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Minireview

Sliding on the surface: bacterial spreading without an active motor

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Summary

Bacteria are able to translocate over surfaces using different types of active and passive motility mechanisms. Sliding is one of the passive types of movement since it is powered by the pushing force of dividing cells and additional factors facilitating the expansion over surfaces. In this review, we describe the sliding proficient bacteria that were previously investigated in details highlighting the sliding facilitating compounds and the regulation of sliding motility. Besides surfactants that reduce the friction between cells and substratum, other compounds including exopolysaccharides, hydrophobic proteins, or glycopeptidolipids where discovered to promote sliding. Therefore, we present the sliding bacteria in three groups depending on the additional compound required for sliding. Despite recent accomplishments in sliding research there are still many open questions about the mechanisms underlying sliding motility and its regulation in diverse bacterial species.

Introduction

Most natural habitats of bacteria include abiotic or biotic surfaces like soil particles, the root mantle or even algal clusters in the ocean. Bacteria have therefore developed different mechanisms to move over such substrates, ranging from active appendage-mediated motility to passive spreading. The first landmark classification of bacterial movement types was conducted by Henrichsen in 1972. He examined the movement of over 30 bacterial species on agar plates and classified them into the distinct types of swarming, swimming, gliding, twitching, sliding and darting, although the latter is not used anymore (Henrichsen, 1972). A major reason why his paper is still cited today despite novel findings regarding the underlying mechanisms is the precise definition he provided for each type of movement. These definitions are still practical even though additional criteria were discovered since. Before Henrichsen's article, many of the movement types were just described as swarming or spreading and it was often not distinguished between the different surface colonization modes that exploit an active motor. In addition to the multicellular flagellum-driven swarming, these include type IV pilus-dependent twitching and focal adhesion complex supported gliding (Kearns, 2010). Further, swarming-based collective motility might be facilitated by additional secreted proteins, promoting wandering colony formation during surface colonization (Kobayashi *et al.*, 2016). Contrary to surface colonization that requires active appendages, sliding is defined as a passive bacterial translocation created by expansive forces accelerated by surfactants that reduce surface tension (Henrichsen, 1972; Kearns, 2010). The original definition of sliding also incorporated colony growth (Henrichsen, 1972). However, it was recently recognized that bacterial sliding necessitates more constituents than previously assumed. In this review, we present an overview of the various components required for expansion and describe bacteria in which sliding was investigated in more detail to divide them in three groups according to the so far characterized sliding facilitating machinery (Fig. 1). We are aware that new discoveries of sliding mechanisms could possibly require regrouping of the below discussed bacteria or result in a system with additional groups. Some of the spreading mechanisms characterized as sliding might even have an underlying active part that is not known yet, as suggested before (Shrout, 2015). However, we provide a first attempt of classifying the sliding proficient

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bacteria since the current knowledge of different sliding mechanisms is still very limited and does therefore not allow a more sophisticated system.

In the first group, we describe bacteria that require only the pushing force of cell division and a secreted surfactant (considering state-of-the-art research). Bacterial sliding that involves additional secreted components like exopolysaccharides is illustrated in group II. Finally, group III is composed of bacteria that necessitate growth and another component, but no surfactant. In this review we focus on the bacterial species in which the sliding mechanism was investigated more extensively. Henrichsen (1972) depicted also other species capable of sliding, however, to our knowledge the sliding behaviour of those bacteria was not examined beyond that article or like in case of *Flavobacterium sp*. and *Acinetobacter calcoaceticus* surface spreading was correctly identified as gliding and twitching respectively (Henrichsen, 1984; Shrivastava and Berg, 2015; Shrout, 2015). In the following sections we will describe the sliding behaviour of different bacteria assigned to the above mentioned groups and elaborate about the so far known requirements and regulatory pathways involved.

Growth and surfactant dependent sliding (group I)

The opportunistic pathogen *Pseudomonas aeruginosa* belongs to group I since until now, rhamnolipid biosurfactants were the only secreted components found to be important for sliding (Fig. 2). Murray and colleagues discovered the sliding ability of *P. aeruginosa* when a *fliC pilA* double mutant, designed to be a negative control that can neither swarm nor twitch, was also spreading on semisolid agar plates (Murray and Kazmierczak, 2008). This type of movement was identified as sliding since swimming, swarming and twitching was not possible due to a lack of flagellin (Δ*fliC*) and type IV pili (Δ*pilA*). The requirement of rhamnolipids was confirmed by using a mutant lacking the gene responsible for the production of the rhamnolipid precursor that showed severely decreased sliding (Murray and Kazmierczak, 2008). The regulatory components important for sliding in *P. aeruginosa* overlap with the regulation of swarming and biofilm formation. The regulators identified were the two component system GacA/GacS (see the overview of regulatory pathways related to sliding in Table 1) that regulates swarming motility for which rhamnolipid production is also necessary. GacA/GacS is proposed to indirectly influence the expression of exopolysaccharide genes during biofilm formation. A random transposon mutagenesis experiment of the *fliC pilA* double mutant resulted in mutants with increased sliding behaviour and tendril formation that harboured transposon insertions in *gacA* and *gacS*. As tests with these hyper-sliders (*fliC pilA gacA* triple mutant) showed no difference in rhamnolipid production and the lack of flagella and pili was confirmed, the response regulator GacA seems to target additional yet unknown genes responsible for the hyperslider phenotype (Murray and Kazmierczak, 2008). The transposon mutagenesis also revealed another regulator, RetS to be involved in sliding. The role of RetS during sliding was not investigated further, however, rhamnolipid production in the *fliC pilA retS* triple mutant was not reduced (Murray and Kazmierczak, 2008). The third type of regulatory pathway discovered to play a role in sliding included cyclic di-GMP. The SadC and BifA enzymes are responsible for cyclic di-GMP synthesis and degradation respectively. Similar to swarming, overexpression of *sadC* inhibited sliding whereas overexpression of *bifA* resulted in increased sliding (Kuchma *et al*., 2007; Merritt *et al*., 2007; Murray and Kazmierczak, 2008). In conclusion, it is possible that additional factors under control of the revealed regulators are also involved in sliding but not identified yet.

Another organism from the same genus, *Pseudomonas syringae* pv. tomato DC3000 is also capable of sliding over semi-solid surfaces with the help of a surfactant. *P. syringae* uses another lipopeptide, syringafactin to reduce the surface tension and thereby facilitate passive movement. While investigating the regulation of motility in *P. syringae*, Nogales *et al*. discovered that a *fleQ* mutant lacking the proposed master regulator of motility can spread over semi-solid agar in a distinct pattern despite lacking flagella (Nogales *et al*., 2015). This spreading was proposed to be sliding motility based on its flagellum-independency. Further, the essentiality of syringafactin was demonstrated using a double mutant (lacking the first gene of the operon encoding enzymes for syringafactin production, *syfA* next to *fleQ*) that was unable to spread. Interestingly, the *fliC* mutant lacking only flagellin was unable to spread in comparison to the *fleQ* mutant which was explained by the difference in syringafactin production level: the authors discovered that the amount of syringafactin is 40% higher in the *fleQ* mutant compared with *fliC* mutant and wild-type (Nogales *et al*., 2015). RNA-seq experiments revealed that the expression of the *syf* operon and of *syfR*, the transcriptional regulator presumably activating the *syf* operon, is upregulated in the *fleQ* mutant. These results suggest a negative regulation of *syfR* and therefore also of the *syf* operon by FleQ. Additionally, in plant experiments the *fleQ syfA* double mutant showed diminished disease symptoms suggesting that sliding contributes to *P. syringae* colonization of the leaf surface, a habitat where flagella-dependent movement might not be optimal (Nogales *et al*., 2015).

Similarly, a *fleQ* mutant of the plant-growth promoting bacterium *Pseudomonas fluorescens* SBW25 was also

Table 1. Known sensing and regulatory components and their targets for several bacteria described in this review.

Organism	Known sensing and regulatory components	Target genes/operons
P. aeruginosa	GacA/GacS two component system	*rhlAB*
	RetS	?
	cyclic di-GMP	
P. syringae pv tomato DC3000	FleQ-dependent inhibition	*syfR* operon
S. marcescens	ExpR	EPSII
B.s subtilis	KinB/C dependent Spo0A phosphorylation	*epsA-O*
S. enterica serovar Typhimurium	PhoP/PhoQ two component system	*pagM*

discovered to exhibit sliding motility on semi-solid medium with the same colony morphology. Here, the sliding facilitating compound was identified to be the surfactant viscosin (Alsohim *et al*., 2014).

Likewise, *Serratia marcescens* is one of the organisms where sliding was found to be dependent on the pushing force of cell division and a secreted surfactant without other sliding facilitating compounds being revealed so far. This ubiquitous gram-negative enteric bacterium was shown to translocate over agar surfaces in the passive manner characteristic for sliding under conditions that do not allow flagellum dependent movement (i.e. high agar concentration)(Matsuyama *et al*., 1992). Matsuyama and colleagues discovered that the movement of *S. marcescens* is dependent on the lipopeptide surfactant Serrawettin since mutants unable to produce it were not able to spread across the surface (Matsuyama *et al*., 1992). Further, they showed that non-flagellated mutants could also spread over plates with a low agar concentration usually used to observe flagellum-dependent movement (Matsuyama *et al*., 1995). Spreading was abolished when the strain was defective for Serrawettin production but was restored with exogenously supplied Serrawettin. Notably, not only Serrawettin promoted spreading but also several surfactants of other bacterial species were able to complement Serrawettin defective *S. marcescens* strains (Matsuyama *et al*., 1995). This suggests a purely functional role of lowering the surface tension to promote movement. As surface colonization was not dependent on flagella or chemotaxis components, the authors suggested a passive type of spreading which is fitting to the definition of sliding.

The gram-negative pneumonia-causing bacterium *Legionella pneumophila* also belongs to the group of bacteria that slide over surfaces with the help of a surfactant. *L. pneumophila* can spread over semi-solid agar plates in a 'lobed, wavelike pattern' as well as many other *Legionella* species (Stewart *et al*., 2009). This behaviour was the first observation of surface translocation in *L. pneumophila* and was evident in the wild-type as well as in single and double mutants lacking the genes for flagellin (*flaA* mutant) and the type IV pilus (*pilE* mutant). These results excluded the contribution of flagella and pili to the observed surface spreading mechanism that was therefore identified as sliding. Additionally, a translucent film was detected for all spreading *Legionella* species including the mentioned *L. pneumophila* mutants advancing well in front of the cells. Extracts of spreading plates with the film showed drop collapse and friction reduction characteristics indicating the presence of a surfactant molecule that presumably facilitates sliding (Stewart *et al*., 2009). However, the composition of this film was not analysed in detail.

Because of the evolutionary relation of the type IV pilus and the type II secretion system (TIIS), mutants lacking different components of the TIIS were also tested for their sliding ability. Interestingly, all TIIS mutants were defective in sliding and did not show the characteristic film (Stewart *et al*., 2009). However, when spotted on the film of the wild-type, sliding was restored suggesting that the TIIS mutants lack the secreted surfactant of *L. pneumophila*. There are several possible links between the TIIS and surfactant secretion: (i) the surfactant is secreted via the TIIS, (ii) the surfactant is modified by an enzyme secreted by the TIIS, and (iii) a regulatory network involved in surfactant production is influenced by the TIIS (Stewart *et al*., 2009). Although the sliding-facilitating surfactant has not been identified yet in this species, it was shown in a subsequent study that *L. pneumophila* excretes a surfactant that exhibits antimicrobial activity towards other *Legionella* species and its production depends on lipid metabolism and the outer membrane protein TolC (Stewart *et al*., 2011).

Exopolysaccharide facilitated sliding (group II)

Sliding motility of *Bacillus subtilis*, a gram-positive soil-dwelling bacterium, depends on a surfactant as well as exopolysaccharides, therefore it is presented in group II. This type of surface motility of *B. subtilis* was discovered while examining rhizosphere derived strains that exhibited a distinct dendritic growth pattern in a flagellum-independent manner (Kinsinger *et al*., 2003; Fall *et al*., 2006). If sufficient amounts of potassium ions were supplied in the medium, initial dendritic growth was followed by planar spreading (Kinsinger *et al*., 2003). This transition

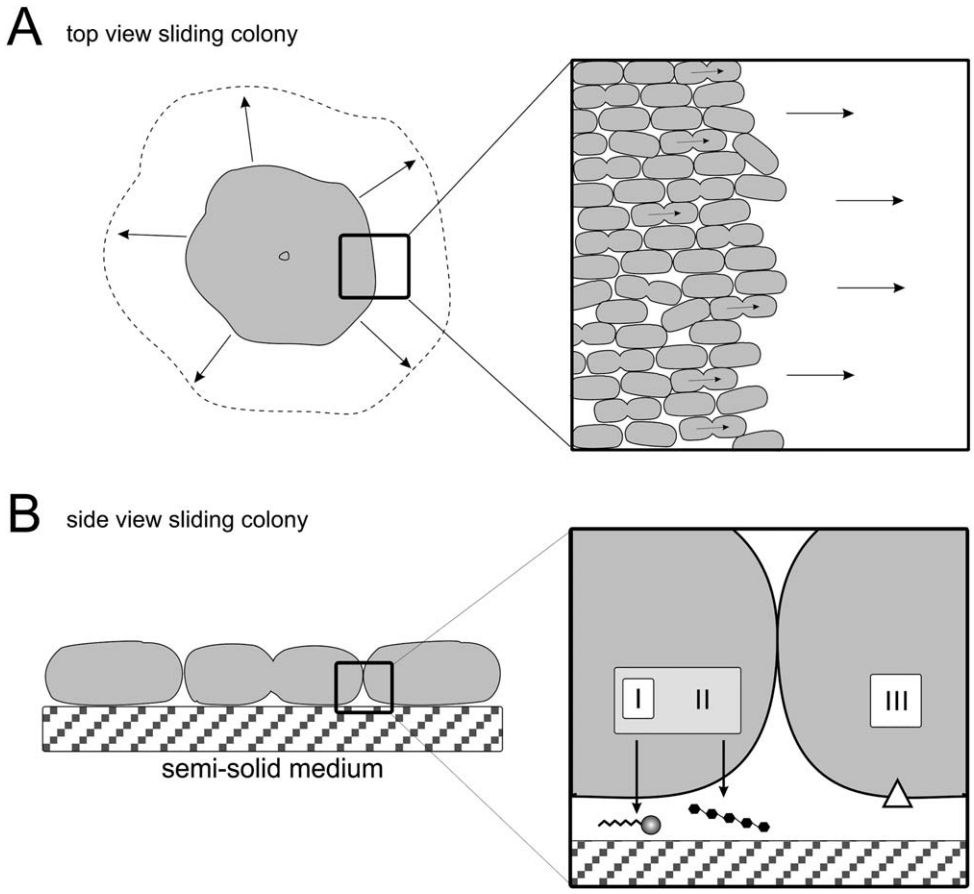


Fig. 1. Mechanism of sliding motility.
(A) Top view of an expanding sliding colony (left). On the right, a magnification of the marked region on the edge of the colony is depicted showing the expansion powered by the pushing force of dividing cells (arrows).
(B) Side view of a monolayer of cells at the edge of a sliding colony (left). The magnification highlights that sliding is promoted by a secreted surfactant (Group I, left), by a surfactant and exopolysaccharides (Group II, middle) or by an additional compound in the absence of surfactant (Group III, right).

to planar growth was also shown to be dependent on sufficient levels of other macro- and micronutrients suggesting a model where tendril sliding occurs at low nutrient concentrations and converts to planar sliding at higher nutrient concentrations (Fall *et al*., 2006). The potassium seemed to stimulate the production of surfactin, a cyclic lipopetide surfactant which was shown to be necessary for *B. subtilis* sliding (Kinsinger *et al*., 2003; Kinsinger *et al*., 2005). Isolation of strains with reduced or diminished sliding using directed and random mutant screens led to the identification of additional genes connected to surfactin biosynthesis, growth or potassium transport, emphasizing the requirement of a surfactant and the pushing force of growing cells (Kinsinger *et al*., 2005).

The requirement of exopolysaccharides (EPS) for *B. subtilis* sliding was recently demonstrated by two independent studies via mutant analysis and microarray experiments. Mutant strains lacking either the complete or part of the *epsA-O* operon whose products are responsible for exopolysaccharide biosynthesis were not able to slide, showing that EPS is essential for sliding (Grau *et al*., 2015; van Gestel *et al*., 2015). Interestingly, the same *eps* gene cluster is also critical for *B. subtilis* biofilm formation (Vlamakis *et al*., 2013).

The study by Grau *et al*. (2015) focussed on identifying the regulatory network governing sliding motility. Spo0A, a master regulator of various cellular processes in *B. subtilis* such as biofilm formation, sporulation, and cannibalism was found to be also the key modulator of sliding motility (Grau *et al*., 2015). In addition to EPS, the microarray approach highlighted the differential expression of the *bslA* gene that encodes a bacterial hydrophobin protein. Mutant analysis proved that BslA is as indispensable for sliding as EPS production and surfactin secretion. The level of phosphorylated, and therefore transcriptionally active Spo0A in *B. subtilis* is modulated by soluble and membrane bound histidine kinases via a phosphorelay. Detailed analysis demonstrated that two of these kinases,

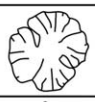
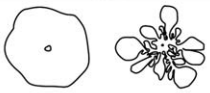
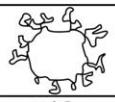
Group	Bacterium	Sliding facilitating component	Sliding morphology
I	*Serratia marcescens*	lipopeptide surfactant serrawettin	
	Pseudomonas aeruginosa	rhamnolipid biosurfactant	
	Pseudomonas syringae pv. tomato DC3000	lipopeptide surfactant syringafactin	
	Pseudomonas fluorescens SBW25	lipopeptide surfactant viscosin	
	Legionella pneumophila	unidentified surfactant	
II	*Bacillus subtilis*	surfactin, exopolysaccharides, protein BslA	
	Sinorhizobium meliloti	siderophore rhizobactin, exopolysaccharide EPSII	
III	*Salmonella enterica* serovar Typhimurium	protein PagM	
	Mycobacterium smegmatis	acetylated glycopeptidolipids	

Fig. 2. Bacteria discussed in this review are depicted that are capable of sliding.
The group species are categorized, the compound(s) that facilitate sliding, and the morphology of the individual sliding colonies are indicated.

KinB and KinC are required for sliding (Grau *et al*., 2015), while KinB is possibly active on the edge of the sliding colony, KinC is rather active in the interior. Further, the level of phosphorylated Spo0A sufficient to activate sliding motility is lower than for triggering biofilm formation and sporulation suggesting that sliding occurs before biofilm formation (Grau *et al*., 2015). Therefore, the fine modulation of active Spo0A-level allows the precise expression of genes leading to the distinct developmental pathways in *B. subtilis* (Kovács, 2016). In addition, the domain of the KinB kinase that resembles the selectivity sequence of the pore loop domain of eukaryotic potassium channels was demonstrated to be essential for sliding in response to the presence of potassium ions whose importance was also shown before (see above).

In the study of Grau *et al*. (2015), sliding was investigated mainly on rich medium with possibly higher potassium concentrations resulting in planar sliding colonies (Grau *et al*., 2015). In contrast, van Gestel and colleagues used a minimal medium with low potassium levels promoting sliding in a dendritic form (Fall *et al*., 2006; van Gestel *et al*., 2015). In this later study, the focus was brought on the differentiation of cells during sliding and how it affects migration. Additionally to EPS and the already known surfactin the authors identified also another component of the *B. subtilis* biofilm matrix, the protein TasA to be necessary for sliding (van Gestel *et al*., 2015). While *tasA* was found to be unnecessary for sliding by Grau *et al*., the differences could originate from different sliding modes. When mixed, different mutants of these components (surfactin, EPS, TasA) were able to at least partially complement each other for sliding, occasionally even performing better than the wild-type demonstrating the advantage of division of labour (van Gestel *et al*., 2015). Using reporter strains, the temporal expression of genes involved in surfactin and matrix production was examined revealing a peak of surfactin producing cells at the early stage of dendrite formation followed by an increase of matrix producers. These cell types showed a distinct spatial arrangement

during the outgrowth of the dendrites. The matrix producers were located in bundles formed by chains of cells (so called 'van Gogh bundles') whereas the surfactin producers surrounded these bundles in a less coordinated form. When mutant strains were mixed to complement the sliding behaviour, the bundles contained only matrix producers, thereby demonstrating the requirement of the matrix producing cell type for bundle formation. The formation of bundles promoted the appearance of larger loops at the rim of the sliding expansion as demonstrated by time lapse experiments. These loops were suggested to facilitate migration in agreement with modelling experiments on the importance of loop formation on spreading (van Gestel *et al*., 2015).

In summary, two hypotheses were proposed on the importance of EPS during sliding: van Gestel and colleagues demonstrated its requirement for bundle formation which in turn allows expansion, whereas Grau *et al*. proposed that EPS promotes spreading by generating osmotic pressure as shown for biofilms (Seminara *et al*., 2012; Grau *et al*., 2015). However, both hypotheses might actually be valid since two slightly different forms of sliding (dendritic and planar) were investigated in these studies. Importantly, both studies highlight the alternative functions of the extracellular matrix that in addition to be essential for biofilm development, also necessary for other processes, including surface spreading via sliding (Dragoš and Kovács, 2017).

In *Sinorhizobium meliloti*, sliding was also reported to facilitate surface movement of this gram-negative soil-dwelling bacterium. During investigations of discrepancies about the requirement of the quorum-sensing transcriptional regulator ExpR for swarming, it was discovered that *S. meliloti* strains harbouring a functional ExpR could spread over semisolid medium in a way atypical for swarming whereas mutants lacking ExpR were not able to spread (Nogales *et al*., 2012). As strains lacking the flagellum behaved similarly, the surface movement was suggested to be sliding. As ExpR is among others responsible for the regulation of exopolysaccharide production in *S. meliloti*, a mutant unable to produce EPSII (galactoglucan) was investigated and found to be deficient in sliding (Nogales *et al*., 2012). Similar to *B. subtilis*, EPSII could generate an osmotic pressure gradient which drives surface spreading in *S. meliloti* (Seminara *et al*., 2012). Additionally, the authors claimed to have identified another type of movement that is independent of ExpR and flagella since an *expR* mutant without flagella was still able to colonize a semisolid minimal medium (Nogales *et al*., 2012). This spreading was found to be dependent on siderophore rhizobactin production since mutation in the corresponding gene abolished spreading. It seems plausible that rhizobactin can act as a wetting agent and thereby contribute to facilitate spreading (Nogales *et al*., 2012). We hypothesize that this second type of movement could be also considered as sliding and while both rhizobactin and EPSII could promote surface colonization, secretion of either components is sufficient for spreading. When ExpR is intact EPSII can be produced and rhizobactin is not necessarily required but when ExpR and therefore also EPSII are missing and the iron concentration is low, rhizobactin can be produced and rescue sliding by acting as a wetting agent.

In addition to the laboratory conditions, the importance of sliding for *S. meliloti* in a natural habitat was demonstrated. *S. meliloti* is one of the bacteria known as rhizobia which can form a symbiotic interaction with legume plants where they fix nitrogen and provide it to the plant in exchange for nutrients. After recognition and entry into the root hair, *S. meliloti* invades the apoplasm of the root hair via so called infection threads (e.g. Gage and Margolin, 2000). Fournier and colleagues found that *S. meliloti* forms clusters in these threads that move forward and become longer over time (Fournier *et al*., 2008). This observation led them to conclude that sliding might facilitate infection thread colonization, additionally supported by the fact that rhizobia in the infection thread lack flagella (Gage and Margolin, 2000; Fournier *et al*., 2008). This study represents one of the few examples where sliding was analysed in a natural setting and shows that there are indeed conditions under which it might be useful for a bacterium to slide.

Surfactant independent sliding (group III)

Bacteria belonging under the last category similarly require the pressure of growing cells for sliding but rather depend on an additional factor and not surfactant. For sliding of *Salmonella enterica* serovar Typhimurium under low Mg^{2+} conditions, the protein PagM was identified by mutant analysis (Park *et al*., 2015). This protein seems to facilitate spreading through a surface protein (i.e. by being a surface protein itself or being connected to a so far unidentified one) since the sliding ability of a *pagM* mutant could be complemented in the presence of a strain with an intact *pagM* gene while proteinase treatment abolished this sliding. PagM is regulated by the PhoP/PhoQ system which is induced under low Mg^{2+} conditions (Park *et al*., 2015). Interestingly, this protein can be identified uniquely in *S. enterica* which suggests a form of sliding that is slightly different from the above described mechanisms.

Another distinct sliding-facilitating mechanism was described for *Mycobacterium smegmatis*. As this gram-positive bacterium belongs to the generally non-flagellated genus of *Mycobacteria*, it was long believed to be impaired in any kind of translocation. Yet, it was

discovered that after several days of incubation, *M. smegmatis* is able to spread over the surface of semisolid plates (Martínez *et al*., 1999). Interestingly, the spreading morphology was dependent on whether the medium was solidified using agar-agar or agarose, resulting in thin finger-like structures or a circular spreading front respectively. Using the circular spreading as a model, electron microscopic analysis revealed a distinct organization of cells in pseudo-filaments that were connected at distinct positions of the cells and not only at the poles. Further, it was confirmed that the spreading is accompanied by growth and almost no rearrangement of the cells was observed in the spreading zone, indicating sliding (Martínez *et al*., 1999). An investigation of different *M. smegmatis* colony variants uncovered the impaired sliding of a rough variant compared with the rather smooth wild-type. This lead to the hypothesis that glycopeptidolipids (GPLs), molecules that are part of the outer layer of some mycobacterial capsules, are connected to sliding since previous studies showed a correlation of a rough phenotype with a reduced amount of GPLs. Lipid extracts were analysed and showed a GPL characteristic pattern for the wild-type which was absent in the rough variant indicating that indeed GPL are facilitating *M. smegmatis* spreading (Martínez *et al*., 1999). In a subsequent study, a transposon mutagenesis resulted in several mutants that lost the ability to slide (Recht *et al*., 2000). All of them showed rough colony morphology and no GPLs could be detected in thin layer chromatography analyses. Almost all of the transposon insertions were located in the *mps* gene encoding a non-ribosomal peptide synthetase involved in GPL biosynthesis thus providing a direct evidence for the importance of GPLs for sliding motility (Recht *et al*., 2000). Only one additional mutant exhibiting a similar phenotype was identified to contain a transposon insertion in a gene coding for a putative membrane transporter (*tmtpC*) that could possibly be

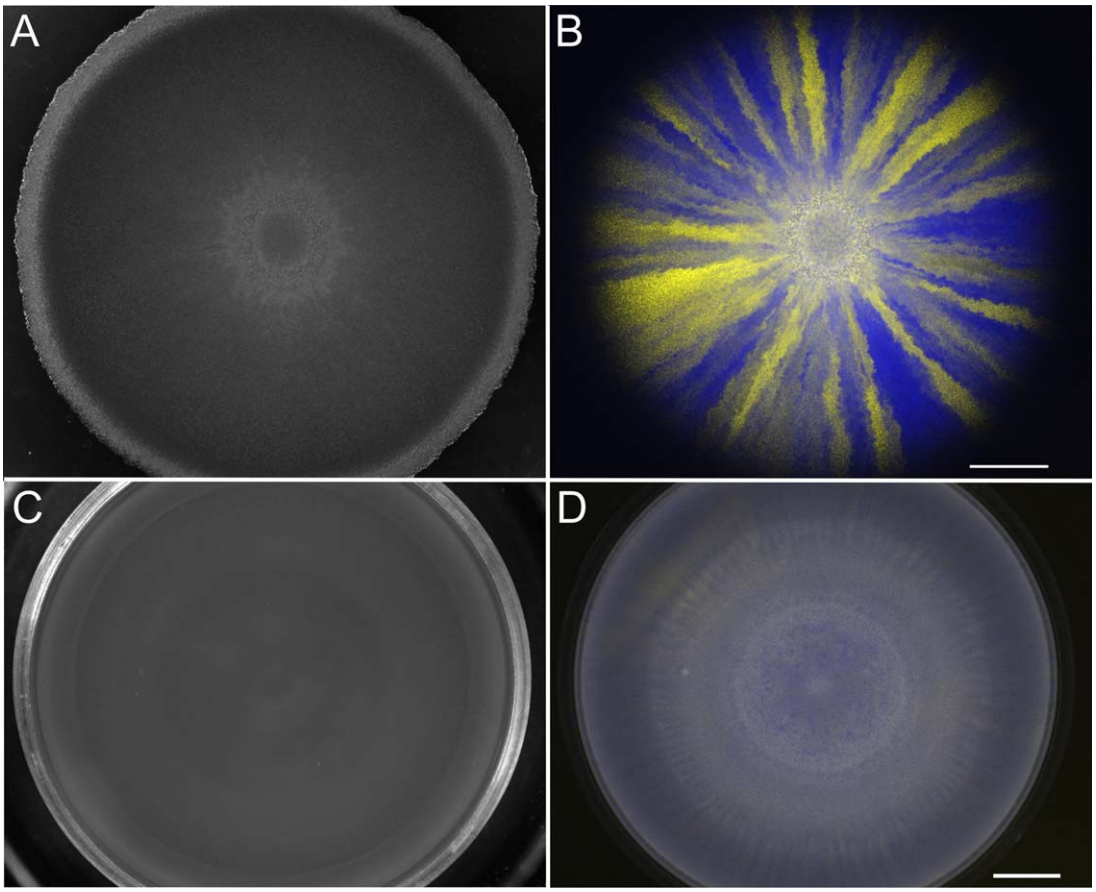


Fig. 3. Dissimilar spatial segregation levels can be appreciated in sliding and swarming colonies of *B. subtilis*.
To initiate sliding, two strains of *B. subtilis hag* mutant with different fluorescent markers were used. Similarly, the two *B. subtilis* strains that were used as inoculum for swarming possessed different fluorescent markers in the wild-type genetic background. Bright field images of sliding (A) and swarming (C) colonies. Overlay of the two fluorescent images with false colours (B, D). The semi-solid agar plates were incubated for 24h and 10h at 37°C for sliding and swarming, respectively, based on the methodology as described in Hölscher *et al*., (2016). The scale bars correspond to 5 mm. [Color figure can be viewed at wileyonlinelibrary.com]

involved in carrying the GPLs across the cytoplasmic membrane. A defect in this type of sliding could not be fully complemented by surfactants like Serrawettin, purified GPLs or the presence of a sliding-proficient strains demonstrating the importance of cell envelope bound GPLs (Recht *et al*., 2000).

Additionally, GPLs seem to be important for biofilm formation since the GPL deficient rough variants were not able to form biofilms attached to a plastic surface (Recht *et al*., 2000). An additional screen of a transposon mutant library for impaired biofilm formation revealed a mutant with an intermediate phenotype in which sliding and biofilm formation were both diminished but not completely abolished (Recht and Kolter, 2001). In this mutant, a transposon was inserted in the *atf1* gene that encodes a putative acetyl transferase and is located in a GPL biosynthesis gene cluster. An analysis of the GPLs suggested that the product of *atf1* is responsible for acetylation of the GPLs. Based on these studies the following model was proposed: GPLs in the outermost layer of the *M. smegmatis* cell envelope increase the cell surface hydrophobicity therefore facilitating sliding and biofilm formation. Without or with non-acetylated GPLs, the cell surface is more hydrophilic leading to abolished or reduced sliding and biofilm formation respectively (Recht and Kolter, 2001).

Concluding remarks

In summary, a number of bacteria have been identified that are capable of passively migrating over surfaces, considered to be sliding. We described here three groups according to the sliding mechanism and necessary components. Notably, it is possible that some of the organisms e.g. from Group I belong actually to Group II, but the additional components contributing to sliding motility are yet to be discovered.

In addition, social interactions during sliding can be compared with swarming and biofilm formation. For example, spatial segregation can be dissimilar (Fig. 3), which might have an ultimately different impact on the adaptation and evolution of bacteria (Hölscher *et al*., 2016; Martin *et al*., 2016).

Many regulators and components important during sliding are also necessary for other processes like surfactants for swarming and exopolysaccharides for biofilm formation. Thus, it is possible that sliding motility represents an intermediate stage between different developmental processes under conditions that favours neither one nor the other and it might be an innovative way to exploit components evolved for other processes. In conclusion, it is very likely that so far we have only seen the tip of the iceberg and many other organisms are able to slide over surfaces.

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Conflict of Interest

The authors declare no conflict of interest.

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