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# Coupling of DNA Replication and Cell Division: *sulB* Is an Allele of *ftsZ*

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Treatments that damage DNA in *Escherichia coli* result in the inhibition of cell division. This inhibition is controlled by the *lexA-recA* regulatory circuit and can be specifically uncoupled by the mutations *sulA* (*sfiA*) and *sulB* (*sfiB*), which map at 21 and 2 min, respectively. Presently it is thought that *sulA* codes for an inducible inhibitor of cell division, the expression of which is controlled directly by the *lexA* repressor. In this report, it is shown that *sulB* is an allele of *ftsZ*, an essential cell division gene. A *sulB* mutation leads to an altered *ftsZ* gene product which is slightly thermosensitive and has an altered mobility on polyacrylamide gels. It is suggested that the altered *ftsZ* gene product is resistant to the *sulA* inhibitor, thus permitting cell division after induction of the SOS response. It is also shown that an increase in the gene dosage of *ftsZ* delays the onset of filamentation after SOS induction.

Damage to DNA in the bacterium *Escherichia coli* results in the induction of a variety of functions (SOS response) that aide in the survival of the cell (15, 21, 25). The molecular mechanism through which this occurs is now fairly well documented. The damaged DNA, detected by the appearance of single-stranded DNA, leads to the activation of the proteolytic activity of *recA* protein (20). The target of *recA* protease is the *lexA* repressor. Cleavage of *lexA* protein leads to the induction of the genes that it controls (13).

Among the responses grouped under the SOS response is the inhibition of cell division. Mutations have been isolated which specifically suppress this aspect of the SOS response, but do not affect the other inducible functions. These mutations, *sulA* which maps at 21 min and *sulB* which maps at 2 min, have been isolated as methyl methane sulfonate, nitrofurantoin, or UV-resistant derivatives of a *lon* mutant (3, 5, 9). A *lon* mutation prolongs the transient filamentation observed in wild-type strains when these treatments are used, and it is lethal (6, 12). Additional mutations, designated *sfiA* and *sfiB*, have been isolated as thermoresistant derivatives of the *tif lon* double mutation (4). These mutations are thought to be allelic to *sulA* and *sulB*, respectively. It is known that the *sulA* locus codes for a nonessential gene, since it has been possible to obtain insertions within the gene with a derivative of the bacteriophage Mu (7). With such an insertion, it has also been demonstrated that *sulA* is under the *lexA-recA* control circuit and is induced by treatments that result in DNA damage. This suggests that the *sulA* gene product is a good candidate for the proposed inducible inhibitor of cell division (4).

In contrast, no insertion mutations have been found in the *sulB* gene. The mutations that have been isolated at the *sulB* locus occur at a 10-fold-lower frequency than at the *sulA* locus. Significantly, the *sulB* mutations affect cell division when growth is at low ionic strength or high temperature. Such defects are not exhibited by *sulA* mutants. Therefore, it has been proposed that the *sulB* gene is a component of the cell division machinery (5, 9).

The *sulB* mutations all map at 2 min on the standard *E. coli* genetic map. This region contains several genes involved in cell division and cell wall biosynthesis (24). The organization of the genes in this area has been determined to a great degree with the aid of λ transducing phages for this region (1, 16–18). In the present study, these transducing phages were used to locate the *sulB* mutation. It was found that *sulB* is an allele of *ftsZ*, a recently discovered cell division gene (17). This finding is incorporated into a model for the inhibition of cell division.

## MATERIALS AND METHODS

**Bacterial and phage strains.** The *E. coli* K-12 strains used in this investigation are listed in Table 1. The λ transducing phages are listed in Table 2. Phage λ16-25 is a deletion derivative of λ16-2 and was described previously (17).

**Media and growth conditions.** LB medium contains Difco yeast extract (Difco Laboratories) (5 g/liter), Difco tryptone (10 g/liter), and NaCl (5 g/liter unless otherwise indicated). This medium was solidified by the addition of Difco agar (15 g/liter) for plates. The

TABLE 1. Bacterial strains

| Strain | Relevant marker | Other markers | Source or reference |
|---|---|---|---|
| AB1157 | | *thr leuB6 proA2 his argE3 thi ara lacY1 galK2 xyl mtl rpsL31 tsx supE44* | 9 |
| PAM660 | *lon-22* | As AB1157, but *met* | 9 |
| PAM161 | *lon-22 sulB25* | As PAM660 | 9 |
| JFL100 | *ftsZ84*(Ts) | *ilv his leu thyA deo ara*(Am) *lacZ125*(Am) *galU42*(Am) *tyrT supFA81*(Ts) | 16 |
| DM511 | *lexA3 tsl-1* | *arg his leu pro thr thi* | 19 |
| TKF12 | *ftsA12*(Ts) | *thr leu thi pyrF thyA ilvA his arg lac tonA tsx* | 24 |
| TOE1 | *ftsQ1*(Ts) | As AB1157, but *thyA* | 1 |
| GIA86 | *envA* | *thr thi pyrF thyA ilvA his arg lac tonA tsx* | 24 |
| RGC103 | *lon* | *leu-6 purE34 trpE38 thi-1 ara-14 lacY1 galK2 xyl-5 mtl-1 tonA23 tsx-67 agi-6 rpsL109 pon supE44* | 3 |
| RGC103-9 | *lon sulB9* | As RGC103 | 3 |
| 159 | | *uvrA gal rpsL* | 16 |

ability of transducing phages to suppress the *lon* phenotype was determined by streaking preconstructed lysogens onto LB plates containing 2 to 4 μg of nitrofurantoin per ml (3). The ability of transducing phages to suppress *lexA*(Ts) was determined by streaking preconstructed lysogens at 44°C and looking for the growth of isolated colonies. The *lexA*(Ts) strain usually grew slightly at the site of the inoculum, but such cells were extremely long filaments and isolated colonies were not formed. The suppressed strains grew very well at 44°C, and examination of cells from individual colonies revealed that they were heterogeneous in size but that very few filaments were present. Complementation of *fts* mutants was assessed by streaking the lysogen to be tested at 44°C.

TABLE 2. Phage strains[a]

| Phage strain | Bacterial genes on phage | Reference |
|---|---|---|
| λ16-2 | *envA ftsZ ftsA ftsQ ddl murC murG* | 16 |
| λ16-3 | As λ16-2 but *envA22* | 16 |
| λ16-6 | As λ16-2 but *ftsZ84*(Ts) | 17 |
| λ16-7 | As λ16-2 but *sulB25* | This study |
| λ16-9 | As λ16-2 but *sulB9* | This study |
| λ16-25 | *envA ftsZ ftsA ftsQ* | 17 |
| λ16-26 | *ftsQ* | This study |
| λJFL65 | As λ16-25 but *sulB25* | This study |
| λ16-25#45 | As λ16-25 but *ftsA*::Tn*5* | This study |
| λ16-25#20 | As λ16-25 but *ftsZ*::Tn*5* | This study |
| λ16-25#9 | As λ16-25 but *envA*::Tn*5* | This study |
| λJFL60 | *ftsQ ftsA* | This study |
| λ540 | | 16 |
| λ616 | | 17 |

[a] The phages with Tn*5* insertions were isolated by growing λ16-25 on strain DB1162-17 (*lacZ*::Tn*5*). The phages that could transduce Kan$^r$ were selected and screened for their ability to complement various mutations. Loss of the ability to complement a given mutation was taken as evidence that the insertion was in the corresponding gene. The complete description of those phages will be presented elsewhere.

In experiments involving the determination of cell number, samples were removed from the culture and fixed in 10% formaldehyde. Cell number was estimated with a Coulter Electronic Particle Counter (model ZB), using a tube with a 30-μm orifice.

**Isolation of new transducing phages.** All methods for the construction of transducing phages in vitro and their analyses by restriction mapping have been described previously (17). Deletion mutants of λ16-7 were isolated in vivo by resistance to EDTA as described previously (17). Isolation and characterization of derivatives of λ16-25 carrying Tn*5* insertions will be described in detail elsewhere. The phages were selected by growing λ16-25 on a bacterial strain carrying a Tn*5* insertion in *lacZ* (DB1167-62) and then transducing for kanamycin resistance. The position of each insertion was determined genetically by complementation and physically by restriction mapping and found to be in agreement.

**Infection and protein labeling in UV-irradiated cells.** A standard procedure was employed for infection and protein labeling (16) and is described briefly as follows. Cells were grown in minimal medium supplemented with maltose (0.4%) and $MgCl_2$ (1 mM) to an optical density at 540 nm of 0.5. The cells were concentrated (10×) and suspended in the same medium but with increased $MgCl_2$ (20 mM). After exposure to a UV dose of 8,000 ergs/mm$^2$, the cells (75 μl) were infected with phage at a multiplicity of 5 to 10 per cell. After 20 min, an additional 200 μl of prewarmed medium was added, and incubation continued for another 20 min, at which time 5 to 10 μCi of [$^{35}$S]methionine (New England Nuclear Corp.) was added. Five minutes later, the cells were harvested and lysed with sodium dodecyl sulfate sample buffer (16), and the proteins synthesized were analyzed by sodium dodecyl sulfate-polyacrylamide gel electrophoresis (12.5%) and autoradiography.

## RESULTS

**Isolation of a *sulB* transducing phage.** The *sulB* mutation *sulB25* was previously shown by P1 transduction to map in the *ftsA-envA* region at ca. 2 min on the *E. coli* genetic map (9). Thus, it seemed likely that specialized transducing phage

TABLE 3. Ability of phages to suppress the *lon* and *lexA*(Ts) phenotypes

| Phage | Suppression of phenotype | | |
|---|---|---|---|
| | *lon*[a] | *lon sulB25*[a] | *lexA*(Ts)[b] |
| | − | + | − |
| λ16-2 | − | + | − |
| λ16-7 | + | + | + |
| λ16-7Δ1 | NT | NT | − |
| λ16-7Δ4 | NT | NT | + |
| λJFL60 | NT | NT | − |
| λ16-25 | NT | NT | − |
| λJFL65 | NT | NT | + |
| λ16-9 | NT | NT | + |

[a] Suppression of the *lon* phenotype was measured as resistance to nitrofurantoin (2 μg/ml) on plates; +, growth; −, no growth; NT, not tested.

[b] Suppression of the *lexA*(Ts) phenotype was determined by streaking each strain on LB plates and incubating them at 44°C; +, growth; −, no growth.

λ16-2 might carry the wild-type *sulB* gene. This phage has a 10-kilobase (kb) *E. coli* DNA insert and is known to carry genes *envA*, *ftsZ*, *ftsA*, *ftsQ*, *ddl*, *murC*, and *murG* (1, 18, 22). To test for the presence of the *sulB* gene, I took advantage of the fact that *lon* mutants are unable to grow on plates containing nitrofurantoin, whereas *lon sulB* double mutants can grow on such plates (3). Nitrofurantoin is particularly useful since it induces filamentation of *lon* mutants but does not induce a resident λ prophage (11). A λ16-2 lysogen of a *lon sulB25* double mutant was found to be resistant to nitrofurantoin (Table 3), suggesting either that λ16-2 does not carry the *sulB*$^+$ gene or that the *sulB25* allele is dominant. If the latter case were true, it should be possible to obtain a λ16-2 *sulB25* derivative and demonstrate this dominance.

Therefore, λ16-3 (a transducing phage carrying the same amount of DNA as λ16-2, but carrying a mutation in the *envA* gene) was grown on strain PAM161 (*lon sulB25*). Eight λ *envA*$^+$ recombinants (Fig. 1) were selected and screened for the presence of *sulB25*. All lysogens of a *lon* mutant constructed with each of these recombinants were able to grow on nitrofurantoin plates (Table 3, λ16-7), indicating that *sulB* is closely linked to *envA* and that the *sulB25* allele is dominant. The presence and dominance of the *sulB25* allele were also tested by examining these phages for their ability to suppress a *lexA*(Ts) mutant strain (DM511). The *lexA*(Ts) strain is thermoinducible for SOS functions (19), which include the *sulA* gene product (7), and it undergoes filamentous death at the nonpermissive temperature. A phage (λ16-7) carrying the *suB25* mutation was able to suppress a *lexA*(Ts) mutant (Table 3). This confirms the dominance of *sulB25* over the wild type gene. Superinfection curing of the lysogen with λ *b2* restored the *lexA*(Ts) phenotype.

The suppression of filamentation is shown more clearly in Fig. 2. In this experiment, cell number was followed after a temperature shift of cultures of the *lexA*(Ts) mutant strain carrying either λ16-2 or λ16-7. The cell number of the *lexA*(Ts)(λ16-2) culture increased for ca. 40 min and then ceased, whereas the cell number of the *lexA*(Ts)(λ16-7) culture was still increasing at the end of the experiment.

**Locating the *sulB25* mutation.** Since *sulB25* was found at high frequency among λ16-3 *envA*$^+$ recombinants (8/8), *sulB25* must be very closely linked to *envA*. The same method has previously been used to determine that the linkage between *ftsA12* and *envA* is 37% and that for *ftsZ84* with *envA* is 90% (16; unpublished data). A more precise location for the *sulB25* mutation relative to *envA* was also determined with the aid of newly isolated deletion mutants of λ16-7. Deletion mutants such as Δ1 (Fig. 3; Table 3), in which the *mur* and *fts* genes are deleted, no longer express *sulB25*, whereas deletion mutants such as Δ4, which has lost only the *mur* genes (Fig. 3; Table 3), can still suppress *lexA*(Ts) and, therefore, still express a *sulB25* mutant product. Thus, *sulB25* must map to the left of the *envA* gene between *envA* and *ddl*.

Previous results (1, 18) suggested that most, if not all, of the coding capacity of the region

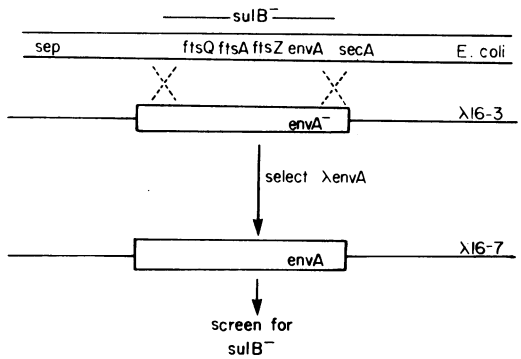


FIG. 1. Selection of λ transducing phage recombinants carrying a *sulB* mutation. The top line shows the relative location of genes in the 2-min region of the *E. coli* genetic map. The genomes of two λ transducing phages are indicated below. λ DNA is represented by a thin line, and the 10-kb *E. coli* DNA insert is represented by a box. Phage λ16-3 was grown on strain PAM161 (*sulB25*), and λ *envA*$^+$ recombinants were obtained from this lysate by selection for complementation of the *envA* mutation in strain GIA86. Transductants were used to lysogenize a *lon* or *lexA*(Ts) strain. These lysogens were then tested for resistance to nitrofurantoin (*lon*) or for temperature resistance (*lexA*(Ts), as an indication of the presence of the *sulB* mutation on the transducing phage.

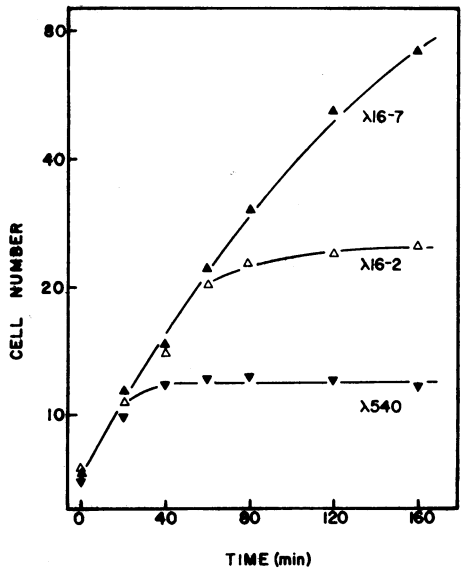


FIG. 2. Suppression of *lexA*(Ts)-induced filamentation. Lysogens of strain DM511 were grown to the exponential phase in L broth at 30°C. At time zero the cells were diluted into prewarmed L-broth at 30 and 44°C. At the indicated times, samples were taken and fixed in 10% formaldehyde, and the cell number was determined. Only samples taken at 44°C are indicated here; at 30°C, the cell number continued to increase exponentially. A value of 10 on the ordinate corresponds to a concentration of $2 \times 10^6$ cells per ml.

between *envA* and *ddl* can be accounted for by the three *fts* genes, *ftsQ*, *ftsA*, and *ftsZ*. The *sulB25* mutation, however, does not map in *ftsQ* or *ftsA*, since the 2.3-kb *Eco*RI fragment from λ16-7 which carries these genes did not suppress the *lexA*(Ts) mutant when it was subcloned into another λ vector (λJFL60, Fig. 3, Table 3). No convenient restriction sites are known for cloning the *ftsZ* gene alone. The 3.5-kb *Hin*dIII fragment carries the entire *ftsZ* gene, but when this fragment is subcloned onto another λ vector, the gene is expressed very weakly and complementation of an *ftsZ*(Ts) mutant is not observed (17). Therefore, λ16-25 (Fig. 3), a deletion mutant of λ16-2 which complements an *ftsZ*(Ts) mutant, was utilized. The left-hand *Hin*dIII site has been removed by this deletion, making it easy to remove the single 3.5-kb *Hin*dIII fragment in vitro (λ16-26) and replace it with the analogous fragment from λ16-7 (Fig. 3). Lysogenization of the *lexA*(Ts) mutant with one such phage, λJFL65, which carries the replacement *Hin*dIII fragment in its original orientation, suppressed the temperature sensitivity (Table 3). Thus, the *sulB25* mutation must map on the 3.5-kb fragment.

***sulB25* is an allele of *ftsZ*.** The above-mentioned results suggested that the *sulB25* mutation maps within the *ftsZ* gene. More direct evidence was obtained by testing the ability of λ16-7 to complement *fts*(Ts) mutations. Phage λ16-7 can complement *ftsQ*(Ts) or *ftsA*(Ts) mutations as well as λ16-2 can (data not shown). However, λ16-7 is impaired in its ability to complement *ftsZ84*(Ts). Although λ16-7 complements this mutation at 42°C, an increase in temperature or decrease in the salt (percent NaCl) concentration results in a loss of *ftsZ84*(Ts) complementation (Table 4). This effect was not seen when λ16-2 was used. One interpretation of this result is that the *sulB25* mutation results in an alteration of the *ftsZ* protein, causing it to be slightly temperature sensitive.

**Characterization of another *sulB* mutation.** A second *sulB* mutation, isolated independently by Gayda et al. (3), was also tested to determine whether it had the same effect as *sulB25*. This *sulB* mutation (*sulB9*) was isolated as a nitrofurantoin-resistant derivative of *lon* (3), whereas *sulB25* was isolated as a methyl methane sulfonate-resistant derivative of *lon* (9). The *sulB9* mutation was crossed onto λ16-3 by the same technique used for *sulB25*. The five λ *envA*$^+$ recombinants selected were all found to suppress the *lexA*(Ts) mutant, indicating that *sulB9* was also dominant. One of these recombinants, λ16-9 (Table 1), was then tested for its ability to complement *ftsZ84*(Ts). Phage λ16-9, like λ16-7, was impaired in *ftsZ84*(Ts) complementing activity (Table 3). Thus, two independently isolated *sulB* mutations behaved very similarly in suppressing filamentous death and in altering *ftsZ* activity. I concluded that *sulB* is an allele of *ftsZ*.

***sulB* mutations alter the *ftsZ* gene product.** The genetic results presented so far argued strongly that *sulB* mutations alter the *ftsZ* gene product. The next step was to demonstrate that the *sulB* mutations do indeed result in a biochemical alteration of the *ftsZ* gene product. The proteins synthesized by each of the transducing phages carrying various *sulB* and *ftsZ* mutations were

TABLE 4. Effect of the *sulB* allele on the phage on complementation of the *ftsZ mutation*[a]

| Phage | Growth | | | |
|---|---|---|---|---|
| | 0.4% NaCl | | 0.1% NaCl | |
| | 42°C | 44°C | 40°C | 42°C |
| None | − | − | − | − |
| λ16-2 | + | + | + | + |
| λ16-7 | + | − | + | − |
| λ16-9 | + | − | + | − |

[a] The strains were streaked on a plate, and the plate was incubated at the indicated temperature; +, growth; −, no growth.

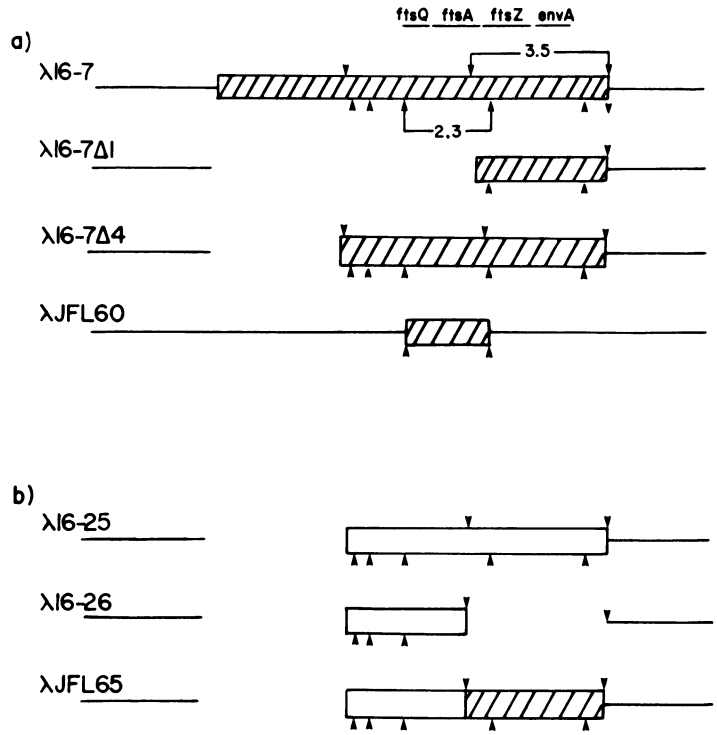


FIG. 3. Diagram of λ phages used in the location of the *sulB* mutation. Phage λ DNA is indicated by a thin line. *E. coli* DNA originating from λ16-2 is indicated by an open box, whereas that from λ16-7 is indicated by a hatched box. (▼) *Hin*dIII, (▲), *Eco*RI. The positions of the relevant genes are indicated above the diagram. (a) Deletion mutants of λ16-7 were obtained by selecting for resistance to EDTA as described previously (17). Two of the mutants, Δ1 and Δ4, are diagramed here. The extent of these deletions was determined by restriction endonuclease mapping. Phage λJFL60 was obtained by subcloning the 2.3-kb *Eco*RI fragment from λ16-7 into an *Eco*RI vector, λ616 (17). (b) DNA from λ16-25 was digested with *Hin*dIII and ligated to obtain a phage, λ16-26, in which the 3.5-kb *Hin*dIII fragment was deleted. The analogous *Hin*dIII fragment from λ16-7 was then isolated from an agarose gel and ligated into λ16-26. A derivative, λJFL65, was then obtained in which the *Hin*dIII fragment is in its original orientation.

labeled after infection of UV-irradiated cells. The identification and position of the *ftsZ* gene product had been determined previously with the use of deletion mutants of λ16-2 (18). The protein has a slightly slower mobility than the λ head protein, the gene *E* product. Both *sulB* mutations tested had the same effect on the *ftsZ* gene product, causing it to migrate more slowly than the wild-type gene product (Fig. 4). The mobility of the gene product of the *ftsZ84*(Ts) allele appeared identical to that of wild type.

**Effect of gene dosage of *ftsZ* on filamentation induced by the SOS response.** In the experiment shown in Fig. 2, the lysogen carrying phage λ16-2 consistently showed more residual cell division after a shift to nonpermissive conditions than did nonlysogens or lysogens carrying control phages. The range in percent increase in cell number was 180 to 240 for such lysogens and 70 to 110 for nonlysogens or control phage lysogens. Since the difference observed was twofold, it seemed possible that a gene dosage effect was responsible. The region of the DNA responsible for this must lie between *envA* and *ftsQ*, since a 240% increase in cell number was observed when a λ16-25 lysogen was tested (Table 5). To determine which of the genes carried by λ16-25 was responsible, advantage was taken of

TABLE 5. Effect of gene dosage on residual cell division

| Phage | Cell no. increase (%)[a] |
|---|---|
| None | 72 |
| λ16-25 | 240 |
| λ16-25#45 | 182 |
| λ16-25#20 | 76 |
| λ16-25#9 | 236 |

[a] Cell number was measured as described in the legend to Fig. 2. The figures represent the percent increases over the initial values and are averages of two determinations.

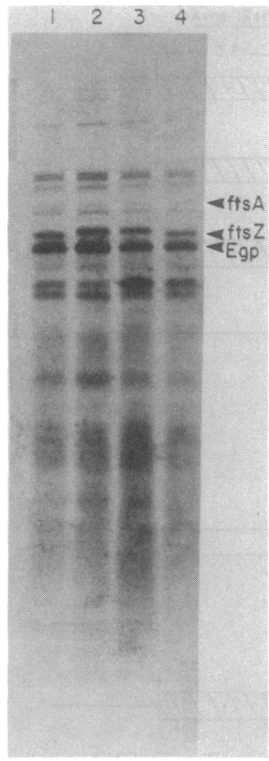


FIG. 4. Effect of *sulB* mutations on the *ftsZ* gene product. A culture of strain 159 was grown in minimal medium, UV-irradiated, and infected with various phages as described in the text. Proteins synthesized were labeled by the addition of [$^{35}$S]methionine and analyzed by gel electrophoresis (12.5% sodium dodecyl sulfate-polyacrylamide gel electrophoresis) and autoradiography. The position of the wild-type *ftsZ* product is indicated by the arrow, and the position of the λ head protein (Egp) is also indicated. The phages were as follows: lane 1, λ16-2(*ftsZ*$^+$); lane 2, λ16-7 (*sulB25*); lane 3, λ16-9(*sulB9*); lane 4, λ16-6(*ftsZ84*).

Tn*5* insertions which had been isolated in this phage. Lyosgens of phages carrying insertions in *ftsA*, *ftsZ*, and *envA* were tested. An insertion in *ftsZ* reduced the amount of residual cell division to the value obtained with a nonlysogen (Table 5). The other two insertions had little effect: residual cell division was approximately twice the nonlysogenic value. Thus, the presence of two copies of the *ftsZ* gene results in delaying the onset of cellular filamentation, which results in a twofold increase in cell number.

## DISCUSSION

One of the many effects of SOS induction is the inhibition of cell division. This inhibition can be specifically uncoupled by mutations at two loci, *sulA*(*sfiA*) and *sulB* (*sfiB*). Previous work (7) has shown that the *sulA* gene product is an inducible component of the SOS response under control of the *lexA-recA* regulatory circuit. This product is the postulated inhibitor of cell division (4). In the present study, I have presented several lines of evidence which indicate that *sulB* mutations map within the essential cell division gene, *ftsZ*. First, recombination and deletion analysis of a phage carrying a *sulB* mutation located it in the vicinity of *ftsZ*. Second, transducing phages carrying *sulB* mutations were impaired in their ability to complement an *ftsZ* mutation (Table 4). Third, the presence of a *sulB* mutation caused the *ftsZ* gene product to exhibit an altered mobility on a sodium dodecyl sulfate-polyacrylamide gel (Fig. 4). Although it is somewhat surprising that a putative missense mutation causes a change in mobility in such a gel system, changes in mobility can also be caused by dominant *lexA* mutations, which can slightly increase or decrease the mobility of the *lexA* protein (14).

The fact that *sulB* mutations map in an essential cell division gene was first suggested by Johnson (9), because *sulB* mutations cause cells to be slightly temperature sensitive for cell division, especially when the salt concentration is low. This same observation and conclusion was made by Gottesman et al. (5) in their analysis of *sulB* mutations. My study confirms their work and extends it by determining the locus affected by the *sulB* mutations.

These results suggest a mechanism for the inhibition of cell division that occurs as a result of SOS induction. After DNA damage, the proteolytic activity of *recA* protein is activated (20) and the repressor of SOS genes, the *lexA* gene product, is cleaved (13). Cleavage of *lexA* repressor results in induction of the cell division inhibitor coded for by the *sulA* gene product (7). The *sulA* protein then inhibits or inactivates the product of *ftsZ*, blocking an essential step in cell division. It is interesting that Walker et al. (23) in their study of an *ftsZ84*(Ts) mutant found that the initiation of septum formation is defective and that the *ftsZ* gene product is probably not required for completion of septation. Burdett and Murray (2), in an electron micrographic analysis of cell division, also concluded that the *ftsZ84*(Ts) mutation affects an early stage of septum formation. An initiator of septum formation would be an expected target for an inhibitor of the process.

Mutations such as *sulB25* and *sulB9* could result in an altered *ftsZ* gene product which is reduced in sensitivity to the *sulA* gene product but is still functional in cell division. In this case, *sulB* mutations might be expected to be dominant, as was observed both in this study and by Gottesman et al. (5). Huisman et al. (8), however, have reported that the *sulB* mutations they

isolated are recessive. It is possible that they isolated a different type of *sulB* mutation, since these investigators isolated mutations that suppress *tif lon* mutation strains, whereas others selected mutations that suppress the nitrofurantoin, UV, or methyl methane sulfate sensitivity of *lon*. Moreover, it should be noted that different tests were applied to determine dominance. Each investigator used the same conditions that were used in the isolation of their *sulB* mutation to test dominance. The data of Huisman and D'Ari (7), in fact, show that thermoinduction of a *tif lon* mutant leads to an induction of the *sulA* gene, which is twofold higher than for the other treatments used. This finding, coupled with the fact that the *sulB* mutations used in this study and by Gottesman et al. (5) may not be completely dominant, might explain the discrepancy. Although the *sulB* mutations used in this study allow *lon* and *lexA*(Ts) mutants to form colonies under restrictive conditions, the cells in these colonies are heterogeneous in size and the formation of filaments is certainly not completely suppressed. Thus, the dominance may depend upon the test used to determine it.

Recently, Ward and Lutkenhaus (unpublished data) have observed that expression of a protein fusion product of *lacZ* and *ftsZ* (which does not possess *ftsZ* activity) inhibits cell division. This supports a model in which *ftsZ* gene product functions as a multimer, which could easily complicate the complementation pattern. If so, this could explain why *sulB* mutant proteins are more thermostable in the presence of a normal *ftsZ*$^+$ product than in the presence of *ftsZ84*(Ts) protein. [Compare Tables 4 and 3. DM511(λ16-7) grows at 44°C, but JFL100(λ16-7) does not.]

This study also shows that the gene dosage of *ftsZ* affects the period in which cell division can continue after thermal inactivation of the *lexA*(Ts) repressor. The simplest explanation of this observation is that the two copies of the *ftsZ* gene provide twice the amount of *ftsZ* product and, therefore, twice as much substrate for the *sulA* inhibitor to inactivate. Recently, Ward and Lutkenhaus have constructed a high-copy-number plasmid containing the *ftsZ* gene to determine the effect of an even higher gene dosage on this system. We found that the higher gene dosage leads to considerably more residual cell division after induction of the SOS response (unpublished data).

If *ftsZ* product is, in fact, inactivated by the *sfiA* product, then a question arises about the recovery of cell division after DNA damage is repaired; i.e., is the inactivation of the *ftsZ* product reversible, or does cell division require the synthesis of new *ftsZ* product? Jones and Donachie (10) found that protein synthesis is required before division can take place after the cells have recovered from SOS induction. Thus, it is possible they detected a requirement for the synthesis of the *ftsZ* product.


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