CHAPTER I: INTRODUCTION

1.1 Background

Foul body odor is a health problem encountered by many people around the world. This problem may affect the inconvenience of the subject and lead to personal judgment from society. This unpleasant body odor may arise due to complex biological interaction between genetics, hormones, and skin microbiome together with physiological conditions. The body odor itself is created by the interaction of sweat which is produced by the apocrine gland with odor-causing bacteria living on the skin. Initially, the apocrine gland secretes sweat with no odor but contains long fatty acids linked to amino acids which are classified as non-volatile compounds. One of the body parts that have an unpleasant odor compared to other body parts is the *axilla*. Bacteria that are abundant in the armpit are *Corynebacterium tuberculostearicum* and *Staphylococcus hominis*. These 2 bacteria come with the same character as they are anaerobic facultative bacteria that fit to live in the armpit area (Callewaert, Lambert & Van de Wiele, 2017). These two abundant types of bacteria in the axillary area will convert odorless substances such as glycerol and lactic acid which are secreted by sweat glands to form thioalcohol and Volatile Fatty Acids (VFAs) as the source of body foul odor via 4 mechanisms: Biotransformation of steroids, breaking of glutamine-conjugates, metallopeptidases and C-S lyses induced glycine-cysteine-(S)-conjugates breakage, and long-Chain Fatty Acid (LCFA) transformation into Volatile fatty acid (VFAs) (Lam et al.,2018; Rudden et al., 2020).

Deodorant is a cosmetic product that is mainly used to prevent body odor which usually contains various compounds to effectively reduce bad body odor. Commercialized deodorant products available in the market contain various compounds that act as an antiperspirant most likely coming from the aluminum salt agent that can reduce sweat gland secretion. Moreover, killing the odor-causing bacteria is also an important aspect of the deodorant product as the metabolism result of bacteria is one of the

major causes of bad body odor (Alzomor, Moharram & Al Absi, 2014). Effective deodorant product contains antimicrobial agents such as quaternary aluminum which is classified as broad-spectrum biocides against gram-positive and gram-negative bacteria to minimize the growth of odor-causing bacteria (Rajkowska et al., 2015). Therefore, an effective deodorant product should contain antimicrobial agents that are able to inhibit or avoid the growth of bad-body odor bacteria to avoid the degradation of eccrine and apocrine gland secretion to bad body odor-causing substances (Susanty et al., 2017).

Bacterial identification is a group of techniques to reveal the causative agent of a disease caused by bacteria. Various identification techniques have been developed nowadays, some common identification methods are microscopic visualization, selective/identification media, biochemical testing, staining, and molecular technique (Braga *et al*., 2013). Mannitol Salt Agar (MSA) is a selective and differential media used mainly to isolate *Staphylococcus spp* as this type of bacteria has several mechanisms that make this bacteria salt-tolerant (Sandle, 2019). Therefore, expected that only *Staphylococcus spp* colonies can grow on MSA media.

Gram staining is a widely used diagnostic technique in microbiology. Most bacteria species can be classified into gram + and gram - bacteria. The gram + bacteria appears as violet under microscope observation due to retaining crystal violet due to the thick peptidoglycan layer at its cell wall of bacteria outside of its cytoplasmic membrane. In contrast, the bacteria cell walls of gram-negative bacteria consist of a thin (single) peptidoglycan layer (10 nanometers) but contain an outer layer that makes them retain counterstain safranin and appear red under microscope observation.

A clinical trial is a study that involves human subjects being exposed to one or more treatment interventions to assess the outcome of the defined product. This study is a compulsory phase before a drug/cosmetic product is approved to be marketed as one regulatory necessity (Sambandan & Turcu-Stiolica, 2019). A clinical trial was conducted to assess the antimicrobial activity of a newly developed daily deodorant product that contained multiple natural extracts that theoretically have an

antimicrobial activity such as S oil, H extract, and B root extract. Therefore, this study was conducted to assess the antimicrobial claim efficiency of the product before being marketed. The product was tested on male university students of a range ages 19-23 years old. Males tend to have stronger body odor related to the production of high testosterone levels which leads to high sebum-producing cells or called sebocyte proliferation and thereby increases sweat secretion (Sorokowska, Sorokowski & Szmajke, 2012). In addition to that, a young adult male will have more active apocrine gland activity induced by hormonal factors which this phenomenon will be declined following adulthood (Lam et al., 2018).

The clinical trial was conducted to observe the antimicrobial activity of newly developed daily deodorant products towards four male university students as the subjects to check the effectiveness of deodorant on microbial growth reduction between treated and untreated armpits. Thus, the clinical trial result can be used as the antimicrobial claim effect of the product in the market. Moreover, the identification of the types of bacteria present in the treated *axilla* of the subject was observed using Mannitol Salt Agar (MSA) selective media towards bad body odor-causing bacteria as well as Gram Staining as the confirmatory method. Lastly, an additional procedure to check the bioburden level of the product was performed to assess the quality of the deodorant product.

1.2 Objectives

The main objective of this clinical trial study is to measure roll-on deodorant X's effectiveness to reduce bad body odor and identification of *axilla* microbiome in male Indonesian University Students by achieving several objectives:

1. Determining reduction of microbial load from treated *axilla* treated with roll-on daily deodorant product compared to control *axilla* that did not receive daily deodorant product via 3 days of clinical trial observation

2. Identifying the remaining bad body odor-causing bacteria in the treated *axilla* of male Indonesian university students using selective media and followed by microscopic observation using Gram Staining as the confirmatory method

1.3 Research Scopes

1. Document ethics and protocol preparation for clinical trials
2. Recruiting male participants among I3l University students (4 subjects) for clinical trials deodorant study
3. Sample collection using swab method to collect microbial samples from armpit at 3 different times (9 am, 1 pm, and 5 pm) for 3 consecutive days
4. Bacterial culture using Miles and Misra Method on Mueller Hinton Agar media
5. Colony Forming Unit (CFU) count to compare the number of bacterial colonies between treated and untreated armpits during 3 days of observation
6. Performing Spread Plate Method to culture the stock solution from treated *axilla* to quantitatively assess the presence of *Staphylococcus spp* bacteria in the treated sample
7. Gram Staining observation of bacterial culture retrieves from clinical trials subject for *axilla* bacteria identification
8. Statistical analysis to determine the significance of the reduction rate to determine the effectiveness of the deodorant product
9. Bioburden evaluation of the tested daily deodorant product using spread plate method and presenting the results using total plate count