



**UNIVERSITI PUTRA MALAYSIA**

***ANTIOXIDANT, ANTIMICROBIAL AND ANTICANCER PROPERTIES  
IN DIFFERENT PLANT PARTS OF *Dendrobium crumenatum* Sw***

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By

**SURIA BINTI JOHARI**

**Thesis Submitted to the School of Graduate Studies, Universiti Putra  
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Master of Science**

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Abstract of thesis presented to the Senate of Universiti Putra Malaysia in fulfillment of the requirement for the degree of Master of Science

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July 2019

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*Dendrobium crumenatum* is commonly known as Pigeon Orchid or *Orkid Merpati*. There is limited information on the phytochemical properties of *D. crumenatum* available at present. With the great development in herbal and pharmaceutical industry, more valuable extracts are required to discover their potential. The objectives of this study were to evaluate the level of antioxidant activity as well as anticancer and antimicrobial activities in different plant parts of *D. crumenatum* by using different plant parts, extracting solvent, drying temperature of plant materials and shaking duration, the optimum antioxidant, antimicrobial and anticancer activity of *D. crumenatum* were determined. The extracts of all tested plant parts with different extracting solvents shows antioxidant activities ranging from 2.3 to 10.3 mg TE/g DW. The highest total antioxidant in for DPPH and FRAP method was found on seven days shaking duration with the value 14.41 mg TE/g DW (pseudobulb dried at 70°C) and 35.51 mg TE/g DW (stem dried at 45°C) respectively. The antimicrobial activity shows significant differences on antimicrobial activity to that of the standard antibiotic against all tested microorganisms with MIC value in range of 0.31 to 10.0 mg/ml. From the *in vitro* cytotoxic bioassay results, the methanolic stem extracts of *D. crumenatum* demonstrated moderate activities against the NCI-H460 (IC<sub>50</sub> value of 64 µg/ml) and MCF-7 (IC<sub>50</sub> value of 77 µg/ml). The results from this bioactivity investigation suggested that pseudobulb and stem of *D. Crumenatum* may have great potential for further development as cancer chemoprevention agents specifically against NCI-H460 and MCF-7 or food supplements for promoting human health.

Abstrak tesis yang dikemukakan kepada Senat Universiti Putra Malaysia  
sebagai memenuhi keperluan untuk ijazah Master Sains

**ANTIOKSIDA, ANTIMIKROBIAL DAN ANTIKANSER DALAM PELBAGAI  
BAHAGIAN TUMBUHAN *Dendrobium crumenatum* Sw.**

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*Dendrobium crumenatum* biasanya dikenali sebagai *Pigeon Orkid* atau Orkid Merpati. Maklumat mengenai sifat fitokimia *D. crumenatum* yang terdapat pada masa ini adalah terhad. Dengan perkembangan yang hebat dalam industri herba dan farmaseutikal, ekstrak yang lebih berharga diperlukan untuk mengetahui potensi mereka. Objektif kajian ini adalah untuk menilai tahap aktiviti antioksidan serta aktiviti anti-kanser dan antimikrobial dalam pelbagai bahagian tumbuhan *D. crumenatum* dengan menggunakan pelbagai bahagian tumbuhan, pelarut pengekstrakan, suhu pengeringan bahan tumbuhan dan tempoh putaran yang berbeza, aktiviti optimum antioksidan, antimikrobial dan antikanser *D. crumenatum* telah ditentukan. Semua ekstrak bahagian tumbuhan yang diuji dengan pelarut pengekstrakan yang berbeza menunjukkan aktiviti antioksidan antara 2.3 hingga 10.3 mg TE / g DW. Jumlah antioksidan tertinggi menggunakan kaedah DPPH dan FRAP didapati pada tujuh hari tempoh putaran dengan nilai masing-masing 14.41 mg TE / g DW (pseudobulb dikeringkan pada 70 ° C) dan 35.51 mg TE / g DW (batang dikeringkan pada 45 ° C). Aktiviti antimikrobial menunjukkan perbezaan aktiviti antimikrobial yang ketara dengan rujukan antibiotik terhadap semua mikroorganisma yang diuji dengan nilai MIC dalam lingkungan 0.31 hingga 10.0 mg / ml. Daripada hasil penyaringan *in vitro* sitotoksik, ekstrak daripada batang metanol *D. crumenatum* menunjukkan aktiviti sederhana terhadap NCI-H460 (IC<sub>50</sub> 64 µg/ ml) dan MCF-7 (IC<sub>50</sub> 77 µg / ml). Hasil daripada penyaringan bioaktiviti ini mencadangkan bahawa pseudobulb dan batang *D. crumenatum* mungkin berpotensi besar untuk pembangunan lebih lanjut sebagai agen pencegahan kemo bagi kanser khususnya terhadap NCI-H460 dan MCF-7 atau makanan tambahan untuk menggalakkan kesihatan manusia.

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This thesis was submitted to the Senate of the Universiti Putra Malaysia and has been accepted as fulfilment of the requirement for the degree of Master of Science. The members of the Supervisory Committee were as follows:

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## LIST OF ABBREVIATIONS

|                                 |                                       |
|---------------------------------|---------------------------------------|
| °C                              | Degree Celcius                        |
| µL                              | Micro litre                           |
| µM                              | Micro molar                           |
| 5-FU                            | Fluoruracil                           |
| AlCl <sub>3</sub>               | Aluminum chloride                     |
| ANOVA                           | Analysis of variance                  |
| CE                              | Catechin equivalents                  |
| ddH <sub>2</sub> O              | Distilled deionized water             |
| DPPH                            | 2,2-diphenyl-1-picrylhydrazyl         |
| DMSO                            | Dimetyl sulfoxide                     |
| DW                              | Dried weight                          |
| EDTA                            | Ethylenediaminetetraacetic acid       |
| FRAP                            | Ferric reducing antioxidant potential |
| FW                              | Fresh weight                          |
| MCF-7                           | Hormone-dependent breast carcinoma    |
| Na <sub>2</sub> CO <sub>3</sub> | Sodium carbonate                      |
| NaOH                            | Sodium hydroxide                      |
| NCI-H460                        | Lung cancer                           |
| GAE                             | Gallic acid equivalent                |
| Nm                              | Nano meter                            |
| RPMI-1640                       | Roswell Park Memorial Institute       |
| SRB                             | Sulforhodamine B                      |
| TPTZ                            | 2,4,6-tri (2-pyridyl)-s-triazine      |

## CHAPTER 1

### INTRODUCTION

#### 1.1 Medicinal Plant Industry in Malaysia

Awareness on traditional knowledge of plants which have been used in various human civilizations around the world for medicinal purposes is very crucial before they are destroyed forever (Fatemeh et al., 2018). Malaysia has the oldest rainforest in the world and one of the 12 countries with mega biodiversities which is estimated to comprise of 1,200 medicinal plants. These plant species were known to possess potential pharmaceutical values (Ali et al., 2014) and have been used for generations in various traditional health care systems (Raja Rina and Mohd Khanapi, 2015).

The application of medicinal plants is still of great importance for therapeutic potential, and nowadays still represent an important pool for the identification of novel drug leads (Atanasov et al., 2015). Therefore, due to the great diversity of plant species in Malaysian rainforests, documentation of ethnobotanical and ethnomedicinal uses of these medicinal plants is required to avoid time consuming experimentations to discover modern therapeutic drugs from plants for pharmaceutical product development.

To date, progressive studies on the medicinal potential of therapeutic plants have been carried out for discovery of natural products and novel compounds. There are great development of synthetic drugs available in the market for treating various types of diseases for instance cancer, heart diseases, diabetes and malaria. However, research in natural products have continued since synthetic products are either too expensive or have negative side effects (Alafiatayo et al., 2014) and many diseases remain incurable (Fernando et al., 2018).

#### 1.2 *Dendrobium crumenatum*

One of the common orchid species within South and Southeast Asia is *Dendrobium crumenatum* which is generally well-known as Pigeon Orchid or *Orkid Merpati*. (Leong et al., 2013). This orchid is a tropical epiphytic orchid which can easily be found on mature trees along roadsides of tropical countries. The distinctive flowering characteristic of *D. crumenatum* is usually triggered by a drop in temperature (Sandrasagaran et al., 2014) and their flowering is synchronous and lasts for only one or two days. The white flowers consists of sepals (elliptical dorsal and two lateral sepals), petals (two lateral petals similar

to dorsal sepal and one labellum of three lobes) and the column (pistils and stamens) (Leong et al., 2013).

*D. crumenatum* was selected in this study due to its potential medicinal properties. According to Hossain (2010) a poultice of *D. crumenatum* leaves was reported for their traditional medical uses to treat boils and pimples. *D. crumenatum* was also reported to have the potential as antimicrobial agent due to the presence of alkaloid and flavonoid compounds (Sandrasagaran et al., 2014). Due to potentiality of this species as sources for drugs, a study on the biochemical evaluation for antioxidant and antimicrobial and anticancer properties in different plant parts of *D. crumenatum* should be carried out.

### 1.3 Problem Statements

There are a number of papers have been reported on the scientific medicinal properties of *Dendrobium* spp. (Moretti et al., 2013, Chinsamy et al., 2014, Jin et al., 2016 and Minh et al., 2016). To date, lots of studies have been carried out on *Dendrobium* spp. due to its attractive potential inhibition and prevention of cancer functions, immunostimulatory and antioxidant activity (Luo et al., 2011). However, there is very limited information on antioxidant properties, biochemical evaluation of different plant parts of *D. crumenatum* available at present. Therefore it is important to discover its potentiality as source for antioxidant and to identify the best plant parts containing high antioxidant, antimicrobial and anticancer activities.

By using different plant parts, extracting solvent, drying temperature of plant materials and shaking duration, the optimum antioxidant activity of *D. crumenatum* were determined. Different plant parts might produce different level of flavonoid, phenolic acid and polyphenol content. The resulting data will contribute to a better understanding on the importance of this species specifically as an antioxidant, antimicrobial and anticancer agents.

### 1.4 Objectives of Research

The general objective of this study was to evaluate the level of antioxidant compounds as well as anticancer and antimicrobial activities in different plant parts of *D. crumenatum*. The specific objectives of this study were:

- i. To determine the suitable extracting solvent, drying temperature and shaking duration (maceration technique) in pseudobulb, leaf and stem of *D. crumenatum*
- ii. To compare the antioxidant activity in pseudobulb, leaf and stem of *D. crumenatum*.



- iii. To determine the total flavonoid , phenolic acid and polyphenol content in pseudobulb, leaf and stem of *D. crumenatum*
- iv. To determine the antimicrobial and anticancer properties in pseudobulb, leaf and stem of *D. crumenatum*



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