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JUDUL: PHYTOCHEMICAL STUDY OF MEDICINAL PLANT - ALOE VERA

Ijazah: SARJANA MUDA SAINS (KIMIA INDUSTRI)

**SESI PENGAJIAN:** 2002/2005

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# PHYTOCHEMICAL STUDY OF MEDICINAL PLANT-*ALOE VERA*

**TEOH CHUN KEAT**

**THIS DISSERTATION IS SUBMITTED TO FULFILL THE REQUIREMENT FOR THE DEGREE OF BACHELOR OF SCIENCE WITH HONOURS**



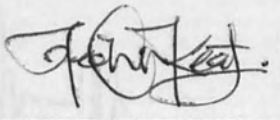
**INDUSTRIAL CHEMISTRY PROGRAMME**
**SCHOOL OF SCIENCE AND TECHNOLOGY**
**UNIVERSITI MALAYSIA SABAH**

**MARCH 2005**

## DECLARATION

I declare that this thesis contains my original research work. Sources of findings reviewed herein have been duly acknowledged.

23 March 2005



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# VERIFICATION

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## ABSTRACT

*Aloe vera* is a perennial, drought-resisting, succulent plant belongs to the Liliaceae family, which historically has been used for variety of medicinal purposes. It has high medicinal value, such as anti-inflammatory, anti-cancer and anti-diabetic. A phytochemical test were carried out of the plant and screened for alkaloids, saponins, terpenes, leucoanthocyanins and tannins. Alkaloids, saponins and tannins were found in the plant meanwhile leucoanthocyanins and terpenes were absent from the plant. Thin layer chromatography shows that the chloroform: methanol ratio of 9:1 is the best solvent mixture to separate the compounds in crude extract.

# ABSTRAK

*Aloe vera* merupakan sejenis tumbuhan yang hidup lama, tahan kemarau, tumbuhan sukulen yang berasal daripada famili Liliaceae. Tumbuhan ini digunakan secara meluas h dalam bidang perubatan, seperti ubat anti-keradangan, anti-kanser dan anti-diabitis dan sebagainya. Penyaringan fitokimia digunakan untuk mengetahui kehadiran sesuatu sebatian seperti alkaloid, saponin, terpena, leukoantosianidin dan tanin. Dalam ujian penyaringan fitokimia, didapati alkaloid, saponin dan tanin hadir dalam tumbuhan ini manakala leukoantosianidin dan terpena tidak dapat dikesan dalam tumbuhan ini. Dalam ujian TLC, didapati bahawa larutan kloroform: metanol pada nisbah 9:1 adalah pelarut yang memberi pemisahan yang paling baik bagi sebatian dalam ekstrak-ekstrak mentah.

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## LIST OF SYMBOL AND ABBREVIATION

| | |
|---|---|
| AIDS | Acquired Immunodeficiency Syndrome |
| CC | Column Chromatography |
| HDL | High Density Lipoprotein |
| HIV | Human Immunodeficiency Virus |
| NAD | Nicotinamide Adenine Dinucleotide |
| NMR | Nuclear Magnetic Resonance |
| PTLC | Preparative Thin Layer Chromatography |
| $R_f$ | Retention factors |
| TLC | Thin Layer Chromatography |
| UV | Ultraviolet |
| β | beta |

## LIST OF APPENDIX

# CHAPTER 1

# INTRODUCTION

## 1.1 PHYTOCHEMICAL STUDY

Phytochemicals, as the word implies, are the individual chemicals from which plant are made (Kaufman *et al.*, 1999). Phytochemistry, or plant chemistry, has developed in between natural product organic chemistry and plant biochemistry and is closely related to both. It is concerned with the enormous variety of organic substances that are elaborated and accumulated by plants and deals with the chemical structures of these substances, the biosynthesis, turnover and metabolism, their natural distribution and their biological function (Kaufman *et al.*, 1999).

In all these operations, methods are needed for separation, purification and identification of the many different constituents present in plants. Thus, advances in our understanding of phytochemistry are directly related to the successful exploitation of known technique and continuing development of new techniques to solve outstanding problems as they appear. One of the challenges of phytochemistry is to carry out all the above operations on vanishingly small amounts of material.

The general categories of plant natural products are lipids, including the simple and functionalized hydrocarbons, as well as the terpenes, which are treated separately. Following this are the unsaturated natural products, including the polyacetylene and aromatic compounds. We then cross over into the realm of the primarily hydrophilic molecules including sugars, and continue with those which can form salts, including alkaloids, the amino acids, and the nucleosides. Overall, this scheme provides a simple organizational pattern for the phytochemicals (Kaufman *et al.*, 1999).

Phytochemical progress has been aided enormously by the development of rapid and accurate methods of screening plants for particular chemicals and the emphasis is inevitably on chromatographic techniques. These procedures have shown that many substances originally thought to be rather rare in occurrence are almost universal distribution in the plant kingdom (Kaufman *et al.*, 1999).

*Aloe vera* is a succulent perennial herb with a rosette of narrow, prickly edged, fleshy leaves filled with bitter juice (Goh *et al.*, 1995). The plant has stiff grey-green lance shaped leaves containing clear gel in a central mucilaginous pulp. *Aloe vera* is a popular house plant and has a long history of use in folk medicine for skin and other disorders which goes back over thousands of years. Two kinds of products are provided from aloe leaves. One is a colourless and tasteless gel obtained from parenchyma cells. The second a yellow exudate from leaves, the active phenolic components of which are abundant in the inner epidermal cell layers (Okamura *et al.*, 1997).

*Aloe vera* gel consists primarily of water, monosaccharides and polysaccharides (glucomannan and other polysaccharides containing arabinose, galactose and xylose). It also contains tannins, sterols, organic acids, enzymes (including cyclooxgenase), saponins, vitamins and minerals (Barnes *et al.*, 2002).

*Aloe vera* is widely used for the external treatment of minor wounds and inflammatory skin disorders. The gel is used in treatment of minor skin irritations, including burns, bruises and abrasions (Barnes *et al.*, 2002). Commercial preparations of *Aloe vera* for medicinal and cosmetic use are available. It is further used in cosmetic industry as a hydrating ingredient in liquids, creams, sun lotions, shaving creams, lip balms, healing ointments and face packs (Bruneton, 1995). People are most familiar with *Aloe vera* use in skin care products but also be used as a beverage. Aloe product for internal use have been promoted for constipation, coughs, wounds, ulcers, diabetes, cancer, headaches, arthritis, immune system deficiencies and many other condition. However, the only substantiated internal use is as a laxative (Grindlay and Reynolds, 1986).

## 1.2 OBJECTIVES OF STUDY

Since *Aloe vera* is well know and grown worldwide for various uses and has a long history as a multipurpose folk remedy. It attracted my interest in doing this research on *Aloe* species. My objectives are listed below:

a. Phytochemical screening of *Aloe vera*

b. Extraction and isolation of chemical compounds that found in *Aloe vera*

## 1.3 SCOPE OF STUDY

The scope of study is to identify the alkaloids, terpenes, saponin, leucoanthocyanin and tannins in *Aloe vera* leaf gel and to separate the constituents by using thin layer chromatography and column chromatography.

# CHAPTER 2

# LITERATURE REVIEW

## 2.1 ALOE VERA

*Aloe vera* is native to North Africa, the Mediterranean region of southern Europe, and to Canary Island. It is now cultivated throughout the West Indies, tropical America, and tropics in general (Ross, 1999). Aloe vera is obtained from the mucilaginous tissue in the centre of the *Aloe vera* leaf and consists mainly of polysaccharides and lipids. It should not be confused with aloes, which is obtained by evaporation of water from the bitter yellow juice that is drained from the leaf. Unlike aloes, *Aloe vera* does not contain any anthraquinone compounds and does not, therefore, exert any laxative action (Barnes *et al.*, 2002).

Aloe is known as "lily of the desert", "the plant of immortality" and the "medicinal plant" (Fetrow *et al.*, 2000). Aloe vera is a short-stemmed succulent perennial herb of the Liliaceae family, the succulent leaves are crowded on the top of their stems, spreading grayish green and glaucous; spotted when young, 20 to 50 cm long, 3 to 5 cm wide at the base, tapering gradually to the pointed tip, 1 to 2.5 cm thick; having spiny

edges and bitter latex inside. Flowers are borne in cylindrical terminal racemes on central flower stalks, 5 to 100 cm high. The yellow perianth is divided into six lobes, about 2.5 cm long with scattered bracts. Each flower has six protruding stamens and three celled ovary with long style. Forms of the species vary in sizes of leaves and colours of flowers (Ross, 1999). Clinical evaluations have revealed that the pharmacological active ingredients have been are concentrated in both the gel and rind of the Aloe vera leaves. These active ingredients have been shown to have analgesic and anti-inflammatory effects (Ross, 1999).

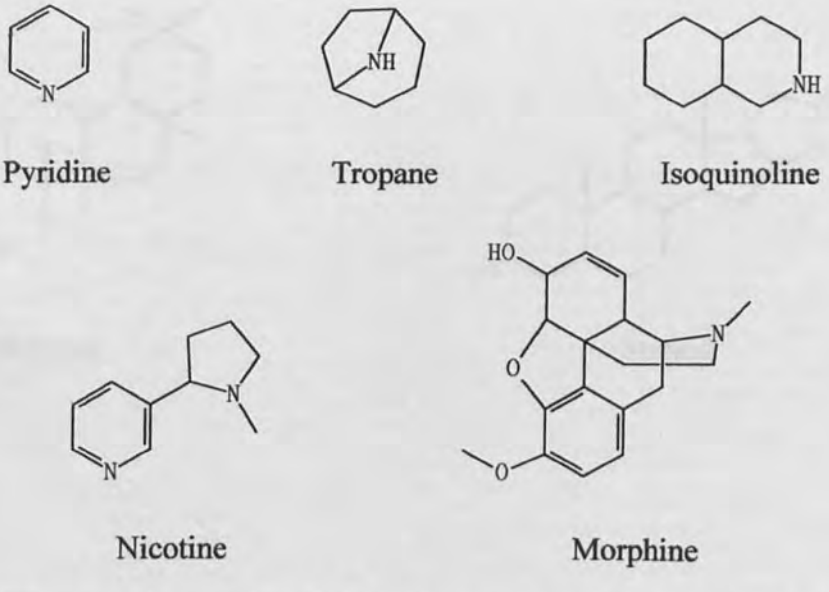
*Aloe vera* is now a familiar ingredient in a range of healthcare and cosmetic products widely available and advertised in shops. The preserved but otherwise untreated gel is also sold as a therapeutic agent in its own right as are various concentrated, diluted and otherwise modified products. This commercial activity has been accompanied by an upsurge of both clinical and chemical research which is reaching more closely towards the active ingredients and their biological activity (Ross, 1999).

Reasons presented for aloe gel efficacy is still varied perhaps there are in fact several different healing activities operating (Capasso *et al.*, 1998). The action of aloe gel as a moisturizing agent is still a popular concept (Briggs, 1995) and may account for much of its effect. *Aloe vera* is also often mentioned are the antibacterial, antifungal and even antiviral properties demonstrated by the gel (Ahmad *et al.*, 1993), while anti-oxidant effects are more becoming of interest.

## 2.2 THE CHEMICAL CONSTITUENTS

### 2.2.1 Alkaloids

The alkaloids include those natural products that contain nitrogen, usually as part of a cyclic system. Compounds of this type are numerous among plants and are perhaps best known for their often potent pharmacological properties. Thus, many of the common drugs are alkaloids based (Kaufman *et al.*, 1999). Relatively mild examples include caffeine, quinine and nicotine. More potent examples include cocaine, morphine and strychnine. Biosynthetically, they may be derived from amino acids, terpenes or aromatics depending on the specific alkaloid structure. Because of this diversity, they are often derived from the plant source rather than being produced synthetically. In some cases, they may be grouped on the basis of the ring system present. Several common ring system used for classification are shown in Figure 2.1 (Kaufman *et al.*, 1999).




Pyridine Tropane Isoquinoline

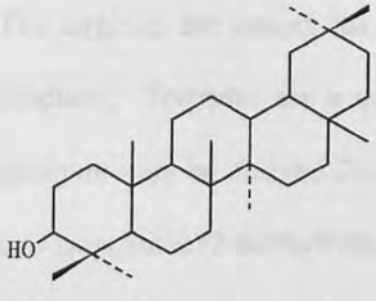
Nicotine Morphine

**Figure 2.1** Classes and examples of alkaloid natural products (Kaufman *et al.*, 1999).

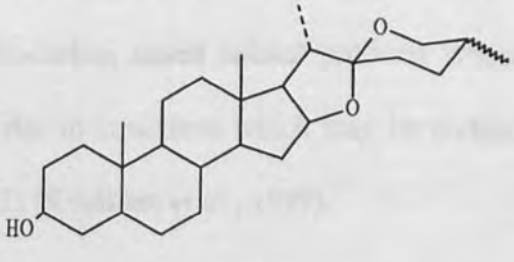
### 2.2.2 Saponins

Saponins are high molecular weight triterpene glycosides containing a sugar group attached to either a sterol or other triterpene. They are widely distributed in the plant kingdom and composed of two parts: glycon (sugar) and aglycone or genin (triterpene) (Kaufman *et al.*, 1999). Typically, they have detergent properties, readily form foams in water, have a bitter taste and are piscicidal (toxic to fish). Many of the plants that contain saponins have been used historically as soaps. The aglycones or genins as they are sometimes called may be of the triterpene, steroid or steroid alkaloid class. Saponins may be monodesmodic or polydesmodic depending on the number of attached sugar moieties (Figure 2.2). Biosynthetically, the saponins are comprised of six isoprene units and are derived from squalene (Kaufman *et al.*, 1999).

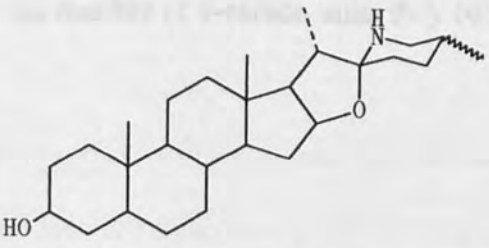
Saponin Classes



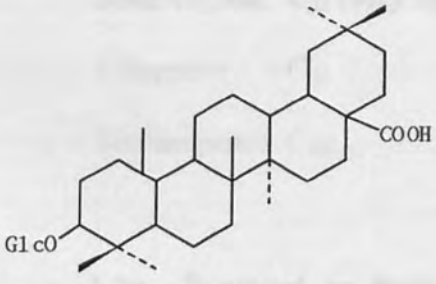

Triterpene



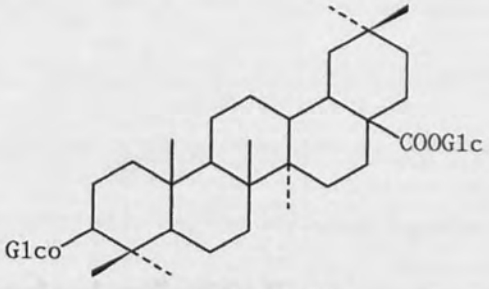

Steriod




Steroid Alkaloid




Monodesmodic

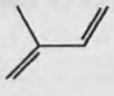



Bidesmodic

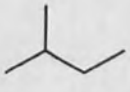
**Figure 2.2** Classification of saponins (Kaufman *et al.*, 1999).

### 2.2.3 Terpenes

The terpenes are among the most widespread and chemically diverse groups of natural products. Terpenes are a unique group of hydrocarbon based natural products whose structure may be derived from isoprene, giving rise to structures which may be divided into isopentane (2-methylbutane) units (Figure 2.3) (Kaufman *et al.*, 1999).



isoprene
(2-methyl-1,3-butadiene)



isopentane
(2-methylbutane)
(isopentane)

**Figure 2.3** The terpenes are comprised of isoprene units (Kaufman *et al.*, 1999).

Terpenes are classified by the number of 5-carbon units they contain:

Hemiterpenes $C_5$

Monoterpene $C_{10}$

Sesquiterpene $C_{15}$

Diterpene $C_{20}$

Sesterterpene $C_{25}$ (very rare)

Triterpene $C_{30}$

Tetraterpenes $C_{40}$

Like all natural products, within this simple classification lies an enormous amount of structural diversity which leads to a wide variety of terpene-like (or terpenoid) compounds. Note that the simplest examples of the terpenes are technically hydrocarbons, though they are considered separately here because of their common structural features. Not surprisingly, the terpenes are of a similar biogenetic origin, in which isopentenyl pyrophosphate and dimethylallyl pyrophosphate combine to yield geranyl pyrophosphate, leading to monoterpenes. Similarly, compounds derived from farnesyl pyrophosphate lead to sesquiterpenes, and triterpenes are formed from two equivalents of farnesyl pyrophosphate. These various combinations and oxidations give rise to a large variety of terpenes (Kaufman *et al.*, 1999).

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