**FORM 2**

**THE PATENT ACT 1970**

(39 OF 1970)

AND

The patent rules, 2003

**COMPLETE SPECIFICATION**

(See section 10: rule 13)

**TITLE OF INVENTION**

**Evaluation of a new prepared medium for identification of *S. aureus***

**APPLICANT (S)**

| Name | Nationality | Address |
|---|---|---|
| Reyam Abdul Khuder Mohammed | Iraq | Assistant Lecturer, Al-Mustaqbal University College, Hilla, Iraq |
| Noor Flayyih Hasan | Iraq | Al zahraa university for women, iraq. karbala |
| Hanan Selman Hessan | Iraq | College of Dentistry, University of Babylon, Babylon province, Iraq |
| Mustafa Salah Hasan | Iraq | College of Vet. Med., Dep. Of Internal and Preventive Medicine, University of Fallujah, Anbar, Iraq |
| LubnaDhari Mohammed | Iraq | Gilgamesh Ahliya University, Iraq |
| Duaa Mohammed Abdulsattar | Iraq | AI_Iraqia University, College of medicine, Iraq |

**PREAMBLE TO THE DESCRIPTION**

**COMPLETE**

Following specification particularly describes the invention and the manner in which it is to be performed.

## DESCRIPTION

## TECHNICAL FIELD OF INVENTION

The present invention in general relates to evaluation of a new prepared medium for identification of S. aureus.

## BACKGROUND OF THE INVENTION

Alexander Ogston's in the 1880swas firstly described *Staphylococcus aureus*. He found a grapelike cluster of pus grapes from postoperative wounds and abscesses(Ogston, 1881). Rosenbach prospered in isolating and developing the bacteria on a solid medium in 1884. The characteristic yellow pigmentation in their colonies gave him the name *Staphylococcus aureus* (Liu et al., 2005). Colonies' yellow pigmentation results from the development of staphyloxanthin carotenoids (Marshall & Wilmoth, 1981).

*Staphylococcus aureus* belongs to human's natural microbial flora. In the body, it can develop as a harmless microbe, or cause infections it can live in different ecological niches (Tong et al., 2015). The anterior nares of healthy adults also have the bacterium (van Belkum et al., 2009). Moreover, *S. aureus* in animals like dogs, cats, and pigs, etc, was also indicated. (Morgan, 2008).

The most public pathogen for humans in the Staphylococcus genus is *S. aureus* and is the causative agent for numerous illnesses.(Lowy, 1998). *S. aureus* caused infections may be divided into (1) local infections linked to skin & soft tissues (2) systematic infections such as sepsis, bacteremia, pneumonia, etc. (Liu, 2009; Kurlenda&Grinholc, 2012).

*S. aureus* diagnosis is done by the culture of wound swabbing in non-specific mediums and the approval of suspicious colonies by biochemical and/or serological tests is normally performed. The most frequently included colonies of staphylococci

for the identification of coagulase, Protein A, and/or clear capsular antigens with sensitized Latex particles (Personne et al., 1997; van Griethuysen et al., 2001).

Colonies of *S. aureus* might be uncharacteristic and hard to distinguish from CNS (Baumert et al., 2002); hence, a great number of agglutination trials might be essential to rule out the occurrence of *S. aureus*. A variety of cultural media has been created to enhance the detection of *S. aureus*, including MSA and Baird-Parker medium (Baird & Lee, 1995; Davies et al., 2000). The use of CHROMagar Staph was a more recent technique that uses chromogeneous enzyme substrates to enable the detection of S. aureus in a selective agar medium (Gaillot et al., 2000), this medium and tests are expensive, for this reason, this study was conducted to evaluate the efficacy of new medium prepared by using abundant materials found in the laboratory to identify *S. aureus*.

**OBJECTIVE OF THE INVENTION**

An objective of the present invention is to attempt to overcome the problems of the prior art and provide a evaluation of a new prepared medium for identification of S. aureus.

**DETAILED DESCRIPTION OF THE INVENTION:**

**Materials and Methods:**

**Isolates:**

Three isolates of *S. aureus* and 3 isolates of coagulase-negative staphylococci (CNS) were used in this study.

**Media preparation:**

the prepared medium contains:

For 1 L :

Agar-Agar 15 gm

Peptone water 15 gm

Bromocresol purple 2% take 1ml

**121c 15 bar autoclave**

Maltose 10 gm (1%) in water bath 90 ċ for 5 min.

**Culturing:**

All bacteria were cultured on the prepared medium by streaking method, then incubated for 24hrs at 37 Ċ (Markey *et al*., 2013).

**Results and discussion:**

The current results showed yellow colonies of *S. aureus* with a zone of inhibition (Figure 1) and white colonies of CNS without inhibition zone (Figure 2).


**Figure 1. *S. aureus* on prepared medium**

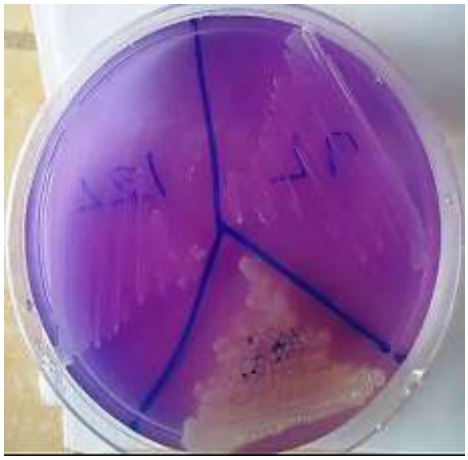

**Figure 2. CNS on prepared medium**

Adegoke finds all the isolates of glucose fermented for 24 hours, 98,8 and 98,7% fermented maltose and sucrose, respectively, in the form of acid processing (Adegoke and Ojo, 1982). All *S. aureus* isolates in an Ajuwape's study were fermented mannitol, glucose & sucrose, also 98.1%, and 89.1% of them fermented trehalose and maltose (Ajuwape and Aregbesola, 2001). The results of these 2 studies are in agreement with our results.

Maddux &Koehne (1982) found that all isolates of *S. aureus* were positive for maltose fermentation on purple agar medium given yellow color. Also, Reddy et al. (2016) tested S. aureus isolated from clinical cases of dogs on purple agar medium supplemented by maltose 1%.

Shrivastava et al. (2018) used a purple agar medium for the rapid identification of S. aureus from milk samples.

Tessema and Tsegaye (2017) reported that *Staphylococcus aureus* digest maltose easily, and the acid metabolic products transform the medium and colonies into yellow by the pH indicator (bromocresol purples).

This was based on *S. aureus* identity maltose and acid metabolic products are easily fermented and change the medium and the colonies to yellow by pH indicator (bromocresol purple), while *S. hyicus* did not ferment maltose but attacked the medium for peptone that created an alkaline response around colonies (a deeper violet) (Markey et al., 2013).

In conclusion, this media can be used for the rapid identification of S. aureus and differentiation from another type of Staphylococcus Spp.

**I/We claim:**

1. A evaluation of a new prepared medium for identification of S. aureus.

5

**Abstract**

*Staphylococcus aureus* is a significant pathogen which is life-threatening in human and animals. This study was conducted to assess the newly prepared medium in identifying *S. aureus*. Three isolates of *S. aureus* and 3 isolates of coagulase-negative staphylococci (CNS) were used in this study. The media contains agar, maltose, peptone water, and bromocresol purple as a pH indicator. The results showed yellow colonies of *S. aureus* with a zone of inhibition and white to purple colonies of CNS without inhibition zone. In conclusion, this medium contains affordable materials and can be used for the rapid identification of *S. aureus*.