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Phytochemical Profiling and Invitro Antioxidant Potential of *Clitoria ternatea* Linn seeds.

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Abstract:
Clitoria ternatea commonly known as Butterfly pea is a medicinal plant belonging to the family Fabaceae. It is used in ayurvedic medicine because of its multipotent bioactive molecules. All parts of the plant have medicinal properties and are used for hepatoprotective, cytotoxic, antifungal, antihyperglycemic, antihyperlipidemic, antimicrobial and antioxidant activity. The present study was aimed to investigate the preliminary phytochemical screening and invitro antioxidant potential of *Clitoria ternatea* seed extracts. For phytochemical profiling ethanol and aqueous solvents were used to detect the various compounds. In aqueous extract of *C. ternatea* Saponins, Qinines, Alkaloids, Flavonoids, Cardiac Glycoside Phenols and Anthocyanin were present. In ethanolic extract Quinines, Flavonoids, Cardiac Glycoside, Terpenoids, Phenols, Coumarin and Betacyanin were present. The presence of different phytocompounds in the plant extracts are responsible for the various pharmacological properties. The antioxidant potential was determined by DPPH free radical scavenging method. The seed extracts of *C.ternatea* showed strong antioxidant activity. The presence of significant amount of phytochemicals, phenolic and flavonoid contents in the seeds are responsible for the in vitro antioxidant activity of *Clitoria ternatea*. The study concluded that the *Clitoria ternatea* seeds could be a potential source of natural antioxidants for pharmacological preparations which can be used to prevent the oxidative stress related degenerative diseases.

Key words: *Clitoria ternatea*, Phytochemicals, Flavonoids, Phenolic compounds and Antioxidant activity.

Introduction

Medicinal plants are very important in health care of individuals and communities in many developing countries. These plants are believed to be much safer and are used in treatment of various ailments. In recent times, the use of medicinal plants is considered as a complementary and alternative therapies in combination with other treatments (Kawamura and Muraoka,2018). Plants contain numerous antioxidants which help to confer protection against free radicals associated diseases. The research on phytochemicals and use of phytochemicals is increasing more because of the harmful side effects of the synthetic compounds. According to World Health Organization (WHO), medicinal plants would be the best source to obtain variety of drugs. About 80% of individuals from developed countries use traditional medicines, which has compounds derived from medicinal plants. However, such plants should be investigated to better understand their properties, safety, and efficiency (Arunkumar and Muthuselvam, 2009).

Clitoria ternatea is commonly known as butterfly pea, belongs to Fabaceae family. It is a tropical plant, perennial twinning herb bearing blue or white flowers (Gomez and Kalamani, 2003). It is used in ayurvedic medicine because of its multipotent bioactive molecules. Many plant extracts medicinal values lies in some of it's chemical substances that produce a definite physiological action on the human body. The most important of these bioactive constituents of plants are alkaloids, tannins, flavonoids, glycosides and phenolic compounds (Ghasemzadeh et al., 2010). In India *Clitoria ternatea* (CT) was traditionally used as medhya rasayan (brain tonic) for neurological disorders during ancient times. The plant extract of *Clitoria* has potential medicinal values such as anti-helmintic (Khadatkar et al., 2008), antipyretic, anti-inflammatory, antibacterial (Kamilla et. al., 2009), analgesic (Devi et. al., 2003), anxiolytic, antidepressant, anticonvulsant, sedative, hypoglycemic, anticancer properties (Jain et al., 2003, Sharma et. al., 1990).

In recent decades, many researchers are interested in medicinal plants for evaluation of antioxidant phytochemicals such as phenols, flavonoids and tannins which have received more attention for their potential role in prevention of human diseases (Upadhyay, et.al.,2010). Hence the present study is aimed to investigate the phytochemical screening and the antioxidant activity of seed extracts of *Clitoria ternatea*.

Materials and Methods

Collection of plant material

The plant *Clitoria ternatea* was collected from the terrestrial garden located at Iyyapanthangal, Chennai. The plant was identified and authenticated by the taxonomist at the Department of Botany, Queen Mary's College, Chennai and the herbarium has been deposited at the college for further reference. The fresh pods from the plant were collected and the seeds were removed from pods, shade dried and ground into a course powder. The powder was stored in an airtight container and used for further analysis.

Preparation of plant extract

About 10g of seed powder was taken and soaked in aqueous and ethanol (75%) extract for 1 minute and stored for 24 hours at room temperature. The extracts were filtered using Whatman filter paper No: 1 in a funnel. Then the extracts were stored in airtight container at 10°C in a refrigerator and kept for further use.

Phytochemical analysis

The aqueous and ethanolic seed extracts of *Clitoria ternatea* were subjected to different tests to identify the nature of bioactive chemical constituents present in the plant material. The crude extracts were screened qualitatively for the phytochemical constituents using the standard procedure of Trease and Evans (2005).

Free Radical Scavenging Activity:

The antioxidant activity of the extracts was evaluated through the DPPH (1,1 diphenyl-2 picryl hydrazyl) radical scavenging assay as described by Lee et.al., (2003).100 µl of seed extract was mixed with 2.7 ml of ethanol and then 200 µl of 0.1% ethanolic DPPH was added. The suspension was incubated for 30 minutes in dark condition. Initially, absorption of blank containing the same amount of ethanol and DPPH solution was prepared and measured as a control. Subsequently, at every 5 minutes interval the absorption

maximum of the solution were measured using a UV double bean Spectra scan (Chemito,India) at 517 nm. The antioxidant activity of the sample was compared with known synthetic standard of (0.16%) of butylated hydroxyl toluence (BHT). The experiment was carried out in triplicates. The capacity of scavenging free radical was calculated as scavenging activity (%) =

$$\frac{\text{(Absorbance of control) - (Absorbance of sample)}}{\text{Absorbance of control}} \times 100$$

Results and Discussion

Phytochemical screening of medicinal plants is very important in identifying new sources of therapeutical and industrial importance (Salhan et al., 2011). The phytochemical composition of seed extracts of *Clitoria ternatea* were given in **Table1**. The results revealed the presence of various secondary metabolites in the seed extracts of *Clitoria ternatea.*

Table:1 Phytochemicals screening of seed extracts of *Clitoria ternatea.*

S.No	Phytochemicals	Different Solvents Used	
		Aqueous	Ethanol
01	Tannin	_	_
02	Saponins	+	_
03	Quinines	+	+
04	Flavonoids	+	+
05	Alkaloids	+	_
06	Glycoside	_	_
07	Cardiac Glycoside	+	+
08	Terpenoid	+	_
09	Phenols	+	+
10	Steroids	_	_
11	Coumarin	_	+
12	Anthocyanin	+	_
13	Betacyanin	_	+

Ethanol and aqueous solvents were used to detect the various compounds. In aqueous extract Saponins, Qinines, Alkaloids, Flavonoids, Cardiac Glycoside Phenols and Anthocyanin were present. In ethanolic extract Quinines, Flavonoids, Cardiac Glycoside, Terpenoids, Phenols, Coumarin and Betacyanin were present. The presence of different phytocompounds in the plant extracst are responsible for the various pharmacological properties. Manjula et al., (2013) were reported that the qualitative analysis of *C. ternatea* showed the presence of many bioactive compounds such as alkaloids, tannins, glycosides, resins, steroids, saponins, flavonoids and phenols. The flower petals also contain a wide variety of polyphenols, the main polyphenol constituents found are anthocyanins (Pasukamonset et al., 2016). Study has also

suggested that its flower possesses many health beneficial properties, such as tranquilizing effect, anti-inflammatory and antipyretic activities (Mukherjee et al., 2008).

All the parts of the herb (leaf, root, shoot and seed) is used as medicine. In traditional Ayurvedic medicine, it has been used for centuries as a memory enhancer, nootropic, antistress, anxiolytic, antidepressant, anticonvulsant, tranquillizing and sedative agent (Mukherjee. et.al.,2008). It is also used in neurological disorders (Gupta, et.al., 2010). Phytochemical screening of the roots shows the presence of ternatins, alkaloids, flavonoids, saponins, tannins, carbohydrates, proteins, resins, starch, taraxerol and taraxerone (Uma et.al., 2009). A wide range of secondary metabolites including triterpenoids, flavonol glycosides, anthocyanins and steroids has been isolated from *Clitoria ternatea* Linn. (Mukherjee et.al.,2008). Phytochemistry helps in standardizing the herbal preparations so as to get the optimal concentrations of known active constituents and in preserving their activities. Shekhawat and Vijayvergia (2010) studied the presence of metabolites in various plant parts of *C. ternatea*.

Antioxidant activity

Medicinal plants are considered as potential sources of antioxidant compounds. There is an increasing interest in the investigation of naturally occurring antioxidants from plants. The ethanolic extract of *Clitoria ternatea* was evaluated for its in vitro antioxidant activities by DPPH free radical method. The results of the study was given in **Table :2**

Table:2 DPPH free radical scavenging activity of *Clitoria ternatea*

Concentration(mg/ml)	Percent inhibition
20	33.33
40	51.51
60	66.66
80	77.77
100	100
IC 50	39.1

The results revealed that the activity was increased by increasing the concentration of the sample extract. The presence of phenols and flavonoids in the seed extracts are responsible for its high radical scavenging activity. An IC_{50} value is the concentration of the sample required to scavenge 50% of the free radicals present in the system. In the present study the IC_{50} value was found to be 39.1mg/ml. The phenols contain hydroxyl that are responsible for the radical scavenging effect mainly due to redox properties (Rice - Evans et.,al,1997). The high phenolic contents of the seed extracts can explain it high free radical scavenging activity. The results revealed that the selected plant materials have free radical scavenging activity and can be used as a source of antioxidants for pharmacological preparations. Phenolic compounds have strong in vitro and in vivo antioxidant activities associated with their ability to scavenge free radicals, break radical chain reactions and chelate metals. Many plant phenolics exhibiting antioxidant properties have been studied and proposed for protection against oxidation (Oktay et.al.,2003). Rai (2010) reported the presence of tannins, flavonoids and steroids in the ethanolic extract of *C. ternatea* that are known to be the reason for the antioxidative potential of the plants. Amol Patil and Vijay Patil (2011) reported the

evaluation of in vitro anti oxidant activity of seeds of blue and white flowered variety of C*litoria ternatea.* Methanol extract of seeds of white flowered variety showed more significant antioxidant activity as compared to blue flowered variety. In this study the seed extracts of C*litoria ternatea* had shown the antioxidant and free radical scavenging activity, which revealed that the plants can be used as a potent source of natural antioxidant.

Conclusion

The study revealed that the phytochemical analysis of seed extracts of *Clitoria ternatea* showed the presence of various phytochemicals such as Quinones, Flavonoids, Alkaloids, Cardio glycosides, Phenols, Coumarin and Betacyanins. Presence of these bioactive compounds are responsible for the medicinal properties of the plant. The seeds of *Clitoria ternatea* also shows prominent antioxidant activity. Thus this paper concluded that *Clitoria ternatea* as promising medicinal plant with wide range of pharmacological properties which could be utilized in several medical applications because of its effectiveness and safety.

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