CHARACTERISTICS OF CULTIVATION ON AGAR – BLOOD AND ON AGAR HYPERCHLORINATED – MANNITOL – RED PHENOL OF STAPHYLOCOCULUI AUREUS

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REVIEW, RESEARCH ARTICLE

Abstract
In the practice of laboratory the culture media are used for the isolation from natural media of different microorganisms to obtain pure cultures, for their cultivation in the purpose of obtaining biomass or for the maintenance of the selected pure culture. These media include beside the nutritive substances also substances with inhibiting effect on the accompanying microorganisms met in the microbiota of which is made the isolation of the culture that we want to select. A selected medium used to determine the coliform bacteria is the broth – gall – lactose – brilliant greed in which the biliary salts inhibits other bacteria, while the coliforms are adapted. In conditions of laboratory are used liquid media, especially for the cultivation of microorganisms facultative anaerobe, for the study of fermentative processes, solid media – bread, sliced potatoes and frequently solidified media obtained by adding in liquid media some agents of solidification

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INTRODUCTION

The bacteria from the Staphylococcus type are pathogen agents of the man and other mammals. Traditionally they were divided in two groups based on their capacity to coagulate the blood plasma. The coagulase-positive staphylococci are considered the most pathogen S aureus species. Coagulase-negative staphylococci are known now as including over 30 of other species. SNC are common commensals of the skin although some species can cause infections. Now it is obvious that the dividing of the staphylococci in positive and negative coagulase is artificial and really misleading in some cases. The coagulase is a marker for S aureus, but there are is no direct evidence that it is a factor of virulence. Also some natural isolates of S aureus are defects in coagulase. Beside all these, the term is still used on a large scale among the clinical microbiologists. S. aureus expresses a variety of extracellular proteins and polysaccharides, from which some are correlated with virulence. The virulence results from the combined effect of many factors expressed during the infection. The antibodies will neutralize the toxins and the staphylococci toxins and enzymes but the vaccines are not available. Both the treatment with antibiotics and the surgical drainage are often necessary to heal the abscesses, large furuncles and the infections of the wounds. The staphylococci are frequent causes of the infections associated with the existent medical devices. They are difficult to be treated only with antibiotics and often needs the elimination of the device. Some strains that infect the patients admitted are resistant to the majority of the antibiotics used to treat infections, vancomycin being the only drug remaining for which it was not developed resistance yet. The test of catalase is important in distinguishing the streptococci staphylococci (catalase-negative) that are positive catalase. The test is made by flooding an inclined agar culture or broth with a few drops of hydrogen peroxide 3%. The test shouldn't be made on agar with blood because the blood in itself will produce bubbles.

MATERIAL AND METHODS

For the performing of the study we used also the archive, registered in the specific program of the computer from the laboratory of

S.C. Diaser, Oradea, in the computerized data base of the unit, respectively.

Necessary materials for the performing of the examination:

■ A recipient of collection (collection recipient with collecting spoon) with transport medium

■ Wooden spatula

■ Latex gloves

For the collection of fecal matter it has to be collected a sample of fecal matter of 5-10g introduced in the collection recipient of fecal matter with transport medium. If the stool is liquid, it will be collected 5 ml. It is recommended to be chosen a liquid, mucous and bloody portion, if there is one. Don't collect quantities larger than 10g because it will reduce the chances of isolating the pathogen bacteria.

RESULTS AND DISCUSSIONS

In regard to the characteristics of cultivation, staphylococci were developed easily, in a time of 18-24 hours, on simple nutritive media, in aerobiosis. Vitamin B1 and nicotinic acid are indispensable factors for growth.

But, *Staphylococcus aureus* has grown in a longer time, in approximately 24-36 hours, in aerobiosis and on the same nutritive media.

Some versions, rare, of *Staphylococcus aureus* were developed only in the presence of CO2 or of other metabolites.

The forms with deficiency of the cellular wall need hypertonic medium in order to survive and multiply.

The optimum temperature of growth of staphylococci is of 37°C, and the optimum ph is of 7,5, but the great variations are tolerated.

After 24 h of incubation of solid media there were colonies of type S developed, creamy, round, with diameter between 2-3 mm, perfect margins, smooth surface, bulged and shiny. In regard to the liquid media they are homogenous cloudy, with granular deposit, in the inferior part of the culture tube.

The strains of staphylococci with deficiency of the cellular wall come from samples that include antimicrobial substance or substances submitted to some thermal treatments, originated on solid media some colonies of G type, shiny, small or G-T, rugose, small, with irregular margins, granulated, without hemolysis. These types of microorganisms don't shake up the liquid medium in which they are seeded and they appear with granular deposit in the inferior part of the culture tube.

Rarely did we meet from sampled cultures, colonies of M type, mucous, characteristic to the encapsulated strains.

On agar culture medium with 5-8% of ram defibrinated blood, it produces a circular area of hemolysis around the colony.

Hemolysis α or *Staphylococcus aureus* determines the total hemolysis with complete clearing of the β hemolysis medium of animals, met occasionally also in human strains, determines hemolysis of "warm-cold" type, areas of incomplete hemolysis at 37°C, which after the exposure for a few hours at 4°C, becomes complete, similar to the one produced by α hemolysis.

Tab.1.

Characteristics of cultivation on agar – blood and on agar hyperchlorinated – mannitol – red phenol

Species	Aspect of colonies on agar - blood	Diameter of colony	Pigment	Aspect of colonies on hyperchlorinated solid medium
S. aureus	Smooth, creamy, transparent, mucous	5 – 10 mm	Golden, citrine or without pigment	Yellow colonies, mannitol positive, turns to yellow on Chapman medium
S. epiderminis	Mat	2,5 – 6 mm	Without pigment, rarely grey yellow	Colonies not pigmented, the medium remains pink

The presence of the carotenoid pigment, orange or citrine yellow, is a characteristic relatively constant of the strains of *Staphylococcus aureus*, but also of some coagulase-negative staphylococci, as *S. saprophyticus, S. hemolyticus, S. hominis.* Not all the strains of S. aureus produce hemolysis or pigment.

CONCLUSIONS

1. It can be hard to differentiate them from micrococci, streptococci although usually streptococci appear more frequently as chains of diplococci.
2. For the bacteriology of the food it is indicated the utilization of hyperchlorinated media as enriching medium.
3. In regard to the characteristics of cultivation, staphylococci were easily developed in 18-24 hours, on simple nutritive media, in aerobiosis.
4. *Staphylococcus aureus*, has grown in a longer period of time, in approximately 24-36 hours, in aerobiosis and on the same nutritive media.

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