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Genetics

Myxococcus cells respond to elastic forces in their substrate

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ABSTRACT **Elasticotaxis describes the ability of *Myxococcus xanthus* cells to sense and to respond to elastic forces within an agar gel on which they rest. Within 5 min of the application of stress, each cell begins to reorient its long axis perpendicular to the stress force. The cells then glide in that direction, and the swarm becomes asymmetric. A quantifiable assay for the strength of elasticotaxis is based on the change in swarm shape from circular to elliptic. By using a collection of isogenic motility mutants, it has been found that the ability to respond to stress in agar depends totally on adventurous (A) motility, but not at all on social (S) motility or on the *frz* genes. In fact, S^- mutants (which are moving only by means of A motility) respond to the applied stress more strongly than does the wild type, despite the fact that their spreading rates are slower than that of the wt strain. Based on the swarming and elasticotactic phenotypes of isogenic *frizzy* strains that were also defective either in A or S motility, *frz* behaves as if part of the S motility system.**

Myxococcus xanthus, a rod-shaped bacterium, moves across solid surfaces by gliding. A gliding cell moves in the direction of its long axis with occasional reversals but without the aid of flagella (1). Although the molecular basis of translocation by gliding is unknown, many genes are known that control gliding behavior (2–5). To date, all such genes belong to one of two independent genetic systems, called adventurous (A) and social (S) (3), and one gene, *mglA*, appears to affect both systems (6). Cells with mutations in either of these systems (A^+S^- or A^-S^+) are still able to swarm by means of the remaining system but at a slower rate than the wild-type (wt) (A^+S^+) strain. Cells bearing mutations in both the A and the S genes (A^-S^- strains) fail to swarm or to glide for any appreciable distance.

Adventurously motile cells (A^+S^-) can move even when they are separated from other cells. Nevertheless, the rate of A motile swarming increases 4-fold with higher cell densities, implying interactions between A motile cells (7). Some A^- mutants (called *cgl* mutants, for *c*onditional *gl*iding) can be extracellularly complemented by cells bearing the corresponding wt allele, and mutations in five different *cgl* genes can be complemented this way (8). The A^- mutants unable to be so complemented are called *agl*, and about 20 different *agl* genes have been distinguished by their map positions (2).

In contrast to A motility, S motility is limited to cells that are within a few micrometers of each other (7). Social motility depends on the activity of type IV pili, polar appendages that emerge in a tuft from a cell end and typically are several micrometers in length (9–11). Also related to swarming, a cluster of *frz* genes controls the frequency at which cells reverse their gliding direction (12). Several of these genes are homologous to the enteric phosphorelay proteins of chemotaxis (13–15). Although *frz* action has been previously considered to stand outside the A and S motility systems, evidence in this paper supports the idea that the *frz* genes belong to the S motility system.

Elasticotaxis is a remarkable attribute of myxobacteria, in which cells respond to elastic forces in the agar that supports them. First discovered by R. Stanier in 1942, elasticotaxis involves cells orienting their movement perpendicular to stress forces within their substrate (16). Observing *Chondrococcus exiguus* (now *Corallococcus exiguus,* a relative of *M. xanthus*), he demonstrated that swarms on the surface of a slab of agar that was subject to tension in some of its parts and to compression in others spread rapidly perpendicular to either force but very slowly parallel to the force. Some years later, Dworkin (17) described particular instances of the directed movement of *M. xanthus* swarms moving rather directly toward small latex or glass beads resting on an agar gel. The glass beads had been freed of organic materials by incineration so that these experiments implied an ability of the swarm to respond to an object not by chemotaxis or by contact, but by a physical force produced by the bead (17). Dworkin suggested that elasticotaxis was responsible and that it would be useful in finding bacterial colonies on which the Myxobacteria would feed.

In the present work, we report a simple assay to quantify the elasticotactic response of *M. xanthus* cells based on the change of the shape of swarms moving over compressed agar. This assay has been used to see how elasticotaxis depends on A motility and S motility.

MATERIALS AND METHODS

Bacterial Strains, Media, and Phages. DK1622 is the wt strain for motility studies. Strains DK1963, DK2730, DK3377, DK3468, DK6204, DK8615, DK9032, DK9033, DK10405, DK10407, DK10409, and ASX1 have been described (9, 18–25). Strains DK1292 (*aglR2*, *sglB3*), DK2214 (*aglB1*, *tgl1*), and DK2232 (*aglB3*) were constructed by D. Morandi and D.K. (unpublished data). The following strains were constructed by using generalized transduction with bacteriophage Mx4, with each transduction represented as transductant (donor × recipient): DK11301 (DK1963 × DK1622KmR A^-), DK11303 (DK2214 × DK1622KmR A^-), DK11304 (DK2232 × DK1292KmR A^-S^-), DK11305 (DK2730 × DK1622KmR A^-), DK11306 (DK11304 × DK1622KmR A^-), DK11316 (DK10407 × ASX1TcR). Several of the resulting transductants were subsequently used as donors to construct double mutants, taking advantage of their kanamycin resistance (KmR) or tetracycline resistance (TcR) determinants. *M. xanthus* cells were grown in casitone-Tris (CTT) complex medium (8). Agar concentration was 1.5% except when noted. Generalized transductions with Mx4*ts18ts27hrm* were performed as described (26). Transductants with different motility phenotypes were easily discernible by visual examination of

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Abbreviations: wt, wild type; E, elasticotaxis coefficient.

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the colonies in 1.5% and 0.5% agar plates and by microscopic examination of the colony edges. The KmR determinant of Tn*5* in strain DK3377 was replaced with the gene conferring TcR from Tn*5–132* as described (26) to generate strain DK11326. For the elasticotaxis assay, gentamicin sulfate (Sigma) at a concentration of 2.5 μg/ml was added to the CTT media to avoid fungal contamination as the agar was manipulated. For *Flavobacterium johnsoniae* and *Lysobacter brunescens*, CYE was used for growth (at 33°C) and PY2 for swarming (at 27°C) (27, 28).

To assay for elasticotaxis, cells were transferred with a micropipette tip from a fresh colony onto a plate in which the agar had been compressed by insertion of a tube. Compression was used to squeeze a small amount of water from the agar. The water was allowed to evaporate before inoculation for several hours at room temperature with the lid closed followed by several minutes at 33°C with the lid off. After inoculation, the plates were incubated at 33°C for 3 days. The diameters of the swarms parallel and perpendicular to the compressing tube were then measured, and the ratio between them (E) was calculated. Control inoculations were made onto nonstressed plates (plates without an inserted plastic tube) in all experiments for comparison with stressed plates and for measurements of the rate of spreading.

Photomicroscopy. To observe the orientation of individual cells in response to stress, a small drop from a liquid culture at 150 Klett units ($\approx 10^9$ cells per ml) was deposited with a glass capillary over the surface of a stressed plate prewarmed to 33°C. The small amount of liquid dried in a few seconds. A section of the spot with the majority of the cells oriented parallel to the stress forces was selected and observed under the microscope (Nikon Eclipse E800) by using phase contrast and a 20× objective. Pictures were taken at 1-min intervals. Recording of images and related methods were as described (29).

RESULTS

Assay for Elasticotaxis. Stanier (16) used the weight of an unsupported slab of agar to generate both tension and compression. Experiments by J. Huang (personal communication) had shown that elasticotaxis in response to compression forces in agar gels was reproduced more readily than to tension forces. Starting from Huang's observation, we developed the following simple and reproducible mechanical arrangement. Agar plates of the type normally used for *Myxococcus* bacteriology were prepared. After the agar had solidified (about 12–24 hr), the agar was cut free from the walls of the dish by sliding a flat flexible plastic spatula between the agar and the edge of the dish, all around the edge of the plate. Once the spatula had returned to its initial position, the agar was gently pressed toward the opposite side of the plate. A 1.5-cm piece of flexible plastic tubing 4 mm in diameter was gently inserted in this space, and the spatula was removed, leaving the plastic tube in place to compress the agar.

Behavior of the wt Strain. To study the behavior of the wt strain, DK1622 cells were deposited at different distances from the plastic tube on the compressed circular plates. Fig. 1 *Top* shows that the compression of the agar produced by the inserted tube induces a strong response in the wt *M. xanthus* strain, as evidenced by the elliptical shape of the swarms on the stressed agar compared with the circular shape of swarms formed on nonstressed control plates. The swarms nearest the tube are arc-shaped, and the swarms farther from the tube have less curvature and less asymmetry. This decrease is the result of the curvature of the surface over which the stress is applied by the short tube inserted between the edge of the round dish and the agar, because that decrease was absent in square dishes with straight surfaces of force application (Fig. 2). Swarms at the bottom of the square plate are as asymmetric as those at the top. The arc shape of swarms in a round plate is apparently a consequence of the application of the stress force along the segment of an arc, because the arc of the response is concentric with the arc of the top surface of the agar. Moreover, the decreasing asymmetry of spots farther from the tube corresponds to the increasing length of arc over which the force is applied. Tubes of different widths were used to compress the agar, and a tube 4 mm wide was adopted, because it induced a strong response in the cells without causing macroscopic disturbances in the agar (such as wrinkles

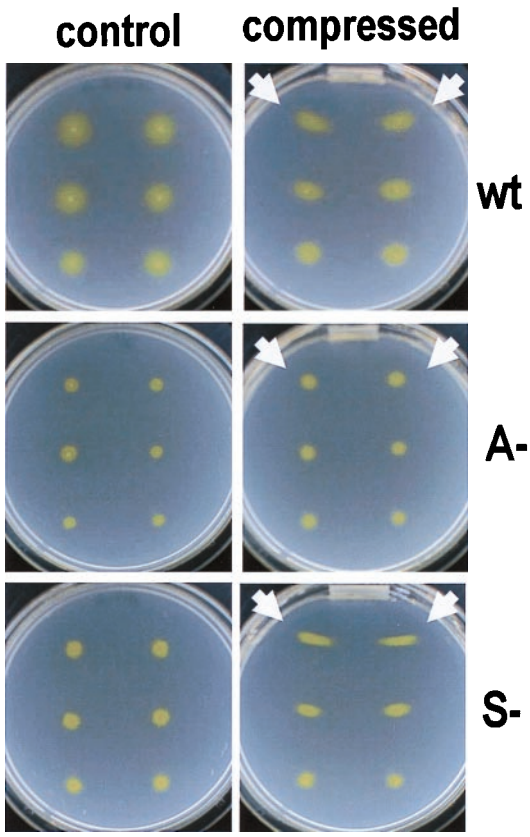

FIG. 1. Swarming of wt and motility mutants in control unstressed (*Left*) and compressed plates (*Right*). Cells were inoculated with a micropippete tip and incubated for 3 days at 33°C. wt, DK1622 (A^+S^+); S^-: DK10407 (A^+S^-); A^-, ASX1(A^-S^+). Arrows near the top of the compressed plates indicate the swarms measured to obtain E for Fig. 3. The inserted tubes are visible at the top.

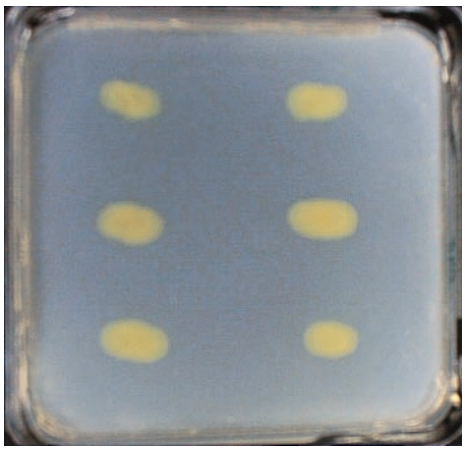

FIG. 2. Elasticotaxis and swarming of the wt strain on stressed agar in a square plate. The plastic tube at the top is longer than the one in Fig. 1, but the same diameter. Cells are as in Fig. 1.

or fractures). Microscopic examination of the agar with phase contrast optics and transmitted light revealed no alterations; neither did an examination of the surface in bright field with low-angle reflected light.

The shapes of the resulting swarms can be recorded for a qualitative assessment. The response can be quantified by measuring the diameters of the elliptical swarm parallel (length) and perpendicular (width), respectively, to the longitudinal axis of the plastic tube. The ratio of the two diameters is defined as an elasticotaxis coefficient (E).

Behavior of Motility Mutants. To investigate how cells detect stress in their substrate, mutants with defective A or S motility were examined. A collection of isogenic single and double mutants, listed in *Materials and Methods*, was assembled so that the genetic background would be the same in all cases. DK1622 is a fully motile strain commonly used as wt (A^+S^+) in motility studies, whose origin and whose circular physical–genetic map have been described (23, 30). All of the mutants used in this study were either generated in the DK1622 background or introduced into it by Mx4 transduction. Many of the mutants affected in S motility genes, in the *frz* system, and in *mgl* had been previously constructed in a DK1622 background. However, some of the mutants affected in A motility had not been isolated in this background. Therefore, four of the five A^- mutations used in this study were first introduced into DK1622, taking advantage of Tn*5* insertions linked to the corresponding A^- point mutations (31).

Values of E for the single and double mutants was calculated in the same manner described for the wt strain, except that only the two spots closest to the plastic tube were used for the determination (at a distance of ≈1.5 cm, see Fig. 1) to maximize the response. Fig. 3 shows E obtained with a collection of 23 single and double motility mutants. Each point in the figure represents the mean of at least 12 different assay spots, plated and measured in (at least) two independent experiments. Two different classes of nonswarming mutants were used in these experiments. One is affected in the gene *mgl,* and the other one is an A^-S^- double mutant. These two

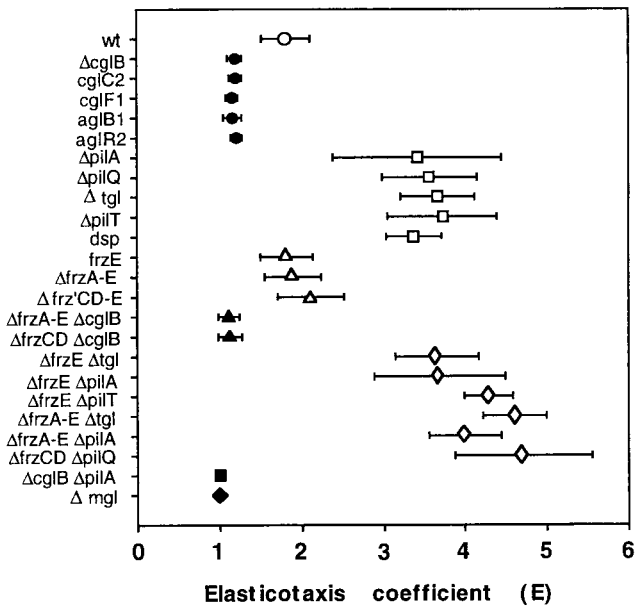

FIG. 3. Values of E are obtained by taking the ratio of the diameters parallel and perpendicular to the plastic tube that compresses the agar. Measured values of the elasticotaxis coefficient are plotted for various motility mutants of *M. xanthus*. ○, wt (A^+S^+); ●, A^-S^+ mutants; □, A^+S^- mutants; △, *frz*$^-$; ▲, *A*$^-$*frz*$^-$; ◇, *S*$^-$*frz*$^-$; ■, *mgl*$^-$; ◆, *A*$^-$*S*$^-$. Error bars indicate one SD on either side of the mean.

strains formed small colonies after 3 days of incubation, but the shape of these colonies was completely circular (and consequently, E = 1). The absence of an elasticotactic response in either strain indicates that the response is not a consequence of stress-oriented cell growth, inasmuch as both of these strains grow normally.

The wt (DK1622) has a coefficient of ≈2. The data summarized in Fig. 3 provide evidence that the A motility system is responsible for the elasticotactic response of the cells (see also Fig. 1), because all five A^-S^+ mutants tested had an E very close to 1. This value is significantly smaller than the E = 2 of the wt strain, as shown by the error bars in Fig. 3. This result is true for all of the mutants affected in A motility regardless of whether they are conditional *(cgl)* or not conditional *(agl),* point or deletion mutants. Because E cannot fall below 1, which indicates a symmetric swarm, and the gap between wt and A^- strains is 1, it was essential to repeat the measurements on the same culture and on independent cultures, as was done for the data of Fig. 3.

The mutants affected in S motility (A^+S^-) are clearly not defective in elasticotaxis (Figs. 1 and 3). In fact, all of them showed an even greater response than the wt strain, a point to be taken up in the *Discussion*. The mutants defective for the *frz* genes exhibit elasticotaxis in a manner similar to the wt strain. This result is in agreement with other observations (32) showing that a *frz* mutant was able to respond to glass beads deposited on agar. Double mutants *frz*$^-$ S^- behave like the A^+S^- mutants in the assay, showing enhanced elasticotaxis (E = 3 to 5); indeed the highest E values in Fig. 3 are *frz*$^-$ S^- strains. These results corroborate the idea that the A system is responsible for the elasticotactic response in *M. xanthus*, because any combination of mutants carrying an A^- mutation is impaired in elasticotaxis, whereas any combination that retains an intact A system is either not defective or hypertactic.

Even though E depends on swarming, there is no correlation between the spreading rate on a nonstressed surface and E (Fig. 4). As an example, the A^+S^- mutants spread less than the wt, yet they have a greater E value than wt.

The Swarm Edge. When observed at a magnification sufficient to resolve individual cells, the edge of a colony of the wt strain shows a characteristic flare morphology, including peninsulas, islands, and a few isolated cells moving outwards from the center of the colony; the average movement is in the radial direction (Fig. 5*A*). This radial pattern was lost when the colony swarmed on a compressed plate. Peninsulas formed by A^+ cells (A^+S^- and A^+S^+ strains) were no longer radial, but parallel. Moreover, stress causes the peninsulas to be longer and narrower. Their direction was always perpendicular to the stress and hence parallel to each other in stressed plates (Fig. 5 *A* and *B*). A^- cells did not extend long peninsulas under these conditions but formed gentle undulations, as is generally observed with S motility (7). Such edges show little if any signs of stress-induced alignment (Fig. 5*C*). In line with Stanier's

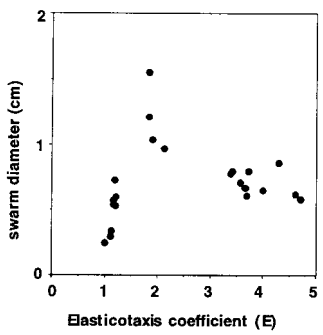

FIG. 4. E versus the swarming rate.

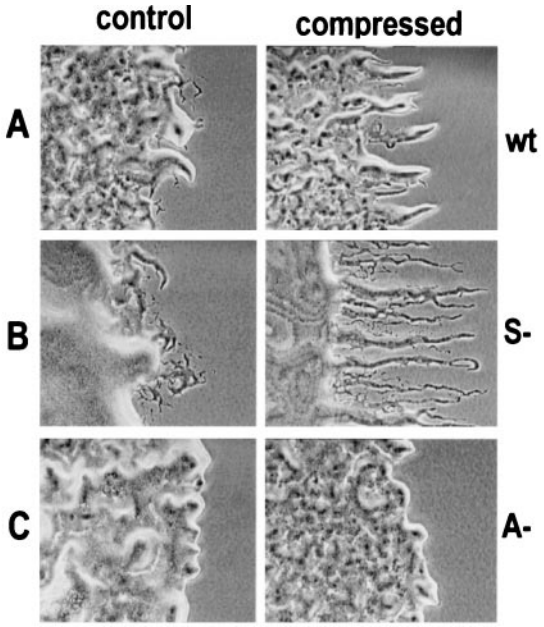

FIG. 5. Colony edges of wt and motility mutants in unstressed (*Left*) and compressed (*Right*) plates. The compressing tube (not visible in this field) is located above the colony, and the stress force is vertically downward. Two microliters of a cell suspension at a calculated density of 1,000 Klett units was spotted in unstressed and compressed plates, and incubated at 33°C for 3 hr. (*A*) wt (A^+S^+, DK1622). (*B*) A^+S^- (DK10407). (*C*) A^-S^+ (ASX1).

hypothesis on elasticotaxis (16), the special pattern of the A^+ swarm edges is because of the ability of individual cells to align their short axis with the direction of the stress force. As long as these asymmetric cells, whose axial ratio is 10:1, move in the direction of their long axis, the net result is a preferential movement of the cells perpendicular to the stress force. Indeed, gliding consists of movement in the direction of the cell's long axis (1). To examine the behavior of individual cells in response to the stress force, they were spotted from liquid cultures at low cell densities. Fig. 6 shows the change in the orientation of individual cells responding to the stress. A field was chosen in which almost all cells happened to be oriented vertically at the start. By 15 min, a large fraction had reoriented toward the horizontal. This alignment was never seen in A^- strains, even when the cells were spotted at a sufficiently high cell density for active social motility and swarming (data not shown).

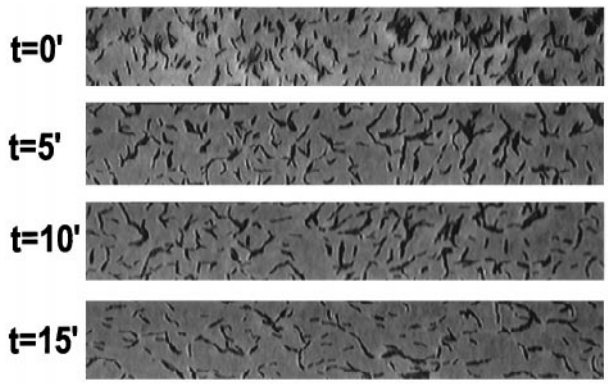

FIG. 6. Time-lapse video micrographs of wt cells on a compressed plate. The cells were spotted in the plate by using a glass capillary. The picture at time 0 was taken immediately after the drop dried. Time is in minutes; the stress force is vertically downward.

Other Gliders. To investigate whether bacteria of other genera can also respond to stress, we examined the behavior of *F. johnsoniae* (wt and two nonmotile mutants) and *L. brunescens,* which belong to different families of gliding bacteria unrelated to *M. xanthus* but whose cells are elongated like *M. xanthus* (27, 28). The elasticotaxis assay was performed on agar as described for *M. xanthus*, except that the nutrient medium and the incubation temperature were adjusted to the requirements of these bacteria. The wt strain of *F. johnsoniae* did respond to the applied stress qualitatively, like *M. xanthus*. Two nonswarming *F. johnsoniae* mutants did not spread and formed small circular colonies, indicating no elasticotaxis. *L. brunescens* spread very quickly but also formed circular colonies (data not shown). Evidently, elasticotaxis is not limited to the myxobacteria, including *C. exiguus* and *M. xanthus*. However, it is not a general property of bacteria that swarm by gliding. Among swimming bacteria, *Escherichia coli* cells on LB plates at 37°C fail to respond to stress forces sufficient to align *M. xanthus* cells; instead, *E. coli* cells form circular colonies on stressed agar.

***frz* Mutants and S Motility.** Investigating the swarm patterns of many double motility mutants for this study, it became apparent that A^-*frz*$^-$ mutants were swarming-defective, as if *frz* genes belonged to the S system (3). Although some A^-*frz*$^-$ mutants have been described as motile, it is significant that those observations were often made on *pilQ1* (formerly *sglA1*) cells, whose S motility is modified by a pilus-assembly point mutation (23). Attempting to clarify the status of *frz* mutants, a series of double mutants A^-*frz*$^-$ and S^-*frz*$^-$, were constructed in a DK1622 background, and their swarming was evaluated. Fig. 7 shows an experiment with *frzE,* and its swarming is strongly reduced in an A^-*frzE* mutant on both 1.5% and 0.5% casitone-Tris agar. Similar reductions in A^-*frz*$^-$ swarming were observed with all A and *frz* alleles tested, including those shown in Fig. 3. When a *frz*$^-$ mutation is combined with an S^- mutation, the spreading rate and the colony morphology are not significantly different from the corresponding single S^- mutants, revealing no deficit in A motility. Moreover, *frz*$^-$ mutants have a lower spreading rate on 0.5% agar than on 1.5% agar, which is characteristic of S^-

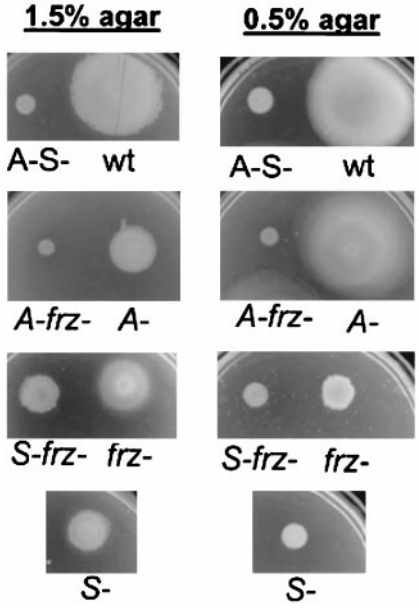

FIG. 7. Pictures of colonies of different strains grown in casitone-Tris plates with 1.5% or 0.5% agar. Drops of cells (2 μl) were deposited on the agar. After the drops had dried, the plates were incubated at 33°C for 4 days. The motility phenotype of the strains is indicated. wt, DK1622; A^-S^-, *cglB*, *pilA*; A^-*frz*$^-$, *aglB1*, *frzE*; A^-, *aglB1*; S^- *frz*$^-$, *pilA frzE*; *frz*$^-$, *frzE*; S^-, pilA.

mutants (33). Microscopic examination of colony edges of all of the *frz*$^-$ single mutant strains used in this study revealed many isolated cells and a paucity of organized groups of cells, as expected from a loss of S motility. Despite this evidence for a swarming defect in *frz*$^-$, the swarm rate of *frz*$^-$ is greater than *pil*$^-$ on both soft and hard agar, suggesting that some S motility remains in a null *frz* mutant. The swarm edge of the double mutants A$^-$*frz*$^-$ did have a narrow fringe of cells similar to the fringe found in an A$^-$*dsp*$^-$ mutant, which is a pil$^+$A$^-$S$^-$ mutant (11). An independent argument that mutations in *frz* decrease S motility but not A motility follows from their enhanced elasticotaxis shown in Fig. 3, where all *frz*$^-$ mutations, alone or combined with other S$^-$ mutations, have an E value equal to or greater than wt.

DISCUSSION

M. xanthus cells respond to elastic forces within the agar on which they move. The response is a reorientation of individual cells followed by gliding movement in the direction of their new orientation. An assay has been developed that measures the strength of this response and thus can be used to compare strains. Compressing an agar gel by less than 1 part in 20 by introducing of a piece of plastic tubing between the agar and the border of a Petri dish is sufficient to elicit a response. Fig. 6 shows how individual cells begin to change their orientation within 5 min and complete that process within 15 min, orienting their long axes perpendicularly to the direction of the applied force. Because gliding cells move in the direction of their long axis, movement after reorientation is also directed perpendicular to the compression force. Despite the relatively slow speed of gliding, the observed changes in movement can be seen with the naked eye, as elongation (perpendicular to the stress) of a swarm, whereas swarms formed on control, nonstressed plates have a circular symmetry.

All of the elasticotactic effects described in wt and mutant cells can be explained in this way, including the asymmetry of swarms on stressed agar. The longer and more narrow peninsulas of A$^+$ cells on stressed as compared with control agar (Fig. 5) is the consequence of the alignment of almost all cells perpendicular to the force, contrasting with more varied orientations and hence more varied initial gliding directions in the unstressed control. According to this view, the directed swarming toward a glass or plastic bead resting on agar, such as that observed by Dworkin (17), should result from the compression of the agar beneath the bead and the consequent stresses in the agar surrounding that object. Cells within a peninsula from a neighboring swarm that crosses the lines of elastic tension radiating from a spherical object would individually reorient, and the new direction points toward the object.

All of the experiments figured here used agar gels, although agarose was observed to perform like agar with respect to elasticotaxis (data not shown). Agar and agarose are composed mainly of copolymer chains of galactosyl and anhydrogalactosyl units whose residues are occasionally modified with methylether, sulfate, or pyruvate groups. Bundles of polysaccharide chains in unstressed fresh gels are presumably oriented equally in all directions. It is possible that compression tends to align polysaccharide bundles perpendicular to the direction of a compressive force and that the cells sense this orientation and align their long axes with the polysaccharide chains.

The experiments described here were carried out on compressed agar that is neither folded nor buckled. Macroscopic and microscopic examination of the plates reveals neither wrinkles nor grooves. However, more extensive compression by tubes of greater width, or of less flexible material, did lead to folding and buckling of the agar. The absence of visible folds in the agar stressed in the standard way is further supported by the parallel pattern of peninsulas in Fig. 5 and the rapidly formed pattern of cell orientation in Fig. 5. Had wrinkles been present in the agar but overlooked, either the peninsulas or the cells would have shown positional preferences generated by the concentrated elastic forces in such wrinkles. Yet, no preferences were observed in these or any of the additional experiments not documented in the figures. The absence of wrinkles or folds suggests that the conditions adopted for the elasticotaxis assay fall within the linear elastic range of the agar gels used.

It is possible that elasticotaxis is the sum of two biological processes: one that "senses" stress and another that "responds" by reorienting the cells. Because the A mutants do not respond, their defects could be in either process. If the A system were required in elasticotaxis only for sensing, it might be possible to isolate mutants that are elasticotaxis-negative but swarm normally. As yet, such mutants have not been sought.

The ability of the cells to sense and respond to compression forces requires a fully functional A system (Fig. 3), implying that the response is not a passive physical effect but an active biological process that involves movement and possibly sensation as well. Mutations in any of the five different A genes examined in this study have, within experimental variation, the same quantitative effect (Figs. 1 and 3). Three of the A genes are stimulatable *(cglB, cglC,* and *cglF)*, which suggests that they encode different cell-surface proteins. A membrane location is further supported by the amino acid sequence deduced from the sequence of the *cglB* gene (25), which is a putative lipoprotein. The two other A genes examined in this study, *aglB and aglR,* are not stimulatable. Apart from the *cglB* mutant, the other four A mutations were chosen only because they were linked to insertions of Tn*5*, which facilitated the necessary transductions of those mutations into a DK1622 background; they were not chosen for any motility-related quality. Four are point mutations, whereas the fifth (*cglB*) is a deletion mutant. Thus, the ability to respond to stress forces does not appear to depend on any particular A gene(s) or on the molecular nature of the mutations.

Elasticotaxis was no less than the wt in five different mutants with defective S motility. These mutants cover a range of recognized types of social-motility genes: pilin synthesis (Δ*pilA*), pilus assembly (Δ*pilQ*, Δ*tgl*), pilus action (Δ*pilT*), and fibril production (*dsp*). All but the *dsp* mutation are large in-frame deletions, and therefore single-gene null mutations. Strikingly, all S motility mutants have larger E values than the wt strain. This observation suggests that S motility may compete with A motility for elasticotaxis. S motility apparently involves one cell orienting its movement parallel to that of another cell. This orientation effect is thought to coordinate the movement of the several cells that make up a raft (7) and to align the cells in a social-motility flare (29). It is shown here that stress forces in the substratum are able to reorient individual cells in a direction perpendicular to a compressive force. A wt cell (A$^+$S$^+$) thus perceives two forces: one tending to orient it parallel to neighboring cells (S motility), and another tending to orient it perpendicular to a compression stress in its substrate (A motility). When the direction of these two orienting forces diverges, competition would be expected. Elimination of S motility by mutation would remove a competitor, resulting in a stronger elasticotactic response.

Is there a role for elasticotaxis in nature? We suppose that elasticotaxis has evolved as a consequence of the feeding habits of myxobacteria. They feed omnivorously on particulate organic matter containing proteins, polysaccharides (cellulose and chitin), lipids, or nucleic acids in the soil, using extracellular hydrolases on these polymers (34). Colonies of prey bacteria containing any of these nutrients might be detected irrespective of their chemical composition by the elastic distortions produced in a gel surface. Exopolysaccharide polymers released from microbes growing in the soil would provide

an elastic surface, and an elasticotactic response ought to enhance the ability of a nearby myxobacterial swarm to make a feeding encounter with a bacterial colony. The experiments of Dworkin (17), in which *M. xanthus* cells perceive the presence of small glass or plastic beads deposited on the agar, show that myxobacteria do have that capacity. McBride and Zusman (32) showed that *frz*$^-$ mutants retain the ability to migrate toward plastic beads on agar. The elasticotaxis detected in *F. johnsoniae*, which also feeds on organic soil particles, suggests that elasticotaxis does increase the feeding efficiency of cells, because *F. johnsoniae* might also benefit from such an advantage.

In addition to feeding, a sensitivity to the orientation of polysaccharides—specifically to those found on the *M. xanthus* cell surface—would enhance fruiting-body formation. Within a nascent fruiting body, cells interact and tend to align with each other (35), presumably by engaging their surfaces. It has been suggested that elasticotaxis is not involved in the choice of the sites that will become aggregation centers for fruiting-body formation (36). However, alignment of adjacent cells within a nascent fruiting body would be favored by the combined orienting power of A and S motility, which in that context would choose the same cell direction, increasing the frequency of end-to-end contacts and thereby the intensity of C-signaling, which is essential for fruiting-body development (37). Contrary to previous suggestions (3, 5), the A$^-$ mutants constructed for this work proved as defective in fruiting-body development as their S$^-$ relatives.

A role for *frz* genes in social motility is also described, and as a consequence, *frz*$^-$ mutants move largely by means of A motility. Should the level of A motility be reduced, either by mutation in a gene of the A system or by a decrease in the agar concentration, the result is a severe defect in swarming. This conclusion follows from the swarming defect of the double *frz*$^-$A$^-$ mutants (Fig. 7), from the reduced spreading rate of the *frz*$^-$ mutants in soft agar (our data, and refs. 15 and 38), and from the absence of S motile organization among cells at the edge of a *frz*$^-$ colony. Although some S motility remains in the *frz* mutants, it appears that the *frz* phosphorelay contributes more to S motility than to A motility.

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