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EXTRACTION AND EFFECT OF SAPONIN EXTRACTED FROM
PEEL OF *Musa acuminata* TOWARDS Sf9 CELLS

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ABSTRACT

Saponins are phytochemicals found in many plants. Their structural diversity make them to have various biological activities. Saponin is produced in various parts of plants such as fruits and fruit peels. Since saponin has been used in many industrial applications, it will be excellent if the source of saponin comes from the fruit.waste as this source is more economical and it can be converted into a useful bioproduct. One of the properties of saponin currently being studied is the insecticidal properties. The aims of this research were to extract saponin from the peel of banana *Musa acuminata* by using three different sample preparation methods, namely dried, fresh, and macerated peels, to determine the saponin content from the peel of banana, and to preliminary screen the insecticidal properties of saponin extracted on the growth of an insect cell line, *Spodoptera frugiperda* (*Sf9*). Fresh and macerated peels were prepared by blending and dried peels were oven-dried at 60°C before blended into powder. Both dried and fresh peels were exposed to solvent for 24 hours while the macerated peels was subjected to 72 hours. The extraction of saponin from the peels was done using two solvents namely, 50% (v/v) ethanol and 85% (w/v) hexane. All extracts were profiled at wavelength ranging from 300 nm to 700 nm. These profiles were compared with an absorbance profile containing 5 µg/mL of quercetin (Qu) which was used as flavonoid standard. Total saponin (TS) content was quantified by treating the extracts with vanillin and sulfuric acid, and the absorbance were recorded at 544 nm. The *Sf9* cells in suspension were treated with fresh-ethanol crude peel extract at a volume-to-volume ratio of 1:1 for 24 hours and the cell growth was measured its absorbance at 630 nm. From the profiling of crude peel extracts, dried extracts in ethanol showed higher absorbance profile than both fresh and macerated extracts in ethanol in which these two extracts shared similar absorbance profiles. In addition, hexane was able to extract saponin in fresh peel better than ethanol by giving higher absorbance profile. This observation of fresh peel extract was in contrast to the macerated peel extract. Based on the total saponin assay, dried peel extracts in ethanol showed the highest saponin content compared to both fresh and macerated peel extracts in ethanol. Total saponin contents in both fresh and macerated extracts were higher when hexane was used as the extraction solvent compared to ethanol. Fresh-hexane extract was found to contain more flavonoids compared to fresh-ethanol, macerated-ethanol and macerated-hexane extracts based on the comparison of peaks at 380 nm of quercetin absorbance profile . The percentages of growth inhibition for *Sf9* cells treated with fresh-ethanol extract and 50% (v/v) ethanol was 8.04% and 7.59% respectively. In conclusion,

dried peel extract in ethanol has higher saponin content compared to both fresh and macerated extracts based on the absorbance profiling and TS assay. Hexane was suggested as the suitable solvent to extract saponin compared to ethanol. The inhibition of *Sf9* cells' growth with fresh-ethanol extract did not show any significant difference with the solvent used.

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LIST OF ABBREVIATIONS

%	percent
°C	degree Celsius
A ₃₈₀	absorbance at 380 nanometer
A ₅₄₄	absorbance at 544 nanometer
A ₆₃₀	absorbance at 630 nanometer
CABI	CAB International
cells/mL	cells per mL
Eds.	editors
et al.	et alia
FAO	Food and Agriculture Organization of the United Nation
g	gram
g/mL	gram per millilitre
HCT-116	human colorectal carcinoma cell line
HIV	human immunodeficiency virus
HT-29	human colorectal adenocarcinoma cell line
JPNPP	Jabatan Pertanian Pulau Pinang
mg	milligram
mg/g	milligram per gram
mg/mL	milligram per millilitre
mL	millilitre
mm	millimeter
MOA	Ministry of Agriculture & Agro- Based Industry Malaysia

nm	nanometer
rpm	revolutions per minute
QS-21	<i>Quilliaja saponin</i>
Qu	Quercetin
<i>Sf</i>	<i>Spodoptera frugiperda</i>
SF-900 II SFM	<i>Sf</i> -900 II Serum-Free Media
sp.	species
subsp.	subspecies
TS	total saponin
UV	ultraviolet
v/v	volume per volume
w/v	weight per volume
µg/mL	microgram per millilitre
µL	microlitre

CHAPTER 1

INTRODUCTION

Fruits, along with vegetables, are one of the important components in food pyramid for a healthy human diet. They are the major source of minerals, vitamins, and fibres that are important for maintaining human health (Mañiyan, John & Mathew, 2015). The goodness of fruits has become a market interest in food industry to produce many fruit-based food products. Most of the food processing industry and also regular fruit consumption by consumers only use the edible parts of the fruits (Deng et al., 2012). A very little number of the fruit wastes such as the cores and the peels are processed to make other types of edible food or for other agricultural purposes. Most of the time, the fruit wastes are usually thrown away as municipal waste. According to Deng et al. (2012), fruit wastes are among the major sources of municipal wastes, raising an environmental issue on waste management. Based on the report published by Food and Agriculture Organization (FAO) of the United Nation in 2011, approximately 20% from total of 50% fruit and vegetables are lost or wasted due to food processing in South and Southeast Asia (FAO, 2011). Due to this, many researches have been done in identifying potential compounds from fruit wastes for industrial applications, converting the waste into a possible value-added bioproducts.

Besides food products, fruits are also used to make nutraceutical products such as health supplements and cosmetics. This is because fruits are also rich in phytochemicals, or bioactive compounds which provide colour, flavour, and as natural protection to the fruits (Maniyan et al., 2015). These compounds possess various promising health benefits such as antioxidants, anticancer, antifungal, and other benefits (Maniyan et al., 2015). An example of a phytochemical is saponin, which is widely distributed in plants (Moses, Papadopoulou & Osbourn, 2014), accumulating in organ and/or tissue-specific manner (Moses et al., 2014). Many plants synthesized and stored saponin in underground organ such as roots (Moses et al., 2014). Besides roots, different types of saponin have been isolated from stems, barks, leaves, seeds, fruits and flowers (Moses et al., 2014). Saponin has been commercially used as foaming agent in food industry, and as dietary supplements in pharmaceutical industry (Moses et al., 2014). Ginseng, soybean, and soapbark are commonly known plants for their saponin content (Choon, Hanaa & Rabiha, 2014).