

**Reclassification of ‘*Corynebacterium haemolyticum*’ (MacLean, Liebow & Rosenberg) in the Genus *Arcanobacterium* gen.nov. as *Arcanobacterium haemolyticum* nom.rev., comb.nov.**

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‘*Corynebacterium haemolyticum*’ (MacLean, Liebow & Rosenberg) differs to such an extent from the type species of *Corynebacterium*, *C. diphtheriae* (Lehmann & Neumann), that it should be removed from this genus. Chemical and numerical phenetic data indicate that ‘*C. haemolyticum*’ is a distinct taxon worthy of generic status. A new genus, *Arcanobacterium*, is described for the species *A. haemolyticum* (MacLean, Liebow & Rosenberg) nov.rev., comb.nov. The genus is tentatively placed within the ‘coryneform group of bacteria’. The type species of the genus is *Arcanobacterium haemolyticum* and the type strain is ATCC 9345.

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The generic assignment of bacteria named ‘*Corynebacterium haemolyticum*’ (MacLean *et al.*, 1946) has always been controversial (Cummins & Harris, 1956; Barksdale *et al.*, 1957; Barksdale, 1970; Harrington, 1966; Cummins *et al.*, 1974; Jones, 1975; Schofield & Schaal, 1981; Collins *et al.*, 1982). In the 8th edition of *Bergey’s Manual of Determinative Bacteriology*, the species is listed as an addendum to the genus *Corynebacterium* (Cummins *et al.*, 1974) and it does not appear in the *Approved Lists of Bacterial Names* (Skerman *et al.*, 1980).

‘*Corynebacterium haemolyticum*’ was originally isolated from infections amongst American soldiers, and assigned to the genus *Corynebacterium* by MacLean *et al.* (1946). However, the species bears little similarity to other corynebacteria and its placement in the genus *Corynebacterium* has been questioned by several workers (Barksdale *et al.*, 1957; Barksdale, 1970; Jones, 1975; Minnikin *et al.*, 1978; Schofield & Schaal, 1981; Collins *et al.*, 1982). Further, the relationship of ‘*C. haemolyticum*’ to the species *C. pyogenes* (Glage) remains unclear. On the basis of cell wall composition, Cummins & Harris (1956) suggested that both ‘*C. haemolyticum*’ and *C. pyogenes* were very closely related to the streptococci. This view was upheld by Barksdale *et al.* (1957) who suggested not only that ‘*C. haemolyticum*’ and *C. pyogenes* should be reclassified in the genus *Streptococcus*, but also that ‘*C. haemolyticum*’ was a mutant form of *C. pyogenes*.

Recent numerical phenetic (Schofield & Schaal, 1981; D. Jones & M. D. Collins, unpublished results) and chemical (Collins *et al.*, 1982) studies have shown that ‘*C. haemolyticum*’ and *C. pyogenes* are two distinct taxa. Collins & Jones (1982) have proposed that *C. pyogenes* should be reclassified in the genus *Actinomyces*, as *Actinomyces pyogenes*. The taxonomic position of ‘*C. haemolyticum*’ is, however, problematical (Collins *et al.*, 1982).

Fatty acid data (Collins *et al.*, 1982) do not support the view of Barksdale *et al.* (1957) that '*C. haemolyticum*' should be reclassified in the genus *Streptococcus*. '*Corynebacterium haemolyticum*' possesses major amounts of monounsaturated fatty acids of the oleic acid series (synthesized via an aerobic pathway) (Collins *et al.*, 1982). In contrast, members of the genera *Streptococcus* and *Lactobacillus* possess monounsaturated fatty acids of the *cis*-vaccenic acid series (synthesized via an anaerobic pathway) (see Kroppenstedt & Kutzner, 1978). The fatty acid data indicate that '*C. haemolyticum*' is more closely related to certain actinomycete and coryneform taxa (Collins *et al.*, 1982). However, on the basis of phenetic, peptidoglycan, fatty acid, menaquinone and DNA data (Schofield & Schaal, 1981; Collins *et al.*, 1982; M. D. Collins, D. Jones & R. M. Kroppenstedt, unpublished results), '*C. haemolyticum*' appears to be quite distinct from all other coryneform and actinomycete taxa examined to date. We therefore consider that bacteria presently designated '*C. haemolyticum*' should be reclassified in a new genus, for which we propose the name *Arcanobacterium*, composed of one species *Arcanobacterium haemolyticum* (MacLean, Liebow & Rosenberg) nom.rev., comb.nov.

*Description of Arcanobacterium gen.nov.*

Ar.can.o.bac.te'ri.um. L. adj. *arcanus* secretive; Gr. neut.dim.n. *bakterion* a small rod; M.L. neut.n. *Arcanobacterium* secretive bacterium.

The salient characters of this genus, based on the literature descriptions of '*Corynebacterium haemolyticum*' (MacLean *et al.*, 1946; Cummins & Harris, 1956; Barksdale *et al.*, 1957; Cummins *et al.*, 1974; Jones, 1975; Schofield & Schaal, 1981; Collins *et al.*, 1982) and our own observations, are as follows.

On blood agar plates slender, irregular, bacillary forms predominate during the first 18 h; many cells are arranged at an angle to give V-formations. As growth proceeds organisms become granular and segmented so that they resemble small and irregular cocci. Both rods and coccoid cells are Gram-positive, non-acid fast and non-motile; endospores are not formed.

Organisms are facultatively anaerobic. Growth is considerably enhanced in an atmosphere of CO<sub>2</sub>. Growth is sparse on ordinary media but enhanced by blood or serum. The optimum temperature for growth is 37 °C. Organisms will not withstand heating at 60 °C for 15 min. Acid is produced from glucose, dextrin, lactose and some other sugars (Schofield & Schaal, 1981).

The cell wall peptidoglycan is based on lysine. Mycolic acids are not present. The long-chain fatty acids are primarily straight-chain and monounsaturated (oleic acid series) acids. The major respiratory quinones are tetrahydrogenated menaquinones with nine isoprene units. The DNA base composition, as estimated from melting point ( $T_m$ ) determinations, is 50–52 mol % G + C. The type species is *Arcanobacterium haemolyticum*.

*Description of Arcanobacterium haemolyticum nom.rev., comb.nov.*

hae.mo.ly'ti.cum. Gr. n. *haema* blood; Gr. adj. *lyticus* dissolving; M.L. neut.adj. *haemolyticum* blood-dissolving, haemolytic.

This description is based on the studies of the type strain ATCC 9345 and strains NCTC 8452, 9697, 10513, 10514 and CCM 5947 (MacLean *et al.*, 1946; Cummins & Harris, 1956; Barksdale *et al.*, 1957; Cummins *et al.*, 1974; Jones, 1975; Schofield & Schaal, 1981; Collins *et al.*, 1982; and our own observations).

Surface colonies on blood agar are small (0.75 mm diam.) after 24 h, becoming large (1.5–2.5 mm diam.) on extended incubation. Colonies are circular discoid and slightly raised, and  $\beta$ -haemolytic. Growth is sparse on ordinary media but is enhanced by blood or serum. Slender, irregular rods predominate during the first 18 h on blood agar; many cells exhibit V-formations. Upon extended incubation, organisms become granular and segmented, and

resemble small irregular cocci. On Loeffler's medium they maintain the slender, irregular, bacillary form, but become pleomorphic at 48 h, with numerous club and comma forms.

*Arcanobacterium haemolyticum* is facultatively anaerobic. Its growth is considerably enhanced in an atmosphere of CO<sub>2</sub>. The optimum temperature for growth is 37 °C. The organism will not withstand heating at 60 °C for 15 min. Acid is produced from glucose, lactose and some other sugars. The catalase reaction is negative. Extracellular DNAase is produced. Gelatin, aesculin and casein are not hydrolysed. β-Galactosidase and N-acetyl-β-glucosaminidase are produced, but β-glucuronidase and α-fucosidase are not. The organism is resistant to oxytetracycline (30 µg per disc).

The cell wall peptidoglycan is based on lysine. The principal menaquinones are tetrahydrogenated menaquinones with nine isoprene units. The fatty acids are mainly straight-chain saturated and monounsaturated acids. The major fatty acids are hexadecanoic, octadecanoic and octadecenoic (C<sub>18:1</sub>, ω9) acids. The G + C content of the DNA, determined by estimation of the melting point, is 50–52 mol %<sup>1</sup>. The type strain is ATCC 9345.

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