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ACTINOMYCETES



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nomycete taxa, the most comprehensive studies having been carried out on streptomycetes (Vickers *et al.*, In: *Biological, Biochemical and Biomedical Aspects of Actinomycetes*. Academic Press, London, 553, 1984; Williams *et al.*, In: *The Microbe*. Cambridge University Press, part II, 219, 1984). Information from the appropriate numerical taxonomic studies was used as a logical basis for the formulation of new media, selective for the isolation of particular species groups of *Nocardia*, *Rhodococcus* and a-wcIV actinomycetes. The principles and preliminary results of this approach are outlined here.

New selective isolation media were formulated with the assistance of the DIACHAR program (Sneath, *Computers and Geosciences*, 6, 21, 1980) which selects the most diagnostic characters for individual clusters within a numerical data matrix as well as indicating the degree of differentiation of each particular species group. The program was thus used to select nutritional or tolerance characters which could usefully be incorporated into new selective isolation media. These objectively devised sets of media, when applied together will provide a more accurate picture of the qualitative nature of soil actinomycete populations rather than the 'general' isolation media presently in use.

Fifty one new media were designed for the selective isolation of eleven species groups of *Nocardia*, *Rhodococcus* and a-wcIV actinomycetes. The selectivity of these media was first assessed by comparing the growth of a range of cluster representatives on these new media and on a control medium (currently used for the isolation of the particular group being tested). Of these fifty one new media, eight were subsequently found to be sufficiently selective for their target clusters. These were then evaluated quantitatively and the most practically selective media will be used in soil isolation studies. These new media will be applied to a variety of soils from diverse habitats with the objectives of isolating unexploited actinomycetes with industrial potential by providing

better quality biological material for screening.

ISOLATION AND IDENTIFICATION OF CELLULOSE- AND LIGNIN-DEGRADING ACTINOMYCETES FROM THE GUT OF HIGHER TERMITES (*TERMITIDAE*)

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From the gut of termite workers (*Macrotermes*, *Armitermes*, *Microcerotermes*, *Odontotermes*) it was possible to isolate lignocellulosolytic actinomycetes by using selective conditions.

These consisted mainly of enrichment cultures in liquid media with cellulose as the sole carbon source followed by inoculation onto the surface of cellulose filters laid on solid media containing mineral salts. Alternatively inoculation was carried out on bilayered solid media plus Avicel as a carbon source.

The temperature for optimal growth of the isolates ranged from 28°C to 46°C; optimal pH for growth was 6.0 to 7.0. All isolates (20/20) were able to grow on pure cellulose, supplied either as filter paper or a microcrystalline substrate (Pasti and Belli, *FEMS Microbiol.Lett.*, 26, 107, 1985), and in the presence of 6.0×10^{-3} mM phenol or 1 mM guaiacol. Although all of them are able to grow on Kraft lignin in the presence of other carbon sources, only three isolates exhibited a strong ability to metabolize the former substrate.

The ability of the isolates to degrade lignin was assessed by monitoring the degradation of labelled substrates and the consequent formation of $^{14}\text{CO}_2$, determining substrate losses during growth on purified lignocellulose, and quantifying the amount of APPL (acid precipitable polymeric lignin) formed during substrate degradation (Pasti *et al.*, paper submitted, 1990).

Most isolates produce extracellular peroxidases and esterases, some of which are inducible in the presence of wheat straw. Twelve strains were identified using the probabilistic matrix of Williams *et al.* (*J.gen.Microbiol.*, **129**, 1815, 1983) and the matrix of Langham *et al.* (*J.gen.Microbiol.*, **135**, 121, 1989). The identified strains were found to belong to the clusters *Streptomyces chromofuscus*, *S.rochei* and *S.diastaticus*.

NEW APPROACHES TO THE
IDENTIFICATION OF
STREPTOMYCETES WHICH
PRODUCE NOVEL BIOACTIVE
COMPOUNDS

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As the number of known antibiotics increases it becomes more difficult for industry to discover new bioactive compounds. Methodologies which enhance the discovery of new antibiotics and improve the identification of those organisms capable of producing them are required to counteract this problem. There are many innovations within our group which aspire to improve the identification of bioactive strains.

Certain physiological characters may be diagnostic for bioactivity, current research involves investigating antibiotic resistance, fatty acid and metabolite profiles as phenotypic markers for predicting bioactivity. The bioactivities of streptomycete isolates are superimposed onto their phenotypic distributions. Progress is being made to show that correlations do indeed exist between these two variables.

Systems for wide gene expression are under development, where strains are exposed to wide ranging environmental conditions, using minimal amounts of time and resource. We also hope to investigate the specificity and application of certain physiological factors to the provocation of wide gene expression and therefore gain better un-

derstanding of environmental factors affecting production.

Removing the ability to produce known broad spectrum compounds may allow isolates to produce new compounds or facilitate the detection of novel narrow spectrum compounds. We are using a model system to investigate these effects and hope to develop repression systems for a particular polyether and ansamycin antibiotics.

SELECTED PROBLEMS AND
APPROACHES TO ACTINOMYCETE
TAXONOMY

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Actinomycete taxonomy, dominated for decades by the use of morphological, physiological and ecological characters and culminating in their statistical processing, entered the molecular level with the elucidation of peptidoglycan structures. These and further chemotaxonomic characters were understood to reflect phylogenetic relationships more closely than the attributes used before. Continuing this line, the base sequences (or their reflections, respectively) of DNA, RNA and particularly 16S rRNA were used to achieve phylogenetic evidence. Both in phenetical and phylogenetical studies the comparisons of the organisms or the macromolecules data sets are presented in the form of trees or other graphical approaches. The trees need to be translated into hierarchical systems as bases of worldwide communication on the microorganisms being and capacities. This means, the levels of ramifications must be defined to determine the nomenclatural ranks, particularly those of the species and the genus. As a matter of principle and of experience, this can only be performed empirically. *I.e.*, the phylogenetic relationships of the organisms will be elucidated more and more in detail, and the systems to be used in biological sciences and in practice will be continuously adopted; but they will always be man-made and, consequently, a subject of controversy. Among the