

## DO R-FACTORS AFFECT THE ABILITY OF HOST BACTERIA TO COMPETE FOR NUTRIENTS?

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Possession of R-factors by bacteria, i.e. the R<sup>+</sup> state, confers an advantage over bacteria lacking them (R<sup>-</sup>), when antibiotics to which the R<sup>+</sup> bacteria are resistant are in use. It is often believed that should the use of antibiotics cease, the occurrence of R-factors would diminish. There is however little evidence to support this view and on the contrary most studies reveal little difference in the growth of R<sup>+</sup> and R<sup>-</sup> bacteria. A major problem with this work is that in mixed cultures the R<sup>+</sup> bacteria can transfer, by mating, the R-factor to R<sup>-</sup> bacteria, thus making results of mixed culture studies difficult to interpret. Consequently most workers have studied the growth of each type of organism individually with the attendant drawback that there is no direct competition between the R<sup>+</sup> and R<sup>-</sup> bacteria.

To avoid this problem, a self-transmissible R-factor, R46, which confers resistance to ampicillin, tetracycline, sulphonamide, streptomycin and UV irradiation was taken and a derivative constructed by P22 transduction in Salmonella typhimurium. This was done because P22 cannot incorporate all the DNA of R46, consequently this procedure caused deletion of parts of the R46 genome. The chosen derivative, R46 tra<sup>-</sup>, possessed all the antibiotic and UV resistance genes but lacked the apparatus responsible for the transmission of normal R46 by mating. Due to its inability to transfer, the R46 tra<sup>-</sup> derivative was inserted into Escherichia coli by transformation using purified R-factor DNA and calcium chloride treated recipients.

Growth studies were carried out using strains possessing the two plasmids and the R<sup>-</sup> strain. Results showed that as far as multiplication was concerned, neither plasmid significantly affected the rate of growth of either R<sup>+</sup> strain grown individually when compared with the R<sup>-</sup> strain grown alone. Even in mixed cultures with the R<sup>-</sup> strain neither plasmid affected the ability of the R<sup>+</sup> bacteria to compete during the growth phase. However when incubation was carried on beyond the stationary phase the presence of these R-factors did affect the proportional survival of the R<sup>+</sup> bacteria. The results with the R46 tra<sup>+</sup> were much more erratic and the difference between it and R46 tra<sup>-</sup> can be ascribed to mating which seems to occur spasmodically during prolonged incubation in laboratory media. In conclusion these studies show that R-factors can affect the ability of R<sup>+</sup> bacteria to survive in the presence of R<sup>-</sup> bacteria, even when both strains grow equally well. These differences either do not occur when the organisms are cultivated alone or they are too small to detect in individual cultures.

The study of population dynamics of mixed cultures of R<sup>+</sup> and R<sup>-</sup> bacteria under non-selective conditions is of increasing relevance to antimicrobial therapy, particularly with regard to prophylactic and preventive treatment. It is believed that the use of tra<sup>-</sup> mutants of R-factors will provide a better simulation of in vivo conditions where mating is known to occur at much lower frequencies than in laboratory media.