

Spontaneous mutagenesis in exponentially growing and stationary-phase, *umuDC*-proficient and -deficient, *Escherichia coli dnaQ49*

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Spontaneous mutations arise not only in exponentially growing bacteria but also in non-dividing or slowly dividing stationary-phase cells. In the latter case mutations are called adaptive or stationary-phase mutations. High spontaneous mutability has been observed in temperature sensitive *Escherichia coli dnaQ49* strain deficient in 3'→5' proofreading activity assured by the ϵ subunit of the main replicative polymerase, Pol III. The aim of this study was to evaluate the effects of the *dnaQ49* mutation and deletion of the *umuDC* operon encoding polymerase V (Pol V) on spontaneous mutagenesis in growing and stationary-phase *E. coli* cells. Using the *argE3*_{OC}→Arg⁺ reversion system in the AB1157 strain, we found that the level of growth-dependent and stationary-phase Arg⁺ revertants was significantly increased in the *dnaQ49* mutant at the non-permissive temperature of 37°C. At this temperature, in contrast to cultures grown at 28°C, SOS functions were dramatically increased. Deletion of the *umuDC* operon in the *dnaQ49* strain led to a 10-fold decrease in the level of Arg⁺ revertants in cultures grown at 37°C and only to a 2-fold decrease in cultures grown at 28°C. Furthermore, in stationary-phase cultures Pol V influenced spontaneous mutagenesis to a much lesser extent than in growing cultures. Our results indicate that the level of Pol III desintegration, dependent on the temperature of incubation, is more critical for spontaneous mutagenesis in stationary-phase *dnaQ49* cells than the presence or absence of Pol V.

Spontaneous mutations occur in exponentially growing bacteria, but also in non-growing or slowly growing cells as a result of expo-

sure to non-lethal selection. The latter mutations, called adaptive, starvation-associated or stationary-phase mutations, differ from

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spontaneous growth-dependent mutations, which appear in the genome randomly before exposure to selection (Cairns *et al.*, 1988; Cairns & Foster, 1991; Hall, 1990; Foster, 1993; Foster, 1999; Bull *et al.*, 2000).

It has been found that mutations in the *dnaE* gene encoding the α polymerizing subunit of *Escherichia coli* DNA polymerase III (Pol III), the enzyme responsible for duplication of the bacterial chromosome, lead to changes in spontaneous mutagenesis in replicating bacteria. It has also been shown that mutations in *dnaE* modify the level of spontaneous mutations in stationary-phase cultures. Foster and coworkers (1995) have shown that *dnaE915*, an antimutator allele of *dnaE*, causes a 3-fold decrease in adaptive Lac⁺ reversion in the FC40 strain. Results obtained by others also support a direct role of the α subunit in adaptive mutagenesis (Harris *et al.*, 1997).

The polymerase activity and processivity of the α subunit are highly stimulated by the ϵ subunit (and *vice versa*). The ϵ subunit, the product of the *dnaQ* gene, contains a 3'→5' exonuclease that functions in proofreading of replication errors but also plays an important structural role within the Pol III core. In *dnaQ*^{TS} mutants (such as *dnaQ49* investigated in this work), at temperatures above 30°C, the ability of ϵ to interact physically with the α subunit is disrupted which leads to instability of the replication complex (Takano *et al.*, 1986; Jonczyk *et al.*, 1998; Taft-Benz & Schaaper, 1998). In *dnaQ49* bacteria replicating at a non-permissive temperature damage to DNA induces the SOS response that in turn switches on several DNA repair error-free and error-prone pathways (Friedberg *et al.*, 1995). In *E. coli* the SOS response involves over 40 unlinked genes (Courcelle *et al.*, 2001; Crowley & Courcelle, 2002) creating the SOS regulon. Expression of these genes is coordinately regulated by the LexA and RecA proteins. Generation of single stranded DNA (ssDNA) fragments by failed attempts to replicate past lesions leads to the formation of

RecA-ssDNA filaments that facilitate proteolytic self-cleavage of LexA. Autodigestion of LexA, that acts as a transcriptional repressor, leads to expression of LexA-regulated genes. Among them are the *umuDC* genes encoding DNA polymerase V which participates in translesion DNA synthesis (TSL) (Walker, 1998; Woodgate, 1999; Friedberg & Gerlach, 1999; Bridges, 1999; Janion, 2001). Replication by Pol V across DNA damage comes at the cost of reduced fidelity leading to formation of mutations even during replication of an undamaged template. Therefore, to limit mutations, expression of the *umuDC* operon is tightly regulated and proceeds at a late stage during the SOS response.

Another member of the SOS regulon, the *sfiA* (*sulA*) gene, is also induced as one of the latest (Kuzminov, 1999). The gene encodes a protein that inhibits cell division which subsequently leads to filamentous growth of cells. Thus, the massive appearance of filaments proves *sfiA* expression, and may indicate the induction of the SOS response (Janion *et al.*, 2002).

The aim of this work was to test the effect of the *dnaQ49* mutation in the ϵ subunit of Pol III, and of the deletion of the *umuDC* operon on spontaneous mutagenesis in exponentially growing as well as stationary-phase bacteria. We examined the frequencies and specificity of spontaneous mutations in *E. coli* AB1157 strain bearing an ochre mutation in chromosomal *argE3* gene. Only *argE3*→Arg⁺ revertants showed the ability to grow into colonies on minimal plates without arginine. In comparison to a *dnaQ*⁺ strain, AB1157*dnaQ49* growing at the non-permissive temperature of 37°C exhibited a high rate of spontaneous Arg⁺ revertants accompanied by SOS induction. The same strain additionally deprived of Pol V (AB1157*dnaQ49*Δ*umuDC*) showed a 10-fold decrease in the level of growth-dependent Arg⁺ revertants, while the level of stationary-phase revertants decreased only twice. The above observation, as well as the differences in the SOS induction levels and in

specificities of the Arg⁺ revertants in growing and stationary-phase cultures, indicate distinct mutagenic pathways in growing and resting bacterial cells.

MATERIALS AND METHODS

Bacterial strains. The bacteria used in this study were *E. coli* K12 strain AB1157 (genotype: F⁻ *thr-1 leuB6 proA2 his4 thi1 argE3 lacY1 galK2 rpsL supE44 ara-14 xyl-15 mtl-1, txs-33* (Bachmann, 1987)) and its *dnaQ49* and Δ *umuDC* derivatives. In the present work AB1157*dnaQ49* was obtained by P1 transduction of *dnaQ49zae-502::Tn10* from the NR9695 into the AB1157 strain (Schaaper & Cornacchio, 1992), whereas AB1157*dnaQ49-ΔumuDC* by transduction of Δ *umuDC595::cat* from RW82 into AB1157*dnaQ49* (Wood-

plemented with glucose (0.5%), casamino acids (0.2%), thiamine (10 μg/ml) and Arg, His, Thr, Pro and Leu, each at 25 μg/ml. E-Arg was the E medium deprived of arginine and solidified with 1.5% Difco Agar.

Mutation experiments. Mutation experiments were performed as described before (Nowosielska & Grzesiuk, 2000). Bacteria were grown overnight at 28°C or 37°C with shaking in E medium, centrifuged, suspended in fresh E medium, and grown for 3 h. Aliquots of 100 μl were plated onto LB plates to estimate the number of viable cells, and onto E-Arg plates to estimate the number of Arg⁺ revertants. The frequency of mutation was the number of Arg⁺ revertants per viable cell. Colonies on LB plates were counted after one day of incubation. Arg⁺ colonies required 2 days to become visible on E-Arg plates. For further incubation under conditions limiting

Table 1. Nucleotide sequence change associated with the Arg⁺ phenotype of revertants of AB1157*argE3* strain

Site of mutation (suppressor)	Lysis of Arg ⁺ revertants by T4 phages*					Mutation
	wt	B17	oc427	ps292	ps205	
<i>sup B</i>	+	+	+	+	+	GC→AT
<i>argE3</i>	+	+	-	-	-	**
<i>supL</i>	+	+	+	+	-	AT→TA
unknown	+	+	+	-	-	AT→TA?

*Lysis (+) or nonlysis (-) was determined by placing 1–1.5 × 10⁴ p.f.u. of phage suspension on bacterial stripe. **Mutations in *argE3* locus (back mutations) could be caused by transversions or transitions at the AT base pairs in the TAA ochre codon.

gate, 1992). All *dnaQ49* strains were stored on minimal medium to limit the appearance of suppressors. For each constructed strain three independently isolated clones were tested. Experiments were repeated 4–6 times, each in duplicate, and standard deviations (S.D.) were calculated.

Media and plates. LB (Luria–Bertani) medium consisted of 1% Bacto-tryptone, 0.5% yeast extract, and 0.5% NaCl. E medium consisted of C-salts (Vogel & Bonner, 1956) sup-

plemented with glucose (0.5%), casamino acids (0.2%), thiamine (10 μg/ml) and Arg, His, Thr, Pro and Leu, each at 25 μg/ml. E-Arg plates were sealed with parafilm and the appearing revertants were counted daily up to the 11th day. The mutation spectra of Arg⁺ revertants were studied as described elsewhere (Śledziwska-Gójska *et al.*, 1992). The suppressor activities of the Arg⁺ revertants were examined by testing their sensitivity to a set of amber (B17) and ochre (oc427, ps292, ps205) mutants of T4 phage. From each mutation experiment 50 colonies, of growth-dependent and station-

ary-phase Arg⁺ revertants, were tested. The results of phage typing revealed the kind of suppressor present and the type of mutation (Table 1).

Microscopic observations. The experiments were performed according to Yigit and Reznikoff (1997). Briefly, bacteria were collected by centrifugation of liquid cultures or by washing off solid medium (E-Arg plates), suspended in 0.9% NaCl, spread onto glass slides, air-dried and fixed with 100% methanol. The bacteria were then stained with acridine orange (0.15 mg/ml) in 10 mM phosphate buffer (pH 7.4) and examined under a

RESULTS

SOS response in AB1157*dnaQ49*

In early studies on the SOS system it was shown that filamentous growth is characteristic for the induction of the SOS response (Witkin, 1967). Taking advantage of this feature, Janion and coworkers demonstrated that in starved bacteria the SOS system is induced by a cAMP-dependent process but only when cells regain their growth (Janion *et al.*, 2002). Here we observed SOS induction in *E. coli* AB1157*dnaQ49* (Fig. 1). Microscopic

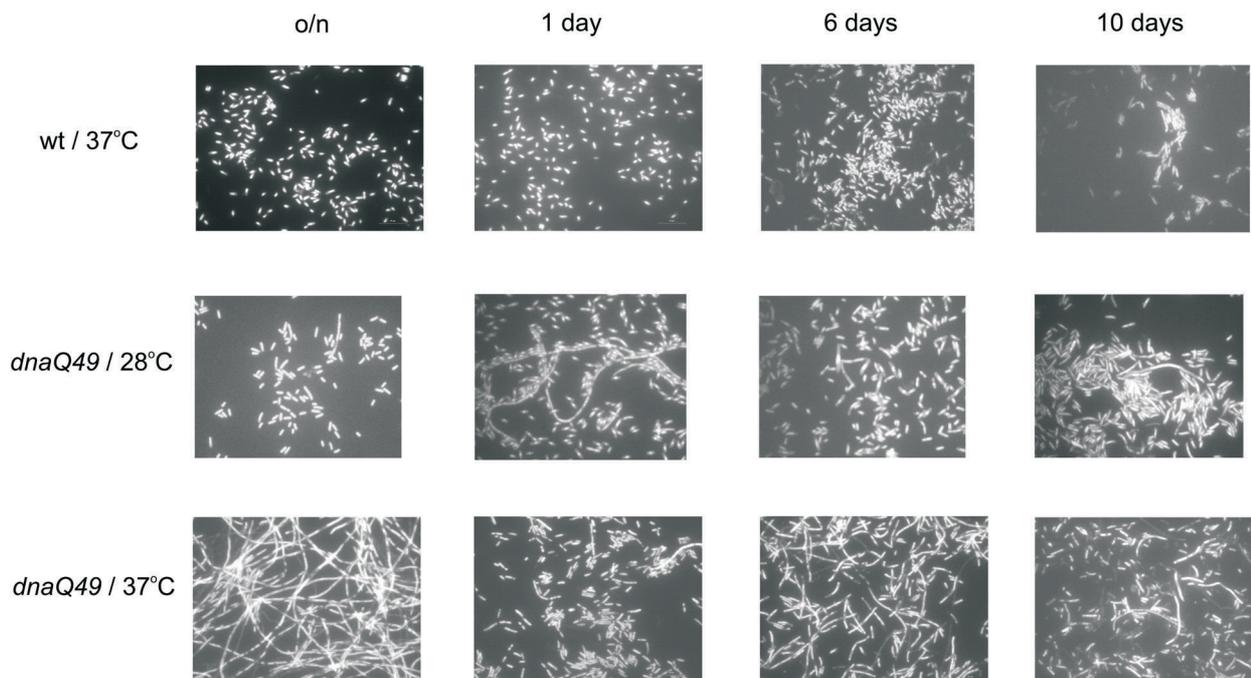


Figure 1. Kinetics of SOS induction in AB1157 strains.

Microscopic preparations were made from wt (AB1157) and *dnaQ49* overnight liquid cultures in E medium – first column (o/n) and from E-Arg plates incubated at the indicated temperatures for the indicated times – columns 2–4.

fluorescence microscope (Nikon Microphot S.A., chroma HQ-FLP 41012 filter) with 100× objective lens. All the photographs were taken under the same conditions and at the same 1800-fold magnification.

preparations were made of overnight liquid *dnaQ49* cultures in E medium and from the suspensions obtained by washing E-Arg plates (incubated for one, six and ten days) with 0.9% NaCl. The wild type strain showed

no filamentation when grown either in liquid or on solid medium, thus indicating no induction of the SOS response. The *dnaQ49* strain incubated at the permissive 28°C showed no filamentation when grown in liquid medium, weak filamentation after one day of incubation on E-Arg plates, and almost normal cell growth after 6 and 10 days of incubation (Fig. 1).

In contrast to the data obtained at 28°C, strong filamentation was observed in *dnaQ49* liquid culture grown overnight at 37°C. Unexpectedly, in preparations of one-day culture on E-Arg plates at 37°C, filaments were rare, but appeared again in 6- and 10-day cultures. However, the degree of filamentation after 10 days of incubation appeared lower than that after 6 days (Fig. 1). In the *dnaQ49ΔumuDC* strain, induction of the SOS response was at a similar level as in *dnaQ49* (not shown).

Growth-dependent and stationary-phase mutations in *dnaQ49* strains

In *E. coli* AB1157 reversion to prototrophy of the *argE3* locus proceeded in E minimal liquid medium with the frequency of 2.7 Arg⁺ colonies/10⁸ cells (Table 2). A similar frequency was observed in AB1157 with deletion

of the *umuDC* operon. Introduction of the temperature-dependent *dnaQ49* mutation into AB1157 dramatically increased the frequency of growth-dependent revertants at the non-permissive temperature of 37°C (870×10^{-8} cells *vs.* 2.7×10^{-8} cells in wt strain). At 28°C the frequency of Arg⁺ revertants was also elevated although to a much lesser degree in comparison to the wild type strain (57 *vs.* 2.7×10^{-8} cells). Additional deletion of the *umuDC* operon in *dnaQ49* bacteria led to a 2-fold (28.2×10^{-8} cells) and over 10-fold (80.2×10^{-8} cells) decrease in the level of the Arg⁺ revertants at 28°C and 37°C, respectively.

When AB1157 bacteria were plated on E-Arg plates and incubated for several days, Arg⁺ colonies that arose were the result of *argE3*→Arg⁺ reversion. The Arg⁺ colonies that appeared between the 4th and 11th day of incubation were regarded as stationary-phase revertants. Frequency of mutations was calculated per viable cell. The number of the cells was determined by washing E-Arg plates after cutting off the colonies of growth-dependent Arg⁺ revertants formed before the 4th day (Nowosielska & Grzesiuk, 2000) Since the number of living cells did not decrease during incubation, the initial values

Table 2. Growth-dependent and stationary-phase Arg⁺ revertants in AB1157 strains

Strain	Growth-dependent mutations			Adaptive mutations	
	c.f.u.* at time of plating × 10 ⁸	Arg ⁺ revertants/plate	Arg ⁺ revertants/10 ⁸ cells	The number of Arg ⁺ colonies/plate/day	Arg ⁺ revertants/10 ⁸ cells
wt [AB1157]	1.68 ± 0.5	4.5 ± 2.0	2.7 ± 0.8	2.2 ± 0.8	1.3 ± 0.5
<i>ΔumuDC</i>	2.4 ± 0.7	6.0 ± 2.7	2.5 ± 0.5	0.4 ± 0.1	0.125 ± 0.1
<i>dnaQ49</i> 28°C	2.0 ± 0.6	114.0 ± 20.5	57.1 ± 13.6	9.5 ± 3.9	4.73 ± 2.4
<i>dnaQ49</i> 37°C	0.3 ± 0.2	261.0 ± 42.5	869.9 ± 111.8	11.7 ± 4.4	39.1 ± 8.4
<i>dnaQ49ΔumuDC</i> 28°C	2.0 ± 0.6	56.4 ± 18.0	28.2 ± 8.4	7.3 ± 0.6	3.65 ± 0.9
<i>dnaQ49ΔumuDC</i> 37°C	1.2 ± 0.4	96.2 ± 21.0	80.2 ± 15.1	23.2 ± 3.4	19.3 ± 3.6

*colony forming units.

were taken for calculation. In the AB1157 and AB1157 Δ *umuDC* strains the rate of stationary-phase Arg⁺ revertants was very low (1.3 and 0.125×10^{-8} cells, respectively). The presence of the *dnaQ49* allele led to an over 3-fold increase in the rate of stationary-phase Arg⁺ revertants at 28°C (4.7×10^{-8} cells) and a 30-fold increase at 37°C (39.1×10^{-8} cells) (Table 2, last column). The absence of Pol V (*umuDC* deletion) in the *dnaQ49* strain was almost without effect on the level of Arg⁺ stationary-phase revertants incubated at 28°C (4.7×10^{-8} cells for *dnaQ49* and 3.7×10^{-8} cells for *dnaQ49\Delta**umuDC*), but led to a 2-fold decrease in the rate of mutations at 37°C (39.1×10^{-8} cells for *dnaQ49* and 19.3×10^{-8} cells for *dnaQ49\Delta**umuDC*).

Specificity of Arg⁺ spontaneous revertants

To determine the presence of suppressors in growth-dependent and stationary-phase Arg⁺ revertants, we used a previously described

revertants), and those which arose between the 4th and the 10th day (stationary-phase revertants). The results of these tests are summarized in Table 3.

Generally, in AB1157 strains Arg⁺ growth-dependent revertants were the result of *supB* and *supL* suppressor formation due to GC→AT transitions and AT→TA transversions, respectively. Some of these reversions arose by formation of an unknown suppressor or by other unidentified pathway(s). The different specificity of the reversions observed at the permissive and the non-permissive temperature in the *dnaQ49* strain is probably the result of disturbed α - ϵ interaction within the Pol III core at 37°C. At this temperature, in comparison to 28°C, a 2-fold decrease in GC→AT transitions, and an additional number of back mutations were observed. Interestingly, the introduction of *umuDC* deletion into *dnaQ49* led to similar proportions of *supB* and *supL* suppressors in *dnaQ49\Delta**umuDC* and in wt strains at 37°C.

Table 3. Specificity of growth-dependent and stationary-phase Arg⁺ spontaneous revertants in AB1157 *dnaQ49* strains

Strain/temp. of incubation	Growth dependent Arg ⁺ revertants (%)*				Stationary-phase Arg ⁺ revertants (%)*			
	back	supB	supL	unknown	back	supB	supL	unknown
wt	0	44	56	0	93	7	0	0
<i>dnaQ49</i> 28°C	0	53	25	22	48	14	3	35
<i>dnaQ49</i> 37°C	26	20	33	21	80	7	0	13
<i>dnaQ49\Delta</i> <i>umuDC</i> 28°C	17	13	51	19	24	0	0	76
<i>dnaQ49\Delta</i> <i>umuDC</i> 37°C	0	45	38	17	76	4	16	4

*Data are means of several experiments (for details see Materials and Methods).

method (Śledziwska-Gójska *et al.*, 1992) based upon estimation of the susceptibility of these revertants to a set of amber and ochre T4 phage mutants (see Methods for details). Two groups of Arg⁺ colonies were tested: the ones that appeared on E-Arg plates after 2 days of incubation (growth-dependent

The Arg⁺ stationary-phase revertants were of a markedly different specificity. In the wt, *dnaQ49*, and *dnaQ49\Delta**umuDC* strains at 37°C they arose mainly by back mutation in the *argE3* locus. In *dnaQ49*, and especially in *dnaQ49\Delta**umuDC* at 28°C, the fraction of Arg⁺ stationary-phase revertants that arose

by the formation of an unknown suppressor was appreciably larger than that found among growth-dependent revertants.

DISCUSSION

In this work we investigated spontaneous mutagenesis in exponentially growing and in growth limited *E. coli* strains deficient in the 3'→5' proofreading activity of Pol III, and additionally proficient or deficient in the SOS-inducible repair polymerase Pol V. The mutagenic pathways were tested in the AB1157 strain with the chromosomal *argE3* ochre mutation. Reversion of the *argE3* mutation to the Arg⁺ phenotype enabled growth on minimal medium deprived of arginine. The Arg⁺ revertants could arise by back mutations at the *argE3* ochre site or by suppressor formation.

Spontaneous mutagenesis in exponentially growing AB1157*dnaQ49*

Errors that occur during normal DNA replication can be a major source of spontaneous mutations in bacteria. In *E. coli* high fidelity of replication is ensured by the α and ϵ subunits of Pol III. The *dnaE*-encoded α subunit matches the nucleotides in the newly synthesized DNA strand to the template DNA. The *dnaQ*-encoded ϵ subunit shows 3'→5' exonuclease activity that removes the incorrect nucleotide from the growing end of nascent DNA. This feature of ϵ is disturbed in the *dnaQ49* strain. The mutator phenotype (a high level of spontaneous mutations) is fully expressed at the non-permissive temperature of 37°C and to a much lesser degree at the permissive 28°C. In the *dnaQ49* strain mutations can arise as a result of: (i) lack of 3'→5' exonuclease function; (ii) induction of the SOS-directed error-prone Pol V, and (iii) disturbed ϵ - α interaction within Pol III at a non-permissive temperature, enabling a better access of SOS-inducible polymerases to

the newly synthesized fragment of DNA. In *dnaQ49* growing at 37°C the high level of growth-dependent Arg⁺ revertants (870 *vs.* 2.7 in wt strain) results from the loss of the proofreading activity of the ϵ subunit and probably saturation of the mismatch repair system (MMR) (Mellon *et al.*, 1996; Schaaper, 1993), as well as from the disturbance in the α - ϵ interaction within the Pol III core, which leads to Pol III destabilization. Filamentous growth of *dnaQ49* in liquid medium at 37°C indicates strong induction of the SOS response. Elimination of *umuDC* from the *dnaQ49* strain led to a 10-fold decrease in the number of growth-dependent Arg⁺ revertants. These results indicate that a large fraction of the growth-dependent mutations in the *dnaQ49* strain at 37°C is due to spontaneous mutagenesis involving the SOS-induced repair polymerase Pol V (UmuD'₂C proteins). On the other hand, since in *dnaQ49* Δ *umuDC* we still observed an elevated level of Arg⁺ growth-dependent revertants, and the same level of SOS induction as in *dnaQ49*, we assume that, in the absence of Pol V, defective polymerases III as well as pol IV or pol II may contribute to DNA mutagenesis (Nowosielska *et al.*, 2004).

Spontaneous mutagenesis in stationary-phase AB1157*dnaQ49*

In spite of extensive studies on the phenomenon of adaptive mutagenesis, the mechanism of spontaneous mutation formation in the absence of DNA replication is still not clear. Studies in the FC40 system have indicated that in adaptive mutagenesis such processes as recombination (Harris *et al.*, 1997), hypermutability (Rosche & Foster, 1999), and amplification (Slecht *et al.*, 2003) of sequences under selection are involved. In contrast, in the *argE3* → Arg⁺ system studied here the main source of stationary-phase mutations in *dnaQ49* cells are DNA lesions left by Pol III defective in correction activity. These Arg⁺ reversions were mostly back mu-

tations in the reporter gene, whereas, the growth-dependent reversions resulted from suppressor formation.

Besides impaired Pol III, Pol V also contributes to the increase of Arg⁺ revertants. However, the action of Pol V seems to have a lesser effect on the level of Arg⁺ revertants in starving than in growing *dnaQ49* cells. The frequency of mutations at 37°C was 2-fold lower in *dnaQ49ΔumuDC* than in *dnaQ49* while in growing cells this rate was about ten times decreased (Table 2).

Additionally, it seems that in the case of the *dnaQ49* mutant, the temperature of incubation, permissive or non-permissive, is more important for the formation of stationary-phase Arg⁺ revertants than the presence or absence of Pol V. Since in the *dnaQ49* strain at 37°C the level of growth-dependent as well as stationary-phase Arg⁺ revertants is much higher than at 28°C, we assume that the destabilization of Pol III core at 37°C is more critical for the occurrence of Arg⁺ revertants than the absence of properly operating 3'→5' proofreading activity of the mutated ε subunit. It is possible that partial disconnection of Pol III at the replication fork facilitates the access of Pol V, Pol IV and Pol II up-regulated within the SOS system. It has been reported that Pol IV is required for adaptive mutations in the *lac* operon of FC40 strain and, together with Pol III, can account for all the adaptive point mutations at *lac* (Tomkins *et al.*, 2003). In our *argE3* → Arg⁺ reversion system Pol IV may play a similar role and to some extent replace Pol V (Nowosielska *et al.*, 2004).

REFERENCES

- Bachmann BJ. (1987) Derivations and genotypes of some mutant derivatives of *Escherichia coli* K-12. In *Escherichia coli and Salmonella typhimurium: Cellular and Molecular Biology*, Neidhardt FC, Ingraham J, Low KB, Magasanik B, Schaechler M, Umberger HE, eds, vol 2, pp 1190–219. AMS Press, Washington, D.C.
- Bridges BA. (1999) DNA repair: Polymerases for passing lesions. *Curr Biol.*; **9**: R475–7.
- Bull HJ, McKenzie GJ, Hasting PJ, Rosenberg SM. (2000) Evidence that stationary-phase hypermutation in *Escherichia coli* chromosome is promoted by recombination. *Genetics.*; **154**: 1427–37.
- Cairns J, Overbaugh J, Miller S. (1988) The origin of mutants. *Nature.*; **335**: 142–5.
- Cairns J, Foster PL. (1991) Adaptive reversion of a frameshift mutation in *Escherichia coli*. *Genetics.*; **128**: 695–701.
- Courcelle J, Khodursky A, Peter B, Brown PO, Hanawalt C. (2001) Comparative gene expression profiles following UV exposure in wild type and SOS deficient *Escherichia coli*. *Genetics.*; **158**: 41–64.
- Crowley DJ, Courcelle J. (2002) Answering the cell: Coping with DNA damage at the most inopportune time. *J Biomed Biotechnol.*; **2**: 66–74.
- Foster PL. (1993) Adaptive mutation: the uses of adversity. *Annu Rev Microbiol.*; **47**: 467–504.
- Foster PL. (1999) Mechanisms of stationary phase mutations: A decade of adaptive mutation. *Annu Rev Genet.*; **33**: 57–88.
- Foster PL, Gudmundsson G, Trimarchi JM, Cai H, Goodman MF. (1995) Proofreading defective DNA polymerase II increases adaptive mutation in *Escherichia coli*. *Proc Natl Acad Sci USA.*; **92**: 7951–5.
- Friedberg EC, Gerlach VL. (1999) Novel DNA polymerases offer clues to the molecular basis of mutagenesis. *Cell.*; **98**: 413–6.
- Friedberg EC, Walker GC, Siede W. (1995) SOS response and DNA damage tolerance in Prokaryotes. In *DNA Repair and Mutagenesis*, Friedberg EC, Walker GC, Siede W, eds, pp 407–34, ASM Press, Washington, DC.
- Hall BG. (1990) Spontaneous point mutations that occur more often when they are advan-

- tageous than they are neutral. *Genetics.*; **90**: 673–91.
- Harris RS, Bull HJ, Rosenberg SM. (1997) A direct role for DNA polymerase III in adaptive reversion of a frameshift mutation in *Escherichia coli*. *Mutat Res.*; **375**: 19–24.
- Janion C. (2001) Some aspects of the SOS response system – a critical survey. *Acta Biochim Polon.*; **48**: 599–610.
- Janion C, Sikora A, Nowosielska A, Grzesiuk E. (2002) Induction of the SOS response in starved *Escherichia coli*. *Envir Molec Mutagen.*; **40**: 129–33.
- Jonczyk P, Nowicka A, Fijalkowska I, Schaaper RM, Ciesla Z. (1998) *In vivo* protein interactions within the *Escherichia coli* DNA polymerase III core. *J Bacteriol.*; **180**: 1563–6.
- Kuzminov A. (1999) Recombinational repair of DNA damage in *Escherichia coli* and bacteriophage. *Microbiol Mol Biol Rev.*; **63**: 751–813.
- Mellon I, Rajpal DK, Koi M, Boland CR, Champe GN. (1996) Transcription-coupled repair deficiency and mutations in human mismatch repair genes. *Science.*; **272**: 557–60.
- Nowosielska A, Grzesiuk E. (2000) Reversion of *argE3* ochre strain *Escherichia coli* AB1157 as a tool for studying the stationary-phase (adaptive) mutations. *Acta Biochim Polon.*; **47**: 459–67.
- Nowosielska A, Janion C, Grzesiuk E. (2004) Effect of deletion of SOS-induced polymerases, pol II, IV and V, on spontaneous mutagenesis in *Escherichia coli mutD5*. *Envir Molec Mutagen.*; **43**: 226–34.
- Rosche WA, Foster PL. (1999) The role of transient hypermutators in adaptive mutation in *Escherichia coli*. *Proc Natl Acad Sci U S A.*; **96**: 6862–7.
- Schaaper RM. (1993) Base selection, proofreading, and mismatch repair during DNA replication in *Escherichia coli*. *J Biol Chem.*; **268**: 23762–5.
- Schaaper RM, Cornacchio R. (1992) An *Escherichia coli dnaE* mutation with suppressor activity toward mutator *mutD5*. *J Bacteriol.*; **174**: 1974–82.
- Slecht ES, Bunny KL, Kugelberg E, Kofoid E, Andersson DI, Roth JR. (2003) Adaptive mutation: general mutagenesis is not a programmed response to stress but results from rare coamplification of *dinB* with *lac*. *Proc Natl Acad Sci USA.*; **100**: 12847–52.
- Śledziwska-Gójska E, Grzesiuk E, Płachta A, Janion C. (1992) Mutagenesis of *Escherichia coli*: A method for determining mutagenic specificity by analysis of tRNA suppressors. *Mutagenesis.*; **7**: 41–46.
- Taft-Benz SA, Schaaper RM. (1998) Mutational analysis of 3'→5' proofreading exonuclease of *Escherichia coli* DNA polymerase III. *Nucleic Acids Res.*; **26**: 4005–11.
- Takano K, Nakabeppu Y, Maki H, Horiuchi T, Sekiguchi M. (1986) Structure and function of *dnaQ* and *mutD* mutators of *Escherichia coli*. *Mol Gen Genet.*; **205**: 9–13.
- Tompkins JD, Nelson JL, Hazel JC, Leugers SL, Stumpf JD, Foster PL. (2003) Error-prone polymerase, DNA polymerase IV, is responsible for transient hypermutation during adaptive mutation in *Escherichia coli*. *J Bacteriol.*; **185**: 3469–72.
- Vogel HJ, Bonner DM. (1956) Acetylo-ornithinase of *Escherichia coli*: Partial purification and some properties. *J Biol Chem.*; **218**: 97–106.
- Walker GC. (1998) Skiing the black diamond slope: progress on the biochemistry of translesion DNA synthesis. *Proc Natl Acad Sci USA.*; **95**: 10348–50.
- Witkin EM. (1967) The radiation sensitivity of *Escherichia coli* B: a hypothesis relating filament formation and prophage induction. *Proc Natl Acad Sci USA.*; **57**: 1275–9.
- Woodgate R. (1992) Construction of a *umuDC* operon substitution mutation in *Escherichia coli*. *Mutat Res.*; **281**: 221–5.

- Woodgate R. (1999) A plethora of lesion-replicating DNA polymerases. *Genes Dev.*; **13**: 2191–5.
- Yigit H, Reznikoff WS. (1997) Examination of the Tn5 transposase overproduction phenotype in *Escherichia coli* and localization of suppressor of transposase overproduction killing that is an allele of *rpoH*. *J Bacteriol.*; **179**: 1704–13.