



Isolation And Identification of Staphylococcus Species

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Introduction

The most typical food-borne infection is caused by a staphylococcus infection. More than 50% of healthy people have these organisms on their skin, in their hair, and in their throats. Any food that must be handled while being prepared is susceptible to contamination from this bacterium. Staphylococci are Gram-positive, aerobic, spherical organisms that are typically organized in clusters that resemble a bunch of grapes. They may ferment a variety of carbohydrates despite not being motile and spore-forming. *Staphylococcus aureus* is the most hazardous bacteria in terms of public health, producing enterotoxins A, B, C, D, E, and F, toxin associated with toxic shock syndrome, epidermolytic toxins A and B, haemolytic toxins alpha, beta, gamma and delta and leucocidin. They are spread through touch, inhalation and eating tainted food. They can result in lesions that suppurate, such as impetigo, boils, pneumonia and abscesses. The presence of oxygen is crucial for growth.

Materials collected for isolation

- Suspected food samples
- Suspected nasal swabs
- Swab from suspected lesion, vomitus etc

Isolation

S.aureus may be isolated from food by plating where number of bacteria is high. Enrichment is done for studies

Enrichment

Alternate trypticose sodium broth with 10% NaCl and 1% sodium pyruvate

Selective plating

Glycerine, potassium tellurite and pyruvate agar are the inhibitors of Baird Parker agar, sometimes known as egg yolk glycerine tellurite pyruvate agar. Because of its great selectivity and capacity to restore stressed cells, Baird Parker agar is the preferred medium. *S. aureus* colonies are 1 to 1.5 mm in diameter, Jet black colonies at centre with narrow white zone around in BP agar. Another media used is mannitol salt agar that gives tiny yellow colonies.

Procedure

Food samples should be inoculated in enrichment broth and incubated for 24 hours at 37°C. Incubate a loop of culture from enrichment broth over BP agar for 24 hours at 37°C (48 hours are required for the establishment of an opaque zone). Pick up the suspension culture and place it in tubes with brain heart infusion broth as an agar slant for 18 to 24 hours at 37°C for 24 hours. Run biochemical testing to provide additional confirmation.

Biochemical tests for *Staphylococcus aureus*:

Gram's staining:	Positive
Catalase:	Positive
Oxidase:	Negative
Indole ring test:	Negative
Methyl red test:	Positive
VP test:	Positive
Coagulase test:	Positive
Urease test:	Positive

Tube test and slide test

Tube test for free coagulase and slide test for bound coagulase

Procedure

Take 0.2 ml of rabbit plasma and 0.8 ml of broth, incubate for 1 hour at 37°C and check for clotting. If there is no clotting, check again in three hours; if the result is negative, store for the night and test.

There are two distinct types of coagulation.

1. Free coagulation: convert fibrinogen to fibrin by triggering a plasma-based coagulase responding factor. Test for free coagulation using a clotting tube
2. Bound or clumpy factor: convert fibrinogen to fibrin without the need for a plasma-based coagulase responding factor.

References:

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