

Streptococcus Selective Supplement (COBA)

PRODUCT INFORMATION

C039-100mg - Colistin Sulfate, Powder, 100mg

C039-1g - Colistin Sulfate, Powder, 1g

O002-5g - Oxolinic Acid, Powder, 5g

O002-25g - Oxolinic Acid, Powder, 25g

DESCRIPTION

Columbia Blood Agar Base with Streptococcus Selective Supplement (COBA) is a selective medium for the isolation of Streptococcus species.

BACKGROUND

Colistin is a polymyxin antibiotic produced by certain strains of *Bacillus polymyxa* var. *colistinus*. Colistin is a mixture of cyclic polypeptides colistin A and B. Colistin is effective against most Gram-negative bacilli and is used as a polypeptide antibiotic.

Oxolinic acid is a quinolone antibiotic. The antibiotic works by inhibiting the enzyme DNA gyrase. It also acts as a dopamine reuptake inhibitor and has stimulant effects in mice.

Mechanism of action

APPLICATION IN COLUMBIA BLOOD AGAR BASE

Streptococcus Selective Supplement is based on the formulation of Petts (COBA Medium) and is recommended for the selective isolation of *streptococci* of medical and veterinary importance.

COBA Medium possesses advantages over other media described for selective isolation of *streptococci*.

Agents previously recommended for inhibition of Gram-positive organisms can be shown to have severe effects on streptococci even at subminimal inhibitory concentrations. The antibiotics gentamicin, amikacin, fucidic acid, neomycin and cotrimoxazole have all been shown to have adverse effects as have the long established inhibitors crystal violet and sodium azide.

Both colistin and oxolinic acid are thermostable and can, if necessary, be stored without refrigeration.

Streptococci are commonly isolated from the upper respiratory tract. They are also often isolated from burns and other sites where frequently there is an abundance of competing organisms. In order to isolate *streptococci*, especially when present in small numbers, it is necessary to inhibit the competing flora without any adverse effect by the selective agents upon the *Streptococcus* species. The selective agents colistin sulphate (10 mg/ml) and oxolinic acid (5 mg/ml) have been found to have no inhibitory effect on Streptococcus species although amongst Group D organisms *Enterococcus faecalis* colonies are somewhat smaller. The combination of these two selective agents results in total inhibition of Gram-negative organisms and almost all non-streptococcal Gram-positive organisms. A very few *staphylococci* and coryneform organisms may grow with reduced colony size. The haemolytic reactions on media containing blood are clearly defined, and the colonial size and growth recovery of streptococcal groups A, B, C, D and G and *S. pneumoniae* are comparable to that on a nonselective medium. The selective agents can also be used with Islam's Medium for the isolation of Group B streptococci without loss of pigmentation occurring.

Content concentrations

Typical Formula*	mg/litre
Columbia Blood Agar Base	
Special peptone	23
Starch	1
Sodium chloride	5
Agar	10
Final pH 7.3 ± 0.2 @ 25°C	
Streptococcus Selective Supplement (COBA)	
Colistin sulphate	10
Oxolinic acid	5
* Adjusted as required to meet performance standards	

Table 1 - Typical Formula for Columbia Blood Agar Base and Streptococcus Selective Supplement (COBA)

METHOD

Preparation

Add appreciate amount of Columbia Blood Agar Base to distilled water. Boil to dissolve and sterilise by

autoclaving at 121°C for 15 minutes. Aseptically add the supplement contents to sterile Columbia Blood Agar Base containing 5% Defibrinated Horse Blood cooled to approximately 50°C. Mix gently and pour into sterile Petri dishes.

Protocol

1. Prepare the medium from Columbia Blood Agar Base, Streptococcus Selective Supplement and Defibrinated Horse Blood, according to the preparation.
2. Inoculate the plates in the normal way and incubate at 35°C overnight in an atmosphere enriched with 5% carbon dioxide or anaerobically.*
3. Confirm that the colonies are *streptococci* by microscopy, biochemical or serological tests.

* Improved haemolytic reactions are achieved by anaerobic incubation. Gram-positive anaerobic cocci (*Peptostreptococcus* and *Peptococcus* species) would be selectively isolated under these conditions.

Quality control

Positive control:

Streptococcus pyogenes ATCC® 19615: Good growth; gas production.

Negative control:

Staphylococcus aureus ATCC® 25923: Inhibited

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