activity (BR regressed) to the secretion of the ecdysone, and others with still-active puffing (BR expanded). The centromeres of the salivary gland chromosomes are represented by blocks (C) of extremely condensed chromatin. They divide the chromosomes into a short left and a long right arm.

One of the 57 mutants, in the transition stage, showed structural heterozygosity in the left arm of chromosome I in all salivary gland cells (fig., b-f) for a complex of an adjoining deletion and an inversion. The location of these rearrangements is shown in the figure, a. The deletion includes 28 bands (1A17-1A71; according to the unpublished chromosome map by Staiber and Behnke, bands with uneven and interbands with even numbers). One of the inversion breakpoints is located within the centromere. As a consequence, a part of the centromeric region is separated and transposed into a euchromatic part of the left arm (adjacent to band 1A73; fig., c). The anterior lobe of the salivary gland of *A. lucidus* consists of 12–16 cells. In one of the salivary glands of the mutation carrying F_1-animal all 16 cells of the anterior lobe could be examined. In 10 cells, in which the BR 3 and BR 4 were fully expanded, the segment formed a BR (fig., b and e), whereas in 6 cells, in which BR 3 and BR 4 were regressed, the segment was present in a slightly puffed or condensed form respectively (fig., c and f). Apparently the segment concerned has the same pattern of activity as the loci of BR 3 and BR 4. So one can suppose that in the new position its activity is under the control of ecdysone.

In all the cells of the main and side lobes the segment was still somewhat disaggregated like a regressing structure of a formerly expanded BR (fig., d).

In the normal position, integrated in the centromere, the segment never shows any puffing or RNA-synthesis (no incorporation of ^{3}H-uridine, no specific RNA-staining by toluidine-blue- or methylgreen-pyronin-staining).

The expression of this special BR is interpreted as a position effect, due to a change of chromosomal structure[6-8].

Evidently, the locus is not able to express a BR in the regular position; in the new position, however, the locus is puffed to a BR. Another case of a position effect in *A. lucidus* has been described by Mechelke[9].

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Host cell reactivation of ozone-treated T3 bacteriophage by different strains of *Escherichia coli*

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Summary. Host cell reactivation capacity for ozone T3 phage was determined for different *E. coli* strains deficient in one or more of the DNA repair mechanisms. The results indicate that DNA polymerase I appears to play a key role in the repair of damage produced on the DNA by ozone while the *lex*A gene product seems to play a minor one.

In the last few years considerable data have been accumulated suggesting that ozone may be radiomimetic[2,3], mutagenic[4,5] and can cause some DNA degradation in different strains of *E. coli*[6-8]. Furthermore, ozone has been reported to cause chromosome breakage in *Vicia faba*[9] and in mammalian cell cultures[10]. There is still however very little information available on the ozone DNA-damage and the repair mechanism(s) in the cell.

Previous workers have defined host cell reactivation (HCR) by the fact that UV-damaged phage lambda has different plating efficiency depending on the genetic background of the host cell in which it multiplies[12]. With the need to determine rapidly the DNA damage repair capacity of different bacterial strains, many researchers have applied this process with T1, T3 and T7[13,14]. Since ozone inactivation of T3 phage is caused in part by DNA damage[15,16], this technique seemed very useful in the possible determination of the ozone DNA-damage repair mechanism(s).

Material and methods. Stocks of T3 phage were prepared by the confluent lysis method with *E. cole* B251[17]. Water suspensions of T3 phage were exposed to 10 ppm of ozone for 10 min as described elsewhere[5]. Samples (0.1 ml) were removed from the phage suspensions before and after treatment and proper dilutions were mixed with a log phase of different bacterial strains (m.o.i = 0.02) and plated on nutrient agar plates (Difco) by the layer method[17]. Plates were incubated at 37 °C and counted the next day. We calculated the fraction of survivors (S/S°) and the reactivation factor (fraction of survivors with the mutant strain/fraction of survivors with the wild type strain) for each bacterial strain. Ozone inactivation curve was done on nutrient agar plates (Difco) as previously described[5,17] and control experiments were carried out with clean air instead of ozone.

Results and discussion. The ozone inactivation curve of T3 phage to 10 ppm of ozone and for different intervals of treatment-time is represented in the figure. According to these results, the inactivation capacity of ozone on this bacteriophage increases with increasing treatment time. This type of linear decrease through the origin during ozonation has been observed with different types of virus[15,18,19].

The host cell reactivation capacity obtained with different strains of *E. coli* are shown in table 2. The host cell strains with no deficiency in any of the DNA repair mechanisms, B251, JG139 and HMS49, should theoretically reactivate the ozone-treated phage. Based on this assumptions, we calculated the host cell reactivation factor for different

mutants of *E. coli*. For better comparison, the reactivation factor was determined for different host cells which are derived from the same wild type strain.

Previous results from this laboratory[20,21] have shown that the *lex*A gene product is essential to the recovery of *E. coli* from the effects of ozone. The host cell reactivation factors for *lex*A mutants were 0.85 (MQ309) and 0.83 (Bs-2). These results confirm the possible role of the *lex*A gene product in the repair of the ozone DNA-damage in *E. coli*.

*Pol*A defective strains repair single-stranded DNA breaks poorly[22,23] and are extremely sensitive to X-rays[22,23] and ozone[24]. Furthermore, *pol*A1 strains degrade their DNA more rapidly and to a greater extent than *pol*$^+$ strains after ozonation[6]. It was found in the present study that host cell reactivation factor in *pol*A strains varied between 0.43 and 0.67, the lowest reactivation factors obtained in this study. On the basis of these results, DNA polymerase I seems to play a major role in the repair of ozone DNA-damage in *E. coli*.

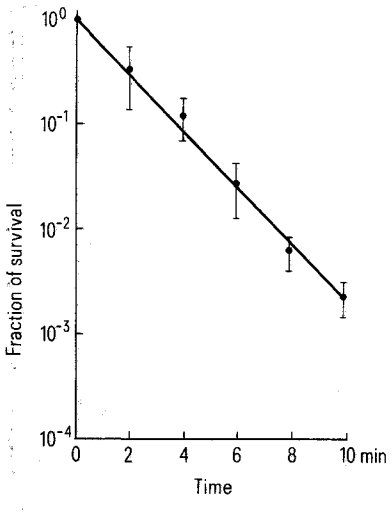
Table 1. Bacterial strains

Strain	Genotype	Source
B251	*Escherichia coli* B wild type	Arber and Morse (1965)
Bs-2	*lex*A *mal*B	Hill and Simpson (1961)
MQ309	*lex*A, derivative of B251	This laboratory
HMS49	*pol*$^+$	Campbell et al. (1972)
HMS83	*pol*A1 *pol*B derivative of HMS49	Campbell et al. (1972)
JG138	*pol*A1 derivative of JG139	Campbell et al. (1972)
JG139	*thy*A *rha lac*Zam	Campbell et al. (1972)
MQ208	*uvr*A *mal*B *met*Av derivative of B251	This laboratory
PE109	*pol*A1 derivative of JG138	Emmerson and Howard-Flanders (1965)

Table 2. Host cell reactivation capacity for the ozone-treated T3 phage

Host cell strain		Reactivation factor
B251	(Wild type)	1
Bs-2	(*lex*A)	0.83 ± 0.07*(5)**
MQ208	(*uvr*A)	3.91 ± 0.35 (3)
MQ309	(*lex*A)	0.85 ± 0.14 (4)
JG139	(*pol*$^+$)	1
JG138	(*pol*A)	0.43 ± 0.07 (3)
PE109	(*pol*A)	0.67 ± 0.12 (3)
HMS49	(*pol*$^+$)	1
HMS83	(*pol*A*pol*B)	0.44 ± 0.02 (3)

*Mean $\pm$ SE; **number of experiments.
Reactivation factor: fraction of survivors with mutant strain/fraction of survivors with wild type strain.



Ozone inactivation curve of T3 phage to a concentration of 10 ppm. Average of 3 experiments.

The *pol*A*pol*B strain (HMS83) showed about the same host cell reactivation capacity for the ozone-treated phage as the *pol*A strains. This is in agreement with the radiation and ozonation data which have indicated that DNA polymerase II plays no major role in the repair of DNA damage[24,25].

We also found that the *uvr*A strain (MQ208) can efficiently reactivate ozone-treated phages. Excision repair defective strains are deficient in the host cell reactivation of UV-treated phages[26], thus their insensitivity to ozone-treated phages suggests that the ozone DNA-damage to be quite different from the UV DNA-damage.

Previous works have suggested that the majority of excision gaps are repaired by the DNA polymerase I dependent short patch repair[27,28]. According to the results presented in this paper, DNA polymerase I appears to play a key role in the repair of damage produced on the DNA by ozone while the *lex*A gene product seems to play a minor one. Furthermore, we found that *uvr*A gene product seems to have a certain role in the repair efficiency of ozone DNA-damage in *E. coli*, but at present it is not possible to draw any conclusions as to the nature of the role of that gene product on the host cell reactivation capacity for ozone-treated phage.

1 This research was sponsored by the National Sciences and Engineering Council of Canada. One of us (PLH) acknowledges a scholarship from the NSERC of Canada.
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