THE EFFECTS OF AN R-FACTOR AND ITS NON-TRANSMISSIBLE DERIVATIVE ON THE SURVIVAL OF BACTERIA IN MEDIA CONTAINING MITOMYCIN C

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R46, termed here R46tra$^+$, is a self-transmissible (tra$^+$), N-incompatibility group R-factor, conferring resistance to: ampicillin; streptomycin; sulphonamides; tetracyclines and ultraviolet (U.V.) irradiation. Alldrick and Smith (1978) described the construction of a non-transmissible (tra$^-$) derivative of this plasmid termed R46tra$^-$, which possessed all the resistance genes, but lacked the necessary genetic information for transfer. It was also demonstrated, that under non-selective conditions, possession of either R-factor (i.e. the R$^+$ state) did not significantly affect the ability of the host to survive, compared with R-factor deficient (R$^-$) strains in single batch cultures. It was however shown that in mixed R$^+$/R$^-$ batch cultures possession of either R-factor significantly affected the host's ability to survive prolonged incubation.

Subsequent studies have been carried out on the effects of these R-factors on their hosts in the presence of antimicrobial agents of known mode of action, and against which the R-factors do not confer resistance. In this communication the effect of mitomycin C, which inhibits DNA synthesis by monofunctional alkylation and by causing cross-linking between DNA stands, was investigated using single batch cultures of the R$^-$ and R$^+$ strains. The results were that all strains exhibited similar inhibitory responses to mitomycin C at concentrations of 0.25, 0.5, or 1.0 μg/ml. However, when mitomycin C was tested at 0.5 μg/ml in mixed R$^+$/R$^-$ cultures it was found that possession of either R-factor, especially R46tra$^-$, conferred a disadvantage to the host, particularly during the initial stages of the growth phase.

There are three possible explanations for these results:

i) the plasmid-mediated U.V resistance mechanism also functions against alkylated bases, and that these functions are absent from R46tra$^-$;
ii) R46tra$^+$ possesses genes that independently confer resistance to mitomycin C which in the case of R46tra$^-$ were lost at the same time as the transfer genes when the mutant R-factor was constructed.
iii) this effect is due to an interaction between R$^+$ and R$^-$ cells, which is enhanced by the presence of mitomycin C and is masked in the R46tra$^+$/R$^-$ mixture due to mating.

Experiments designed to test these hypotheses showed that both R$^+$ strains were similarly more resistant to U.V. irradiation than the R$^-$ strain, while minimum inhibitory concentration (M.I.C.) experiments with mitomycin C showed that possession of neither R-factor affected the M.I.C. compared with the R$^-$ strain. It therefore seems that mitomycin C enhances competitive effects between R$^+$ and R$^-$ bacteria when grown together in batch culture particularly during the early phases of growth. Moreover, it would seem that these effects can be obscured by mating. Whether this phenomenon is the same as during prolonged incubation of R$^+$/R$^-$ mixtures is not known. However, these results emphasise the need for the use of tra$^-$ R-factors when studying interactions of this kind.

Alldrick, A.J. & Smith, J.T. (1978) J.Pharm.Pharmacol. 30 (Suppl.) 18p