

aortas and is highest in the coronary arteries of subjects who died of coronary heart disease.

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Computer Classification of Streptococci, mostly of Oral Origin

Streptococcus is a well established bacterial genus, but its sub-division into species has always been controversial. Wilson and Miles¹ expressed their dissatisfaction with existing classification schemes for streptococci, revision of which was long overdue.

An objective method of classification was introduced by Sneath², who pioneered the use of computers in the taxonomy of microorganisms. His approach has been used by Colman³, who examined Lancefield groupable streptococci, and by Raj and Colwell⁴, who found that the existing classification of enterococci left much to be desired. Carlsson⁵ has recently examined the taxonomy of oral streptococci, which have never been satisfactorily classified.

The discovery and isolation of caries-inducing streptococci have emphasized the need for a suitable classification, and have led Guggenheim⁶ to appeal for such a scheme, which would embrace organisms of oral origin and award a taxonomic status to the cariogenic streptococci.

To meet this need, we began the work described here. We noted 178 characteristics for each of 252 streptococcal isolates. Our collection of streptococci included sixty-three type culture strains and forty-seven rat faecal isolates, the remainder being human oral isolates collected at Liverpool. Characteristics observed included cellular and colonial morphology; inhibition by antibiotics, dyes, inorganic substances, and synthetic derivatives; sugar fermentation; polysaccharide synthesis; cell wall analysis; amino-acid metabolism; possession of various enzymes. The 50,000 results we obtained had to be handled by a computer, for which we used a program devised by Harrison⁷. This program differed from most programs previously used by other workers, for objects were clustered not in accordance with their overall similarity based on characteristics of equal weight, but according to the probability of combinations of characteristics occurring together.

The IBM computer used was programmed to examine the experimental data and print out clusters of like-

organisms which formed at various levels of significance and the shared characteristics of which could not be explained by random distribution.

By examining (1) the overlap of strains found in any two clusters, (2) the overall similarity of the central strains in each cluster, and (3) the more highly significant characteristics displayed by clusters, it was possible to merge similar clusters, so creating twelve phenons. Two phenons displayed combinations of characteristics, not ascribable to any species of *Streptococcus* described in Bergey's manual⁸. These phenons will be fully discussed in a future publication. Five phenons were found to contain representatives of the enterococcus group; in keeping with the findings of Raj and Colwell⁴, we found that members of any particular designated species of faecal streptococci did not cluster together. A typical phenon contained two strains of *Strep. faecalis*, one each of *faecium*, *durans*, *faecalis* var. *zymogenes*, and *faecalis* var. *liquefaciens*. To resolve this impasse, the five phenons can be provisionally termed *Strep. bovis*, and *Strep. faecalis* varieties I, II, III and IV.

Phenons corresponding to the well established species *Strep. mitis*, *Strep. salivarius* and *Strep. sanguis* were found. Representatives of Lancefield groups F, G, M, O and P were found in a single phenon and resembled the species *Strep. anginosus* described in Bergey's manual⁸. Only one member of each of these Lancefield groups had been included in this study, and if more representatives had been examined, sub-division of this phenon might have occurred.

Finally, a very highly significant phenon was formed, consisting of only five strains, all of which were known caries-inducing microorganisms. These strains were AHT, BHT, E49, FA-1 and OMZ61, to which the name *Strep. mutans* may be provisionally assigned as suggested by Carlsson⁵. Our results lead us to suppose that *Strep. mutans* is a clearly defined, distinct species and not synonymous with *Strep. bovis* or *Strep. sanguis* as de Stoppelaar⁹ and others have suggested.

The characteristics of *Strep. mutans* are now being examined in detail, and ultimately it may be possible to detect caries-inducing or cariogenic streptococci by simple means, with obvious consequences for caries prevention.

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Metabolism of Coumarin in Man

COUMARIN is a naturally occurring constituent of many plants, and has been used extensively as a flavouring material, although its use for this purpose is now banned in many countries. Its toxic effects on many species of animal have been reported¹⁻⁶ and its metabolism in rats and rabbits has been investigated⁷⁻⁹. No study has