

# *Staphylococcus aureus* Can Produce Catalase Enzyme When Adding to Human WBCs as a Source of H<sub>2</sub>O<sub>2</sub> Productions in Human Plasma or Serum in the Laboratory

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Received 31 October 2014; revised 28 November 2014; accepted 17 December 2014

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## Abstract

**Background:** *Staphylococcus aureus* is one of the most virulent gram positive bacteria. It produces a lot of toxins and enzymes, most of which are virulent factors. Among the enzyme that produces is the catalase which is very useful in differentiating *staphylococci* from *streptococci* [1]. Catalase is nearly ubiquitous among some of organisms that can grow in the presence of oxygen (air). It promotes the conversion of hydrogen peroxide, a powerful and potentially harmful oxidizing agent, to water and molecular oxygen; so the major function of catalase within cells is to prevent the accumulation of toxic levels of hydrogen peroxide formed as a by-product of metabolic processes—primarily that of the electron transport pathway. **Objectives:** The main aim of this study is to prove that human WBCs can produce H<sub>2</sub>O<sub>2</sub>. This H<sub>2</sub>O<sub>2</sub> when reacting with catalase producing *S. aureus* can easily be degraded to H<sub>2</sub>O + O<sub>2</sub>. **Methodology:** In this study a total of 40 subjects were included. Aliquots of 2.5 ml of venous blood were collected by venous puncture after disinfecting the site of collection with 70% alcohol and the collected blood was drawn into EDTA containers (20 subject) and anticoagulant free containers (other 20 subject), centrifugation for 5 minute at 1500 RPM. The separated sera and plasma were converted to new sterile eppendorf tubes and freezing until used (we leaved the eppendorf tubes that contained sera and plasma at room temperature before using it for DE freezing). Standard catalase producing *S. aureus* were used by taking 1 colony from Macconkey media by using applicator wooden stick, and inserted in eppendorf tube, then air bubbles would appear to indicate occurrence of the reactions. **Results:** According to this study, it was proved that WBCs in human plasma or serum can produce H<sub>2</sub>O<sub>2</sub>; this H<sub>2</sub>O<sub>2</sub> was reacted with catalase enzyme produce from colony of *S. aureus* to produce air bubbles and water. There were no differences between using H<sub>2</sub>O<sub>2</sub> or human plasma/serum that contains WBCs to detect and identify *S. aureus* by both techniques. **Conclusion:** Based on the results of this study, we can use WBCs that are found in human plasma or serum to identify catalase producing *S. aureus*.

**How to cite this paper:** Mustafa, H.S.I. (2014) *Staphylococcus aureus* Can Produce Catalase Enzyme When Adding to Human WBCs as a Source of H<sub>2</sub>O<sub>2</sub> Productions in Human Plasma or Serum in the Laboratory. *Open Journal of Medical Microbiology*, 4, 249-251. <http://dx.doi.org/10.4236/ojmm.2014.44028>

## Keywords

WBCs, Catalase Enzyme, *S. aureu*

## 1. Introduction

Catalase is an enzyme present in the cells of plants, animals and aerobic (oxygen requiring) bacteria (Orent, 2006). In order to survive, *S. aureus* has many defense mechanisms such as catalase enzyme which facilitates cellular detoxification; it neutralizes the bactericidal effects of  $H_2O_2$  [2] [3].

In 1893, a publication by Gottstein brought attention to bacterial catalase, making it one of the first bacterial enzymes to be described. After 30 years, McLeod and Gordon developed and published what was thought to be the first bacterial classification scheme based on catalase production and reactions, and so on; catalase becomes the most useful technique in medical microbiology to identify and differentiate between *Staphylococcal* species especially *S. aureus* (catalase positive) and *Streptococcal* species (catalase negative) [3].

## 2. Material & Method

This is a prospective experimental study which was conducted in The Asbab Hospital, from 1/5/2014-31/5/2014. Aliquots of 2.5 ml of venous blood were collected by venous puncture after disinfectant the site of collection with 70% alcohol and then the collected blood was drawn into EDTA containers (20 subject) and anticoagulant free containers (Other 20 subject), centrifugation for 5 minute at 1500 RPM, then the separated sera and plasma were converted to new sterile eppendrof tubes and freezing until used (before used we will leaves the eppendrof tubes at room temperature for DE freezing).

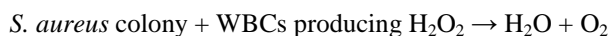
Standard catalase producing *S. aureus* were used by taking 1 colony from Mac-conkey media by using applicator wooden stick, and inserted in eppendrof tube, then after that waited for the appearance of air bubbles to indicate occurrence of the reactions.

The principle of catalase enzyme as following:



## 3. Results

*S. aureus* can produce catalase enzyme to react with  $H_2O_2$  from WBCs in human plasma or serum Principle:



There were no differences between using  $H_2O_2$  or human plasma/serum that contains WBCs for detections and identifications of *S. aureus* by both techniques, as in image below:



The image appears air bubbles, that produced when *S. aureus* producing catalase enzyme react with WBCs producing  $H_2O_2$  to indicate occurrence of the reaction.

## 4. Discussion

According to this study of using human plasma or sera that contains WBCs to detect presence of catalase enzyme by *S. aureus*, it gives good results that consist with each founding in microbiology books for isolation and identification of microorganism even in case of weak reaction [1] [4].

Furthermore if buffy coat just used the reaction cannot occur or weak after coagulation of plasma first.

## 5. Conclusion

Based on the results of this study, we can use human plasma or serum that contains WBCs to identify catalase producing *S. aureus*, and so on to differentiate between *Staphylococcus* and *Streptococcus*.

## 6. Recommendations

I would like to recommended using of human plasma not only for coagulation test but also for catalase test, we also needed a further studies on it. My concern now why we cannot invent something that can measured  $H_2O_2$  especially in case of inflammation due to rise on it, in case of inflammation, to become as indicator for inflammation, beside this we can also extract the hem ring of catalase enzyme to be used as iron supplement, we can benefit from natural reaction if it is endothermic or exothermic in industry filed (heating and cooling process), we can found some way to mimic this reaction by using continuous culture media to produce water and to collect  $O_2$  for  $O_2$  cylinder to be used in ventilation, We can benefit from the belching characteristic of catalase and hydrogen peroxide mixture in industry and others different things even in detection for elements.

## Acknowledgements

I would like to acknowledge the inspirational instruction and guidance of Dr. Shamsoun Khamis Kafi, also I would like to thank my father for always encourage and supporting me, thanks to the family of the College of Medical Laboratory Science, The National Ribat University and the patients without whom this research would have never seen the light.

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