

Letters to the Editor

Sensitivity of 100 strains of group A β -haemolytic streptococcus

	Room air		5% CO ₂		Anaerobic	
	Choc	BA	Choc	BA	Choc	BA
No of strains	9	2	7	0	2	6
Sensitive	91	98	93	100	98	94
Resistant						

Choc = chocolate agar.

BA = blood agar.

Sensitive = zone of inhibition >14 mm diameter.

Resistant = zone of inhibition <14 mm diameter.

Optochin sensitivity of β -haemolytic streptococci group A

Optochin (ethyl hydrocuprein hydrochloride) sensitivity testing is routinely performed by most laboratories for the differentiation of *Streptococcus pneumoniae* from other streptococci.

In our laboratory all streptococci, seen by Gram staining of blood cultures, are tested for sensitivity to optochin. The optochin test results are read after 4–6 h incubation at 36°C, along with other antibiotic sensitivity tests.

This procedure was followed on a set of blood cultures from a patient who was subsequently shown to have a β -haemolytic streptococcus group A infection; however, there was a zone size >14 mm round the optochin disc on the original chocolate agar plate. On overnight testing β -haemolysis was apparent, but the isolate still gave a zone >14 mm on a fresh chocolate agar plate.

We have investigated the phenomenon of optochin sensitive β -haemolytic streptococci group A and our results are presented.

Material and methods

One hundred strains of group A β -haemolytic streptococcus from various clinical sites were used. The isolates were each streaked on to three sets of chocolate agar (Oxoid) and Columbia 5% horse blood agar (Oxoid) plates. Two or three colonies of each group A β -haemolytic streptococcus were streaked on to one quarter of the test medium and a 6 mm optochin disc (Mast) was placed on the innoculum. The plates were then incubated at 36°C for 18–24 hours under varying conditions: (a) one set in a normal aerobic incubator, (b) one set in a 5% CO₂ incubator, and (c) one set in an anaerobic incubator (Don Whitley Scientific).

The original optochin sensitive group A β -haemolytic streptococcus and a fresh laboratory isolate of *Str pneumoniae* as a control were also included.

After incubation zones of inhibition were measured. A zone size >14 mm was recorded as sensitive. All zone sizes <14 mm were discounted.¹

The table records the sensitivity of the 100 strains tested.

The zones of inhibition shown by group A β -haemolytic streptococci were comparable to that of the *Str pneumoniae* control. With the exception of blood agar in 5% CO₂, zones of inhibition were shown on both blood agar and chocolate agar under different atmospheric conditions.

Four strains were sensitive to optochin only on blood agar in anaerobic conditions. Another four strains showed sensitivity only on chocolate agar in aerobic conditions. Other strains were sensitive to optochin randomly on blood agar or chocolate agar or both in aerobic, CO₂, or anaerobic conditions.

There did not seem to be any correlation between optochin sensitivity and the type of medium used or the conditions of incubation.

None of the sensitive strains was bile soluble.

Discussion

Optochin sensitivity has been described for use in the identification of *Str pneumoniae* by several workers.^{2–4} Although reports of false sensitivity of the α -haemolytic streptococci do occur (<1%), there have been no such reports with β -haemolytic strains. The largest zone size suggested by Austrian⁴—that is, 14 mm—was used to define a sensitive strain.

We have found that the incidence of β -haemolytic strains sensitive to optochin is much higher than that reported for α -haemolytic streptococci.

The effect of a CO₂ environment in reducing the zone size of some strains is also mimicked by *Str pneumoniae*. Ragsdale and Stanford⁵ described how the presence of 5% CO₂ diminished the zone size of *Str pneumoniae* to optochin and suggested that strains that do not grow in atmospheric conditions should be tested by the bile solubility technique.

We offer no explanation of the variation from strain to strain within the group A

β -haemolytic streptococci and cannot suggest a mode of action for optochin. Kreger *et al*⁶ showed that electron dense aggregates appeared in the cytoplasm of treated cells, but this was found to be a non-specific effect.

Although we do not believe that there are many laboratories that would mistake a β -haemolytic streptococcus group A for a *Str pneumoniae* on an overnight culture, on rapid 4–6 h testing the β -haemolysis may not be evident and another character, such as microscopical appearance, must also be used before presumptive identifications are made.

GH BURGESS

J KHOR

Department of Microbiology,
St James Hospital,
London SW12

References

- 1 Lennette EH, Balows A, Hausler WJ, Truant JP, eds. *Manual of Clinical Microbiology*. 3rd ed. Washington DC: American Society for Microbiology, 1980.
- 2 Wilson GS, Miles AA. *Topley and Wilson's Principles of bacteriology and immunology*. 3rd ed. London: Edward Arnold, 1946.
- 3 Bowers EF, Jeffries LR. Optochin in the identification of *Streptococcus pneumoniae*. *J Clin Pathol* 1955;8:58–60.
- 4 Austrian R. *Diplococcus pneumoniae* (pneumococcus). In: Blair JE, Lennette EH, Truant JP, eds. *Manual of clinical microbiology*. Bethesda, Md: American Society for Microbiology, 1970.
- 5 Ragsdale AR, Sanford JP. Interfering effect of incubation in carbon dioxide on the identification of pneumococci by optochin discs. *Appl Microbiol* 1971;22:854–5.
- 6 Kreger AS, Swartzendruber DC, Olsen RH. Alteration in bacterial morphology by optochin and quinine hydrochlorides. *J Bacteriol* 1969;97:362–6.