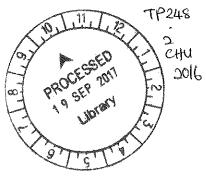
ISOLATION OF ANTIBIOTIC RESISTANT BACTERIA FROM HEALTHY STUDENTS AND STAFF OF INTI INTERNATIONAL UNIVERSITY

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DISSERTATION SUBMITTED IN PARTIAL FULFILLMENT OF THE REQUIREMENTS FOR THE DEGREE OF BACHELOR OF BIOTECHNOLOGY (HONOURS)



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ABSTRACT

Disease related to antibiotic resistance in Malaysia is at a critical level because of the frequent misuse of antibiotics and this caused the emergence of antibiotic-resistant bacteria. It is important to isolate the antibiotic resistant bacteria and create more awareness towards antibiotic resistant bacteria among public. Methicillin resistant Staphylococcus aureus (MRSA) is one of the more serious antibiotic-resistant bacteria. Thus, the objectives of this project were to isolate antibiotic resistant bacteria from the nasal cavity and axilla of healthy individuals. Samples collected were cultured on nutrient agar and the single colonies were sub-cultured to obtain pure cultures. As all isolates were Gram positive, they were grown on Mannitol Salt Agar (MSA) and subjected to catalase test. Antibiotic susceptibility testing was conducted to determine the susceptibility of bacteria towards antibiotics by using the disc diffusion method. Bacteria that were Gram-positive, cocci with cluster-like cell arrangement, was catalase positive, and resistant to oxacillin, were further subjected to inoculation on Brilliance MRSA 2 agar. This is because these bacteria indicate the likely presence of MRSA. In this study, 24 isolates were MRSA while 12 isolates are likely to be MRSE. However, the strains of the MRSA and MRSE could not be determined conclusively. Further tests such as DNA sequencing are required to confirm the identity of bacteria. This finding is important so that the staff and students of INTI International University can be alerted to prevent any possible outbreak of MRSA or MRSE-related diseases.

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LIST OF ABBREVIATIONS

A Axilla

AIDS Acquired Immune Deficiency Syndrome

BSAC British Society for Antimicrobial Chemotherapy

CA-MRSA Community-Associated Methicillin-resistant S. aureus

CFU/mL Colony forming units/ milliliter

DA Clindamycin

DNA Deoxyribonucleic acid

H₂O₂ Hydrogen peroxide

HA-MRSA Healthcare-Acquired Methicillin-resistant S. aureus

KZ Cephazolin

L Liter

M May

mL Millilitre

mm Millimetre

MRSA Methicillin-resistant Staphylococcus aureus

MRSE Methicillin-resistant Staphylococcus epidermidis

MSA Mannitol Salt Agar

N Nasal cavity

OX Oxacillin

P. acnes Propionibacterium acnes

rpm Revolutions per minute

S. aureus Staphylococcus aureus

S. epidermidis Staphylococcus epidermidis

URTI Upper respiratory tract infections

VRSA Vancomycin-resistant Staphylococcus aureus

 $^{\circ}\mathrm{C}$ Degree celcius $^{\mu}\mathrm{L}$ Microlitre

CHAPTER 1

INTRODUCTION

Antibiotic-resistant bacteria means that the bacteria has acquired resistance mechanisms to overcome the effects of antibiotics. This can happen through mutation or obtaining resistance traits from other bacteria. Overuse or misuse of antibiotics can increase the acquisition antibiotic-resistant traits by these bacteria (World Health Organization, 2015) which can then spread worldwide due to poor hygiene practice and improper food preparation (Better Health Channel, 2015).

The emergence of antibiotic-resistant bacteria is a major problem worldwide resulting in limited choices of antibiotics that can be used to treat infectious diseases (Centers for Disease Control and Prevention, 2015). Methicillin-resisitant *Staphylococcus aureus* (MRSA) is one of the more serious types of antibiotic-resistant bacteria. This bacterium causes reddish-rash, boils, and cellulites (Better Health Channel, 2015). Methicillin-resisitant *Staphylococcus aureus* is resistant to methicillin and beta-lactam antibiotics. This makes it hard to treat MRSA infections. People with weak immune system easily acquire MRSA infections (Centers for Disease Control and Prevention, 2015).

In this study, antibiotic-resistant bacteria were isolated from the nasal cavity and axilla of healthy individuals. Studies show that antibiotic-resistant bacteria such as MRSA can be isolated from healthy individuals. Knowing this is important as healthy individuals can be the carriers of antibiotic-resistant bacteria as well. Some of the research on the isolation of antibiotic-resistant bacteria from the nasal cavity of healthy individuals has been done in United States (Rath, Christmas, Picardo, & Westbay, 2015) and Nigeria (Ugwu, Anie, Ibezim, & Esimone, 2016). However, there are very few studies on the isolation of antibiotic-resistant bacteria from axilla of healthy individuals. Therefore, the objective of this study was to isolate antibiotic-resistant bacteria from the nasal cavity and axilla of healthy individuals in INTI International University.

CHAPTER 2

LITERATURE REVIEW

2.1 ANTIBIOTIC-RESISTANT BACTERIA

Antibiotic-resistant bacteria are bacteria that have the ability to resist the effects of antibiotics. Hence, antibiotics cannot be used to kill them. Antibiotic-resistant bacteria will develop and proliferate when there is misuse or repeated use of antibiotics (Centers for Disease Control and Prevention, 2015). Bacteria that are resistant to many antibiotics are called as multi-drug resistant bacteria. As a result, stronger antibiotics have to be prescribed to overcome the infection and kill the antibiotic-resistant bacteria.

Antibiotic resistance should be prevented because this is a very serious health problem worldwide (Better Health Channel, 2015). If bacteria are able to resist all antibiotics, then, it will be a problem for clinicians to treat the patients effectively as this can cause severe effects or even death in patients (Centers for Disease Control and Prevention, 2015). Furthermore, these bacteria can cause serious problems in public health as they are resistant to readily available antibiotics that are currently prescribed (Better Health Channel, 2015). This means research on new antibiotics is needed in order to eliminate these antibiotic-resistant bacteria.

Usually, bacteria develop resistance to antibiotics by mutation or acquiring resistant traits from other bacteria (Alliance for the Prudent Use of Antibiotics, 2014). Genetic mutations enable bacteria to produce enzymes that can inactivate antibiotics. For example, certain antibiotic-resistant bacteria are able to produce beta-lactamase to inactivate beta-lactam antibiotics which include monobactams and penicillins. The decrease in permeability, efflux pumping mechanisms, inactivation of drugs and, alteration in drugs' structure are four other mechanisms which can cause antibiotic resistance (Sun, Deng & Yan, 2014). Efflux pumping mechanism is critical for the survival and proliferation of multi-drug resistant bacteria. These bacteria can eliminate the effects of antibiotics by pumping the antibiotics out of the cell, thereby

rendering them ineffective against these pathogens (Alliance for the Prudent Use of Antibiotics, 2014).

However, some bacteria can transfer genes encoding resistance to other bacteria causing more bacteria to acquire resistance to multiple antibiotics. Resistant traits can be passed from viruses to bacteria as well. The resistant traits are packed in virus and can be injected when bacteria have been attacked (Alliance for the Prudent Use of Antibiotics, 2014). For example, the resistance traits of bacteria are packed in the head portion of virus, the virus can then attack the new bacteria and inject the resistance traits to them (Alliance for the Prudent Use of Antibiotics, 2014).

In order to avoid misusing antibiotics, we should know that antibiotics can only be used to treat bacterial infections and not viral infections. Examples of viral infections include flu, sore throats, and ear infections (Centers for Disease Control and Prevention, 2015). Doctors should prescribe antibiotics that are beneficial to the patients and encourage them to finish the prescribed medication within the given period (Better Health Channel, 2015).

Furthermore, antibiotic resistance cases in Malaysia are at a critical level because of the widespread and misuse of antibiotics. This has resulted in the risk of spreading of bacteria and difficulty in the treatment of these infections. Colistin, which is known as a last-resort drug to fight against serious diseases, was shown to be ineffective to bacterial samples in Malaysia (Malay Mail Online, 2015). The rapid emergence of antibiotic-resistant bacteria caused the challenges to the healthcare department. This problem was solved by broad spectrum antibiotics previously but not effective nowadays (Arumugam, 2016).

Some of the more prevalent bacteria that are multi-drug resistant include MRSA, VRSA, and MRSE (Better Health Channel, 2015).