THE PREVALENCE AND FACTORS ASSOCIATED WITH CRUDE HERBS USE AMONG PATIENTS WITH CHRONIC DISEASES: A CROSS-SECTIONAL SURVEY IN COMBINATION WITH LABORATORY ANALYSIS

By

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ABSTRACT

THE PREVALENCE AND FACTORS ASSOCIATED WITH CRUDE HERBS USE AMONG PATIENTS WITH CHRONIC DISEASES: A CROSS-SECTIONAL SURVEY IN COMBINATION WITH LABORATORY ANALYSIS

DEEPA RAJENDRAN

The use of crude herbs by patients in treatment and management of chronic diseases is prevalent, globally. The bioactive compounds are responsible for the medicinal properties exhibited by the crude herbs. This study was conducted to identify the prevalence and associated factors of crude herbs use among patients with chronic diseases and to screen the bioactive compounds present in the crude herbs used by them. A cross-sectional study was conducted in Klinik Kesihatan Kampar, Perak among 242 patients with chronic diseases. A self-administered questionnaire designed based on previous literatures were used to conduct the survey. The herbs consumed by patients were identified and extracted using aqueous and standard solvent methods. The extracts were screened for eight bioactive compounds, namely phenols, quinones, tannins, terpenoids, saponins, flavonoids, glycosides, and alkaloids. The response rate was 75.2% with a total of 48 male patients (39.7%) and 73 female patients (60.3%). The prevalence of crude herbs use was 36.4%. Factors significantly associated with crude herbs use were females (p=0.035), Indians (p=0.047) and hypertension (p=0.049). The types of crude herbs commonly used by the patients were *Psidium guajava*

(4.1%) and *Coriandrum sativum* (3.3%). The qualitative phytochemical screening of the herbs aqueous and solvent extracts revealed presence of eight phytochemical compounds in varying intensity. Overall, the aqueous extracts exhibited phytochemicals in higher intensity. The use of crude herbs concurrently with conventional medicines among patients with chronic diseases must be taken into account to ensure patient-centred care is warranted.

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DECLARATION

I hereby declare that the project is based on my original work except for quotations and citations which have been duly acknowledged. I also declare that it has not been previously or concurrently submitted for any other degree at UTAR or other institutions.

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DEEPA RAJENDRAN

APPROVAL SHEET

This project report entitled "**THE PREVALENCE AND FACTORS ASSOCIATED WITH CRUDE HERBS USE AMONG PATIENTS WITH CHRONIC DISEASES: A CROSS-SECTIONAL SURVEY IN COMBINATION WITH LABORATORY ANALYSIS**" was prepared by DEEPA RAJENDRAN and submitted as partial fulfilment of the requirements for the degree of Bachelor of Science (Hons) Biomedical Science at Universiti Tunku Abdul Rahman.

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PERMISSION SHEET

It is hereby certified that **DEEPA RAJENDRAN** (ID No: **15ADB01238**) has completed this final year project/ dissertation/ thesis* entitled "THE PREVALENCE AND FACTORS ASSOCIATED WITH CRUDE HERBS USE AMONG PATIENTS WITH CHRONIC DISEASES: A CROSS-SECTIONAL SURVEY IN COMBINATION WITH LABORATORY ANALYSIS" under the supervision of Dr. ANNALETCHUMY LOGANATHAN from the Department of Biomedical Science, Faculty of Science, and Ms. SANGEETHA ARULLAPPAN from the Department of Biomedical Science, Faculty of Science.

I hereby give permission to the University to upload the softcopy of my final year project in pdf format into the UTAR Institutional Repository, which may be made accessible to the UTAR community and public.

Yours truly,

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(DEEPA RAJENDRAN)

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LIST OF ABBREVIATIONS

CCl_4	Carbon tetrachloride
CAM	Complementary and alternative medicine
HCI	Hydrochloric acid
NaCl	Sodium chloride
NaOH	Sodium hydroxide
SPSS	Statistical Package for the Social Sciences
H_2SO_4	Sulphuric acid
TLC	Thin layer chromatography
TM	Traditional medicine
UV	Ultraviolet

CHAPTER 1

INTRODUCTION

1.1 Research Background

The utilisation of herbal medicines for the curation and prevention of infectious and non-infectious diseases has been gaining popularity in recent years (Rivera et al., 2013). Herbal medicines encompass herbs, herbal materials, herbal preparation and finished herbal products (World Health Organization, 2017). Herbs are the most crucial element needed in the preparation of herbal medicines and they are broadly defined as any plants that are used for their flavours, scents, and medicinal properties in the making of cosmetics, fragrances, and pharmaceutical products (Harris et al., 2014).

Crude herbs are subgroups classified under herbal medicines. They are termed as "a vast range of plant substances such as leaves, flowers, fruits, seeds, trunks, woods, rind, roots or any other parts of a plant that is whole, fragmented, powdered or cut" (World Health Organization, 2017). Crude herbs are raw, pure, unprocessed, or sometimes minimally processed, preserved, yet fresh at times (Nahler, 2017).

The use of herbs has been increasing worldwide. The prevalence of herbs use among the population from different countries varies from 30% to 70% due to

the different definitions used in the studies. In developed countries, such as, America have reported a prevalence of 35% of herbal medicine use (Rashrash et al., 2017). In Ethiopia, 48.6% of pregnant women consumed herbal medicines to medicate common cold and infection (Mekuria et al., 2017). Issa and Basheti (2017) reported 65.1% of herbal medicine use in the capital city of Jordan.

Cambodia, Thailand, and Vietnam reported 44.5%, 34.6%, and 42.9% prevalence of herbs use respectively (Peltzer et al., 2016b). Siti et al. (2009) reported a prevalence of 29.6% of raw herbs use in Malaysia. The population of Penang Island consumed herbal drinks (27%), raw herbs (12.5%), and processed herbs (15.5%) either for health maintenance or disease treatment (Dawood et al., 2017).

Previous studies mentioned the use of herbs to be evident among patients with chronic diseases (Baharom et al., 2016; Rashrash et al., 2017). According to Centres for Disease Control and Prevention (2018), chronic disease occurs when an individual experiences a persistent medical condition for at least a year; needs continuous inspection by a doctor; and has to minimise daily activities. Some examples of chronic diseases are diabetes, hypertension, stroke, hypercholesterolemia, respiratory diseases, cardiovascular disease, and many more (World Health Organization, 2018).

Globally, 58% of the elderly population of 65 years old and above were diagnosed with hypertension, followed by 27% of elderly with diabetes and 47% of the elderly with high cholesterol level (National Council on Aging, 2017). A nationwide study conducted in Malaysia, reported about 30.3%, 17.5%, and 47.7% of the elderly population were suffering from hypertension, diabetes, and hypercholesterolemia respectively (Ministry of Health, 2015).

Increasing statistics of persistent illness rendered people to reach out for traditional medicine (TM), complementary and alternative medicine (CAM), herbal medicines or herbs to reduce the effect of chronic diseases. Patients with chronic diseases consumed herbs either with conventional medicine or without conventional medicine (Dawood et al., 2017). Patients consumed herbs along with conventional medicine to complement the medication rather than as an alternative option (Huri, 2009). The concomitant use of herbs with conventional medicine may lead to herb-drug interaction. The interaction between herbs and drugs may result in therapeutic effects or toxic effects (Gupta et al., 2017).

In the United States, the major herbs' consumers have a history of medications: cancer (43%), diabetes (41%), arthritis (43%), heart diseases (43%), stroke (48.7%), and breathing-related disease (41%) (Rashrash et al., 2017). In a study conducted in Jordan, reported that 58.5% out of 200 patients with chronic diseases were found using herbs (Issa and Basheti, 2017). In Malaysia, Baharom et al. (2016) reported a prevalence of 40.6% of herbs use among diabetic

patients. Similarly, Ching et al. (2013b) reported a prevalence rate of 50% of herbs use by patients with diabetes mellitus. In national survey conducted in Malaysia, also reported herbs use among patients with other chronic diseases, such as, asthma (16.9%), cancer (40.2%), and hypertension (46.8%) respectively (Alshagga et al., 2011; Farooqui et al., 2016; Abdullah et al., 2018).

Collectively, the prevalence of herbs is high throughout the world because of the terminology itself. The term "herbs" or "natural" always means pure or safe to the consumers, thus encouraged them to consume it without doctor's advice (Seeman et al., 2018). Some patients believed and viewed herbs, as an optimistic choice compared to modern medicine due to the adverse effects of drugs (Ekor, 2014). The consumers or patients trust that herbs are much effective and safe compared to conventional medicines.

In addition, herbs possess bioactive compounds that exhibit medicinal properties (Wink, 2015). Bioactive compounds are secondary metabolites produced from the end product of primary metabolites. Secondary metabolites cannot partake in normal biological processes of a plant (Kumar et al., 2015; Tiwari and Rana, 2015). Wink (2015) added that secondary metabolites play a role as a defense system to the plants and act as a cue for insects and animals to aid in reproduction. Secondary metabolites also act as an antioxidant, confer UV protection, and remove free radicals (Zimdahl, 2018). Terpenoids, alkaloids, and phenolics are generally classified as secondary metabolites and categorised

further into their respective classes (Kabera et al., 2014). On the contrary, the primary metabolites are essential compounds needed by a plant to undergo maturation and differentiation; for instance, carbohydrates, fats, proteins, and nucleic acids (Sharma and Batra, 2016). Primary metabolites do not contribute to the medicinal properties exhibited by the plants.

1.2 Problem statement

In general, there is a scarce of research focusing on the prevalence of crude herbs use and its associated factors among patients with chronic diseases. Most studies investigated the prevalence of CAM, TM, and herbal medicines. To our knowledge, there is a lack of studies reported solely on use of crude herbs among patients with chronic diseases in Malaysia. Previous studies reported herbs to be the highest used type of CAM. Thus, it is essential to investigate the high use of crude herbs by patients without being monitored by physicians. This is to bring awareness as well as to disseminate information on the related side effects that crude herbs may cause (Ekor, 2014). To our knowledge, there are no studies that incorporate both cross-sectional survey and phytochemical screening on crude herbs used by patients.

This study is done to fill three important gaps in the literature: (1) it investigates the prevalence of crude herbs use among the patients with chronic diseases (2) it investigates the phytochemicals present in the commonly used crude herbs among the patients with chronic diseases (3) it incorporates the literatures of

crude herbs, chronic diseases, and phytochemical compounds present in crude herbs.

1.3 Objectives

Therefore, the objectives of this study are as follows:

i. To identify the prevalence of crude herbs use among patients with chronic diseases.

ii. To identify the type of crude herbs used by patients with chronic diseases.

iii. To analyse the association between socio-demographic factors and crude herbs use among patients with chronic diseases.

iv. To analyse the association between types of chronic diseases and crude herbs use among patients with chronic diseases.

v. To investigate the phytochemical compounds present in the commonly used crude herbs by patients with chronic diseases.

CHAPTER 2

LITERATURE REVIEW

2.1 Medicinal plants found in Malaysia and their benefits

Globally, medicinal plants gained popularity over the past few years with 80% of the population found consuming herbs based products. The consumers believed that herbs based products are safe because of the terms used, such as, natural and herbal (Seeman et al., 2018). Besides, natural products also have been integrated into modern medicine, thus increasing consumer's trust in them. Malaysia is a tropical country rich with many species of medicinal plants. Approximately, 1300 discovered medicinal plants were used to cure diseases (Alsarhan et al., 2014).

An example of medicinal plant, *Andrographis paniculata* is used to treat flu, chest pain as well as an antihypertensive agent and antidiarrhoeal agent. To alleviate bronchitis, asthma, gastritis and dysentery, *Centella asiatica* or 'pegaga' is eaten as a salad (Mitra et al., 2007). *Eurycoma longifolia* or 'Tongkat Ali' is widely used by the Malaysian men to increase their sexual drive or to treat impotence. Roots and barks of *Eurycoma longifolia* cure diarrhoea, fever, consistent coughing, and bleeding gums. Mitra et al. (2007) reported antimalarial and anti-inflammatory properties of the plant.

Clinacanthus nutans or ‘Sabah snake grass’, enhances the immune system, promote peristalsis, treat diabetes, and cancer (Kong et al., 2016). Other examples of medicinal plants include *Hibiscus rosa-sinensis* and *Moringa oleifera,* reported to treat high blood pressure (Krishnaiah et al., 2009). In the subsequent chapters, the taxonomies and medicinal values of the common crude herbs used by patients to treat their chronic medical conditions are discussed.

2.2 *Psidium guajava*

2.2.1 General description

Psidium guajava (*P.* guajava) is commonly known as guava and ‘jambu batu’ by the Malaysians. *P. guajava* is a dicotyledonous shrub about 3 to 10 metres (m) in height. The barks of a guava tree are smooth, vary from light brown to reddish brown in colour, and often flake (Centre for Agriculture and Bioscience International, 2019b). The roots of this species are extensive whereas the leaves are pale green in colour opposite, oblong, thick, and leathery with distinguishable veins underneath. The flowers are white in colour with three to five petals whereas the fruits are green in colour and round, ovoid or pear-shaped. The fruits are either with seeds or seedless, depending on the strain of the species. The seeds are surrounded by edible and fleshy pulp (Centre for Agriculture and Bioscience International, 2019b).

The taxonomical classification and the picture of *P. guajava* are shown in **Table 2.1** and **Figure 2.1** respectively. Guava is grouped under the family of Myrtaceae with the genus of *Psidium* and species of *Psidium guajava* (Centre for Agriculture and Bioscience International, 2019b).

Table 2.1: Taxonomical classification of *Psidium guajava.*

Division	**Class**
Kingdom	Plantae
Family	Myrtaceae
Subfamily	Myrtoideae
Genus	*Psidium*
Species	*Psidium guajava*

Figure 2.1: *Psidium guajava* leaves and young fruits (Joseph et al., 2016).

2.2.2 Geographical distribution

P. guajava originated from Northern Mexico and Central America. After the natives of Spain and Portugal introduced them to other continents, they are now widely found in other tropical countries, such as Malaysia, Vietnam, Thailand, and India (Singh, 2011; Diaz de Cerio et al., 2017). The cultivation of guava in other tropical countries was encouraged because it adapts easily to a new environment (Singh, 2011).

2.2.3 Presence of bioactive compounds and medicinal uses

Different phytochemicals were found in various parts of guava (Joseph et al., 2016). Nevertheless, different solvent extracts of guava exhibit nuances of phytochemicals present; the hexane extract of guava revealed tetracosane, α-copaene, γ-sitosterol, vitamin E, and squalene whereas the chloroform extract of guava revealed the presence of palmitic acid (Ashraf et al., 2016).

In addition, guava consists of phenols, essential oils, saponins, triterpenoids, and tannins that are responsible for its therapeutic properties. The leaves of guava constitute of alkaloids, essential oils, saponins, tannins, flavonoids, phenols, gallic acids, and catechins. The phytochemicals aforementioned possess antitumour, anti-inflammatory, and anti-microbial properties. Isolated quercetin from the guava leaves is responsible for preventing myocardial damage and relieving muscle spasm (Ravi and Divyashree, 2014).

Vitamin C and beta-carotenes found in the pulp of guava possess antioxidant properties and lower down glucose level in the blood (Ravi and Divyashree, 2014; Joseph et al., 2016). A high amount of tannins, flavonoids, phenols, and saponins were present in aqueous, acetone, and ethanol extracts of the barks and leaves of guava. The isolated compounds are responsible for the antioxidant properties of guava (Rafiqkhan et al., 2014).

2.2.4 In vitro studies

Few studies have investigated the therapeutic effects of various parts of guava, mainly the leaves. One of the investigations used albino rats to study the effect of guava leaves on three types of inflammations: acute, subacute, and chronic inflammation. Guava leaves impeded the inflammatory response by restraining the swelling of the rat's paw and the formation of exudate (Dutta and Das, 2010).

The effect of guava leaves on reducing lipid profile in hepatotoxic rats was studied. Quercetin isolated from the leaves was used to treat the rats injected with carbon tetrachloride (CCl_4). The rats exhibited a lower total cholesterol and triglycerides as well as higher high density lipoproteins (Vijayakumar et al., 2018). The blood glucose level of alloxan-induced diabetic rats reduced after administration of ethanolic guava extracts. Aqueous extracts showed a 9% reduction in the blood glucose level of the rats as well (Shukla and Dubey, 2009). The hexane extract of guava leaves was established as the most formidable killer of *Agrobacterium tumefaciencs*, a type of bacteria that causes tumour in plants.

Guava showed anticancer properties by reducing the development of malignant tumour cells on *Agrobacterium tumefaciencs* (Ashraf et al., 2016).

2.3 *Coriandrum sativum*

2.3.1 General description

Coriandrum sativum (*C. sativum*) is known as coriander, Chinese parsley, and cilantro. In Malaysia, it is more commonly known as 'ketumbar' (Centre for Agriculture and Bioscience International, 2019a). The name of coriander originated from 'koris', known as a bug in Greek word. It is named such because the fruits smell like bedbug when it is not riped yet. Once the seeds ripe, the whole plant produces lemon-like scent (Government of Alberta, 2016). Coriander is used as spices and herbs in cooking as well as in medications for many years (Hosseinzadeh et al., 2014).

It is an annual and perennial tree that can grow up to 60 centimetre (cm). The plant possesses solid stem with many branches and a taproot system. The leaves of coriander are hairless, smooth, and green in colour. In addition, the leaves have a waxy texture underneath them and the shape varies, from broad lobed at the bottom to feathery at the top. The flowers are either white or pale pink in colour, asymmetrical in shape with petals pointing away from the centre of umbel (Government of Alberta, 2016). The fruits produced by this tree are small and oval with an aromatic scent. They are cremocarp, yellowish brown in colour

and the seed's size is approximately 3 to 5 millimetres (mm) in diameter with wavy longitudinal ridges (Mia, 2016).

The taxonomical classification and the picture of *C. sativum* are shown in **Table 2.2** and **Figure 2.2** respectively. Coriander or cilantro is grouped under the family of Apiaceae (Umbelliferae or carrot family) with the genus of *Coriandrum* and species of *Coriandrum sativum* (Centre for Agriculture and Bioscience International, 2019a).

Table 2.2: Taxonomical classification of *Coriandrum sativum* (Adapted from United States Department of Agriculture, 2019).

Division	**Class**
Kingdom	Plantae
Family	Apiaceae
Subfamily	Apioideae
Genus	*Coriandrum*
Species	*Coriandrum sativum*

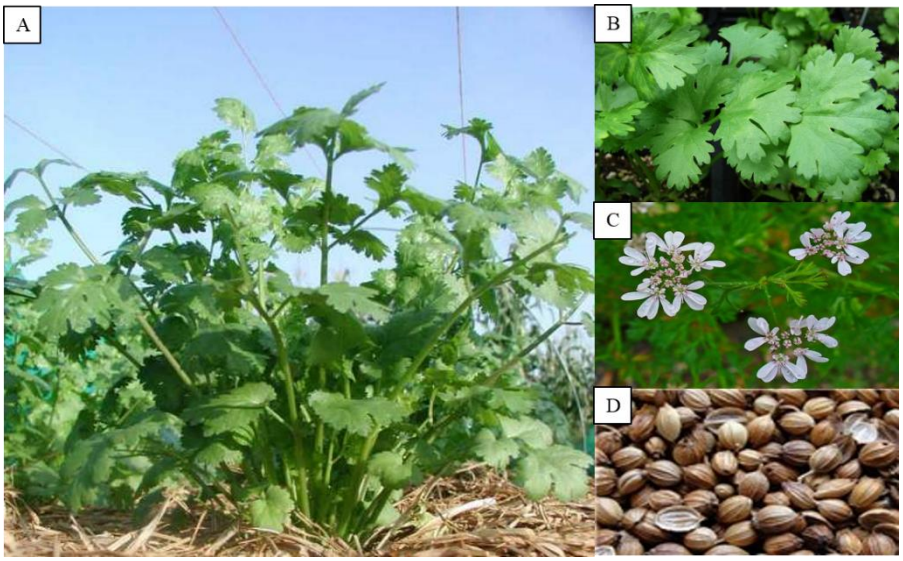

Figure 2.2: *Coriandrum sativum* [A] plant, [B] leaves, [C] flowers, and [D] seeds (Adapted from Goyal, 2012; Allied Botanical Corporation, 2019; Centre of Agriculture and Bioscience International, 2019a).

2.3.2 Geographical distribution

Coriander originated from the Mediterranean region and is grown widely in Europe, Asia, North Africa, and South America. It was introduced as a spice to Great Britain by the Romanians. Then, it was brought over to America and then cultivated across the globe to Asian countries and the United Kingdom (Maroufi et al., 2010). Coriander grows best on loam soil that is either sandy or well drained at 18°C.

2.3.3 Presence of bioactive compounds and medicinal uses

The ethanolic extract of coriander roots revealed the presence of alkaloids, phenols, flavonoids, saponins, and terpenoids. However, there was no any tannins or quinones present in the roots of coriander (Kumar et al., 2014). The seeds of coriander exhibited alkaloids, phenols, tannins, flavonoids, glycosides,

and coumarins when extracted using methanol and acetone (Thangavel et al., 2015). Tannins, alkaloids, reducing sugar, flavonoids, and phenolic compounds are present in coriander leaves extracted using methanol (Kumar et al., 2018).

All three methanolic, ethanolic, and aqueous extracts of coriander seeds revealed the presence of saponins, tannins, and cardenolides (Ahmed et al., 2018). Coriander seeds contain high amount of essential oils (Hwang et al., 2014). In general, there are no tannins in the roots but the seeds and the leaves of coriander showed the presence of tannins. Alkaloids, flavonoids, and phenols were present in all the parts despite any extraction medium used.

The presence of flavonoids in coriander is associated with its antitumour and antioxidant activity. It removes any abnormal cells by causing cell death and stopping the cell cycle. Presence of tannins and glycosides are correlated with its antimicrobial activity. Anthraquinones present in coriander leaves are responsible for toning up smooth muscles. Alkaloids present in coriander leaves are able to dilate blood vessels and treat arrhythmia condition (Kumar et al., 2018). Coriander is also used as a remedy for dysentery, indigestion, and abdominal pain. They possess antidiabetic and hypolipidaemic properties (Maroufi et al., 2010).

2.3.4 In vitro studies

Scientific evidences reported the medicinal effects of coriander. Antimicrobial activity of coriander roots suppressed the proliferation of *Klebsiella* and *Bacillus cereus* (Kumar et al., 2014). Hosseinzadeh et al. (2014) reported that powdered coriander induced better digestion in broiler chicks due to the presence of essential oil (linalool). The aqueous extracts of coriander reduced blood glucose level more actively in the broiler chicks compared to the powdered extracts. Chicks fed with coriander at a concentration of 2% showed a significant reduction in total cholesterol level (±96.00) compared to other treatment groups and control (±111.25).

2.4 Prevalence and associated factors of crude herbs use among patients with chronic diseases

Many studies reported a high prevalence of herbs use, ranging from 29% to 76%. In Malaysia, the prevalence of herbs use was 40.5% and 50.0% among diabetic patients and hypertensive patients respectively (Ching et al., 2013a; Baharom et al., 2016). Baharom et al. (2016) found that 66.7% females used herbs compared to males (33.3%). In Malaysia, Malays (55.7%) were the predominant race to use herbs. Likewise, Ching et al. (2013a) reported that being female (61.6%) and Malay (63.9%) were correlated with the use of herbs. The patients consumed bitter gourd and cat's whiskers to cure their underlying medical condition.

In Sri Lanka, 76% of diabetic patients was using herbs. In the study, females were the significant users, $p < 0.01$. The patients used bitter gourd (50.8%), ivy gourd (44.8%), crepe ginger (36.5%), and fenugreek (2%) to cure diabetes. Crepe ginger was associated with hypoglycaemic condition (P=0.039) (Medagama et al., 2014). In Myanmar, Peltzer et al. (2016a) reported a prevalence of 53.2%. The use of herbs was significant among patients from the rural area, with no educational background, and diagnosed with co-morbidities.

In Turkey, Mollaoglu and Aciyurt (2013) revealed 55.9% of herbs use among patients with chronic diseases, particularly, hypertension (63.8%). In another study, conducted by Tulunay et al. (2015) revealed 29% of herbal medicine use. The most commonly consumed herbs by hypertensive, diabetic, and hyperlipidaemic patients were lemon (39.6%), cinnamon (12.7%), and walnut (6.3%) respectively (Tulunay et al., 2015). Females were significantly associated with herbs use in both studies, p-value of 0.011 and 0.040 respectively. The previous study revealed the determinants of herbs use as higher education (p=0.013), marital status (0.022), and housewives (0.008) (Mollaoglu and Aciyurt, 2013).

CHAPTER 3

METHODOLOGY

3.1 Overview of the experimental procedure

The study comprised of two phases. The first phase involved a cross-sectional survey based on a self-administered questionnaire in Klinik Kesihatan Kampar. The second phase involved phytochemical screening and thin layer chromatography of the selected crude herbs consumed by the patients with chronic diseases from Klinik Kesihatan Kampar.

3.2 Cross-sectional survey

The cross-sectional survey was conducted in Klinik Kesihatan Kampar from October 2018 to December 2018. The target group involved only patients who had been diagnosed with chronic diseases. The collected data were analysed using the Statistical Package for the Social Sciences (SPSS) version 21.

3.2.1 Study site

The study was conducted in Klinik Kesihatan Kampar, located in Kampar district of Kinta valley in the state of Perak, Malaysia. Kampar is about 43 kilometres (km) away from Ipoh, the capital city of Perak. Kampar district was notable for its tin mining reserves back in the 19th century (Pejabat Daerah dan Tanah Kampar, 2018). Kampar is divided into two small towns, known as the

"old" town and the "new" town. The "old" town consists of banks, a local market, a train station, and a bus station. The "new" town has two tertiary education centres, Universiti Tunku Abdul Rahman and Tunku Abdul Rahman College.

According to the population statistics, there are about 107, 000 population with an equal number of male and female. About 73, 900 of the population fall within the age group of 15 to 64 and approximately 12, 900 of the population are 65 years old and above (Department of Statistics, 2018). The percentage of distribution according to races is as follows: Malays (33%), Chinese (52%) and Indians (11%) with Chinese being the majority of the population (Pejabat Daerah dan Tanah Kampar, 2018).

Health facilities in Kampar have been built to ensure proper treatment and care are given to the patients. In total, there are one district hospital, a Satu Malaysia clinic, a primary care clinic, and three community clinics, namely, Klinik Desa Bukit Pekan, Klinik Desa Jeram and Klinik Desa Mambang Di Awan (Ministry of Health, 2018).

This study was conducted in Klinik Kesihatan Kampar, a primary care clinic located at Jalan Degong, Kampar (Ministry of Health, 2018), precisely at the latitude of 4.310422 and longitude of 101.153348. The location and image of

Klinik Kesihatan Kampar are illustrated in **Figure 3.1** and **Figure 3.2**. Out of all these health facilities, Klinik Kesihatan Kampar has been chosen as our study site since it caters patients with different diseases, particularly, chronic diseases that are the main focus of our study. The patients who visit Klinik Kesihatan Kampar are those perceived to be with lower socio-economic status, attending the clinic from sub-rural to rural areas. In addition, Klinik Kesihatan Kampar was also chosen because it is within the reach of the town centre.

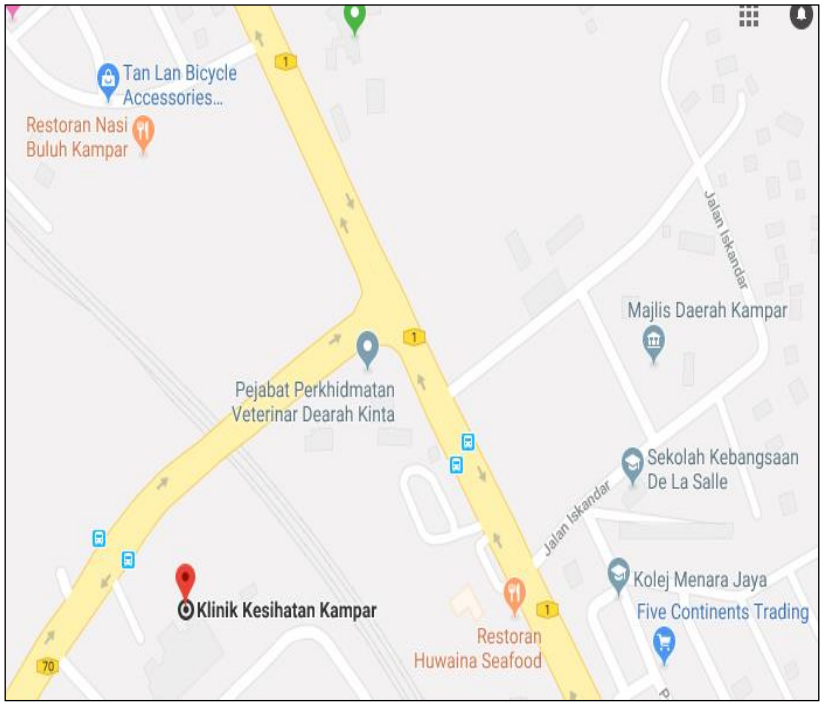

Figure 3.1: The location of Klinik Kesihatan Kampar (Obtained from Google maps, 2018).

Figure 3.2: The image of Klinik Kesihatan Kampar (Obtained from Google images, 2018).

3.2.2 Study design

In this study, a cross-sectional based study design was used to investigate the prevalence of crude herbs use among patient with chronic diseases attending Klinik Kesihatan Kampar. The cross-sectional study design enabled measurement of exposure and outcomes to certain risks or diseases computed simultaneously and at a single point of time (Sedgwick, 2014). Follow up session with the participants was not required and the participants were chosen based on the specification needed for the present study (Setia, 2016).

Cross-sectional study is beneficial because it is economical and rapid. It is a constructive study to be conducted prior to cohort studies. However, cross-sectional cannot be used to study the cause and effect relationship due to one-time data collection (Sedgwick, 2014). In addition, the differences between

partakers and non-partakers may interpret the outcomes as a non-response bias, and the sample may not represent the entire population (Pandis, 2014).

3.2.3 Target population

The population intended for this study was: (1) adults that are aged 18 and above (2) diagnosed with chronic diseases, such as, diabetes, hypertension, and cardiovascular diseases (3) attending official appointment at Klinik Kesihatan Kampar (4) willing to provide informed consent to take part in the study (5) no physical or mental deformity (6) able to speak in either Malay or English language.

3.2.4 Data sampling and sample size

In this study, the data were collected by using convenience sampling. The participants were approached while they were waiting to meet doctors. The patients were included in the study after informed consent was obtained (Sedgwick, 2013). Convenience sampling method was employed as this study is still in its preliminary stage. This preliminary study is significant in providing enough information to allow future studies with well-designed models to be conducted.

In addition, time constraints and financial limitation were also other factors that contributed to this decision. Although biasness is a limitation in this type of

sampling, this sampling would not be much of hassle compared to random sampling. Convenience sampling does not involve reaching back the non-respondents to ensure an adequate number of samples attained to represent the entire population (Elsayir, 2014). The sample size was calculated based on the Cochrane formula and obtained as 242 based on a prevalence of 19.6% from a previous study conducted in Penang (Omar and Jazouly, 2015). The sample size was divided among my project mate and I, thus each of us interviewed 121 patients. A dropout rate of 24.8% was used to calculate the actual sample size.

$$n_0 = \frac{Z^2 pq}{e^2}$$

n_0 = sample size

Z = critical value of desired confidence interval

p = prevalence from previous study (19.6%)

q = 1-p

e = precision level of interest

The calculation was done as shown below:

$$n_0 = \frac{(1.96)^2(0.196)(0.804)}{(0.05)^2}$$

$$n_0 = \frac{0.6054}{0.0025}$$

$$n_0 = \frac{242}{2}$$

n_0 = 121 patients

$$\text{Actual sample size} = \frac{\text{Sample size x } 100\%}{100\% - \text{dropout rate}}$$

$$\text{Actual sample size} = \frac{121 \text{ x } 100\%}{100\% - 24.8\%}$$

$$\text{Actual sample size} = 161 \text{ patients}$$

3.2.5 Ethical considerations

Ethical approval to conduct this study were obtained from Universiti Tunku Abdul Rahman (U/SERC/84/2017), Ethics Committee of National Malaysia Research Registry (NMRR-17-2591-38273 (IIR)) and Pejabat Kesihatan Daerah Kampar. The ethical approvals are as shown in **Appendices A**, **B**, and **C** respectively. Only eligible patients that gave informed consent were included in the study.

3.2.6 Data collection tool

The data for this research were collected using a self-designed questionnaire. The questionnaire shown in **Appendix D** was developed based on wide literature searches involving herbal medicine or CAM use among patients with chronic diseases in Malaysia and neighbouring countries. The questionnaires were developed in two languages, Bahasa Malaysia and English. The entire questionnaire consists of three sections: (Part A) sociodemographic characteristics such as age, gender, ethnicity, monthly income, education level, and status of employment (Part B) medical history (Part C) present use of any

type of crude herbs, part of plant used, preparation method, purpose, and duration of consumption as well as source of information regarding crude herbs.

3.2.7 Data analysis

The collected data were analysed using the SPSS version 21 software. Descriptive analysis was used to describe the categorical variables in a systemic data. Descriptive analysis is mostly used in medical field to study a particular population (Kaur et al., 2018). Chi-square test was performed to test the association between two variables and to compare the observed data with the expected data (Rana and Singhal, 2015).

3.3 Laboratory analysis

Based on preliminary analysis done by the SPSS software, the selected crude herbs were subjected to laboratory analysis. The laboratory analysis consists of sample collection and preparation, plant extraction based on aqueous and solvent method, phytochemical screening, and thin layer chromatography. The chemicals and solvents used to conduct the analysis are as shown in **Appendix E**. The equipments used is listed in **Appendix F**. The details of each process are discussed in the subsections below.

3.3.1 Sample collection and preparation

The types of herbs used by the participants of this study were chosen based on preliminary analysis done by the SPSS software. The selected plants were *Psidium guajava* and *Coriandrum sativum*. The purchased plant samples from local markets were washed with running tap water and immediately air dried at room temperature. The amount used for the aqueous and solvent extraction was weighed respectively.

3.3.2 Plant extraction

Two types of extraction were performed on the plants, namely, aqueous extraction and solvent extraction via freeze-drying and rotary evaporation process respectively. At the end of each process, powdered extracts and gel-like extracts were collected.

3.3.2.1 Aqueous extraction

The plant samples were cut into small pieces. The cut plant samples were placed in a beaker and added with distilled water with the ratio of 1:2. The plant samples were heated for approximately 30 minutes at 37°C on a hot plate. The first filtrate was filtered and allowed to cool down in a conical flask. The remaining plant sample in the beaker was added with distilled water with the ratio of 1:2 and the heating procedure was repeated for second time and followed by third time. The three filtrates obtained were mixed together in a beaker and stored in a 500 mL conical flask inside a chiller at a temperature of 4°C (Rao et al., 2016).

The mixture was then transferred into a round bottom flask up to one quarter level and freezed in a freezer at -20°C prior to freeze drying. The mouth of the round bottom flask was covered with a parafilm to prevent sample loss. Some holes were poked on the parafilm to allow escape of vapour. Next morning, the frozen sample was fixed to a freeze dryer.

To operate the freeze dryer, the drain valve was switched on to release the residual water from the condenser chamber. Once the water was released, the drain valve was switched off. This is to ensure the ice formed inside the condenser chamber was removed to enable efficient removal of moisture from the samples. Then, the machine was set to warm up by switching on the vacuum pump and refrigeration compressor for duration of 30 minutes. The gas ballast valve was turned open to allow oozing of the air and the condenser vapour through the exhaust pipe. The frozen samples were fitted to the sample valve using rubber adapters (Gaidhani et al., 2015).

To start the freeze drying process, the main drying option was chosen on the system. The aqueous samples were subjected to freeze drying for about 6 to 8 hours everyday. The samples were removed by slowly twisting the sample valve to release the vacuum. The gas ballast valve was turned close and the vacuum pump was switched off prior to 30 minutes of machine warm down. The main power system was switched off after that (Gaidhani et al., 2015). The aqueous samples were subjected to freeze drying until they become powdered or

crystallised. The dried extracts were collected and kept into a sample vial (weighed) for the phytochemical screening. The sample with the extracts collected were weighed and subtracted with the sample vial to get pure weight of the extracts. The percentage yield of the sample was calculated using the formula shown as following:

$$\text{Percentage of yield} = \frac{\text{Dry weight (g)}}{\text{Total weight (g)}} \times 100\%$$

Freeze drying consists of two main processes, which are the primary drying and the secondary drying. In the primary drying, sublimation takes place whereby the solid state (frozen sample) changes directly to vapour state without involving liquid state under pressure and temperature (-20°C to -30°C) that is lower than the critical point. The frozen sample containing liquid sublimes, releasing the water molecules as vapour leaving behind the dried sample (Nireesha et al., 2013; Gaidhani et al., 2015). In the secondary drying, which is desorption state, the unfrozen water molecules are removed. This is a vital stage to preserve the desired biological component for long-term storage. In this stage, the temperature is raised to ensure any physicochemical bonds between the water molecules and the frozen molecules are broken (Trelea et al., 2016).

Freeze drying method was chosen to dry the aqueous samples instead of other methods because it stabilises and preserves most of the bioactive compounds

(Kushwaha, 2012). Even though freeze drying takes a longer time to dry the samples, but lesser water is retained compared to microwave drying, thus producing samples which are more powdered. Freeze dried samples is denser compared to samples dried by microwave means (Annie et al., 2017).

3.3.2.2 Solvent extraction

The plant samples were cut and dried in an oven at 37°C for 1 to 2 days. The dried plant sample was crushed into smaller pieces using an electronic grinder. The crushed plant sample was placed into a 250 mL conical flask. Ethanol with a concentration of 95% was added to the conical flask with the ratio of 1:2. The conical flask was sealed with aluminium foil and parafilm to prevent any leakage during the shaking process. The conical flask filled with plant sample in ethanol was placed on a shaker at 100 rpm for 5 days. After 5 days, the sample was filtered using cotton wool. The filtrate obtained was kept in a conical flask and stored in a chiller. The remaining plant residue was added with 95% ethanol and installed on the shaker again for 3 days as the second filtration. The procedure was repeated again but was shaken for only a day for the third filtration. All the filtrates obtained were mixed and kept in a conical flask and stored in a chiller (Alebiosu and Yusuf, 2015).

The extracts were transferred at a one-quarter level of a round bottom flask. The sample was subjected to rotary evaporation in order to obtain gel extract. To operate the rotary evaporator, the chiller was switched on for 20 minutes. The

water bath was set to 40°C. The collecting flask was fixed to the condenser outlet whereas the evaporating flask with the sample was fixed to the rotary evaporator via Keck clip. The aspirator vacuum was switched on after that and the stopcock on the condenser was closed. The flask was lowered down into the water bath. Once the solvent inside the evaporating flask has evaporated, leaving behind the plant extracts, the stop button was pressed and the vacuum pump was switched off. The stopcock was then opened and the evaporating flask was raised from the water bath using the adjustment lever. The evaporating flask was removed and the gel extract was measured in order to obtain the dry weight and calculate the percentage yield of the sample using the formula as shown below. Any remaining gel residues in the flask was rinsed off using 95% ethanol and transferred into a sample vial. The gel extract was placed into a sample vial and stored in the oven at 37°C for further screening of phytochemicals and thin layer chromatography (Chemistry LibreTexts Library, 2017).

$$\text{Percentage of yield} = \frac{\text{Dry weight (g)}}{\text{Total weight (g)}} \times 100\%$$

3.3.3 Phytochemical screening

One gram of plant extract was added to a 15 mL centrifuge tube and the solvent extract was added with 95% ethanol whereas the aqueous extract was added with distilled water. The extracts were then sonicated using a sonicator to ensure all the plant compounds and the mediums (ethanol and distilled water) were mixed thoroughly.

3.3.3.1 Test for phenols

About 2 mL of solvent extract and aqueous extract each was added into two test tubes respectively. Few drops of 5% aqueous ferric chloride were added into each tube. Formation of deep blue or black colour indicated the presence of phenols in the plant samples (Ugochukwu et al., 2013).

3.3.3.2 Test for quinones

Few drops of concentrated (1 M) hydrochloric acid (HCI) were added into the test tubes containing 2 mL of plant extracts for both aqueous and solvent extraction respectively. Presence of quinones in plant samples was determined by the formation of yellow precipitate or colouration (Ugochukwu et al., 2013).

3.3.3.3 Test for tannins (Braymer's test)

Two millilitres of the both aqueous and ethanol extracts were treated with 1% gelatin followed by 10% sodium chloride (NaCl). The formation of a white precipitate was observed to determine the presence of tannins in the plant extracts (Dhawan and Gupta, 2017).

3.3.3.4 Test for terpenoids (Salkowki's test)

Few drops of concentrated 1 M sulphuric acid (H_2SO_4) were added into the test tubes containing aqueous and solvent extracts respectively. Presence of

terpenoids in the extracts was determined by the formation of reddish-brown precipitate (Ugochukwu et al., 2013).

3.3.3.5 Test for saponins (Foam test)

Two millilitres of the respective aqueous and solvent extracts were added with 12 mL of distilled water and shaken vigorously until persistent foam was observed on top of the mixture (Dhawan and Gupta, 2017).

3.3.3.6 Test for flavonoids (Alkaline reagent test)

Few drops of 10% sodium hydroxide (NaOH) solution were added into 2 mL of both aqueous and solvent extracts respectively. Then, 10% HCl was added to both of the extracts. The presence of flavonoids was indicated by the change of colour from yellow to colourless (Ugochukwu et al., 2013).

3.3.3.7 Test for glycosides

Bromine water was added to 2 mL of both aqueous and solvent extracts. Formation of yellow precipitate was observed to indicate the presence of glycosides (Ugochukwu et al., 2013).

3.3.3.8 Test for alkaloids

Two millilitres of Wagner's reagent was added to 2 mL of aqueous and solvent extracts. The mixtures were observed for the formation of either reddish-brown precipitate or brown coloured solution indicating the presence of alkaloids (Ugochukwu et al., 2013).

3.3.4 Thin layer chromatography (TLC) analysis

One gram of solvent extract was weighed and placed into a 15 mL centrifuge tube. Three millilitres of 95% ethanol was added to the gel solvent extract and the centrifuge tube was sonicated to dissolve the extract. If the dissolved extract is too thick, it was added with more 95% ethanol to dilute the extract in order to get a distinct separation of spots. Silica-coated aluminium sheet thin layer chromatography (TLC) plates were cut into the size of 5 cm x 10 cm. The origin for each plate was marked about 1 cm from the bottom of the plate and 1 cm from the top of the plate was marked as the solvent front using a pencil with a blunt end. The plates were labelled with the name of the extracts and the type of solvent systems to avoid misinterpretation (Karthika et al., 2014).

After labelling, about 3 µL of guava and coriander solvent extracts were loaded onto each TLC plates respectively using the 10 µL micropipette. The TLC plates were then allowed to air dry for approximately three to five minutes in the fume hood. The plates were immersed in developing chambers containing the mixture of hexane: acetone with the ratio of 70:30 respectively.

The plant extracts were also tested with different solvent systems such as 70:30 and 60:40 of hexane: ethyl acetate as well as 70:30 and 60:40 of toluene: ethyl acetate to determine which one gives the best separation of spots. The developing chamber was closed to allow the saturation of chamber with solvent vapour. The solvent front was allowed to rise about 1 cm from the top of the plate (Karthika et al., 2014). The TLC plates were removed and the solvent front was marked with a blunt pencil in the fume hood before the solvent dries off. After the solvent dries off, the plates were viewed under the (ultraviolet) UV light with a short wavelength (254 nm). The spots seen were marked with a pencil. Then, the TLC plates were kept into an iodine vapour chamber for approximately 15 minutes and the spots observed were traced with the pencil (Karthika et al., 2014).

TLC works based on the adsorption between the stationary phase and the mobile phase. A solid adsorbent material coated with glass plates is used as the stationary phase (Coskun, 2016). There are various types of adsorbents such as silica, alumina and cellulose but in this study, silica was chosen as the stationary phase. The movement of the mobile phase through the plate is due to capillary action that separates and appears as spots on the plate depending on the polarity of the compounds being tested. Spots that appear closer to the origin indicate that the compounds have moved lesser and possess higher polarity whereas compounds that are less polar move farther from the origin (Musa et al., 2017). TLC was chosen to separate the compounds present in the plant samples because it is both cost and labour saving method (Altemimi et al., 2017).

CHAPTER 4

RESULTS

4.1 Cross-sectional survey

The details pertaining to the cross-sectional survey such as socio-demographic characteristics, medical history and crude herbs use of the patients with chronic diseases in this study are discussed in the subsequent sections. The association between crude herbs use and socio-demographic characteristics and medical history of the patients are also discussed respectively.

4.1.1 Socio-demographic characteristics

A total of 121 out of 161 patients approached took part in this study and thus the response rate was 75.2%. The study population involved more females (60.3%) compared to males (39.7%). Crude herbs were used by 44 patients (36.4%). The socio-demographic characteristics of study population are tabulated in **Table 4.1** for users and non-users of crude herbs respectively. Majority of the patients who were consuming crude herbs belong to the age group between 60 to 69 years old (45.5%) whereas the least crude herbs users were from the age group of 80 years and above (4.5%). Higher number of female patients (72.7%) consumed crude herbs compared to male patients (27.3%). The use of crude herbs were high among Indian patients (56.8%), followed by Malay patients (38.6%) and Chinese patients (4.5%). All the crude herbs users were found to have a minimum wage of RM 3000 and below monthly. Patients with secondary school

education (52.3%) and housewives or unemployed patients (63.6%) were more likely to use crude herbs to treat their chronic medical conditions.

Table 4.1: Socio-demographic characteristics of the crude herbs users and non-users attending Klinik Kesihatan Kampar.

Socio-demographic characteristics	Crude herbs users		Crude herbs non-users		Total
	F	%	F	%	
Age Groups (Years)					
49 and below	4	9.1	8	10.4	12
Between 50 to 59	12	27.3	23	29.9	35
Between 60 to 69	20	45.5	28	36.4	48
Between 70 to 79	6	13.6	15	19.5	21
80 and above	2	4.5	3	3.9	5
Gender					
Male	12	27.3	36	46.8	48
Female	32	72.7	41	53.2	73
Race					
Malay	17	38.6	35	45.5	52
Chinese	2	4.5	13	16.9	15
Indian	25	56.8	27	35.1	52
Others	0	0.0	2	2.6	2

Continued Table 4.1: Socio-demographic characteristics of the crude herbs users and non-users attending Klinik Kesihatan Kampar.

Socio-demographic characteristics	Crude herbs users		Crude herbs non-users		Total
	F	%	F	%	
Salary range					
Below RM 3000	44	100	72	93.5	116
Between RM 3000 to RM 6000	0	0.0	5	6.5	5
Above RM 6000	0	0.0	0	0.0	0
Educational level					
No formal education	7	15.9	13	16.9	20
Primary education	13	29.5	27	35.1	40
Secondary education	23	52.3	37	48.1	60
Tertiary education	1	2.3	0	0.0	1
Employment status					
Housewife or unemployed	28	63.6	33	42.9	61
Employed as general staffs	9	20.5	24	31.2	33
Intellectual professionals	0	0.0	1	1.3	1
Retired	7	15.9	19	24.7	26

*F = Frequency of socio-demographics characteristics of study population
% = Percentage of socio-demographics characteristics of study population

4.1.2 Medical history information

Table 4.2 depicts the chronic diseases suffered by both crude herbs users and non-users of crude herbs. Most of the hypertensive patients (75%) were more likely to use crude herbs followed by patients with diabetes mellitus (72.7%) and hyperlipidaemia (22.7%).

Table 4.2: Medical history information of the crude herbs users and non-users attending Klinik Kesihatan Kampar.

Medical conditions	Crude herbs users		Crude herbs non-users		Total
	F	%	F	%	
Asthma	2	4.5	6	7.8	8
Cancer	0	0.0	0	0.0	0
Cardiovascular disease	1	2.3	5	6.5	6
Diabetes mellitus	32	72.7	43	55.8	75
Hyperlipidaemia	10	22.7	14	18.2	24
Hypertension	33	75.0	44	57.1	77
Hyperuricaemia	1	2.3	0	0.0	1
Kidney disease	0	0.0	0	0.0	0
Leukaemia	0	0.0	0	0.0	0
Hepatitis, jaundice, liver disease	0	0.0	0	0.0	0
Migraine or recurrent headaches	0	0.0	0	0.0	0
Muscle pain	0	0.0	0	0.0	0

Continued Table 4.2: Medical history information of the crude herbs users and non-users attending Klinik Kesihatan Kampar.

Medical conditions	Crude herbs users		Crude herbs non-users		Total
	F	%	F	%	
Obesity	0	0.0	0	0.0	0
Parkinson's disease	0	0.0	0	0.0	0
Peptic ulcer	0	0.0	0	0.0	0
Stroke	1	2.3	0	0.0	1
Thyroid disease	0	0.0	0	0.0	0
Urinary infection	0	0.0	1	1.3	1
Falls or injury in the past 6 to 12 months	0	0.0	0	0.0	0
Osteoporosis	0	0.0	0	0.0	0
Others	0	0.0	1	1.3	1

*F = Frequency of medical conditions suffered by study population
% = Percentage of medical conditions suffered by study population

4.1.3 Prevalence of crude herbs use

Table 4.3 shows prevalence rate of crude herbs used among patients with chronic diseases. In this study, a prevalence of 36.4% of crude herbs use among the patients was recorded. About 77 (63.6%) patients were not using any crude herbs to treat their chronic diseases.

Table 4.3: The prevalence of crude herbs use among the patients with chronic diseases attending Klinik Kesihatan Kampar.

Groups	Prevalence	
	F	%
Crude herbs users	44	36.4
Crude herbs non-users	77	63.6

*F = Frequency of crude herbs use by study population
% = Percentage of crude herbs use by study population

4.1.4 Types of crude herbs

Table 4.4 depicts the the types of crude herbs used by the patients with chronic diseases in this study. The most commonly used crude herbs were *Trigonella foenum-graecum* (9.9%) followed by *Cuminum cyminum* (5.8%) and *Phaleria marcocarpa* (5.0%). Other herbs that were also used by the patients were *Citrus limon* (4.1%), *Psidium guajava* (4.1%), *Centella asiatica* (3.3%) and *Coriandrum sativum* (3.3%).

Table 4.4: The various types of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Types of crude herbs	F	%
Trigonella foenum-graecum	12	9.9
Cuminum cyminum	7	5.8
Phaleria macrocarpa	6	5.0
Psidium guajava	5	4.1
Citrus limon	5	4.1
Coriandrum sativum	4	3.3
Centella asiatica	4	3.3
Momordica charantia	3	2.5
Moringa oleifera	3	2.5
Nigella sativa	3	2.5
Orthosiphon aristatus	3	2.5
Acanthus ilicifolius	2	1.7
Pandanus amaryllifolius	2	1.7
Zingiber officinale	2	1.7
Allium sativum	1	0.8
Cardiospermum halicacabum	1	0.8
Carica papaya	1	0.8
Catharanthus roseus	1	0.8
Cinnamomum verum	1	0.8
Cosmos caudatus	1	0.8
Cuminum cyminum + Nigella sativa	1	0.8
Cuminum cyminum + Piper betle	1	0.8

Continued Table 4.4: The various types of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Types of crude herbs	F	%
Cuminum cyminum + Piper nigrum + Zingiber officinale	1	0.8
Cuminum cyminum + Prunus avium + Piper betle	1	0.8
Malus pumila + Apium graveolens	1	0.8
Orchis mascula	1	0.8
Pandanus conoideus	1	0.8
Senna auriculata	1	0.8
Solanum procumbens	1	0.8
Swietenia macrophylla	1	0.8
Trachyspermum ammi	1	0.8
Trigonella foenum-graecum + Cuminum cyminum	1	0.8
Ziziphus mauritiana	1	0.8

*F = Frequency of type of crude herbs used by study population
% = Percentage of type of crude herbs used by study population

4.1.5 Parts of crude herbs used

Table 4.5 depicts the parts of plants used for each type of crude herbs consumed by patients with chronic diseases. The parts of plants consumed were leaves (24.7%) followed by seeds (23.8%) and fruits (16.5%). The less commonly used parts of plants were shoot (5.8%), flowers (2.4%) and bark (0.8%).

Table 4.5: The parts of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Parts of crude herbs used											
	Leaves		Seed		Shoot		Flower		Fruit		Bark	
	F	%	F	%	F	%	F	%	F	%	F	%
Acanthus ilicifolius	2	1.7	-	-	-	-	-	-	-	-	-	-
Allium sativum	-	-	-	-	-	-	-	-	1	0.8	-	-
Cardiospermum halicacabum	1	0.8	-	-	-	-	-	-	-	-	-	-
Carica papaya	-	-	-	-	1	0.8	-	-	-	-	-	-
Catharanthus roseus	-	-	-	-	-	-	1	0.8	-	-	-	-
Centella asiatica	4	3.3	-	-	-	-	-	-	-	-	-	-
Cinnamomum verum	-	-	-	-	-	-	-	-	-	-	1	0.8
Citrus limon	-	-	-	-	-	-	-	-	4	3.3	-	-
Coriandrum sativum	4	3.3	-	-	3	2.5	-	-	-	-	-	-
Cosmos caudatus	1	0.8	-	-	1	0.8	-	-	-	-	-	-
Cuminum cyminum	-	-	7	5.8	-	-	-	-	-	-	-	-

Continued Table 4.5: The parts of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Parts of crude herbs used											
	Leaves		Seed		Shoot		Flower		Fruit		Bark	
	F	%	F	%	F	%	F	%	F	%	F	%
Cuminum cyminum + Nigella sativa	-	-	1	0.8	-	-	-	-	-	-	-	-
Cuminum cyminum + Piper betle	1	0.8	1	0.8	-	-	-	-	-	-	-	-
Cuminum cyminum + Piper nigrum + Zingiber officinale	-	-	1	0.8	-	-	-	-	1	0.8	-	-
Cuminum cyminum + Prunus avium + Piper betle	1	0.8	1	0.8	-	-	-	-	-	-	-	-
Malus pumila + Apium graveolens	1	0.8	-	-	-	-	-	-	1	0.8	-	-
Momordica charantia	-	-	-	-	-	-	-	-	3	2.5	-	-
Moringa oleifera	3	2.5	1	0.8	2	1.7	-	-	-	-	-	-
Nigella sativa	-	-	3	2.5	-	-	-	-	-	-	-	-
Orchis mascula	-	-	-	-	-	-	1	0.8	-	-	-	-

Continued Table 4.5: The parts of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Parts of crude herbs used											
	Leaves		Seed		Shoot		Flower		Fruit		Bark	
	F	%	F	%	F	%	F	%	F	%	F	%
Orthosiphon aristatus	3	2.5	-	-	-	-	-	-	-	-	-	-
Pandanus amaryllifolius	2	1.7	-	-	-	-	-	-	-	-	-	-
Pandanus conoideus	-	-	-	-	-	-	-	-	1	0.8	-	-
Phaleria macrocarpa	-	-	-	-	-	-	-	-	6	5.0	-	-
Psidium guajava	5	4.1	-	-	-	-	-	-	-	-	-	-
Senna auriculata	-	-	-	-	-	-	1	0.8	-	-	-	-
Solanum procumbens	1	0.8	-	-	-	-	-	-	-	-	-	-
Swietenia macrophylla	-	-	-	-	-	-	-	-	1	0.8	-	-
Trachyspermum ammi	-	-	1	0.8	-	-	-	-	-	-	-	-
Trigonella foenum-graecum	-	-	12	9.9	-	-	-	-	-	-	-	-

Continued Table 4.5: The parts of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Parts of crude herbs used											
	Leaves		Seed		Shoot		Flower		Fruit		Bark	
	F	%	F	%	F	%	F	%	F	%	F	%
Trigonella foenum-graecum + Cuminum cyminum	-	-	1	0.8	-	-	-	-	-	-	-	-
Zingiber officinale	-	-	-	-	-	-	-	-	2	1.7	-	-
Ziziphus mauritiana	1	0.8	-	-	-	-	-	-	-	-	-	-
Total	**30**	**24.7**	**29**	**23.8**	**7**	**5.8**	**3**	**2.4**	**20**	**16.5**	**1**	**0.8**

*F = Frequency of parts of crude herbs used by study population
% = Percentage of parts of crude herbs used by study population

4.1.6 Preparation methods of crude herbs

Table 4.6 depicts the methods used by the patients to prepare the crude herbs for consumption. The most commonly used method to prepare the crude herbs were, boiling (35.7%) and soaking (12.2%). Patients also prepared the crude herbs by blending (4.2%), eat raw crude herbs (4.2%), and roasting (0.7%).

Table 4.6: The preparation method of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Methods of preparation											
	Boiling		Raw		Blend		Decoction		Soaking		Roasting	
	F	%	F	%	F	%	F	%	F	%	F	%
Acanthus ilicifolius	2	1.4	-	-	-	-	-	-	-	-	-	-
Allium sativum	-	-	-	-	-	-	-	-	-	-	1	0.7
Cardiospermum halicacabum	1	0.7	-	-	-	-	-	-	-	-	-	-
Carica papaya	1	0.7	-	-	-	-	-	-	-	-	-	-
Catharanthus roseus	1	0.7	-	-	-	-	-	-	-	-	-	-
Centella asiatica	3	2.2	1	0.7	-	-	-	-	-	-	-	-
Cinnamomum verum	1	0.7	-	-	-	-	-	-	-	-	-	-
Citrus limon	-	-	-	-	-	-	-	-	4	2.9	-	-
Coriandrum sativum	4	2.9	-	-	2	1.4	-	-	-	-	-	-
Cosmos caudatus	-	-	1	0.7	-	-	-	-	-	-	-	-
Cuminum cyminum	6	4.3	-	-	-	-	-	-	1	0.7	-	-

Continued Table 4.6: The preparation method of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Methods of preparation											
	Boiling		Raw		Blend		Decoction		Soaking		Roasting	
	F	%	F	%	F	%	F	%	F	%	F	%
Cuminum cyminum + Nigella sativa	1	0.7	-	-	-	-	-	-	-	-	-	-
Cuminum cyminum + Piper betle	1	0.7	-	-	-	-	-	-	-	-	-	-
Cuminum cyminum + Piper nigrum + Zingiber officinale	-	-	-	-	1	0.7	-	-	-	-	-	-
Cuminum cyminum + Prunus avium + Piper betle	1	0.7	-	-	-	-	-	-	-	-	-	-
Malus pumila + Apium graveolens	-	-	-	-	1	0.7	-	-	-	-	-	-
Momordica charantia	1	0.7	-	-	1	0.7	1	0.7	-	-	-	-
Moringa oleifera	2	1.4	1	0.7	-	-	-	-	-	-	-	-
Nigella sativa	3	2.2	-	-	-	-	-	-	-	-	-	-
Orchis mascula	1	0.7	-	-	-	-	-	-	-	-	-	-

Continued Table 4.6: The preparation method of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Methods of preparation											
	Boiling		Raw		Blend		Decoction		Soaking		Roasting	
	F	%	F	%	F	%	F	%	F	%	F	%
Orthosiphon aristatus	3	2.2	-	-	-	-	-	-	-	-	-	-
Pandanus amaryllifolius	2	1.4	-	-	-	-	-	-	-	-	-	-
Pandanus conoideus	1	0.7	-	-	-	-	-	-	-	-	-	-
Phaleria macrocarpa	6	4.3	-	-	-	-	-	-	-	-	-	-
Psidium guajava	5	3.6	-	-	-	-	-	-	-	-	-	-
Senna auriculata	1	0.7	-	-	-	-	-	-	-	-	-	-
Solanum procumbens	-	-	-	-	1	0.7	-	-	-	-	-	-
Swietenia macrophylla	-	-	1	0.7	-	-	-	-	-	-	-	-
Trachyspermum ammi	-	-	1	0.7	-	-	-	-	-	-	-	-
Trigonella foenum-graecum	1	0.7	-	-	-	-	-	-	11	7.9	-	-

Continued Table 4.6: The preparation method of crude herbs used by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Name of crude herbs	Methods of preparation											
	Boiling		Raw		Blend		Decoction		Soaking		Roasting	
	F	%	F	%	F	%	F	%	F	%	F	%
Trigonella foenum-graecum* + *Cuminum cyminum	1	0.7	-	-	-	-	-	-	-	-	-	-
Zingiber officinale	1	0.7	-	-	-	-	-	-	1	0.7	-	-
Ziziphus mauritiana	-	-	1	0.7	-	-	-	-	-	-	-	-
Total	**50**	**35.7**	**6**	**4.2**	**6**	**4.2**	**1**	**0.7**	**17**	**12.2**	**1**	**0.7**

*F = Frequency of preparation method of crude herbs used by study population
% = Percentage of preparation method of crude herbs used by study population

4.1.7 Perceived medicinal values of crude herbs

Table 4.7 depicts the medicinal benefits of the crude herbs as perceived by the patients with chronic diseases in this study. The data shows that majority of the patients consumed *Trigonella foenum-graecum* (9.1%), *Cuminum cyminum* (5.0%), *Psidium guajava* (4.1%), *Centella asiatica* (3.3%), and *Phaleria marcocarpa* (3.3%) to reduce their blood glucose level (42.1%). Crude herbs such as *Phaleria marcocarpa* (5.1%) and *Trigonella foenum-graecum* (3.3%) were commonly used to reduce blood pressure level (24.0%). To reduce blood cholesterol level (5.0%), herbs such as *Coriandrum sativum* (1.7%), *Cuminum cyminum* (0.8%) and *Orthosiphon aristatus* (0.8%) were consumed by the patients. Other crude herbs that were consumed by the patients include *Centella asiatica* (0.8%), *Cardiospermum halicacabum* (0.8%) and *Solanum procumbens* (0.8%) for overall health maintenance (3.2%), to reduce wheezing (1.6%) and to treat gout disease (0.8%) respectively.

Table 4.7: The medicinal values of crude herbs as perceived by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Crude herbs	Perceived medicinal values of crude herbs											
	Reduce blood glucose level		Reduce blood pressure level		Reduce blood cholesterol level		Health maintenance		Treat gout		Reduce wheezing	
	F	%	F	%	F	%	F	%	F	%	F	%
Acanthus ilicifolius	-	-	2	1.7	-	-	-	-	-	-	-	-
Allium sativum	1	0.8	-	-	-	-	-	-	-	-	-	-
Cardiospermum halicacabum	-	-	-	-	-	-	-	-	1	0.8	-	-
Carica papaya	-	-	1	0.8	-	-	-	-	-	-	-	-
Catharanthus roseus	1	0.8	-	-	-	-	-	-	-	-	-	-
Centella asiatica	4	3.3	-	-	-	-	1	0.8	-	-	-	-
Cinnamomum verum	-	-	1	0.8	-	-	-	-	-	-	-	-
Citrus limon	2	1.7	2	1.7	-	-	-	-	-	-	-	-
Coriandrum sativum	-	-	2	1.7	2	1.7	-	-	-	-	-	-
Cosmos caudatus	-	-	-	-	-	-	1	0.8	-	-	-	-

Continued Table 4.7: The medicinal values of crude herbs as perceived by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Crude herbs	Perceived medicinal values of crude herbs											
	Reduce blood glucose level		Reduce blood pressure level		Reduce blood cholesterol level		Health maintenance		Treat gout		Reduce wheezing	
	F	%	F	%	F	%	F	%	F	%	F	%
Cuminum cyminum	6	5.0	-	-	2	1.7	-	-	-	-	-	-
Cuminum cyminum + Nigella sativa	1	0.8	-	-	-	-	-	-	-	-	-	-
Cuminum cyminum + Piper betle	1	0.8	-	-	-	-	-	-	-	-	-	-
Cuminum cyminum + Piper nigrum + Zingiber officinale	-	-	-	-	-	-	-	-	-	-	1	0.8
Cuminum cyminum + Prunus avium + Piper betle	1	0.8	-	-	-	-	-	-	-	-	-	-
Malus pumila + Apium graveolens	-	-	1	0.8	-	-	-	-	-	-	-	-
Momordica charantia	3	2.5	-	-	-	-	-	-	-	-	-	-

Continued Table 4.7: The medicinal values of crude herbs as perceived by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Crude herbs	Perceived medicinal values of crude herbs											
	Reduce blood glucose level		Reduce blood pressure level		Reduce blood cholesterol level		Health maintenance		Treat gout		Reduce wheezing	
	F	%	F	%	F	%	F	%	F	%	F	%
Moringa oleifera	2	1.7	2	1.7	-	-	-	-	-	-	-	-
Nigella sativa	3	2.5	-	-	-	-	-	-	-	-	-	-
Orchis mascula	-	-	1	0.8	-	-	-	-	-	-	-	-
Orthosiphon aristatus	2	1.7	2	1.7	2	1.7	-	-	-	-	-	-
Pandanus amaryllifolius	-	-	2	1.7	-	-	-	-	-	-	-	-
Pandanus conoideus	-	-	1	0.8	-	-	-	-	-	-	-	-
Phaleria macrocarpa	4	3.3	5	4.1	-	-	-	-	-	-	-	-
Psidium guajava	5	4.1	-	-	-	-	-	-	-	-	-	-
Senna auriculata	1	0.8	-	-	-	-	-	-	-	-	-	-
Solanum procumbens	-	-	-	-	-	-	-	-	-	-	1	0.8

Continued Table 4.7: The medicinal values of crude herbs as perceived by the patients with chronic diseases attending Klinik Kesihatan Kampar.

Crude herbs	Perceived medicinal values of crude herbs											
	Reduce blood glucose level		Reduce blood pressure level		Reduce blood cholesterol level		Health maintenance		Treat gout		Reduce wheezing	
	F	%	F	%	F	%	F	%	F	%	F	%
Swietenia macrophylla	1	0.8	1	0.8	-	-	-	-	-	-	-	-
Trachyspermum ammi	1	0.8	-	-	1	0.8	1	0.8	-	-	-	-
Trigonella foenum-graecum	11	9.1	4	3.3	1	0.8	-	-	-	-	-	-
Trigonella foenum-graecum + ***Cuminum cyminum***	1	0.8	1	0.8	-	-	-	-	-	-	-	-
Zingiber officinale	-	-	1	0.8	-	-	-	-	-	-	-	-
Ziziphus mauritiana	-	-	-	-	-	-	1	0.8	-	-	-	-
Total	**51**	**42.1**	**29**	**24.0**	**6**	**5.0**	**4**	**3.2**	**1**	**0.8**	**2**	**1.6**

*F = Frequency of perceived medicinal values of crude herbs used by study population
% = Percentage of perceived medicinal values of crude herbs used by study population

4.1.8 Duration of crude herbs use

Table 4.8 shows the duration of crude herbs use by the patients with chronic diseases. About 19.0% of patients used crude herbs for less than a year to treat their underlying medical conditions and 14.0% of patients use crude herbs for a period of 1 to 3 years followed by 3.3% of patients used it for more than 3 years.

Table 4.8: The duration of crude herbs use by patients with chronic diseases attending Klinik Kesihatan Kampar.

Duration	F	%	Pearson Chi-square values (χ^2)	p-value
Less than a completed year	23	19.0	12.864	0.002
1 to 3 completed years	17	14.0		
More than 3 completed years	4	3.3		

*F = Frequency of duration of crude herbs use by study population
% = Percentage of duration of crude herbs use by study population

4.1.9 Patients' perceptions on crude herbs use

Table 4.9 depicts the perceptions of patients with chronic diseases on crude herbs use in this study. Most patients were using crude herbs due to recommendation from their family or friends (15.7%), belief on the efficacy of crude herbs (12.4%), no side effects caused by crude herbs (2.5%), health improved (1.7%) and other reasons such as crude herbs were recommended by physicians, herbs are natural and good for health, to assist conventional medications, conventional medications have side effects and do not want to

depend on conventional medications alone with each of these reasons denoting 0.8% of patients.

Table 4.9: The perceptions of patients with chronic diseases on crude herbs use.

Perceptions	**F**	**%**	**Pearson Chi-square values (χ^2)**	**p-value**
Recommended by family/relatives/friends	19	15.7	79.545	<0.001
Belief on the efficacy of crude herbs	15	12.4		
There is no side effects caused by crude herbs	3	2.5		
Health improved after taking crude herbs	2	1.7		
Recommended by physicians/doctors/pharmacists/medical officers	1	0.8		
Herbs are natural and good for health	1	0.8		
To assist conventional medicines	1	0.8		
Conventional medicines have side effects	1	0.8		
Do not want to depend on conventional medicines	1	0.8		

*F = Frequency of perceptions on crude herbs use
% = Percentage of perceptions on crude herbs use

4.1.10 Source of information on crude herbs

Based on **Figure 4.1**, most of the patients obtained information about the crude herbs to treat their diseases from their friends or neighbours (20.9%) followed by relatives (12.2%), television (4.3%) and Internet websites (3.6%). Patients less likely obtained the information on crude herbs from their doctors (1.4%), spouse (1.4%) and newspaper (0.7%).

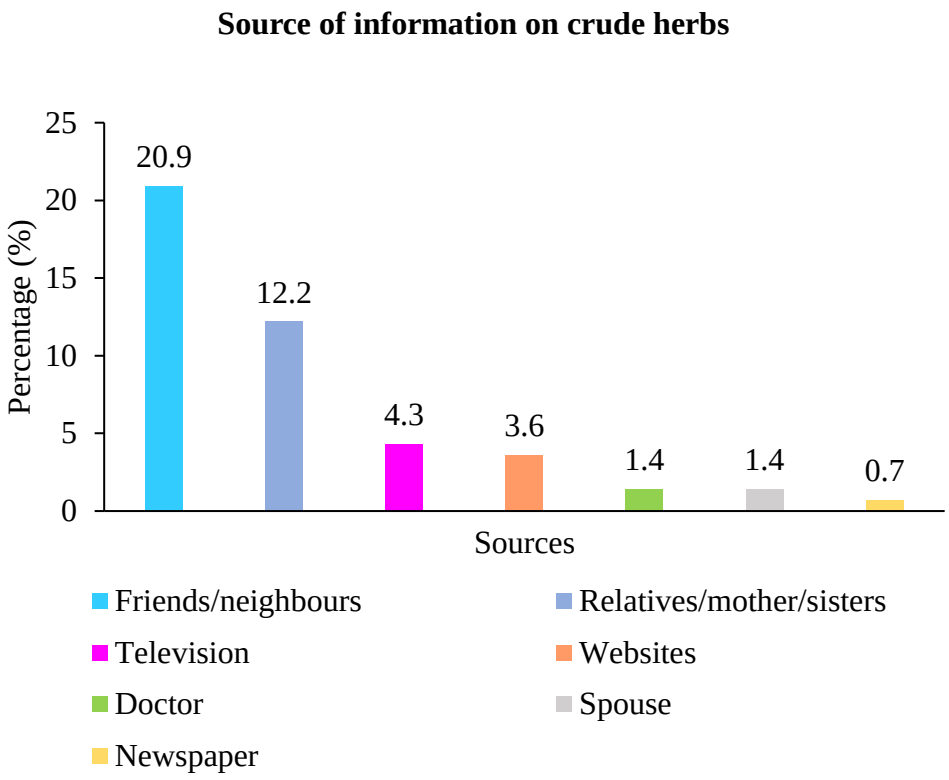

Figure 4.1: Source of information regarding crude herbs to treat chronic diseases.

4.1.11 The association between the socio-demographic characteristics and the crude herbs use

In the following subsections, the association between socio-demographic characteristics and the crude herbs use are analysed and demonstrated. Pearson Chi-square analysis was conducted to determine if any of the socio-demographic factors such as age, gender, race, salary range, education level, and employment status influence the crude herbs use among the patients with chronic diseases in this study. A null hypothesis (H_0) was assumed that there is no significant association between the socio-demographic characteristics and the use of crude herbs. If the p-value obtained is less than 0.05, then H_0 is rejected, whereas if $p > 0.05$, H_0 is accepted.

4.1.11.1 The association between the age of patients and the crude herbs use

A null hypothesis (H_0) was assumed that there is no significant association between the age groups of patients and the use of crude herbs. Based on the Pearson Chi-Square test showed in **Table 4.10**, the values obtained were $\chi^2 = 1.276$, $df = 4$ and $p > 0.05$, therefore H_0 is accepted and there is no significant association between age groups of patients and the crude herbs use.

Table 4.10: Cross tabulation analysis between the age groups of patients and the crude herbs use.

Age groups (Years)	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
49 and below	4	4.4	-0.4	1.276	0.865
Between 50 to 59	12	12.7	-0.7		
Between 60 to 69	20	17.5	2.5		
Between 70 to 79	6	7.6	-1.6		
80 and above	2	1.8	0.2		

Note: The degree of freedom (df = 4) and the 95% confidence interval reflects a significance level of 0.05.

4.1.11.2 The association between the gender of patients and the crude herbs use

A null hypothesis (H_0) was assumed to have no any significant association between the gender of patients and the use of crude herbs. **Table 4.11** showed the Pearson Chi-Square test and the values obtained were $\chi^2 = 4.440$, df = 1 and $p < 0.05$, therefore H_0 is rejected and gender of patients is significantly associated with crude herbs use.

Table 4.11: Cross tabulation analysis between the gender of patients and the crude herbs use.

Gender	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
Male	12	17.5	-5.5	4.440	0.035
Female	32	26.5	5.5		

Note: The degree of freedom (df = 1) and the 95% confidence interval reflects a significance level of 0.05.

4.1.11.3 The association between the race of patients and the crude herbs use

A null hypothesis (H_0) was assumed to have no any significant association between the race of patients and the use of crude herbs. **Table 4.12** showed the Pearson Chi-Square test and the values obtained were $\chi^2 = 7.967$, df = 3 and $p < 0.05$, therefore H_0 is rejected and there is a significant association between race of patients and the crude herbs use.

Table 4.12: Cross tabulation analysis between the race of patients and the crude herbs use.

Race	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
Malay	17	18.9	-1.9	7.967	0.047
Chinese	2	5.5	-3.5		
Indian	25	18.9	6.1		
Others	0	0.7	-0.7		

Note: The degree of freedom (df = 3) and the 95% confidence interval reflects a significance level of 0.05.

4.1.11.4 The association between the salary range of patients and the crude herbs use

A null hypothesis (H_0) was assumed to have no any significant association between the salary range of patients and the use of crude herbs. Based on **Table 4.13**, the Pearson Chi-Square test, the values obtained were $\chi^2 = 2.980$, df = 1 and $p > 0.05$, therefore H_0 is accepted and there is no any significant association between the salary range of patients and the crude herbs use.

Table 4.13: Cross tabulation analysis between the salary range of patients and the crude herbs use.

Salary range	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
Below RM 3000	44	42.2	1.8	2.980	0.084
Between RM 3000 to RM 6000	0	1.8	-1.8		

Note: The degree of freedom (df = 1) and the 95% confidence interval reflects a significance level of 0.05.

4.1.11.5 The association between the education level of patients and the crude herbs use

A null hypothesis (H_0) was assumed to have no any significant association between the level of education of patients and the use of crude herbs. Based on the Pearson Chi-Square test showed in **Table 4.14**, the values obtained were $\chi^2 = 2.125$, df = 3 and $p > 0.05$, therefore H_0 is accepted and there is no any significant association between the education level of patients and the crude herbs use.

Table 4.14: Cross tabulation analysis between the education level of patients and the crude herbs use.

Level of education	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
No formal education	7	7.3	-0.3	2.125	0.547
Primary education	13	14.5	-1.5		
Secondary education	23	21.8	1.2		
Tertiary education	1	0.4	0.6		

Note: The degree of freedom (df = 3) and the 95% confidence interval reflects a significance level of 0.05.

4.1.11.6 The association between the employment status of patients and the crude herbs use

A null hypothesis (H_0) was assumed to have no any significant association between the employment status of patients and the use of crude herbs. Based on the Pearson Chi-Square test showed in **Table 4.15**, the values obtained were χ^2 = 5.150, df = 3 and $p > 0.05$, therefore H_0 is accepted and there is no any significant association between the employment status of patients and the crude herbs use.

Table 4.15: Cross tabulation analysis between the employment status of patients and the crude herbs use.

Employment status	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
Housewife/ unemployed	28	22.2	5.8	5.150	0.161
General staff	9	12.0	-3.0		
Intellectual professionals	0	0.4	-0.4		
Retired	7	9.5	-2.5		

Note: The degree of freedom (df = 3) and the 95% confidence interval reflects a significance level of 0.05.

4.1.12 The association between chronic diseases and the crude herbs use

In the following subsection, the association between chronic diseases and the crude herbs use are analysed and demonstrated. Pearson Chi-square analysis was conducted to determine if any of the chronic diseases influence the crude herbs use among the patients in this study. A null hypothesis (H_0) was assumed that there is no significant association between the chronic diseases and the use of crude herbs. If the p-value obtained is less than 0.05, then H_0 is rejected, whereas if $p > 0.05$, H_0 is accepted.

4.1.12.1 The association between hypertension and the crude herbs use

A null hypothesis (H_0) was assumed to have no any significant association between hypertension and the use of crude herbs. Based on the Pearson Chi-Square test showed in **Table 4.16**, the values obtained were $\chi^2 = 3.858$, df = 1 and $p < 0.05$, H_0 is rejected and thus, patients with hypertension are significantly associated with crude herbs use.

Table 4.16: Cross tabulation analysis between the chronic disease of patients and the crude herbs use.

Chronic disease	Observed count	Expected count	Residual	Pearson Chi-square values (χ^2)	p-value
Hypertension	33	28.0	5.0	3.858	0.049
No hypertension	11	16.0	-5.0		

Note: The degree of freedom (df = 1) and the 95% confidence interval reflects a significance level of 0.05.

4.2 Plant extraction

In phase 2, the laboratory analysis was carried out using the selected crude herbs. The crude herbs for extraction were selected based on the preliminary SPSS analysis. The highly used crude herbs by the patients with chronic diseases were *Trigonella foenum-graecum* (9.9%) followed by *Cuminum cyminum* (5.8%) and *Phaleria marcocarpa* (5.0%). However, the phytochemical compounds present in the three crude herbs aforementioned have been screened by our seniors. In this study, we have selected other crude herbs that were also consumed by the patients, namely, *Psidium guajava* (4.1%) and *Coriandrum sativum* (3.3%).

4.2.1 Percentage of yield for solvent extraction

The percentage yield obtained for the two chosen crude herbs are as shown in **Table 4.17**. The weight of each herb was recorded before subjecting the herbs to ethanol extraction and the dry weights of the herbs were recorded after the extraction was completed. The highest percentage of yield for ethanol extraction was obtained for *Psidium guajava* (59.6%) followed by *Coriandrum sativum* (29.6%). The percentage of yield was calculated using formula as shown below:

$$\text{Percentage of yield} = \frac{\text{Dry weight of crude herbs (g)}}{\text{Total weight of crude herbs (g)}} \text{ x } 100\%$$

Table 4.17: The respective total weight, dry weight and the percentage of yield for solvent extracts of each chosen crude herbs.

Crude herbs chosen	Total weight (g)	Dry weight (g)	Percentage of yield (%)
Psidium guajava	74.18	44.18	59.6
Coriandrum sativum	250.00	73.98	29.6

4.2.2 Percentage of yield for aqueous extraction

The percentage yield obtained for the two chosen crude herbs are as shown in **Table 4.18**. The weight of the herbs was recorded before subjecting the herbs to aqueous extraction and the dry weights of the herbs were recorded after the extraction was completed. The highest percentage of yield for aqueous extraction was obtained for *Psidium guajava* (1.3%) followed by *Coriandrum*

sativum (0.9%). The percentage of yield was calculated using formula as shown below:

$$\text{Percentage of yield} = \frac{\text{Dry weight of crude herbs (g)}}{\text{Total weight of crude herbs (g)}} \text{ x } 100\%$$

Table 4.18: The respective total weight, dry weight and the percentage of yield for aqueous extracts of each chosen crude herbs.

Crude herbs chosen	Total weight (g)	Dry weight (g)	Percentage of yield (%)
Psidium guajava	79.95	1.00	1.3
Coriandrum sativum	250.12	2.31	0.9

4.2.3 Qualitative phytochemical screening

Table 4.19 depicts the intensity of the phytochemical compounds that are present in each of the selected crude herbs, both in solvent and aqueous extracts. For solvent extraction, there were seven phytochemical compounds present in *Psidium guajava* whereas *Coriandrum sativum* exhibited six compounds. Both *Psidium guajava* and *Coriandrum sativum* exhibited phenol, tannins, saponins, flavonoids, glycosides and alkaloids in varied intensity. Terpenoids were present in solvent extracts of *Psidium guajava*. Quinones were absent in the solvent extracts of both of the herbs.

For aqueous extraction, there were seven phytochemical compounds present in *Psidium guajava* and six phytochemical compounds present in *Coriandrum sativum*. Both *Psidium guajava* and *Coriandrum sativum* exhibited phenol, quinones, saponins, flavonoids, glycosides and alkaloids in varied intensity. Terpenoids were absent in aqueous extracts of both *Psidium guajava* and *Coriandrum sativum*. Tannins were absent in the aqueous extract of *Coriandrum sativum*.

Table 4.19: Qualitative phytochemical screening of the two selected crude herbs using solvent and aqueous extraction methods.

Extraction medium	Crude herbs	Phenols	Quinones	Tannins	Terpenoids	Saponins	Flavonoids	Glycosides	Alkaloids
Solvent	*Psidium guajava*	+++	-	++	++	++	+++	++	+
	Coriandrum sativum	+	-	+	-	++	++	++	++
Aqueous	*Psidium guajava*	+++	+++	+++	-	++	+++	+++	+++
	Coriandrum sativum	++	+	-	-	++	++	++	+

Note: +++ indicates high intensity of phytochemical compounds; ++ indicates medium intensity of phytochemical compounds; + indicates low intensity of phytochemical compounds.

4.2.3.1 Qualitative phytochemical screening of standards

Figure 4.2 shows the results of phytochemical screening of the standards. The formation of dark blue colour indicated the presence of umbelliferone, which represents the presence of phenols [A]. The presence of tannins [B] was determined by the formation of white precipitate or cloudy appearance. The presence of saponins [C] in the extract was determined by the formation of persistent foam after the standard was shaken with distilled water. The formation of quercetin which represents flavonoids was indicated by the colour change from concentrated yellow [D] to colourless [E] upon addition of dilute hydrochloric acid. Alkaloids [F] were indicated by the formation of reddish brown colouration in the test tube when tested using caffeine as the standard.

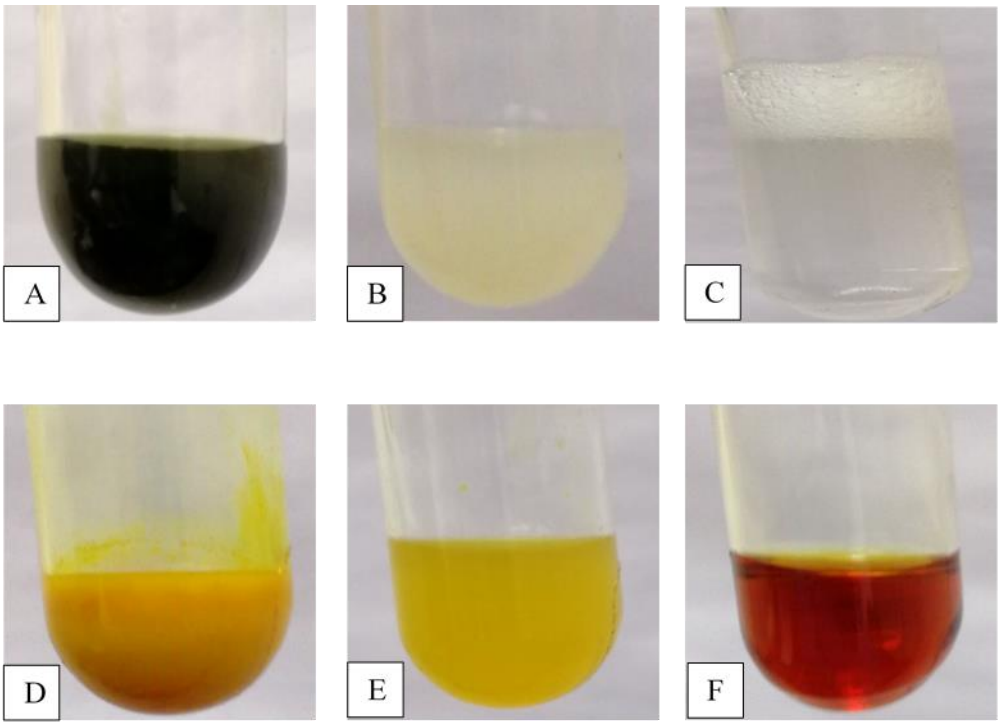

Figure 4.2: The phytochemical screening of standards [A] Phenols; [B] Tannins; [C] Saponins; [D] Flavonoids before adding dilute hydrochloric acid; [E] Flavonoids after adding dilute hydrochloric acid; [F] Alkaloids.

4.2.3.2 Qualitative phytochemical screening for solvent extraction

Figure 4.3 illustrates the results of phytochemical screening of *Psidium guajava* solvent extracts. The presence of phenols [B] in high intensity (+++) was revealed by the formation of a dark blue colour. There is no formation of yellow precipitate (-) for quinones [C]. The presence of white precipitate (++) indicates the presence of tannins [D] in the solvent extracts of *Psidium guajava*. Terpenoids [E] presence was indicated by the formation of reddish brown precipitate at the junction (++) and the presence of saponins [F] was determined by the formation of persistent foam (++). The presence of flavonoids [G] was indicated by the colour change from yellow to colourless (+++). The formation of yellow precipitate in medium intensity (++) indicates the presence of glycosides [H] in *Psidium guajava* solvent extract whereas alkaloids [I] were indicated by the formation of reddish brown colour (+).

Figure 4.4 illustrates the results of phytochemical screening of *Coriandrum sativum* solvent extracts. The slight colour difference of extracts added with phenols [B] and tannins [D] from original solvent extract [A] indicate the presence of phenols and tannins in low intensity (+). The absence of yellow precipitate (-) and reddish brown layer (-) indicate the absence of quinones [C] and terpenoids [E]. The formation of persistent foam for saponin test [F] and colourless solution for flavonoid test [G] indicate the presence of saponins and flavonoids in medium intensity (++). The formation of yellow precipitate and reddish brown precipitate indicate the presence of glycosides [H] and alkaloids [I] in medium intensity (++).

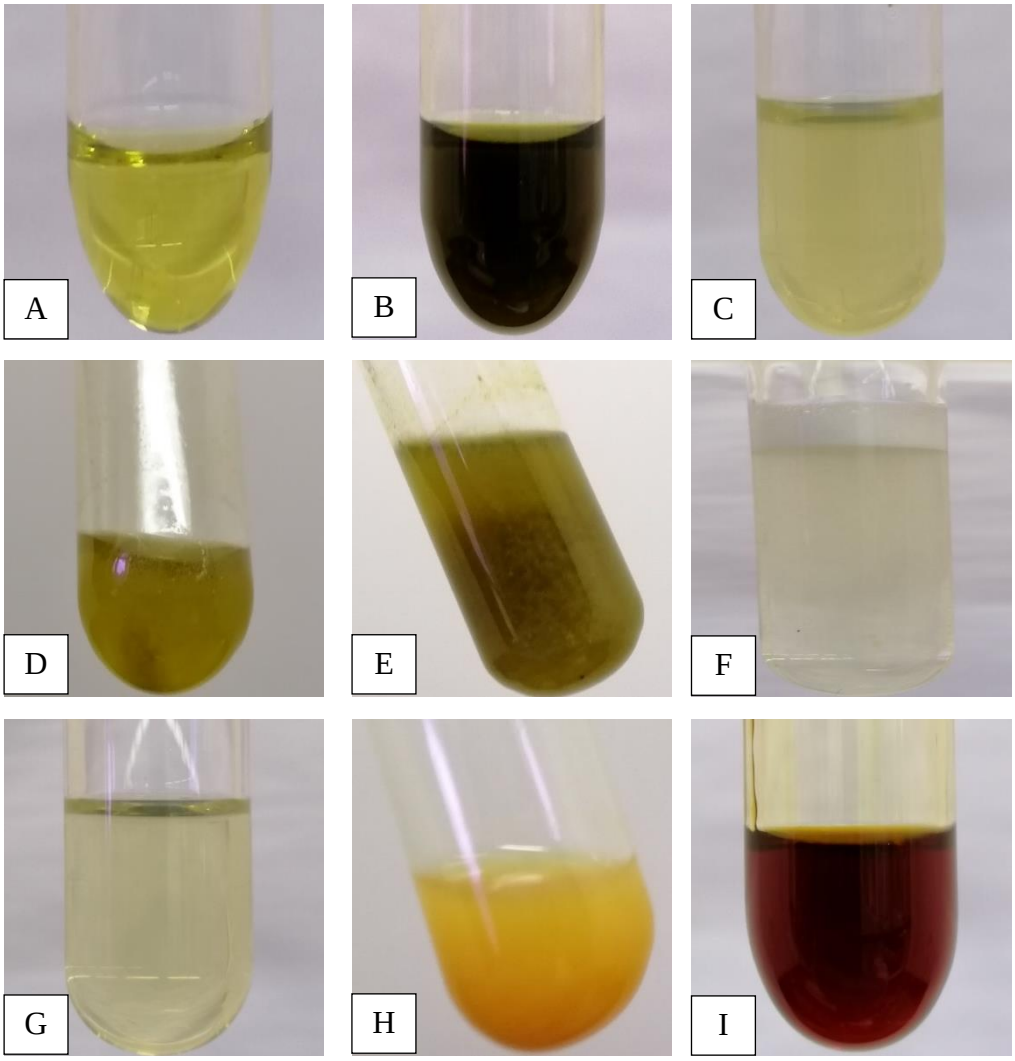

Figure 4.3: The phytochemical screening of solvent extract of *Psidium guajava*: [A] Original solvent extract; [B] Phenols; [C] Quinones; [D] Tannins; [E] Terpenoids; [F] Saponins; [G] Flavonoids; [H] Glycosides; [I] Alkaloids.

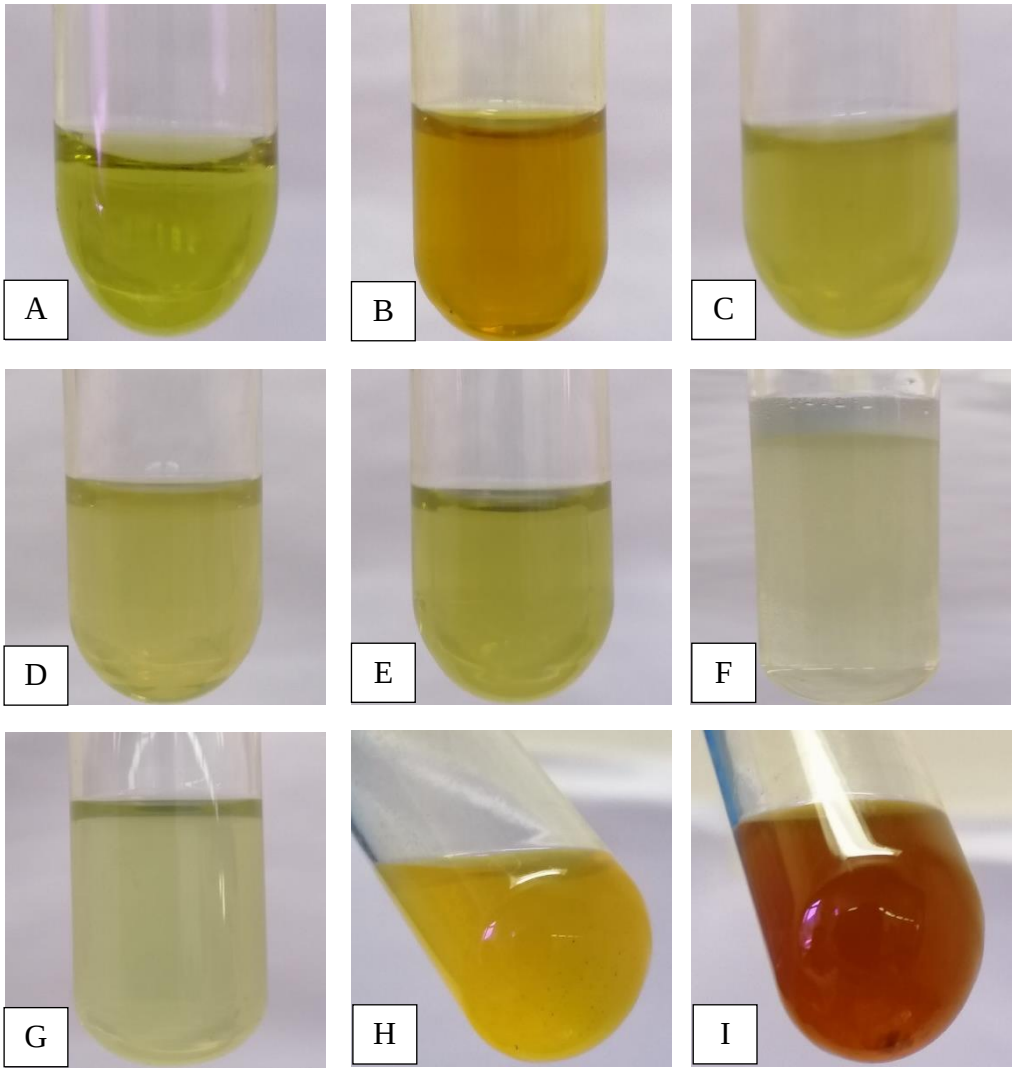

Figure 4.4: The phytochemical screening of solvent extract of *Coriandrum sativum*: [A] Original solvent extract; [B] Phenols; [C] Quinones; [D] Tannins; [E] Terpenoids; [F] Saponins; [G] Flavonoids; [H] Glycosides; [I] Alkaloids.

4.2.3.3 Qualitative phytochemical screening for aqueous extraction

Figure 4.5 illustrates the results of phytochemical screening of the aqueous extracts of *Psidium guajava*. The presence of phenols [B], quinones [C], and tannins [D] was confirmed by the formation of a black colour solution, yellow precipitate, and white precipitate in high intensity (+++) respectively. There was no formation of a reddish brown layer (-), indicating the absence of terpenoids [E] in *Psidium guajava* aqueous extract. The formation of persistent foam in medium intensity (++) was revealed for the saponin test [F]. The presence of flavonoids [G], glycosides [H], and alkaloids [I] was confirmed by the formation of a colourless solution, yellow precipitate and reddish brown precipitate in high intensity (+++) for each test.

Figure 4.6 illustrates the results of phytochemical screening of the aqueous extracts of *Coriandrum sativum*. The formation of dark blue colour in medium intensity (++) were observed for phenol [B] test. The slight colour change from the original solvent extract [A] revealed the presence of quinones [C] in low intensity (+). There was an absence of tannins [D] and terpenoids [E], confirmed by the absence of white precipitate (-) and a reddish brown layer (-) formation respectively. Saponins [F], flavonoids [G], and glycosides [H] were present in medium intensity (++), confirmed by the formation of persistent foam, colourless solution, and yellow precipitate for each of the test. The formation of a reddish brown solution without precipitate indicates the presence of alkaloids [I] in low intensity (-).

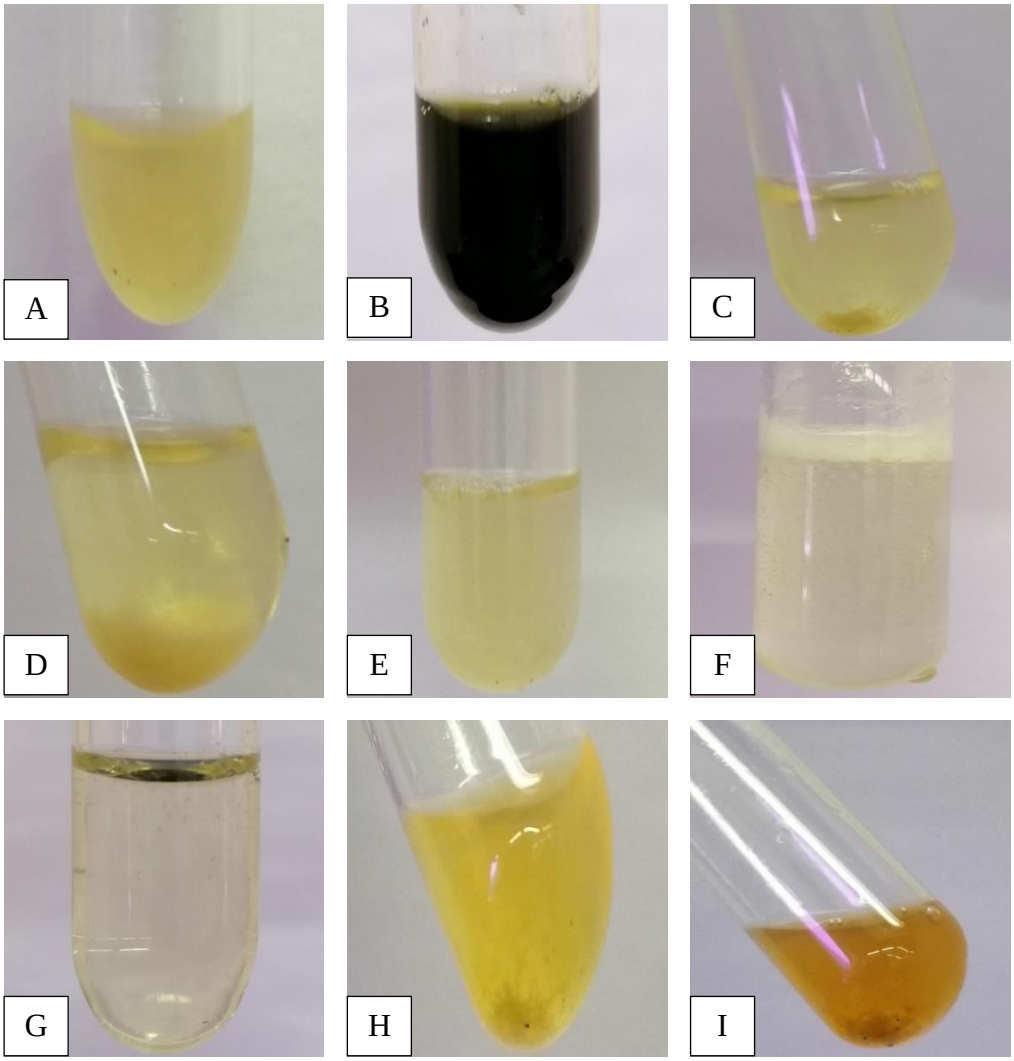

Figure 4.5: The phytochemical screening of aqueous extract of *Psidium guajava*: [A] Original aqueous extract; [B] Phenols; [C] Quinones; [D] Tannins; [E] Terpenoids; [F] Saponins; [G] Flavonoids; [H] Glycosides; [I] Alkaloids.

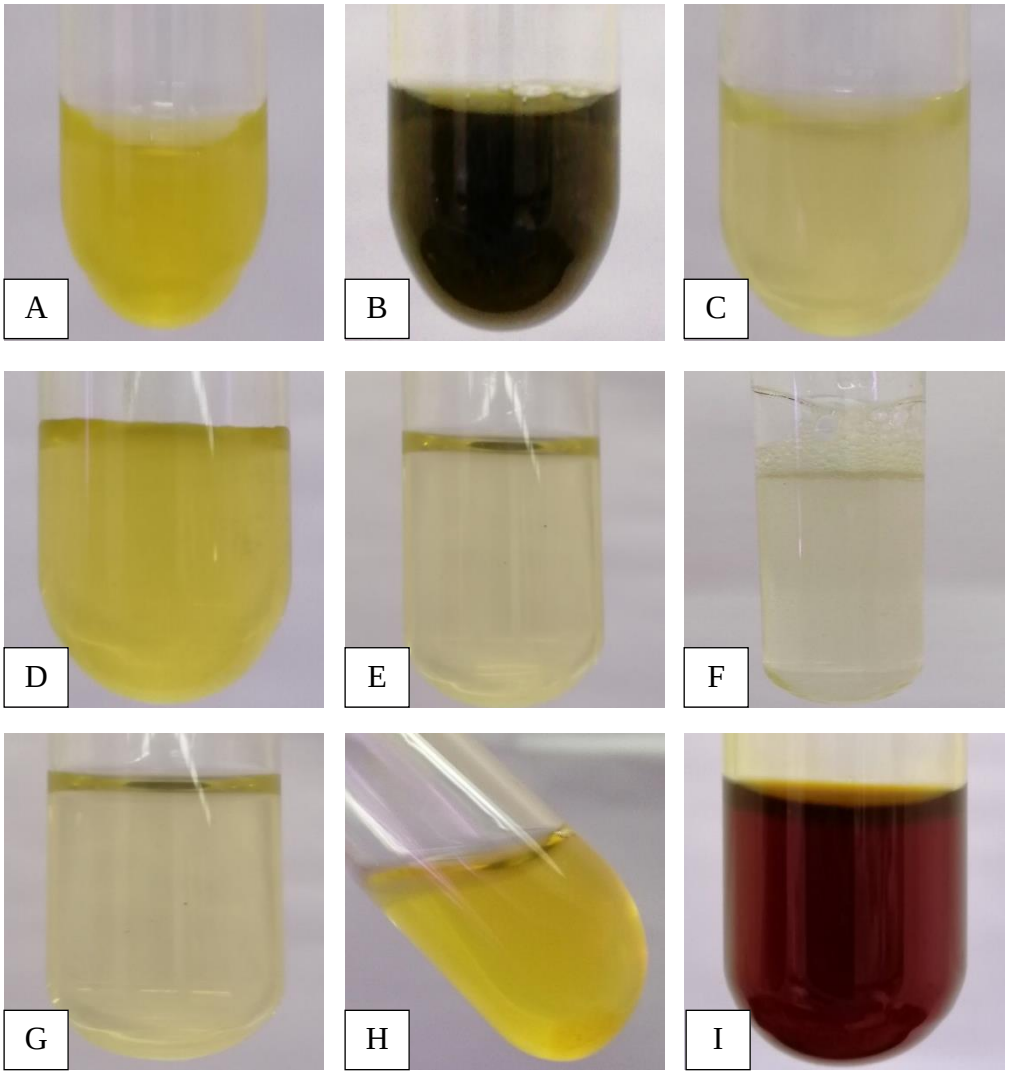

Figure 4.6: The phytochemical screening of aqueous extract of *Coriandrum sativum*: [A] Original aqueous extract; [B] Phenols; [C] Quinones; [D] Tannins; [E] Terpenoids; [F] Saponins; [G] Flavonoids; [H] Glycosides; [I] Alkaloids.

4.2.4 Thin layer chromatography

Table 4.20 depicts the different solvent system that were used for each type of herbs. The number of spots observed on TLC plate of *Psidium guajava* and *Coriandrum sativum* are 16 and 17 spots respectively. The respective optimised solvent systems used for *Psidium guajava* and *Coriandrum sativum* were toluene: ethylacetate (60:40) and hexane: ethylacetate (70:30). **Figure 4.7** illustrates the presence of phytochemical compounds in *Psidium guajava* and *Coriandrum sativum* observed via the thin layer chromatography (TLC) procedure.

Table 4.20: The number of spots observed for *Psidium guajava* and *Coriandrum sativum* in TLC.

Crude herbs	Solvent system	Ratio	Number of spots
Psidium guajava	Toluene: Ethylacetate	60:40	16
Coriandrum sativum	Hexane: Ethylacetate	70:30	17

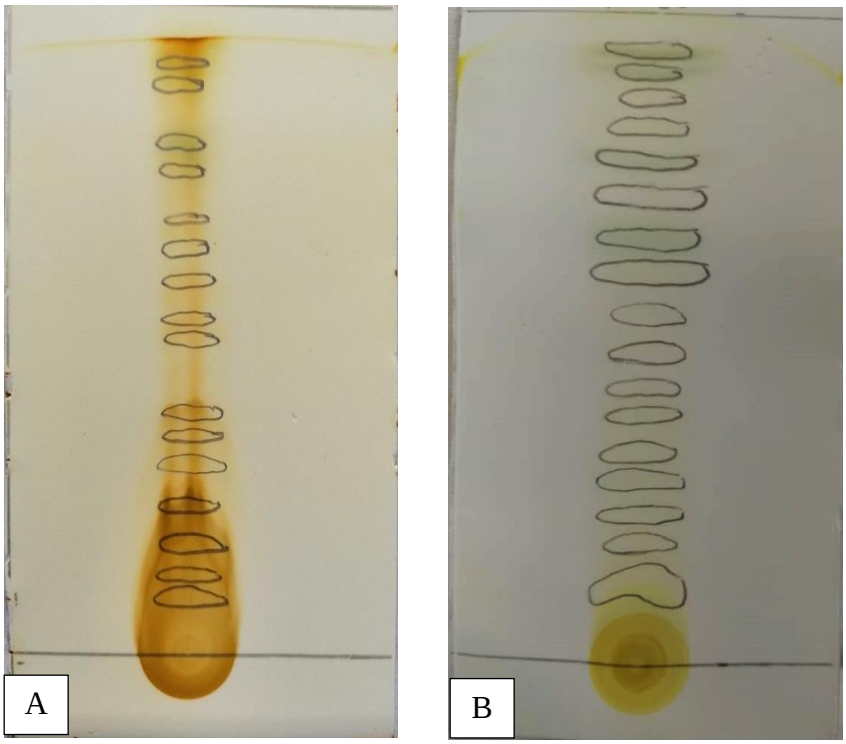

Figure 4.7: The presence of phytochemical compounds on the TLC silica plate after treated with 60:40 (toluene: ethylacetate) and 70:30 (hexane: ethylacetate) and iodine vapour test for [A] *Psidium guajava* and [B] *Coriandrum sativum*.

CHAPTER 5

DISCUSSION

5.1 Socio-demographic characteristics

In this study, the prevalence of crude herbs use was 36.4%. The finding of the present study was comparable to the usage of herbal medicine by the Thailand population (34.6%) and the American population (35%) (Peltzer et al., 2016b; Rashrash et al., 2017). The prevalence in our study was higher than the prevalence of herbal medicine use in Turkey, which was 29% (Tulunay et al., 2015). The current usage is lower compared to other studies in Saudi Arabia (67.7%) and Vietnam (43.6%) (Alghamdi et al., 2018; Peltzer et al., 2017). Our results is still lower when compared to other local studies conducted among the cancer patients (40.2%) and the diabetes patients (40.6%) (Farooqui et al., 2016; Baharom et al., 2016). The differences in the prevalence are due to the varying definitions of herbs based products used and the larger number of sample size involved in the previous studies (Medagama et al., 2014).

The use of crude herbs in the current study was popular among the age group 50 to 59 years old (27.3%) and 60 to 69 years old (45.5%) which is consistent with a study done by Peltzer et al. (2017). Patients aged 60 and above actively use crude herbs as they are highly self conscious and worried about their health (Huri et al., 2009). On the contrary, Ali and Mahfouz (2014) reported high use of herbal medicine by patients aged from 41 to 50 years old (49.0%). Another study

conducted in Sabah reported that younger population aged 18 to 30 years old (30.6%) and 31 to 40 years (47.2%) were more evident in using herbs (Chang et al., 2015). Thus, it is evident that herbs use is prevalent despite any age groups.

The current study revealed that more females (60.3%) than males (39.7%) were using crude herbs, which is supported by other studies (Baharom et al., 2016; Farooqui et al., 2016; Abdullah et al., 2018). A significant association between female and crude herbs use (p=0.035) was demonstrated in this study. Comparably, two other studies showed that female patients are associated with herbs use (Fan et al., 2013; Medagama et al., 2014). The findings suggest that females are more concerned about their health condition compared to males due to the societal pressure (Baharom et al., 2016).

Contrary to other studies (Ching et al., 2013a; Kew et al., 2015), where Malays were the predominant users of herbs, Indian patients (56.8%) were more likely to use crude herbs in the current study. Likewise, findings of Hasan et al. (2009) agree to the current study. The lower number of Chinese patients consuming crude herbs in this study is due to the existence of traditional Chinese medicine, in which they may have stronger belief in (Huri et al., 2009). A significant association between Indian patients and crude herbs use was established in this study (p=0.047). Most of the Indian patients attending Klinik Kesihatan Kampar were from the lower socioeconomic background. They could afford crude herbs

as an alternative treatment option compared to other herbal medicines, as crude herbs are much cheaper (Mollaoglu and Aciyurt, 2013).

In the present study, all the crude herbs' users earn lower than RM 3000 monthly (100%) which is similar to Ching et al. (2013b) findings. Kew et al. (2015) found that herbal medicine use was high among patients with higher income. Similarly, Aziz and Tey (2009) reported high utilisation of herbal medicine among individuals with high income. However, unlike herbal medicines, crude herbs are much cheaper alternative treatment, which explains the reason patients with low income were more likely to use crude herbs in the present study (Mollaoglu and Aciyurt, 2013).

The use of crude herbs was high among the patients that have attained secondary education (52.3%). This is in agreement with studies by Hasan et al. (2009) and Abdullah et al. (2018). The higher usage of crude herbs among the secondary school leavers compared to other education level is due to higher health awareness. In addition, they may have higher access to information of medicinal benefits of crude herbs (Abdullah et al., 2018).

Few studies performed found that housewives were more likely to use herbs (Mollaoglu and Aciyurt, 2013; Tulunay et al., 2015; Pearson et al., 2018). Likewise, the present study found those being housewives or unemployed

(63.6%) to be prevalent in using crude herbs. There are two possibilities for the high use of crude herbs among the housewives or unemployed patients in this study. Firstly, the low cost of crude herbs and secondly, the high concern of housewives about health and wellbeing of family (Mollaoglu and Aciyurt, 2013; Tulunay et al., 2015).

The use of crude herbs were significantly associated with hypertensive patients in this study (p=0.049). Pearson et al. (2018) have also revealed a significant association between hypertension and the use of herbs (p=0.04). However, the findings of the current study is not comparable to other studies that involved complementary and alternative medicine (CAM) use which is a wider scope compared to crude herbs (Mollaoglu and Aciyurt, 2013).

The types of crude herbs consumed by the patients in this study were *Trigonella foenum-graecum* (9.9%), *Cuminum cyminum* (5.8%), *Phaleria marcocarpa* (5.0%), *Citrus limon* (4.1%), *Psidium guajava* (4.1%), *Centella asiatica* (3.3%) and *Coriandrum sativum* (3.3%). Previous studies in Malaysia showed herbs such as *Momordica charantia*, *Orthosiphon stamineus* and *Allium sativum* were used (Ching et al., 2013b; Baharom et al., 2016). In Sri Lanka, herbs such as *Momordica charantia*, *Coccinia grandis* and *Costus speciosus* were predominantly used (Medagama et al., 2014). However, none of the studies showed a similar types of herbs as the studies were conducted in different

locations. This gives a conclusion that crude herbs use varies among patients from different cultural background (Medagama et al., 2014).

Consistent to findings in previous studies (Ching et al., 2013a; Fan et al., 2013), the present study found friends (20.9%) and family (12.2%) being the most common source of information for crude herbs use. In the current study, it is revealed that recommendations of family and friends were the main reason to use crude herbs (15.7%). Family and friends are the closest relationship to the patients, thus rendering patients to believe them more (Huri et al., 2009). High number of patients believed the efficacy of crude herbs (12.4%) which is similar to previous studies (Ching et al., 2013b).

5.2 Phytochemical compounds screened in the crude herbs

In this study, the ethanolic extracts of *Psidium guajava* leaves have revealed the presence of phenols and flavonoids in high intensity. Other phytochemical compounds revealed were tannins, terpenoids, saponins, glycosides, and alkaloids. These findings are similar to another study that revealed the presence of flavonoids, tannins, terpenoids, and alkaloids (Kenneth et al., 2017) in the *Psidium guajava* leaves extracted using ethanol. In the current study, there were no quinones present in the leaves of *Psidium guajava* extracted via ethanol which contradicts the results reported by Gayathri and Kiruba (2014) in India. The variations in the phytochemical compounds revealed are due to the

difference in soil type and weather as both of the studies were carried out in different countries (Inbathamizh and Padmini, 2013).

In this study, the aqueous extracts of *Psidium guajava* revealed the presence of phenols, quinones, tannins, flavonoids, glycosides, and alkaloids in high intensity. Saponins was revealed in a medium intensity. However, terpenoids were absent in the aqueous extracts as compared to the ethanolic extracts. Terpenoids constitute of hydrocarbon chains, which makes them non-polar (Jiang et al., 2016). Thus, extraction using water, which is a polar compound, failed to elute out the terpenoids present in *Psidium guajava* leaves. The current findings are consistent with a study by Sukanya et al. (2017). Phytochemical compounds, such as phenols and flavonoids possess antidiabetic properties (Joseph and Jini, 2013). The high intensity of both of these compounds in the aqueous extracts evinces the high use of *Psidium guajava* (5.1%) among the patients with diabetes mellitus to reduce their blood glucose level (5.1%).

In the current study, the ethanolic extracts of *Coriandrum sativum* leaves and shoots exhibited phytochemical compounds, such as, phenols, saponins, flavonoids, glycosides, and alkaloids. Quinones and terpenoids were absent in the extracts. Paul et al. (2013) reported a similar finding to the current study. However, tests for quinones and terpenoids were not performed by Paul et al. (2013). Gayathri et al. (2016) reported the absence of terpenoids, which is similar to the current study. The ethanolic extracts of coriander seeds revealed

the presence of quinones in their study that contradicts our results. However, the comparison is unclear as the plant's parts used were not same. On the contrary, the methanolic extracts of *Coriandrum sativum* leaves revealed the presence of terpenoids (Patel and Vakilwala, 2016; Ashika et al., 2018). Quinones were absent in all three extracts of *Coriander sativum* leaves, namely, methanol, acetone, and benzene (Patel and Vakilwala, 2016). Collectively, all the solvent extracts used in the previous studies and the current study were unable to reveal the presence of quinones in the leaves of *Coriandrum sativum*.

The aqueous extracts of *Coriandrum sativum* leaves revealed the presence of phenols, quinones, saponins, flavonoids, glycosides, and alkaloids. However, tannins and terpenoids were not revealed in the aqueous extracts. The findings of the present study are comparable to a study done by Chauhan et al. (2012) in India. However, Chauhan et al. (2012) has revealed the presence of both tannins and terpenoids. The differences in the phytochemical compounds between the previous study and the current study are due to the geographical locations of the plants. In this study, reported that patients used *Coriandrum sativum* (3.3%) to reduce their blood pressure level (1.7%). The presence of alkaloids in the leaves extracts are responsible for antihypertensive effects by dilating the blood vessels (Kumar et al., 2018). Moreover, coriander posses diuretic effects, which causes frequent urination. Excess sodium in the blood is ultra-filtrated by the kidney and released through the urine (New Delhi Television Limited, 2018). Phenols and flavonoids are responsible for antioxidant properties that regulate the lipid metabolism (Ramadan et al., 2008; Rao et al., 2016). This could be the reason

of *Coriandrum sativum* use by the patients to reduce their cholesterol level (1.7%).

5.3 Thin layer chromatography

Thin layer chromatography (TLC) was carried out in the current study to screen additional phytochemical compounds that are present in the crude herbs that were not revealed in the eight phytochemical tests. Different solvent systems in different proportions were used to elute the phytochemical compounds present in *Psidium guajava* and *Coriandrum sativum*. *Psidium guajava* revealed 16 spots using toluene: ethylacetate (60:40) whereas *Coriandrum sativum* revealed 17 spots using hexane: ethylacetate (70:30). Each phytochemical compound has different functional group that determines its polarity.

As the mobile phase moves up the mixture of phytochemical compounds, the polar compound will stay close to the origin and the non-polar compounds will move up the plate with the mobile phase. However, if a highly polar mobile phase is used, the polar compound will move along with the mobile phase. In this study, a mixture of non-polar (hexane and toluene) and polar (ethylacetate) mobile phase was used to ensure better separation (Bele and Khale, 2011). The use of high proportion of ethylacetate to elute the phytochemicals present in the *Psidium guajava* suggest that most of the compounds were polar. The use of high proportion of hexane to elute out the phytochemicals present in the

Coriandrum sativum shows that the compounds were non-polar (Bele and Khale, 2011).

5.4 Strengths and limitations of study

The study focused on the use of crude herbs specifically, rather than on complementary and alternative medicine (CAM) in general. The study takes place in a clinical setting, which saved time in recruiting patients with chronic diseases. Face-to-face interviews increased the response rate and reliability of answers from patients.

5.5 Future studies

To obtain a better detail of crude herbs use, patients from other government clinics in Kinta valley should be recruited. Participation of more patients can be ensured with the recommendation from hospital's management.

CHAPTER 6

CONCLUSION

The prevalence of crude herbs use among the patients with chronic diseases in the present study is high. The study has found that variables such as gender, race, and hypertension are significantly associated with crude herbs use. The most commonly used crude herbs among the patients were *Psidium guajava* and *Coriandrum sativum*. Different phytochemical compounds were present in the crude herbs when screened qualitatively. The aqueous extracts were able to reveal more phytochemicals when compared to solvent extracts. The richness of phytochemical compounds in the crude herbs suggest the reason behind their high use among the patients with chronic diseases. Patients with chronic diseases consuming crude herbs should be monitored. Physicians should be vigilant about the crude herbs use among patients with chronic diseases. Further studies should be conducted in other government health clinics as well as in the private setting to determine the other associated factors, such as expenditure, compliance, belief, and perceptions on crude herbs use for a better management of chronic diseases.

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APPENDICES

APPENDIX A

UNIVERSITI TUNKU ABDUL RAHMAN

Wholly Owned by UTAR Education Foundation (Company No. 578227-M)

Re: U/SERC/84/2017

30 October 2017

Ms Annaletchumy a/p Loganathan
Department of Biomedical Science
Faculty of Science
Universiti Tunku Abdul Rahman
Jalan Universiti
Bandar Baru Barat
31900 Kampar
Perak

Dear Ms Annaletchumy,

Ethical Approval For Research Project/Protocol

We refer to your application dated 14 September 2017 which was circulated for the consideration of the UTAR Scientific and Ethical Review Committee (SERC). We are pleased to inform that your application for ethical approval for your research project involving human subjects has been approved by SERC.

The details of your research project are as follows:

Research Title	Biochemistry Analysis of Patients with Chronic Diseases Consuming Crude Herbs: A Cross Sectional Study in Combination with Laboratory Analysis
Investigator(s)	Ms Annaletchumy a/p Loganathan (PI) Ms Sangeetha a/p Arullappan Hamsananthini a/p Selvaraju (UTAR Undergraduate Student) Chan Yun Li (UTAR Undergraduate Student) Afiqah Zulaikha bt. Roslan (UTAR Undergraduate Student) Alicia Loo Mei Ling (UTAR Undergraduate Student) Chow Zhee Shah (UTAR Undergraduate Student) Wong Tzu Ping (UTAR Undergraduate Student)
Research Area	Science
Research Location	Klinik Kesihatan Kampar
No of Participants	323 participants (Age: 18 - 20)
Research Costs	Self-funded
Approval Validity	30 October 2017 - 29 October 2018

Address: Jalan Sg. Long, Bandar Sg. Long, Cheras, 43000 Kajang, Selangor D.E. **Postal Address:** P O Box 11384, 50744 Kuala Lumpur, Malaysia
Tel: (603) 9086 0288 **Fax:** (603) 9019 8868 **Homepage:** http://www.utar.edu.my

The conduct of this research is subject to the following:

(1) The participants' informed consent be obtained prior to the commencement of the research.

(2) Confidentiality of participants' personal data must be maintained; and

(3) Compliance with procedures set out in related policies of UTAR such as the UTAR Research Ethics and Code of Conduct, Code of Practice for Research Involving Humans and other related policies/guidelines.

Should you collect personal data of participants in your study, please have the participants in the research signed the attached Personal Data Protection Statement for your records.

The University wishes you all the best in your research.

Thank you.

Yours sincerely,

Professor Ir Dr Lee Sze Wei
Chairman
UTAR Scientific and Ethical Review Committee

c.c Dean, Faculty of Science
Director, Institute of Postgraduate Studies and Research

APPENDIX B

JAWATANKUASA ETIKA & PENYELIDIKAN PERUBATAN
(*Medical Research & Ethics Committee*)
KEMENTERIAN KESIHATAN MALAYSIA
d/a Institut Pengurusan Kesihatan
Jalan Rumah Sakit, Bangsar
59000 Kuala Lumpur

Tel.: 03-2287 4032/2282 0491/2282 9085
03-2282 9082/2282 1402/2282 1449
Faks: 03-2282 0015

Ruj.Kami : KKM.NIHSEC.P18-386(5)
Tarikh : 8-Mac-2018

DR. ANNALETCHUMY A/P LOGANATHAN
TUNKU ABDUL RAHMAN UNIVERSITY COLLEGE - PERAK CAMPUS

YBhg. Dato' / Tuan / Puan,

SURAT KELULUSAN ETIKA: NMRR-17-2591-38273 (IIR)
THE PREVALENCE AND FACTORS ASSOCIATED WITH CRUDE HERBS USE AMONG PATIENTS WITH CHRONIC DISEASES: A CROSS-SECTIONAL SURVEY IN COMBINATION WITH LABORATORY ANALYSIS.

Lokasi Kajian:
KLINIK KESIHATAN KAMPAR

Dengan hormatnya perkara di atas adalah dirujuk.

2. Jawatankuasa Etika & Penyelidikan Perubatan (JEPP), Kementerian Kesihatan Malaysia (KKM) tiada halangan, dari segi etika, ke atas pelaksanaan kajian tersebut. JEPP mengambil maklum bahawa kajian tersebut hanya melibatkan pengumpulan data melalui:

i. Borang soal selidik

3. Segala rekod dan data subjek adalah **SULIT** dan hanya digunakan untuk tujuan kajian ini dan semua isu serta prosedur mengenai *data confidentiality* mesti dipatuhi.

4. Kebenaran daripada Pegawai Kesihatan Daerah/ Pengarah Hospital dan Ketua-Ketua Jabatan atau pegawai yang bertanggungjawab disetiap lokasi kajian di mana kajian akan dijalankan mesti diperolehi sebelum kajian dijalankan. YBhg. Dato' / Tuan / Puan perlu akur dan mematuhi keputusan tersebut. Sila rujuk kepada garis panduan Institut Kesihatan Negara mengenai penyelidikan di Institusi dan fasiliti Kementerian Kesihatan Malaysia (Pindaan 01/2015) serta lampiran *Appendix 5* untuk templet surat memohon kebenaran tersebut.

5. Adalah dimaklumkan bahawa kelulusan ini adalah sah sehingga **7-Mac-2019**. YBhg. Dato'/ Tuan/ Puan perlu menghantar dokumen-dokumen seperti berikut selepas mendapat kelulusan etika. Borang-borang berkaitan boleh dimuat turun daripada laman web Jawatakuasa Etika & Penyelidikan Perubatan (JEPP) (http://www.nih.gov.my/mrec).

.../2-

i. ***Continuing Review Form*** selewat-lewatnya dalam tempoh 1 bulan (30 hari) sebelum tamat tempoh kelulusan ini bagi memperbaharui kelulusan etika.

ii. ***Study Final Report*** pada penghujung kajian.

iii. Mendapat kelulusan etika sekiranya terdapat pindaan keatas sebarang dokumen kajian/ lokasi kajian/ penyelidik.

6. Sila ambil maklum bahawa sebarang urusan surat-menyurat berkaitan dengan penyelidikan ini haruslah dinyatakan nombor rujukan surat ini untuk melicinkan urusan yang berkaitan.

Sekian terima kasih.

"BERKHIDMAT UNTUK NEGARA"

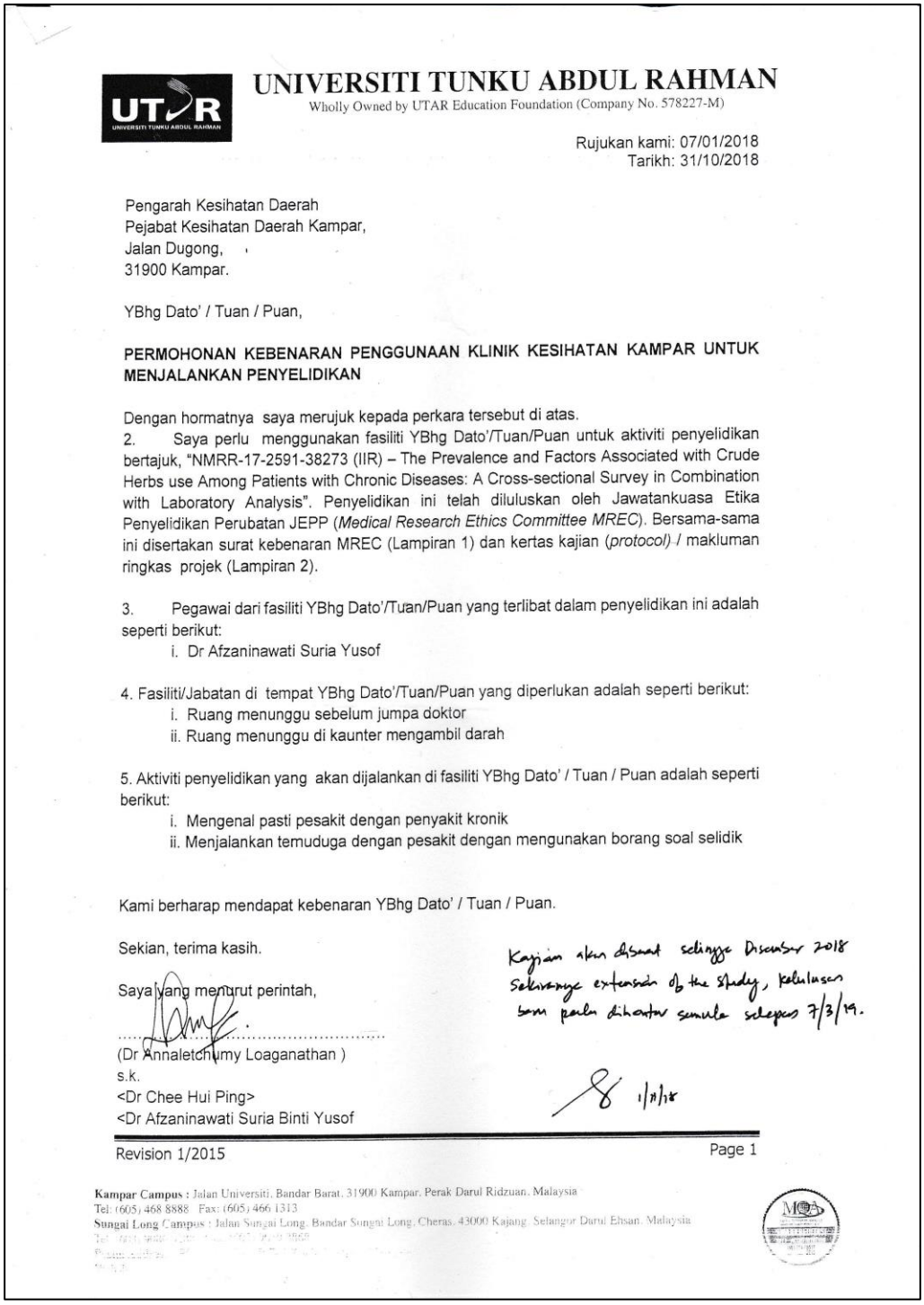
Saya yang menurut perintah,

(DR HJH SALINA ABDUL AZIZ)
Pengerusi
Jawatankuasa Etika & Penyelidikan Perubatan
Kementerian Kesihatan Malaysia
mrecsec@nih.gov.my
03-2282 9085

s.k.: HRRC HOSPITAL RAJA PERMAISURI BAINUN

HM/Approval2017/Mrecshare

APPENDIX C



UNIVERSITI TUNKU ABDUL RAHMAN
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Rujukan kami: 07/01/2018
Tarikh: 31/10/2018

Pengarah Kesihatan Daerah
Pejabat Kesihatan Daerah Kampar,
Jalan Dugong,
31900 Kampar.

YBhg Dato' / Tuan / Puan,

PERMOHONAN KEBENARAN PENGGUNAAN KLINIK KESIHATAN KAMPAR UNTUK MENJALANKAN PENYELIDIKAN

Dengan hormatnya saya merujuk kepada perkara tersebut di atas.

2. Saya perlu menggunakan fasiliti YBhg Dato'/Tuan/Puan untuk aktiviti penyelidikan bertajuk, "NMRR-17-2591-38273 (IIR) – The Prevalence and Factors Associated with Crude Herbs use Among Patients with Chronic Diseases: A Cross-sectional Survey in Combination with Laboratory Analysis". Penyelidikan ini telah diluluskan oleh Jawatankuasa Etika Penyelidikan Perubatan JEPP (*Medical Research Ethics Committee MREC*). Bersama-sama ini disertakan surat kebenaran MREC (Lampiran 1) dan kertas kajian (*protocol*) / makluman ringkas projek (Lampiran 2).

3. Pegawai dari fasiliti YBhg Dato'/Tuan/Puan yang terlibat dalam penyelidikan ini adalah seperti berikut:
   i. Dr Afzaninawati Suria Yusof

4. Fasiliti/Jabatan di tempat YBhg Dato'/Tuan/Puan yang diperlukan adalah seperti berikut:
   i. Ruang menunggu sebelum jumpa doktor
   ii. Ruang menunggu di kaunter mengambil darah

5. Aktiviti penyelidikan yang akan dijalankan di fasiliti YBhg Dato' / Tuan / Puan adalah seperti berikut:
   i. Mengenal pasti pesakit dengan penyakit kronik
   ii. Menjalankan temuduga dengan pesakit dengan mengunakan borang soal selidik

Kami berharap mendapat kebenaran YBhg Dato' / Tuan / Puan.

Sekian, terima kasih.

Kajian akan dimulai selingga Disember 2018
Sekiranya extension of the study, kelulusan
baru perlu dihantar semula selepas 7/3/19.

Saya yang menurut perintah,

.....
(Dr Annaletchumy Loaganathan)
s.k.
<Dr Chee Hui Ping>
<Dr Afzaninawati Suria Binti Yusof

1/11/18

Kampar Campus : Jalan Universiti, Bandar Barat, 31900 Kampar, Perak Darul Ridzuan, Malaysia
Tel: (605) 468 8888 Fax: (605) 466 1313
Sungai Long Campus : Jalan Sungai Long, Bandar Sungai Long, Cheras, 43000 Kajang, Selangor Darul Ehsan, Malaysia

MAKLUMBALAS PERMOHONAN KEBENARAN PENGGUNAAN KLINIK KESIHATAN KAMPAR UNTUK MENJALANKAN PENYELIDIKAN

Tajuk Penyelidikan : The Prevalence and Factors Associated with Crude Herbs use Among Patients with Chronic Diseases: A Cross-sectional Survey in Combination with Laboratory Analysis

Nama dan Jabatan Ketua Penyelidik : Dr. Annaletchumy Loganathan

Pihak hospital/institusi dengan ini membuat keputusan seperti berikut : -

☑ Membenarkan projek penyelidikan dijalankan

☐ Tidak membenarkan projek penyelidikan dijalankan

"BERKHIDMAT UNTUK NEGARA"

Saya yang menurut perintah

..... 1/11/18

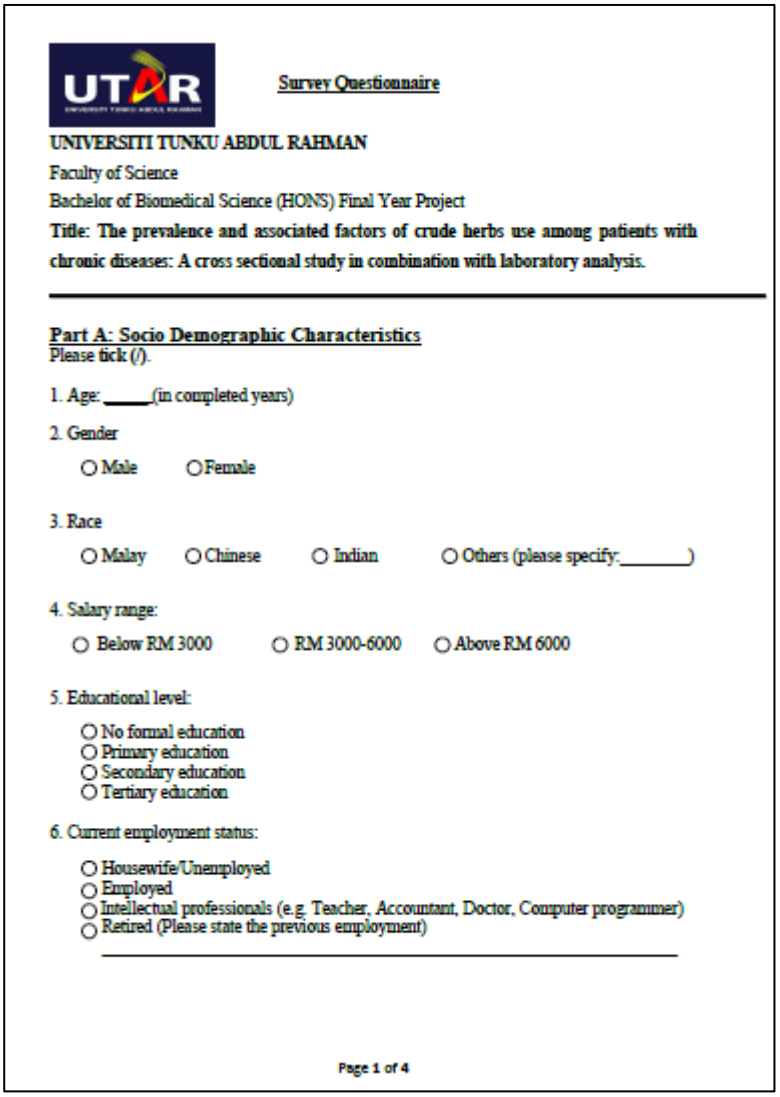
(<Ketua Jabatan di mana penyelidikan akan dijalankan>)

DR AFZANINAWATI SURIA BINTI YUSOF
No. Pendaftaran: 40322
Pakar Perubatan Kesihatan Awam Gred UD54
Pengawai Kesihatan Daerah Kampar
Pejabat Kesihatan Daerah Kampar

.....

(<Nama Pengarah >)

APPENDIX D



Survey Questionnaire

UNIVERSITI TUNKU ABDUL RAHMAN

Faculty of Science

Bachelor of Biomedical Science (HONS) Final Year Project

Title: The prevalence and associated factors of crude herbs use among patients with chronic diseases: A cross sectional study in combination with laboratory analysis.

Part A: Socio Demographic Characteristics

Please tick (/).

1. Age: _____(in completed years)

2. Gender

○ Male ○ Female

3. Race

○ Malay ○ Chinese ○ Indian ○ Others (please specify:_____)

4. Salary range:

○ Below RM 3000 ○ RM 3000-6000 ○ Above RM 6000

5. Educational level:

○ No formal education
○ Primary education
○ Secondary education
○ Tertiary education

6. Current employment status:

○ Housewife/Unemployed
○ Employed
○ Intellectual professionals (e.g. Teacher, Accountant, Doctor, Computer programmer)
○ Retired (Please state the previous employment)

_____

Page 1 of 4

Part B: Medical History

Please **circle** the answer that best represents your medical history*.

No.	Medical History	Yes	No	Not sure
1	Asthma	Y	N	NS
2	Cancer (Type of cancer:_____)	Y	N	NS
3	Cardiovascular disease (e.g. *Heart disease*)	Y	N	NS
4	Diabetes mellitus (*High blood glucose*)	Y	N	NS
5	Dyslipidemia (*Abnormal amount of lipids (cholesterol/fat) in blood*)	Y	N	NS
6	Hypertension (*High blood pressure*)	Y	N	NS
7	Hyperuricaemia (*High level of uric acid in blood*)	Y	N	NS
8	Kidney disease (*Kidney do not work effectively*)	Y	N	NS
9	Leukemia	Y	N	NS
10	Hepatitis, Jaundice, Liver disease	Y	N	NS
11	Migraine or recurrent headaches	Y	N	NS
12	Muscle pain (*Due to tension, over work, or muscle injury from exercise*)	Y	N	NS
13	Obesity (*High body fat. BMI ≥ 30*)	Y	N	NS
14	Parkinson's disease (*Tremor, muscular rigidity, and slow, imprecise movement*)	Y	N	NS
15	Peptic ulcer (*Burning stomach pain*)	Y	N	NS
16	Stroke	Y	N	NS
17	Thyroid disease	Y	N	NS
18	Urinary infection (Chronic type: *Infection involving the kidneys, ureters, bladder, or urethra*)	Y	N	NS
19	Falls/with injury in the past 6 to 12 months (an unexpected event in when a person comes to rest on the ground, floor or lower level).	Y	N	NS
20	Osteoporosis	Y	N	NS
21	Others (Please list out):			

** The chronic diseases were adopted from related literature in conjunction with the objective of this study.*

Part C: The use of crude herbs

1. Are you taking any medicines prescribed by the physician/pharmacist?
   ○ Yes ○ No

2. Are you consuming any crude herbs for treatment purposes?
   (Crude herbs: the raw plant, before it is processed or dried)
   ○ Yes (Proceed to question 3 and the subsequent questions)
   ○ No (Thank you for your participation)

3. Reason for taking crude herbs?
   ○ Recommended by physicians/doctors/pharmacists/medical officers
   ○ Recommended by family/relatives/friends
   ○ Belief on the efficacy of crude herbs
   ○ There is no side effects caused by crude herbs
   ○ Others (Please state): _____

4. In conjunction with your presribed medicines and crude herbs, are you taking any dietary supplements? (e.g. herbal medicine, vitamins, bird nest, ginseng extract)
   ○ Yes ○ No

5. What kind of crude herbs are you taking currently? (the common)

Crude herbs	Herbs' part	Preparation methods	Reasons
i._____	_____	_____	_____
_____	_____	_____	_____
_____	_____	_____	_____
ii. _____	_____	_____	_____
_____	_____	_____	_____
_____	_____	_____	_____

6. How long have you been using the crude herbs?

   ○ Less than a completed year
   ○ 1-3 completed years
   ○ More than 3 completed years

7. How do you get the information on the use of crude herbs to control the chronic diseases? (You may tick more than one)
   - ○ Books/Magazines
   - ○ News paper
   - ○ Television
   - ○ Internet websites
   - ○ Pharmacists
   - ○ Store clerk/sale assistant
   - ○ Specialist in Chinese/Malay/Indian traditional medicine
   - ○ Friend/Neighbour
   - ○ Spouse
   - ○ Relatives/mother/sisters
   - ○ Doctor
   - ○ Others
     Please state: _____

Thank you for participating in our research.

The End

APPENDIX E

Table 3.1: The list of chemicals and solvents used for the study.

Chemicals and Solvents	**Company, Country**
Acetone	QRec, Thailand
Bromine water	Bendosen, Malaysia
Ethanol	RCI Labscan, Malaysia
Ethyl acetate	RCI Labscan, Malaysia
Hexane	QRec, Thailand
Hydrochloric acid	GENE Chemical, Malaysia
Iodine	Nacalai Tesque, Japan
Iron (III) chloride powder	GENE Chemical, Malaysia
Potassium iodide powder	GENE Chemical, Malaysia
Sodium hydroxide powder	GENE Chemical, Malaysia
Sulphuric acid	DUSKAN Pure Chemical, India

APPENDIX F

Table 3.2: The list of equipments used for the study.

Laboratory Equipment	Company, Country
Blender	WARING, United States
Electronic balance	OHAUS Scot Pro, United States
Fluorescence analysis cabinet	Spectroline, United States
Freezer (-20°C)	Toshiba, Malaysia
Fume hood	Qinos, Malaysia
Hot plate stirrer	LMS, Japan
Incubator	Memmert, German
Refrigerator (4°C)	Samemax, Malaysia
Rotary vacuum evaporator	BUCHI, India
Shaker incubator	YIHDER, Taiwan
Sonicator	Elmasonic, United States