

# Antioxidant and Anti-Cancer Properties of Underutilized Fruits and Herbs in Kuala Kurau, Perak, Malaysia

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**ABSTRACT:** Plants, particularly fruits and vegetables, have many phytochemicals that possess various bioactivities, including antioxidant and anticancer properties. Malaysian population generally consume a lot of tropical and sub-tropical fruits in their daily diet that are reported to be high in antioxidant components with strong potential scavenging activities. However, there are some underutilized fruits that grow abundantly in the region of Peninsular Malaysia, Sabah and Sarawak which may have potential benefits towards human health. Underutilized fruits and herbs have potential functional properties but generally lack national recognition or attention. Reasons why these fruits are classified as underutilized are due to the lack of promotion, minimal planting area and having an economic potential that has not been fully explored. Therefore, extensive efforts have been made by researchers in promoting the consumption of fruits and vegetables due to the many health advantages, including prevention of chronic diseases such as cancer. Evidence shows that free radicals are responsible for the damage of lipids, proteins and nucleic acid in cells could lead to these common diseases. According to Malaysian Food Pyramid, individuals are encouraged to consume at least 2 servings of fruit per day. The replacement of synthetic antioxidants by natural sources may play a role in maintain health and have benefits for emulsions in food system. Therefore, probably it would be possible to discover more sources of natural antioxidant and anti-cancer properties with the work done on selected underutilized fruits and herbs.

**KEYWORDS:** antioxidant, anti-cancer, underutilized, fruits, herbs

## 1. INTRODUCTION

Fruits and vegetables have been a major part of food science as it conveys numbers of health benefits to human (Prasad et al., 2008). According to IPGRI (2002), underutilized plants are usually known as 'minor crops' as they are reduced in importance compared to main crops in the view of commercialization. In the other hand, underutilized can be described as per their potential to contribute to economic in overall. Sometimes, these species plant often said 'neglected' because they receive minor attention from research and development and few scientific approach about them (Osewa et al., 2013).

Recently, the study on free radical chemistry field has received continuous attraction. Those free radicals reactive oxygen species (ROS) and reactive nitrogen species are produced by the body via several ways. A free radical is defined as molecule which contains an unpaired electron in its outer orbital. These radicals are unstable and highly reactive. They become an oxidant or reductant by donating or accepting an electron by other molecules (Lobo et al., 2010). Antioxidant is known as a substance which delays or inhibits the oxidation of the substrate when present in low concentration compared to the oxidisable substrate. The function of antioxidant is to prevent cellular damage which occurs due to free radicals chemical reactions (Young and Woodside, 2001). Cancer is becoming more viral in worldwide. It is a condition of abnormal cells growth in body which can cause death. The pathogenesis of cancer cells are often attack and kill the normal cells (Prakash et al., 2013). Anticancer is a compounds that can treat or prevent types of cancer. The process of cancer is complex and composed of different steps which are the initiation, promotion and progression. The initiation phase is where the carcinogenic agent enters the cell mainly in nucleus and DNA which will results in mutation and causing toxic effect. This stage is irreversible but can be prevented by phase I and phase II metabolizing enzymes which converts the carcinogen to less toxic soluble products. Followed, the promotion phase which is the multiplication of cancer initiated cells. This phase is reversible and gives rise to pre-cancerous cells. The major part of this phase is the cell multiplication and apoptosis. The last stage is the cancer progression stage whereby the cells slowly convert to malignant form. During this stage, metastasis and invasion also occurs through angiogenesis process with the help of specific growth factors and receptors which leads to the spread of cancer (Dayem et al., 2016).

The characteristics of cancer cells in being highly concentrated with reactive oxygen species (ROS) and the altered redox action have been an added advantage for the medicinal benefits in current trends. At the late stage of cancer, the cells often display various genetic alteration and oxidative stress. This condition brought the researcher to find ways to eliminate the cell by pharmacological activity which is the ROS scavenging systems (Trachootham et al., 2009).

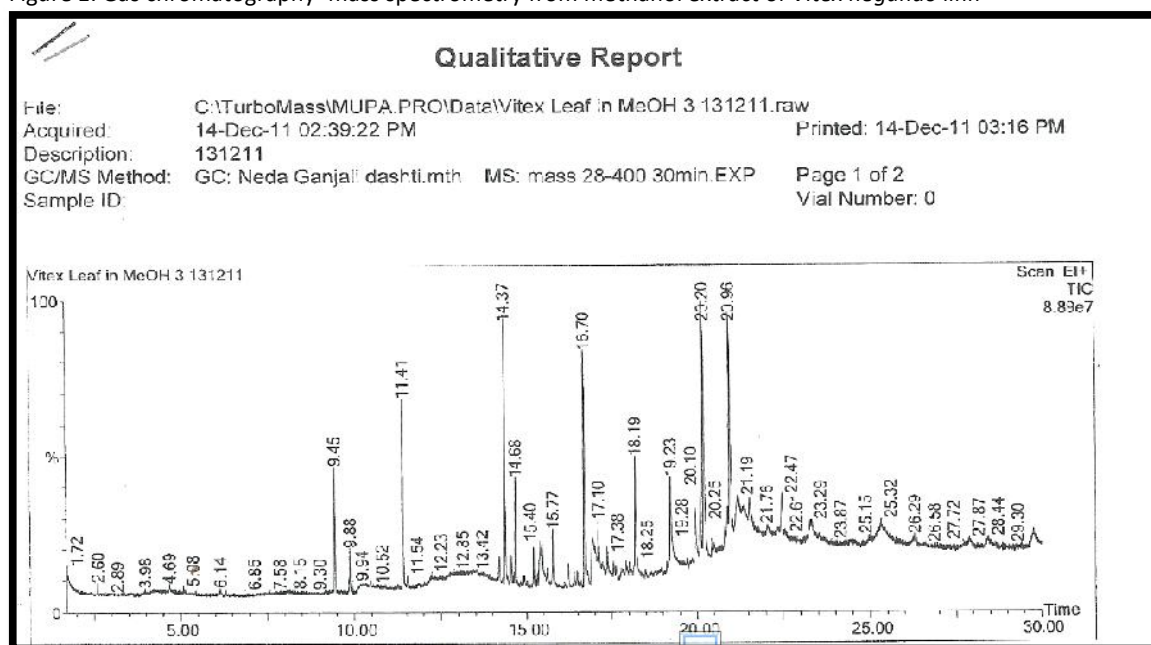
**Medicinal plant**

Medicinal plants have received vast interest due to its unique characteristic of being medically and biologically important in these decades. Literatures have been collected on the application of medicinal plant in traditional and modern medicines field (Shukla and Mehta, 2015). The presence of bioactive compounds such as phenolic acids, flavanoids, lignans, tannins, coumarins, alkaloids, terpenoids and quinones in medicinal plants aids in treatment of cancer where it promotes significant antioxidant activities.

***Vitex negundo***

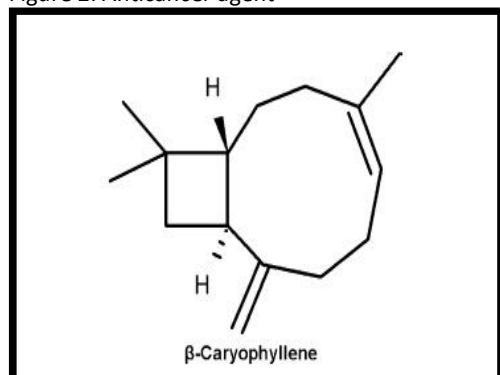
The plant *Vitex negundo* is an aromatic, large and sometimes small tree which measure 3 meters in height and it belongs to Verbenaceae family. This plant is rich with various polyphenolic compounds such as terpenoids, glycosidic iridoids and alkaloids (Kumar et al., 2010). Many in vitro studies have been done to determine the antioxidant capacity of this plant since numbers of literature reported that polyphenolic compounds are responsible for antioxidant activity (Tagne et al., 2014).

Figure 1. Gas chromatography–mass spectrometry from methanol extract of *Vitex negundo linn*



The result shows present of caryophyllene, D-viridiflorol,  $\beta$ -ionone methyl, (E) - phytol and 1-triacontanol.

Figure 2. Anticancer agent



$\beta$ -Caryophyllene is compound in group of sesquiterpene bioactive compound. This component often found in essential oils of medicinal plant and due to its woody and spicy odour, it is commonly used as fragrance and flavouring agents in many different formulations years ago. Besides, it also has contributed to medical field as an anticancer agent due to its properties to stimulate the detoxifying enzymes or to improve the natural killer cell activity (Di Sotto et al., 2010).

*Vitex negundo* can be a choice for cancer treatment since the compounds found in the extract shows the presence of caryophyllene, D-viridiflorol,  $\beta$ -ionone methyl, (E)- phytol and 1- triacontanol. Based on previous literature and other literature, presence of caryophyllene and other compounds such as phytol and linolenic acid could promote the anticancer activity (Kumar et al., 2010; Jananie et al., 2011). Moreover, the presence of  $\beta$ -ionone in the extract also suggest that the plant could be an alternative for cancer treatment whereby the in vitro and in vivo study done by Janakiram et al., (2008) stated that  $\beta$ -ionone possess anticarcinogenic properties and can further develop for treatment and prevention of colon cancer.

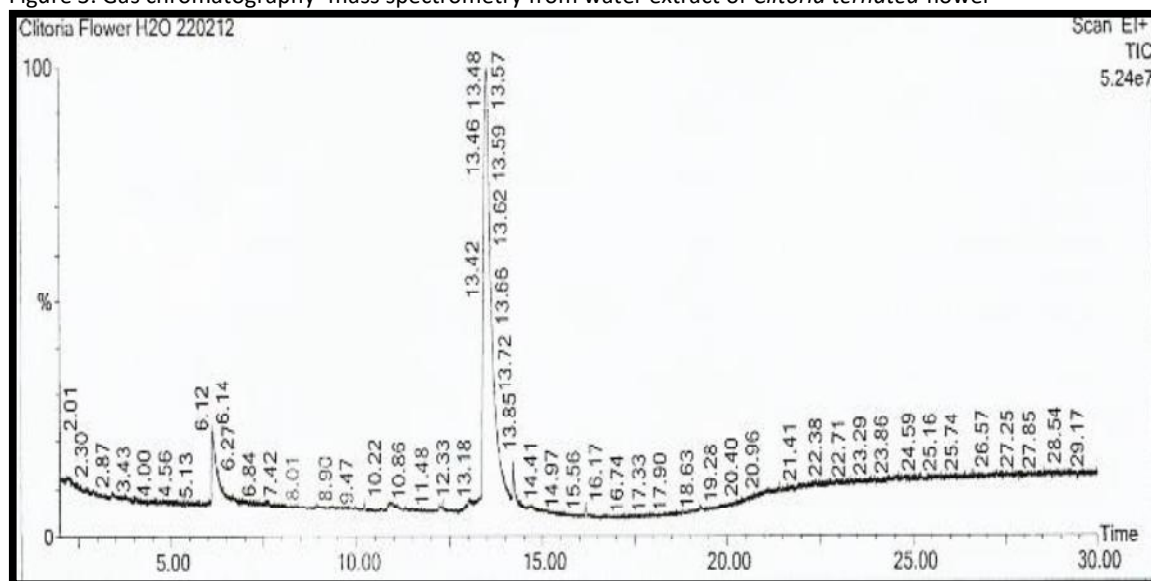
#### ***Clitoria ternatea***

The flower from the family of Fabaceae, butterfly pea or blue pea shaped flower which is vivid blue in colour size ranging from 1 to 2 inches long composed of wavy-rimmed standard and white centre. The flower *Clitoria ternatea*, also locally called as Bunga Telang and commercially used as food colouring in Nasi kerabu, signature food at Kelantan, Malaysia and some desserts called Kueh Tekan in Baba and Nyonya culture. The colour of the flowers was used as colouring purpose without knowing its medicinal value (Rabeta and Nabil, 2013).

#### Antioxidant properties study

Rabeta and Nabil, (2013) have conducted a study on flowers and leaves of *Clitoria ternatea* and *Vitex negundo* Linn to determine the total phenolic compounds (TPC) and 1, 1-diphenyl-2-picrylhydrazyl (DPPH) scavenging activity. Two types of extracts were used in this study. Those are methanol and water extraction. The result of TPC shows that methanol extract is more effective on extracting phenolic compounds compared to water. Furthermore, the *Vitex negundo* Linn is richer with TPC when compared to *Clitoria ternatea* but similarly, both plant leaves contained high amount of TPC when compared with the flowers. Water soluble natural antioxidant, the phenolic compound usually an aromatic ring composed of one or more hydroxyl substituent. The amount of TPC content in plant may be due to presence of several bioactive compounds which are often phenolic acid, flavanoids and anthocyanins. The higher TPC content in both plants is probably due presence of anthocyanin since it is a crucial water-soluble pigments in plants. DPPH assay is a method used to study the antioxidant level of a plant and it contains a nitrogen free radical which can be scavenge by the antioxidant in extract by donating the electron to make the molecule stable. The antioxidant result of both *Vitex negundo* and *Clitoria ternatea* shows that the *Vitex negundo* extract possess high scavenging activity towards DPPH radicals compared to *Clitoria ternatea* extract. The difference may be due to the presence of high TPC amount in *Vitex negundo* plant.

Figure 3. Gas chromatography–mass spectrometry from water extract of *Clitoria ternatea* flower



The result shows that presence of pentanal (14.3%) and mome inositol (38.7)

### Anticancer properties study

Neda et al., 2013 have done study on *Clitoria ternatea* flower for anti-proliferative and to find out the chemical composition. The cytotoxic effect was studied using six normal and cancer- origin cell line. The compositions of the water extract of the flower using GCMS showed that the extract contain high amount of pentanal and mome inositol. The anticancer activity was observed in water extract which support that this may due to the presence of the compound pentanal and mome inositol.

Comparatively, study done by Vucenic and Shamsuddin, (2003) on cancer inhibition by inositol and combination form, state that the combination of inositol and inositol hexaphosphate (IP<sub>6</sub>) could enhance the anticancer activity by reducing the cell proliferation and increases the cancer cells differentiation which results in conversion of normal cell.

### ***Ocimum tenuiflorum***

Throughout the old world and especially in the tropics *Ocimum tenuiflorum* otherwise known as Tulsi or Holy Basil is cultivated abundantly for religious and medicinal purposes (Kumar et al., 2013). *Ocimum sanctum* L. (Tulsi) is an erect, much branched sub-shrub 30–60 cm tall, with simple opposite green or purple leaves that are strongly scented and hairy stems. Leaves have petiole and are ovate, up to 5 cm long, usually somewhat toothed. Flowers are purplish in elongate racemes in close whorls (Pattanayak et al., 2010).

### Antioxidant properties study

Rabeta and Lai, (2013) have done study to evaluate the effect on of drying, fermented and unfermented tea of *Ocimum tenuiflorum* on antioxidant capacity. Different drying methods like freeze drying and vacuum drying of fermented and unfermented tea was done. Then, the samples were studied for their antioxidant properties using ferric reducing antioxidant power (FRAP) and DPPH assay. Besides, the TPC, total flavanoid content and condensed tannin content were also studied. The results are shown below in table.

Table 1. Antioxidant properties using ferric reducing antioxidant power (FRAP) and DPPH assay of *Ocimum tenuiflorum*

Sample	Fresh	Freeze	Vacuum	Fermented Tea	Unfermented Tea
DPPH (% inhibition)	38.09 <sup>b</sup> ±0.24 <sup>b</sup>	74.97 <sup>e</sup> ±0.47	24.16 <sup>a</sup> ±1.15 <sup>a</sup>	47.67 <sup>c</sup> ±0.16	72.20 <sup>d</sup> ±0.32
FRAP (µM Fe(II)/g) <sup>A</sup>	33.18 <sup>a</sup> ±0.53 <sup>a</sup>	98.41 <sup>b</sup> ±1.20	601.91 <sup>d</sup> ±5.36	371.24 <sup>c</sup> ±3.62	2912.8 <sup>e</sup> ±7.81
Total Phenolic Content (mg GAE/100 g) <sup>B</sup>	273.40 <sup>a</sup> ±2.01	1142 <sup>b</sup> ±0.06	5957.1 <sup>e</sup> ±5.85	3216.7 <sup>c</sup> ±11.02	4416.1 <sup>d</sup> ±15.76
Total Flavonoid Content (mg CE /100 g) <sup>C</sup>	109.02 <sup>a</sup> ±3.12	378.81 <sup>b</sup> ±9.17	3918 <sup>d</sup> ±12.51	2411.1 <sup>c</sup> ±2.46	4429.2 <sup>a</sup> ±16.65
Total Condensed Tannins (mg CE /100 g) <sup>C</sup>	51.73 <sup>a</sup> ±5.98	140.15 <sup>c</sup> ±5.44	117.14 <sup>b</sup> ±3.45	108.8 <sup>b</sup> ±7.76	344.37 <sup>d</sup> ±7.20
Total Anthocyanins (mgc-3-gE/100 g) <sup>D</sup>	16.29 <sup>b</sup> ±2.39	n.d	0.44 <sup>a</sup> ±0.10	n.d	n.d

\*Letters followed the same letter are not statistically significant from each other at  $P < 0.05$ ; n.d not detected; means  $\pm$  standard deviation; not detected. A Iron (II) equivalents; B Gallic acid equivalents; C Catechin equivalents; D cyanidin-3-glucoside equivalents.

Based on the results obtained, the antioxidant activity of the dried leaves both freeze and vacuum dry are higher compared to the fresh leaves. This may due to, the water evaporates at lower temperature and the truth about oxygen-free condition which also being a reason that oxidation process is less likely to occur thus preserving the antioxidant compounds.

Anticancer properties study

*Ocimum tenuiflorum* exhibited the anticancer activity against some cancer origin cell lines especially MCF-7 (hormone dependent breast cancer cell line). It can be explained through the significantly decreased cell viability of MCF-7 with the treatment of methanolic plant extraction at particular concentrations. The estrogen-dependent proliferation of MCF-7 can be influenced by polyphenols (compounds found abundantly in natural plant food sources that have antioxidant properties).

#### **Fruits**

The botanical view of fruits, means fleshy or dry ripened ovary of a plant enclosing the seed. But fruits usually understood as the ripened ovaries that are sweet which frequently consumed as an appetizer or desserts. Decades ago, fruits were only ate for its sweetness and believe to be less toxic because its not bitter and the sugar molecules (glucose, fructose and sucrose) present make the fruits sweet and also provide energy (Mann and Truswell, 2007).

#### ***Artocarpus camansi***

The fruit *Artocarpus camansi*, also known as breadnut usually considered to be in the form of seeded breadfruit, *Artocarpus altilis*. The fruit is oblong or round sometime has spiky skin with little pulp inside. The fruit are well recognized for the nutritious seeds (Rabeta and Syafiqah, 2016).

Antioxidant properties study

Recently, Rabeta and Syafiqah, (2016) conducted a study to evaluate the antioxidant activity in conjunction with the TPC of the core, pulp and seed of the breadnut. The parts of the breadnut were extracted with methanol and TPC and DPPH assay were done. The results showed that the seeds extract of methanol has the higher amount of TPC (22.1 mg GAE per gram seed) compared to other parts. Likely, the antioxidant activity of the seed is higher than other which means, this probably due to the TPC in the seeds.

#### ***Manilkara zapota***

This fruit is an underutilized tropical fruit generally known as “sapota” in India and “chiku” in Malaysia is the member of *Sapotaceae* family. The fruits have two distinct features which are hard, gummy and tannin rich when immature and soft, juicy and sweet when ripe. Most of this fruits are eaten fresh in Malaysia (Salleh et al., 2016).

Antioxidant properties study

Woo et al., (2013) have done study to evaluate the effect of extraction conditions on the antioxidant properties and TPC of the fruits. The study was done by subjecting the fruit samples to different concentrations of ethanol (0 – 100%), the extraction periods (1 – 5 h) and the temperature (25 – 60 °C). Based on the evaluation, the optimal conditions for the extraction was 4 h time at 60°C using 40% ethanol for pulp and 2 h extraction at 40°C using 80% ethanol for peel. This optimum condition respectively gives out best antioxidant activity and high TPC in pulp and peel.

#### ***Syzygium species***

*Syzygium* is a plant genus from the family of Myrtaceae. This family includes many famous species that cultivated for the delicious fleshy fruits. The fruits from this family usually consumed fresh or made into fruit salads or also cooked or preserved for individual use. The focused species are three types, which are the *S.aqueum*, *S. malaccense* and *S.malaccense* L (Rabeta et al., 2013).

#### ***Syzygium aqueum***

*S. aqueum* is the local fruits in Malaysia and Indonesia from the family of Myrtaceae. The local name of the fruit is the water apple or *jambu air* (Manaharan et al., 2012).

#### ***Syzygium malaccense***

*Syzygium* means yoked together commonly referred to the paired leaves, is derived from Greek word through Latin. Locally, in Malaysia, *Syzygium malaccense* fruit is called as jambu susu (Rabeta et al., 2013).

#### ***Syzygium malaccense* L**

*Malay apple* is the local name, native from India and Malaysia. This fruit is the member of Myrtaceae family (Savitha et al., 2011).

#### Antioxidant properties study

Lim and Rabeta, (2013) done a study to find out the antioxidant capacity of three different fruits which the water apple, milk apple and malay apple. DPPH and FRAP assay methods were used in this to determine the antioxidant capacity. The samples used were freeze dried and were extracted with two different solvent acetone and water. Among this, the milk apple of freeze dried, acetone extracted possesses high antioxidant capacity in both DPPH and FRAP assay compared to the water extracted. The lowest antioxidant activity can be observed in malay apple water extract for both DPPH and FRAP assay. Thus, this suggesting, this sample could be used as natural antioxidant as an alternative to the synthetic antioxidant.

#### Anticancer properties study

Rabeta et al., 2013 have studies the anticancer activity of the *Syzygium* fruits mainly the *S.aqueum*, *S. malaccense* and *S.malaccense* L. the anticancer properties were studied using two types of cancer-origin cell. The extraction used were the aqueous and methanol. This study was done by using colorimetric MTT assay over 24, 48 and 72 hours. There were no significant change during the 24 and 28 hours but, antiproliferation activity was observed in water apple and malay apple at the 72 hours. But, no activity was observed in milk apple on both cancer and non cancer cell lines in 72 hours. Thus, the results suggesting that, the anticancer activity may be due to presence of compound which responsible for anticancer properties and numerous studies shows that plant bioactive compound, polyphenol could prevent the cancer metastasis via common pathway.

Similarly, Wongwattanasathien et al., (2010) have done a research to study the antimutagenicity. The result obtained by Rabeta et al., (2013) supports the findings that malay apple has the antiproliferative properties on the same cell line, MCF-7.

#### ***Cynometra cauliflora***

Locally known as Nam-nam fruit or some called as "katak puru" because of the physical appearance of being rough and wrinkled is the member of the Fabaceae family (Rabeta and Faraniza, 2013).

#### Antioxidant properties study

Rabeta and Faraniza, (2013) done a research on determining the TPC and antioxidant level using FRAP assay of the fruit *Cynometra cauliflora*. The fruit was extracted using two solvents, methanol and distilled water. The results obtained shows that, the TPC of the fruits are higher in methanol extract compared to the water extract. The antioxidant assay using FRAP method also supports the TPC value where, the methanol extract exhibit potent antioxidant activity with high value compared to the water extract. It can be said that the antioxidant activity are dependent on TPC.

## **2. CONCLUSION**

It could be an added advantage for medical field if researcher focus to find out more health benefits of underutilized plant which available worldwide as it can be promising sources in future drugs development.

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