in *Escherichia coli*. Proc Natl Acad Sci 95:2568–2573
57. Sar N, Mccarter L, Simon M, Silverman M (1990) Chemotactic control of the two flagellar systems of *Vibrio parahaemolyticus*. J Vacteriol 172:334–341
58. Jiang Z, Gest H, Bauer CE (1997) Chemosensory and Photosensory Perception in Purple Photosynthetic Bacteria Utilize Common Signal Transduction Components. 179:5720–5727
59. Williams FD, Anderson DM, Hoffman PS, Robert H, Leonard S (1976) Evidence against the involvement of chemotaxis in swarming of *Proteus mirabilis*. J Bacteriol 127:237–248
60. Dauparas J, Lauga E (2016) Flagellar flows around bacterial swarms. Phys Rev Fluids 1:1–26
61. Turner L, Ryu WS, Berg HC (2000) Real-time imaging of fluorescent flagellar filaments. J Bacteriol 182:2793–2801
62. Damton NC, Turner L, Rojevsky S, Berg HC (2010) Dynamics of bacterial swarming. Biophys J 98:2082–2090
63. Morrison RB, Scott A (1966) Swarming of proteus–a solution to an old problem? Nature 211:255–257
64. Connelly MB, Young GM, Sloma A (2004) Extracellular proteolytic activity plays a central role in swarming motility in *Bacillus subtilis*. J Bacteriol 186:4159–4167
65. Copeland MF, Flickinger ST, Tuson HH, Weibel DB (2010) Studying the dynamics of flagella in multicellular communities of *Escherichia coli* by using biarsenical dyes. Appl Environ Microbiol 76:1241–1250
66. Ariel G et al (2015) Swarming bacteria migrate by Lévy Walk. Nat Commun 6:8396
67. Tremblay J, Déziel E (2010) Gene expression in *Pseudomonas aeruginosa* swarming motility. BMC Genom 11:587
68. Freitas C, Glatter T, Ringgaard S (2019) The release of a distinct cell type from swarm colonies facilitates dissemination of *Vibrio parahaemolyticus* in the environment. ISME J. https://doi.org/10.1038/s41396-019-0521-x
69. Poudel S et al (2019) Integrated proteomics and lipidomics reveal that the swarming motility of *Paenibacillus polymyxa* is characterized by phospholipid modification, surfactant deployment, and flagellar specialization relative to swimming motility. Front Microbiol 10:1–16
70. Lai S, Tremblay J, Déziel E (2009) Swarming motility: a multicellular behaviour conferring antimicrobial resistance. Environ Microbiol 11:126–136
71. Hola V, Peroutkova T, Ruzicka F (2012) Virulence factors in Proteus bacteria from biofilm communities of catheter-associated urinary tract infections. FEMS Immunol Med Microbiol 65(2):343–349. https://doi.org/10.1111/j.1574-695X.2012.00976.x
72. Overhage J, Bains M, Brazas MD, Hancock REW (2008) Swarming of *Pseudomonas aeruginosa* is a complex adaptation leading to increased production of virulence factors and antibiotic resistance. J Bacteriol 190:2671–2679
73. Kelly SA, Panhuis TM, Stoehr AM (2012) Phenotypic plasticity: molecular mechanisms and adaptive significance. Compr Physiol 2:1417–1439

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