

Isolation of *Neisseria lactamicus* from the nasopharynx

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SYNOPSIS During 1971 and 1972, 71 cultures of neisseriae that attacked lactose were received by this laboratory. All strains except one from an eye swab were from the nasopharynx of healthy subjects. Nineteen similar strains from the nasopharynx were isolated in this laboratory.

The characteristics of these strains were compared with those of *Neisseria meningitidis*, *Neisseria pharyngis*, *Neisseria catarrhalis*, and *Neisseria lactamicus*. The 90 strains under investigation closely resembled *Neisseria meningitidis* but could be differentiated by production of acid from lactose and beta-galactosidase activity and were therefore classified as *Neisseria lactamicus*.

Amongst cultures of presumptive meningococci sent to this laboratory in 1971 and 1972 for typing, we have identified 71 cultures of neisseriae that attacked lactose. Nineteen similar strains were isolated in this laboratory. All strains except one from an eye swab were isolated from the nasopharynx of healthy subjects during the course of meningococcal carrier surveys or as a follow up of contacts of cases of meningococcal infection. All nasopharyngeal strains were isolated initially on selective medium (Thayer and Martin, 1966). The characteristics of these 90 strains were compared with those of *N. meningitidis* and a number of commensal neisseriae.

Materials and Methods

Ninety strains of lactose-positive neisseriae detailed above, three strains each of *N. meningitidis* types A and B, one strain of *N. meningitidis* type C, three strains of *N. pharyngis*, and seven strains of *N. catarrhalis* were studied. All these strains had been isolated in this laboratory or received here during the past two years and preserved on Dorset egg slopes at 30°C.

In addition, a number of freeze-dried cultures were examined: three strains of *N. lactamicus* (NCTC 10616, 10617, and 10618), *N. catarrhalis* (NCTC 3622), *N. pharyngis* (NCTC 4590 and 4591), and type strains of *N. meningitidis* types A, B, and C (M 1027, M 993, and M 1628) originally obtained from Sara Branham.

A proportion of strains were tested for growth at 37 and 25°C. Two media were used: 10% horse

blood agar (Oxoid CM 271) and nutrient agar (Oxoid CM 3). Incubation was carried out both aerobically and in a candle jar for five days.

Acid production from four carbohydrates—glucose, maltose, sucrose, and lactose—was tested, using BBL cystine trypticase (no. 01-174) agar slopes, with 1% added carbohydrate. Slopes were incubated aerobically at 37°C for seven days before the final reading.

Beta-galactosidase activity was tested by the method of Lowe (1962) modified by substituting Difco Mueller-Hinton broth for peptone water. A heavy inoculum was used and results read after three and 24 hours' incubation at 37°C.

Serotyping was carried out by the agglutination method of Slaterus (1962) using antisera to *Neisseria meningitidis* types A, B, C, X, Y, and Z.

Results

All strains were oxidase-positive, Gram-negative diplococci. Colonies of the lactose-positive strains and those of the three NCTC strains of *N. lactamicus* were often indistinguishable from those of *N. meningitidis*. Strains of *N. catarrhalis* and *N. pharyngis* all grew well on nutrient agar at 25°C. Growth of the lactose-positive strains and *N. meningitidis* at this temperature was variable according to the strain tested; some lactose-positive strains and some strains of *N. meningitidis* produced growth when a heavy inoculum was used and incubation carried out in a candle jar.

Acid production from carbohydrates was as shown in the table.

Strain	No. Tested	Action on Carbohydrates ¹				
		Glucose	Maltose	Sucrose	Lactose	ONPG
<i>N. catarrhalis</i>	8	-	-	-	-	-
<i>N. pharyngis</i>	5	+	+	-	-	-
<i>N. meningitidis</i>	10	+	+	-	-	-
<i>N. lactamicus</i> (NCTC strains)	3	+	+	-	+	+
Lactose-positive strains	90	+	+	-	+	+

Table Distinguishing biochemical reactions of neisseria strains tested

¹ + = positive, acid produced, - = negative, no acid produced

The NCTC strains of *N. lactamicus* and the lactose-positive strains under investigation consistently fermented lactose, usually within 48 hours, and showed beta-galactosidase activity. No other strains tested fermented lactose or gave a positive ONPG.

All the lactose-positive strains and the three NCTC strains of *N. lactamicus* were untypable with antisera to *N. meningitidis* types A, B, C, X, Y and Z and more than 50% were autoagglutinable.

Discussion

Production of acid from lactose and beta-galactosidase activity are characteristic of *N. lactamicus* (Mitchell, Rhoden, and King, 1965; Hollis, Wiggins, and Weaver, 1969). The strains under investigation also showed these properties and were therefore classified as *N. lactamicus*. Morphologically and in its action on other carbohydrates, this organism may be indistinguishable from *N. meningitidis*. Weiss, Wilson, Schramek, and Hill (1971) found that the DNA of *N. lactamicus* showed partial reassociation with the DNA of *N. meningitidis* (75 to 56%) and more extensive but variable degrees of reassociation with other strains of *N. lactamicus* (98 to 72%). They suggest that *N. lactamicus* can be distinguished from *N. meningitidis* on the basis of DNA reassociation.

N. lactamicus is frequently isolated in carrier

studies. Jephcott and Morton (1972) have recently described the isolation of this organism from a cervical swab. As the clinical significance of *N. lactamicus* has not yet been established, it is important to distinguish it from *N. meningitidis*. Acid production from lactose and beta-galactosidase activity by the ONPG reaction are the most useful means of identifying *N. lactamicus* (Hollis *et al.*, 1969).

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