

FUNCTIONAL ORGANISATION OF THE *nif* GENES CLUSTER
OF *KLEBSIELLA PNEUMONIAE*.

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In order to study the functional organisation of the *nif* genes of *Klebsiella pneumoniae*, genetic and biochemical properties of both chromosomal and plasmid point mutants, prophage Mu insertion mutants and deletion mutants were examined.

Genetic analysis was performed with diploids by recombination or complementation using *rec A* recipient strains. A fine structure map of the *nif* cluster was established.

Analysis of ¹⁴C pulse-labelled proteins synthesized by either Nif⁺ or Nif⁻ strains was achieved by comparison of two-dimensional gel autoradiograms. *In vitro* complementation tests for nitrogenase activity, using crude extracts of different mutants, were done either with purified nitrogenase components or by mixing crude extracts from different mutants. In addition, we looked for CRM with specific antisera obtained against Kp₁ or Kp₂. We thus were able : 1) to characterize some of the *nif* genes products, 2) to underline the negative pleiotropic effect of *nif*⁻ A mutations on the expression of the other *nif* genes, 3) to identify Nif⁻ cryptic mutants which exhibit nitrogenase activity *in vitro*. Finally, both genetic and biochemical analysis of polar mutants lead to the conclusion that the 14 *nif* genes are organised into 8 independent transcription units.

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