



#### Article

# Environmental and Medicinal Importance of Butterfly Pea (*Clitoria ternatea* L.). A Review

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**Abstract**: *Clitoria ternatea* is rich in a wide range of phytochemicals that contribute to its medicinal properties. These compounds play a key role in the plant's biological activities, it is contained tannins, phlobatannin, carbohydrates, saponins, triterpenoids, phenols, flavanoids, flavonol glycosides, proteins, alkaloids, antharaquinone, anthocyanins, cardiac glycosides, Stigmast-4-ene-3,6-dione, volatile oils and steroids. Traditional medicine systems have used this plant to treat a variety of ailments such as indigestion, constipation, arthritis, skin diseases, liver and intestinal problems. The flowers of *C. ternatea* are used worldwide as ornamental flowers and traditionally used as a food colorant. *C. ternatea* flower extracts were found to possess antimicrobial, antioxidant, anti-inflammatory, cytotoxic and antidiabetic activities which are beneficial to human health. This review will highlight the environmental, chemical constituents and pharmacological effects of *Clitoria ternatea*.

**Key words**: Butterfly Pea, blue tea, *Clitoria ternatea*, flavonoids, anthocyanins, antimicrobial, antioxidant, anti-inflammatory, anticancer.

#### 1. Introduction

*Clitoria ternatea*, commonly known as butterfly pea; blue tea or blue pea, is a flowering plant celebrated for its vibrant blue flowers and various medicinal uses. This perennial herb belongs to the Fabaceae family and is native to tropical regions. *Clitoria ternatea* L., is classified according to **Kartesz (1994)** as follows in Table (1):

Kingdom: Plantae	Subclass: Rosidae
Subkingdom: Tracheobionta	Order: Fabales
Superdivision: Spermatophyta	Family: Fabaceae Lindl.
Division: Magnoliophyta	Genus: Clitoria L
Class: Magnoliopsida	Species: Clitoria ternatea L.

#### **Native Habitat**

*Clitoria ternatea* L. is believed to be native to the Indonesian island of Ternate, where it was first documented by botanist Carl Linnaeus. However, its exact origin is somewhat ambiguous, as it has been widely cultivated and naturalized across tropical and subtropical regions globally, including parts of Africa, Southeast Asia, and the Indian subcontinent (**Oguis et al., 2019**).

The plant thrives in a variety of environments, particularly in disturbed areas and human-altered landscapes. It is often found in sandy soils and savannas, demonstrating resilience to drought conditions. Its ability to self-pollinate and fix nitrogen makes it adaptable to various ecological settings, contributing to its widespread distribution (Fantz, 1981; Govaerts, 1999 and Oguis et al., 2019). *Clitoria ternatea* thrives in a range of environments, particularly in tropical and subtropical regions. It is often found in disturbed areas and is known for its adaptability to various soil types (Pengelly and Conway, 2000).

#### Morphological description of Clitoria ternatea

*Clitoria ternatea* features pinnately compound leaves that are arranged alternately along the stem. Each leaf typically consists of 5 to 7 leaflets, which can reach lengths of up to 5 cm. The leaflets are generally ovate to elliptic in shape, with a rounded apex and base. They exhibit a smooth texture and are glabrous (hairless) on both surfaces. The leaves display reticulate venation, which means that the veins form a network pattern. The upper epidermis is covered by a thick cuticle, providing protection, while the lower epidermis contains paracytic stomata, allowing for gas exchange. Beneath the upper epidermis, there is a layer of palisade cells and lignified xylem, contributing to the leaf's structural integrity and function in photosynthesis. *Clitoria ternatea* possesses a branched taproot system that allows it to access water and nutrients from deeper soil layers. This extensive root system is crucial for the plant's survival, especially during periods of drought, as it can endure dry conditions for 7 to 8 months. It has a taproot system with many slender lateral roots. Additionally, the roots are known to form nodules, which are essential for nitrogen fixation. These nodules host symbiotic bacteria that convert atmospheric nitrogen into a form that the plant can utilize, enhancing soil fertility and supporting the plant's growth in nutrient-poor conditions (**Tjitrosoepomo, 1985; Rugayah et al., 2004; Morris, 2009; Gupta, et al., 2010; Bishoyi and Geetha, 2013; Suarna and Wijaya 2021; Surya, et al., 2022 and Hasanah et al., 2023)** 

The flowers of *Clitoria ternatea* are zygomorphic (bilaterally symmetrical) and consist of five petals arranged in a distinctive pattern Fig (1) The calyx is composed of five sepals that are fused at the base, forming a tubular structure that supports the flower. The corolla features five petals, the largest petal, often dark blue, serving as the banner. Two smaller petals that resemble wings, typically less than half the size of the banner petal. Two petals that are fused together, forming a protective structure for the reproductive organs. While the predominant color is a striking blue, flowers can also be found in shades of white, purple, and pink, often with a gradient effect. The flower contains ten stamens, nine of which are fused (forming a tube) and one that is free. Each stamen has a pollen-bearing anther, typically white and consisting of four lobes. The ovary is monocarpellary, containing several ovules, and is topped by a long style with a bent tip. The fruit is a narrow, flattened legume that typically measures 40–130 mm in length and contains 6-10 seeds. The seeds are oblong and flattened, varying in color from olive brown to nearly black, and are shiny The seed is oval in shape and has a blackish or yellowish brown colour with a length range of 4.5-7.0 mm and 3-4 mm wide (**Mukherjee et al., 2008; Kosai et al., 2015; Jeyaraj et al., 2021 and Sarma et al., 2023).** 



Fig. (1). *Clitoria ternatea* flower

#### Ornamental importance of Clitoria ternatea

*Clitoria ternatea* L. is increasingly recognized for its ornamental value in gardens and landscapes (Jeyaraj, et al., 2021 and Islam et al., 2023). Here's an overview of its importance as an ornamental plant, supported by the references as follows:

# 1- Aesthetic appeal

*Clitoria ternatea* is particularly valued for its striking flowers, which can be deep blue, white, or even pink. The vivid colors and unique shape of the flowers make it a popular choice for decorative gardens and landscaping. The plant's ability to bloom throughout the year adds to its appeal, providing continuous color and interest in garden settings (**Staples, 1992**).

# 2- Versatile growth habit

This plant is a perennial climber that can grow up to 10 meters in length, making it suitable for various vertical gardening applications. Its climbing nature allows it to be used on trellises, fences, and arbors, enhancing the vertical space in gardens. Additionally, it can serve as ground cover, helping to suppress weeds and improve soil quality through its nitrogen-fixing properties (**Reid and Sinclair, 1980 and Staples, 1992**).

# 3- Low maintenance

*Clitoria ternatea* is known for its resilience and low maintenance requirements. It thrives in a variety of soil types and conditions, requiring minimal care once established. This makes it an ideal choice for gardeners looking for attractive yet easy-to-care-for plants (**Staples, 1992**).

#### Environmental Importance of Clitoria ternatea

As a leguminous plant, *Clitoria ternatea* contributes to:

1. Nitrogen fixation

As a member of the legume family (Fabaceae), *Clitoria ternatea* has the unique ability to fix atmospheric nitrogen into a form that plants can use, thanks to its symbiotic relationship with nitrogen-fixing bacteria (Rhizobia) in its root nodules. This process enhances soil fertility, making it beneficial for agricultural practices. By enriching the soil, it helps improve the growth of subsequent crops and reduces the need for synthetic fertilizers, which can have detrimental effects on the environment (**Oblisami, 1974; De Souza et al., 1996; Alderete-Chávez, et al., 2011 and Oguis et al., 2019).** 

#### 2. Soil erosion control

*Clitoria ternatea* is often used as a cover crop due to its vigorous growth and ability to form a dense mat. This characteristic helps prevent soil erosion, particularly in areas susceptible to runoff and degradation. By stabilizing the soil, it protects against the loss of topsoil, which is crucial for maintaining soil health and agricultural productivity (**Dayal et al., 2015**).

#### 3. Biodiversity support

The plant provides habitat and food for various pollinators, including bees and butterflies, contributing to local biodiversity. Its flowers attract these beneficial insects, which play a crucial role in pollination, thereby supporting the ecosystem's balance. The presence of diverse plant species, including *Clitoria ternatea*, can enhance ecosystem resilience against pests and diseases (**Raju and Ramana, 2021**).

# 4. Revegetation and restoration

*Clitoria ternatea* is utilized in ecological restoration projects, particularly in disturbed areas such as coal mines and degraded lands. Its ability to thrive in poor soil conditions and its nitrogen-fixing properties make it an ideal candidate for revegetation efforts. By improving soil quality and providing ground cover,

it aids in the recovery of ecosystems that have been damaged by human activities (Gamage et al., 2021).

# 5. Pest management

The plant has been noted for its potential as a natural insecticide. Extracts from *Clitoria ternatea* have shown antimicrobial and insecticidal properties, which can help manage pest populations without the adverse effects associated with chemical pesticides. This aspect is particularly important in sustainable agriculture, where reducing chemical inputs is a priority, cyclotide was an example of a protein-derived bioactive compound that was found almost in every part of butterfly pea and used as a natural pesticide (**Nguyen et al., 2011 and Oguis et al., 2019**).

# 6. Carbon sequestration

By promoting healthy soil and vegetation, *Clitoria ternatea* contributes to carbon sequestration. Healthy plants absorb carbon dioxide from the atmosphere, helping mitigate climate change. The plant's robust growth and ability to cover large areas enhance its capacity to sequester carbon effectively (**Gew et al., 2024**).

# **Phytochemical constituents**

Nutritional analysis of *C. ternatea* flowers identified the percentage of protein, fibre, carbohydrate and fat to be 0.32, 2.1, 2.2 and 2.5% respectively while the moisture content was found to be 92.4%. The flower was also found to have high content of calcium (3.09 mg/g), magnesium (2.23 mg/g), potassium (1.25 mg/g), zinc (0.59 mg/g), sodium (0.14 mg/g) and iron (0.14 mg/g) (Neda et al., 2013). The preliminary phytochemical screening showed that the plant contained tannins, phlobatannin, carbohydrates, saponins, triterpenoids, phenols, flavanoids, flavonol glycosides, proteins, alkaloids, antharaquinone, anthocyanins, cardiac glycosides, Stigmast-4-ene-3,6-dione, volatile oils and steroids (Kamilla et al., 2009, Rai et al., 2015 and Mukherjee et al., 2008). The fatty acid content of *Clitoria ternatea* seeds includes palmitic, stearic, oleic, linoleic, and linolenic acids. Seeds also contained cinnamic acid, anthoxanthin glucoside, a highly basic small protein named finotin, water-soluble mucilage, delphinidin 3, 3', 5'-triglucoside and beta-sitosterol (Kelemu et al., 2004, Husain et al., 1998, Macedo et al., 1992, Sinha, 1960 and Ripperger, 1978).

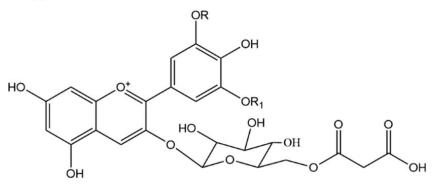


Fig. (2). Delphinidin 3-malonyl glucoside

Several studies investigated, identified and isolated the bioactive compounds from *C. ternatea* flower. The anthocyanin known as ternatins are blue in colour and are acylated based on delphinidin (Fig. 2). Their structures were characterised as malonylated delphinidin 3,3',5'-triglucosides having 3',5'-side chains with alternative D-glucose and p-coumaric acid units at R and R1 with a total of 15 (poly) acylated delphinidin glucosides identified in all the blue petal lines namely ternatins A1-A3, B1-B4, C1-C4 and D1-D3 while some studies have identified several other delphinidin derivatives (**Zakaria et al., 2018; Shen et al., 2016 and Nair et al., 2015**). Ternatins A1, A2, B1, B2, D1 and D2 are the six major anthocyanins present in the flowers (**Mukherjee et al., 2008 and Terahara et al., 1998**).

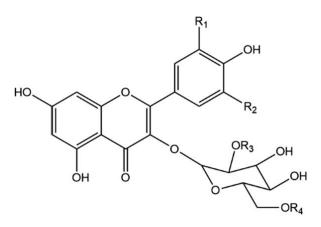


Fig. (3). Flavonol glycosides

The flavonols (Fig. 3) identified in the petals are fourteen kaempferol, quercetin and myricetin glycosides which consist of H or OH at R1 and R2 and with H, rhamnosyl or malonyl at R3 and R4 (**Mukherjee et al., 2008 and Kazuma et al., 2003**). Shen et al., (2016) identified various lipophilic compounds from *C. ternatea* being fatty acids (palmitic acid, stearic acids, petroselinic acids, linoleic acid, arachidic acid, behenic acid and phytanic acid), phytosterols (campesterol, stigmasterol,  $\beta$ -sitosterol and sitostanol) and tocols ( $\alpha$ -tocopherol and -tocopherol). Several other components such as mome inositol, pentanal, cyclohexen, 1-methyl-4-(1-methylethylideme) and hirsutene were identified by Neda et al. (2013). In addition to the identification of various anthocyanins and flavonol glycosides, other components such as 6"-malonylastragalin, phenylalanine, coumaroyl sucrose, tryptophan and coumaroyl glucose were determined (Zakaria et al., 2018).

#### Pharmacological effects

*Clitoria ternatea* flower contains a significant amount of phytochemicals which exhibits great antioxidant, antimicrobial, antidiabetic, anti-inflammatory and antiproliferative/anticancer properties (Lo´pez Prado et al., 2019; Mahmad et al., 2018; Nair et al., 2015; Rajamanickam et al., 2015 and Neda et al., 2013). Acute toxicity study using albino Wistar rats treated orally with aqueous ethanol extract (2000 mg/kg bodyweight) of the flower showed no signs of mortality or abnormality and there was no significant difference in the haematological values. The extract did not display acute toxicity effects and are safe for consumption (Srichaikul, 2018). *Clitoria ternatea* flowers can potentially be utilised as a functional food incorporated into various food products or even as a pharmaceutical supplement/drug combined with commercial drugs to improve treatment efficacy of patients.

#### Antioxidant activity

Oxidative stress plays a part in the development of chronic and degenerative illness such as cancer, autoimmune disorders, cardiovascular and neurodegenerative diseases. The discovery of antioxidants from natural sources is beneficial to human health (Admassu et al., 2018 and Pham-Huy et al., 2008). Various studies investigated the antioxidant activity of *C. ternatea* flowers using antioxidant assays such as 2,2-diphenyl-1-picrylhydrazyl radical (DPPH) radical scavenging, ferric reducing antioxidant power (FRAP), hydroxyl radical scavenging activity (HRSA), hydrogen peroxide scavenging, oxygen radical absorbance capacity (ORAC), superoxide radical scavenging activity (SRSA), ferrous ion chelating power, 2,2'-azino-bis(3-ethylbenzthiazoline-6-sulphonic acid) (ABTS) radical scavenging and Cu<sup>2+</sup> reducing power assays. *Clitoria ternatea* flower has been shown to have potent antioxidant activity (Table 2).

In the DPPH assay, 100% methanol extract of *C. ternatea* flower extract was found to be more potent than vitamin E (Nithianantham et al., 2013), whereas the water extract was found to be lower than ascorbic acid (vitamin C) (Chayaratanasin et al., 2015; Phrueksanan et al., 2014 and Iamsaard et al., 2014) Some studies investigated and compared the antioxidant activity (DPPH assay) of the extracts using different solvents in which the water extract was found to be more potent than 100% ethanol extract at 15 min extraction time (Kamkaen and Wilkinson, 2009). However, in another study the best extraction time was determined (6 h) for the water extract, 100% and 50% methanol extract in which the water extract and

50% methanol were found to be equally potent and had a higher activity than 100% methanol extract (Lo'pez Prado et al., 2019). The optimum condition was investigated using water extract in another study with and without ultrasound at a fixed temperature and liquid-solvent ratio in which the extraction with ultrasound was found to have higher antioxidant activity (Mehmood et al., 2019). The in vitro chemical assays to measure antioxidant activity (Table 2) need to be carefully interpreted as they bear no similarity to biological systems including the absorption of antioxidants by the human body (Gengatharan et al., 2015). In a cell based study, the water extract was found to potently inhibit 2,2'-azobis-2-methyl-propanimidamide dihydrochloride (AAPH)-induced hemolysis and oxidative damage of canine erythrocytes (Phrueksanan et al., 2014). In another study, the pre-treatment of human HaCaT keratinocytes with the water extract reduced UV-induced mitochondrial DNA damage (Zakaria et al., 2018). In a randomized crossover study, acute ingestion of *C. ternatea* flower extract/beverage was found to have increased plasma antioxidant capacity and the effect was further enhanced when consumed together with sucrose in healthy men (Chusak et al., 2018). These studies attribute the flavonols and anthocyanins for the antioxidant activity.

Extract	Antioxidant assay	Results	References
Water extract and 100% ethanol extract	DPPH radical scavenging	Water extract IC <sub>50</sub> =1 mg/mL Ethanol extract IC <sub>50</sub> =4 mg/mL Water extract in gel formulation for inhibition of DPPH reduction=28% at 0.5 mg/mL	Kamkaen and Wilkinson (2009)
100% Methanol extract	DPPH radical scavenging	IC <sub>50</sub> =327 µg/mL	Nithianantham et al. (2013)
Citrate buffer extract	DPPH radical scavenging and FRAP	EC <sub>50</sub> =0.49 mg/mL 13.3 mM/g based on trolox equivalent antioxidant capacity (TEAC)	Siti Azima et al. (2014)
Water extract	DPPH radical scavenging FRAP	IC <sub>50</sub> =84.15 µg/mL 0.33 mmol/mg ascorbic equivalent	Iamsaard et al. (2014)
Water extract	DPPH radical scavenging Oxygen radical absorbance capacity (ORAC) Reduction of free radicalinduced erythrocyte hemolysis (4 h) Inhibition of lipid peroxidation (4 h)	IC <sub>50</sub> =470 μg/mL 17.54 g trolox equivalents/mg extract 96.3% at 400 μg/mL 72.7% at 400 μg/mL	Phrueksanan et al. (2014)
Water extract	DPPH radical scavenging Trolox equivalent antioxidant capacity (TEAC) Ferric reducing antioxidant power (FRAP) HRSA SRSA Ferrous ion chelating power	$\begin{array}{l} \text{IC}_{50}{=}0.47 \text{ mg/mL} \\ 0.17 \text{ mg trolox/mg dried} \\ \text{extract} \\ \end{array}$ $\begin{array}{l} 0.38 \text{ mmol FeSO}_4{/}\text{mg dried} \\ \text{extrac} \\ \text{IC}_{50}{=}19.2 \text{ mg/mL} \\ \text{IC}_{50}{=}26.3 \text{ mg/mL}[ \\ {>}10^3 \text{ mg EDTA/mg dried} \\ \text{extract} \end{array}$	Chayaratanasin et al. (2015)

Table (2). The antioxidant activity of C. ternatea flowers from various research studies

Extract	Antioxidant assay	Results	References
Methanol/acetone/water	ORAC	490.7 mol trolox	Nair et al.
(5:4:1) extract		equivalent/g extract	(2015)
95% methanol extract	DPPH radical scavenging	IC <sub>50</sub> =95.3 μg/mL	Rajamanickam
			et al. (2015)
Water extract	ABTS radical scavenging	4.2 μM trolox equivalent/g	Azima et al.
	DPPH radical scavenging	extract	(2017)
	ORAC extract	EC <sub>50</sub> =0.76 mg/mL	
		15.8 µmol trolox	
		equivalent/g	
Water extract	ABTS radical scavenging	IC <sub>50</sub> =42.9 μg/mL	Zakaria et al.
	DPPH radical scavenging	IC <sub>50</sub> =195.5 μg/mL	(2018)
Water extract with	DPPH radical scavenging	US=931.5 µg trolox	Mehmood et al.
ultrasound assistance	2 · · · · · · · · · · · · · · · · · · ·	equivalent/g extract	(2019)
(US)	ABTS radical scavenging	AGE=764.3 µg trolox	(=01))
and water extract with		equivalent/g Extract	
heat assistance at 50 °C	FRAP		
(AGE)		US=13,488 µg trolox	
(102)		equivalent/g extract	
	Reducing power	AGE=11,720.3 µg trolox	
	Reducing power	equivalent/g	
	Cu <sup>2+</sup> reducing power	Extract	
	Cu reducing power	US=5834.6 µg trolox	
		equivalent/g extract	
	Xanthine oxidase inhibition		
	Xantinne oxidase minoritori	AGE=4195.3 $\mu$ g trolox equivalent/g extract	
		equivalent/g extract	
		US=4539.0 µg trolox	
		10	
		equivalent/g extract AGE=6154.1 µg trolox	
		18	
		equivalent/g	
		Extract	
		US=12,696 µg trolox	
		equivalent/g extract	
		AGE=9549 µg trolox	
		equivalent/g extract	
		$US=1.01 \text{ mg/mL} (IC_{50})$	
W/ / / / 1000/ 1		AGE=1.22 mg/mL (IC <sub>50</sub> )	
Water extract, 100% and	DPPH radical scavenging	Water extract=11.7 mM	Lo´pez Prado et al.
50% methanol extract		trolox equivalent/g extract	(2019)
at 6 h (best condition)		100% methanol extract	
		=6.99 mM trolox	
	Inhibition of cholesterol	equivalent/g extract	
	oxidation	50% methanol extract =12.2	
		mM trolox	
		equivalent/g extract	
		Water extract=79.8%	
		100% methanol	
		extract=49.7%	
		50% methanol	
		extract=89.8%	
Water extract	DPPH radical scavenging	$EC_{50} \text{ of } 12.47 \pm 2.96 \text{ mg/mL}$	Goh, et al. (2022)

# Cont. Table (2).

# Antibacterial activity

Different extracts of *Clitoria ternatea* showed inhibitory effects against *Pseudomonas eruginosa*, Escherichia coli, Klebsiella pneumonia, Bacillus subtilis, Aeromonas formicans, Aeromonas hydrophila and Streptococcus agalactiae. Several studies investigated on the antibacterial potential of C. ternatea flowers. The methanol extract of C. ternatea flower was tested against 12 bacterial species (Bacillus cereus, Bacillus subtilis, Bacillus thuringiensis, Staphylococcus aureus, Streptococcus faecalis, Escherichia coli, Klebsiella pneumoniae, Pseudomonas aeruginosa, Salmonella typhi, Enterobacter aerogens, Proteus mirabilis and Herbaspirillum spp.) and was found to have the most potent activity against Bacillus thuringiensis with a minimum inhibitory concentration (MIC) of 12.5 mg/mL and minimum bactericidal concentration (MBC) of 25 mg/mL with an inhibition zone of 15.7 mm using agar disc diffusion technique (Kamilla et al., 2009). In another study, the water, methanol, petroleum ether, hexane and chloroform extract of C. ternatea flower (4 mg) were tested against E. coli, K. pneumoniae, S. enteritidis, S. typhimurium and P. aeruginosa to determine its antibacterial activity. The methanol extract was found to have the highest activity when tested using agar disc diffusion technique with an inhibition zone range of 16–26 mm in E. coli, K. pneumonia and P. aeruginosa but had no activity against S. typhi and S. enteritidis. The highest zone of inhibition 26 mm was observed against K. pneumonia and P. aeruginosa (Uma et al., 2009). Leong et al. (2017) determined the antibacterial activity of anthocyanins of C. ternatea flower ethanol extract paste against B. cereus, B. subtilis, S. aureus, B. subtilis subsp. spizizenii, Proteus mirabilis, K. pneumoniae, Yersinia enterocolitica and E. coli. The extract was found to have good antibacterial activity against B. cereus, B. subtilis, S. aureus, P. mirabilis and K. pneumonia with the most potent activity against K. pneumonia with a MIC of 1.6 mg/mL and minimum lethal concentration (MLC) of 25 mg/mL while in another study, the anthocyanin fraction obtained from the ethanol extract of C. ternatea flower had the best effect against *B. subtilis* with a disc diffusion inhibition zone of 10 mm (Mahmad et al., 2018). The findings from these studies suggest the potential of the anthocyanins for its antibacterial activity.

# Antifungal activity

A rise in resistance towards most antifungal agents in diverse pathogens which calls for the need to identify new therapeutic agents (**Perfect, 2016**). The methanol extract of *C. ternatea* flower (100 mg/mL) tested against *Candida albicans, Rhizopus* and *Penicillium spp.* had the highest activity against *Candida albicans* with an inhibition zone of 19 mm in agar disc diffusion. However, in broth dilution method, it only had activity against *Penicillium spp.* and *Rhizopus* with similar MIC value of 0.8 mg/mL and MFC value of 1.6 mg/ml (**Kamilla et al., 2009**). The anthocyanin fraction obtained from the ethanol extract of *C. ternatea* flower tested against *Fusarium sp., A. niger* and *Trichoderma sp.* had the highest activity against *Fusarium sp. with* an inhibition zone of 10 mm in agar disc diffusion technique (**Mahmad et al., 2018**). The anthocyanins of *C. ternatea* flower ethanol extract paste (50 mg/mL) tested against *A. niger, P. expansum* and *R. stolonifera* only exhibited activity against *P. expansum* with an inhibition zone of 15.5 mm in agar disc diffusion while it had an MIC value of 12.5 mg/mL and MLC value of 25 mg/mL. The mode of action for the antifungal activity against *P. expansum* was investigated and found to be mediated by the alteration of morphology of *P. expansum* fungal hyphae which had flattened empty hyphae resulting from cell wall disruption and damage of conidiophore. The germination of *P. expansum* conidia was completely inhibited with suppressed conidial development (**Leong et al., 2017**).

#### Anti-inflammatory activity

The current available non-steroidal anti-inflammatory drugs (NSAIDs), including acetaminophen and aspirin are associated with side effects, particularly gastrointestinal and cardiovascular effects as they are known to affect both COX-1 and COX-2. The discovery of new or alternate strategies is needed to reduce the risks associated with NSAIDs while achieving sufficient pain relief (**Brune and Patrignani**, **2015**). The petroleum ether extract of *C. ternatea* flowers was evaluated for anti-inflammatory activity using carrageenan paw edema method with healthy albino rats of either sex. The extract (200 and 400 mg/kg) significantly inhibited paw edema compared to control untreated group while in Eddy's hot plate method, the treatment group (400 mg/kg) had significant increased reaction time (time recorded when animals licked their fore or hind paws or jump response, whichever appear first) compared to control untreated group. The

study suggests the possibility of the extracts to have a protective effect against the release of prostaglandins, kinnins and other substances in carrageenan induced edema (**Shyamkumar and Ishwar, 2012**). In another study, the anthocyanin and flavonol fraction obtained from *C. ternatea* flower extract (extracted in a mixture of MeOH/acetone/H2O at ratio of 5:4:1) were investigated for its anti-inflammatory potential. In the lipopolysaccharides (LPS)-induced inflammation in RAW-264.7 macrophage cells, the flavonols had mild suppression of ROS while the anthocyanins had no effect on ROS production. The anthocyanins were also found to have higher inhibition of nitric oxide production compared to the flavonols. In western blot studies, only the anthocyanins inhibited nuclear factor-B translocation and iNOS protein expression whereas the flavonols significantly inhibited COX-2 expression but not the anthocyanins (**Nair et al., 2015**).

# Anticancer activities

Chemotherapy, radiation therapy and targeted therapy are among the approaches used for the treatment and management of cancer but they are not able to provide a permanent cure and have been associated with various side effects and toxicities (Curigliano et al., 2012). Thus, new agents that are safe, available and effective are urgently needed. Several studies investigated the anticancer potential of C. ternatea flower extracted using different solvents. The 100% petroleum ether extract (IC<sub>50</sub>=36 µg/mL) was found to be more potent than the 100% ethanol extract (IC<sub>50</sub> value of 57  $\mu$ g/mL) in the in vitro cytotoxic assay against Dalton's lymphoma ascites (DLA) cells at 3 h which could be due to different phytochemical composition in both extracts. The petroleum ether extract was found to have presence of saponins, tannins, steroids and triterpernoids while the ethanol extract had flavonols only (Kumar and Bhat, 2011). In another study, the water extract was more potent than the methanol extract having much lower  $IC_{50}$  values with activity against hormone dependent breast cancer cell line (MCF-7), non-hormone-dependent breast cancer cell line (MDA-MB-231), human ovary cancer cell line (Caov-3), and human liver cancer cell line (HepG2) at 72 h. However, the methanol extract had activity against MCF-7 and MDA-MB-231 cells only with higher IC<sub>50</sub> values. The extracts were not toxic against the normal cell line (Hs27). The study suggests the aqueous extract to have more significant anti-proliferative activity than the methanol extract as it may have more active compounds (flavonoids) present (Neda et al., 2013). Shen et al. (2016) found the anticancer effect of the hydrophilic (100% methanol) extract to be more potent than the lipophilic (hexane:ethyl acetate, 1:1) extract on human epithelial laryngeal carcinoma (Hep-2) cell line. The potent active compounds identified in the hydrophilic extract were mainly ternatins, kaempferol and quercetin responsible for the antiproliferative effect as opposed to the lipophilic extract which constitutes of fatty acids, phytosterols and tocopherols.

#### Antidiabetic activity

Herbal based medications are worth exploring for potential use in the management of diabetes as they are considered to be safer and may have reduced side effects (Borikar et al., 2018). Several studies investigated the in vitro and in vivo potential of C. ternatea flower extract for antidiabetic activity. The water extractreduced the formation of fluorescent advanced glycation end products having the highest activity at day 28 (49.4% at 1 mg/mL) as well as significant reduction in fructose amine level (14.47-36.66%) in glycated bovine serumalbumin. The study suggests the potential of the extract in the prevention of the formation of advanced glycation end products to be mediated through its free radical scavenging ability mainly attributed to the active compounds present being the ternatin anthocyanins, delphinidin derivatives and kaempferol (Chayaratanasin et al., 2015). In vivo studyfor antidiabetic activity in alloxaninduced diabetic rats (wistar albino) by Rajamanickam et al. (2015) utilizing 95% methanol, ethyl acetate and chloroform extract were found to have significantly reduced blood glucose level, increased serum protein levels and restored serum albumin to normal levels. The extracts also significantly decreased serum urea, creatinine, cholesterol and triglyceride levels compared to control untreated diabetic rats. A similar trend was also observed in the in vivo studies by Borikar et al. (2018) utilising 100% methanol extract and water extract in the study by Daisy and Rajathi (2009). In a randomized crossover study, acute ingestion of C. ternatea flower extract/beverage was found to have suppressed postprandial plasma glucose and insulin levels when consumed with sucrose in healthy men (Chusak et al., 2018). Overall, these studies suggested the hypoglycemic activity may be exerted by the flavonoid principles (flavonol glycosides and anthocyanins) and alkaloids present in the extract which may involve the potentiation of insulin secretion from the  $\beta$ -cell or by enhancement of the transport of blood glucose from plasma to peripheral tissues.

# Antiparasitic and insecticidal effects

The ethanolic extract of *Clitoria ternatea* (100mg/ml) bring paralysis within 15-20 min and bring death within 28-30 min to the Indian earthworm Pheritima posthuman Shekhawat and Vijayvergia (2011). However, the anthelmintic activity of ethanolic extracts of flowers, leaves, stems and roots of Clitoria ternatea were also evaluated on adult Indian earthworms Pheretima posthuma. Results showed that roots of the Clitoria ternatea took less time to paralyze and death of the earthworms. Roots were further extracted successively with petroleum ether, chloroform, ethyl acetate and methanol and these extracts were screened for anthelmintic activity. Results showed that methanol extract of *Clitoria ternatea* root is the more potent (Nirmal et al., 2008) The in vitro comparative study of anthelmintic activity of aqueous and ethanolic extracts of leaves of Clitoria ternatea was carried out against Eisenia foetida at three different concentrations (100, 50, 25 mg/ml). The study involved the determination of time of paralysis and time of death of the worms. At the concentration of 100 mg/ml both the ethanolic and the aqueous extracts showed very significant anthelmintic activities as compared to the standard drug, levamisole (0.55 mg/ml). In case of aqueous extract, the time of paralysis and death time was observed as  $18 \pm 1.57$  min and  $53.33 \pm 0.33$ min, and in case of ethanolic extracts  $12.33 \pm 0.80$  min and  $32.33 \pm 0.71$  min respectively (Salhan et al., 2008) The mosquito larvicidal activity of *Clitoria ternatea* was investigated against three major mosquito vectors Aedes aegypti, Culex quinquefasciatus, and Anopheles stephensi. Among the methanol extracts of *Clitoria ternatea* leaves, roots, flowers, and seeds, the seed extract was effective against the larvae of all the three species with LC<sub>50</sub> values 65.2, 154.5, and 54.4 ppm, for A. stephensi, A. aegypti, and C. quinquefasciatus, respectively. Among three tested plant species, Clitoria ternatea was showing the most promising mosquito larvicidal activity (Mathew et al., 2009).

# Other biological activities of phytochemicals studied in C. ternatea flowers

Clitoria ternatea flower has shown to have potential antioxidant, antimicrobial, anticancer and antidiabetic activity. However, there are also various studies which have looked into its potential for other beneficial activities. Adhikary et al. (2018) found the 100% methanol extract of C. ternatea flower and its purified compound quercetin-3β-D-glucoside for its anti-arthritic potential in a mice model. Quercetin-3β-D-glucoside was found to be more potent than the extract to significantly reduce myeloperoxidase activity, decrease in release of pro-inflammatory cytokines, chemokines, reactive oxygen species (ROS)/reactive nitrogen species production. It also significantly reduced tumor necrosis factor  $\alpha$ -receptor 1, toll-like receptor 2, inducible isoform of nitric oxide synthase, COX-2 and matrix metalloproteinase-2 expression. The anti-allergy effects of *C. ternatea* flower extract was also found in a study by **Singh et al. (2018)**. The 98% ethanol extract was able to attenuate histamine-induced contraction in both goat tracheal chain and isolated guinea pig ileum preparations. The extract was also found to attenuate histamine-induced dyspnoea and ovalbumin-induced changes of various inflammatory cytokines in animal models. The extract also displayed antitussive activity in sulfur dioxide- and citric acid-induced cough in experimental animals and attenuated inflammation in carrageenan and acetic acid challenged rodents. Clitoria ternatea flower extract was also found to have other beneficial effects in various other studies such as anti-aging (Zakaria et al., 2018), hepatoprotective (Nithianantham et al., 2013), testicular damage protection (Iamsaard et al., 2014), antiadipogenesis (Chayaratanasin et al., 2019) and starch digestion activity (Chusak et al., 2019).

# Side effects and toxicity

LD<sub>50</sub> of ethanol extract of *Clitoria ternatea* root was more than 1,300 mg/kg in mice (**Kelemu et al., 2004**). Acute oral toxicity study showed that there was no mortality up to 3000mg/kg in mice (**Deka and Chandra, 2011**). After single dose 1000 mg/kg in rats, no death or any other disorders up to 72 h (**Taur and Patil, 2011**). The extract wass found safe even at the dose of 2000 mg/kg body weight in rats (**Nawaz et al., 2004**). There was no mortality observed at doses up to 2 g/kg (po) of the ethanol extract of the aerial parts of *Clitoria ternatea* in rats. During observation, the animals exhibited decreased mobility but no signs of convulsions or loss of writhing reflex. This result indicates that *Clitoria ternatea* has a low toxicity profile (**Verma et al., 2013**). The mutagenic effect of the aqueous extract of *Clitoria ternatea* Linn was assessed

by three test methods, Bacillus subtilis rec assay, Salmonella typhimurium Ames' test and micronucleus test. The aqueous extract gave negative results, no mutagenic activities in both bacterial and mammalian cells (**Punjanon and Arpornsuwan, 2009**).

#### Applications of *Clitoria ternatea* in Industry

#### **Application in Traditional Food and Food Industry**

Nowadays, *C. ternatea* attracts a lot of interest due to its potential applications in traditional and modern medicine, cosmetics, agriculture, and the food industry as a source of natural food colorants and antioxidants. *C. ternatea* has been cultivated for a long time as a fodder and forage crop, and previous studies observed the plant for these purposes (**Oguis et al., 2019**). Parts of the plant are widely used for disease prevention, health promotion, and because they are believed to promote memory and intelligence in the Indian system of medicine, particularly in Ayuverda (**Mukherjee et al., 2007**) Differently in Malaysia, the flowers are consumed to make Nasi Kerabu blue in color, which is a famous local dish (**Neda et al., 2013**). Some sweets, namely kuehs in Malaysia, are colored blue for specific religious occasions. Meanwhile, the use of the flower as a food and drink colorant is currently becoming more popular in Indonesia. In Myanmar, *C. ternatea* flowers are dipped in batter, fried, and eaten as snacks (**Ravindran, 2017**). In Thailand, the common Thai drink named Nam Dok Anchan is colored with butterfly pea flower and served with pandan-flavored syrup and lime juice (**E.F.S.A., 2022**). The blue petals are also used to decorate and garnish dishes such as salads, soup, and rice.

#### Application in removal of environmental pollutants

This study has designed a green synthesis of magnetic (cobalt oxide) and noble (gold) nanoparticles from the fresh flowers of *Clitoria ternatea*. The flavonoids and polyphenols in the extract decreased the energy band gap of cobalt oxide and gold nanoparticles; hence, the capping of the natural constituents in Clitoria ternatea helped form stable metal nanoparticles. The cobalt oxide and gold nanoparticles are evaluated for their potential for eliminating organic pollutants from industrial effluent. The novelty of this present work represents the application of cobalt oxide nanoparticles in the removal of organic pollutants and a comparative study of the catalytic behaviour of both metal nanoparticles. The degradation of bromophenol blue, bromocresol green, and 4-nitrophenol in the presence of gold nanoparticles was completed in 120, 45, and 20 min with rate constants of  $3.7 \times 10-3$  /min,  $6.9 \times 10-3$  /min, and  $16.5 \times 10-3$ 3 /min, respectively. Similarly, the photocatalysis of bromophenol blue, bromocresol green, and 4nitrophenol in the presence of cobalt oxide nanoparticles was achieved in 60, 90, and 40 min with rate constants of  $2.3 \times 10-3$  /min,  $1.8 \times 10-3$  /min, and  $1.7 \times 10-3$  /min, respectively. The coefficient of correlation (R2) values justify that the degradation of organic pollutants follows first-order kinetics. The significance of the study is to develop green nanomaterials that can be used efficiently to remove organic pollutants in wastewater using a costeffective method with minimal toxicity to aquatic animals. It has proved to be useful in environmental pollution management (Nishigandha et al., 2024).

#### Extraction of flavonoids from Clitoria ternatea flower as carbon steel corrosion inhibitor

The present study emphasizes the extraction of flavonoids from the Butterfly blue pea (*Clitoria ternatea*), a flower containing very strong antioxidant properties. The performances of mild steel to prevent corrosion in 3.5 % NaCl medium doped with CT extracts (50, 100, 250, 500, 750, 1000 ppm) were ascertained by carrying out electrochemical measurements. It is apparent that CTW is superior in terms of corrosion inhibition of mild steel in NaCl medium under CO<sub>2</sub> environment with respect to CTET extract (% IE<sub>CTW</sub>=89.91% >% IE<sub>CTET</sub>=85.53%). Adsorption of both inhibitors on the surface of MS obeyed Langmuir isotherm. Therefore, it can be inferred from the findings that CT extracts were adsorbed *via* both physical and chemical adsorption, hence showing excellent corrosion resistance behaviour, thus indicating its potential to be an alternative source of natural antioxidants potent CO<sub>2</sub> corrosion inhibitor (**Azahar et al., 2024**).

#### Conclusion

*Clitoria ternatea* has a long history of use in traditional medicine, and recent studies suggest that it is promising medicinal plant with wide range of pharmacological activities which could be utilized in

several medical applications because of its effectiveness and safety. More research is needed to further explore the therapeutic potential of the plant, but it remains a promising natural remedy for several conditions.

# References

Adhikary, R., Sultana, S., and Bishayi, B. (2018). *Clitoria ternatea* flower petals: effect on TNFR1 neutralization via downregulation of synovial matrix metalloproteases. J. Ethnopharmacol, 210: 209–222. https://doi.org/10.1016/j.jep.2017.08.017

Admassu, H., Gasmalla, M.A. and Yang, R. (2018). Bioactive peptides derived from seaweed protein and their health benefits: antihypertensive, antioxidant, and antidiabetic properties. J. Food Sci, 83: 6–16. <u>https://doi.org/10.1111/1750-3841.14011</u>

Alderete-Chávez, A., Brito, R., Gelabert, R., Nunez, E. and Amador-del Ángel L.E. (2011). Evaluation of *Clitoria ternatea* L. in relation with fertility in tropical soils. Journal of Applied Sciences, 11(6): 1044-1048. <u>https://doi.org/10.3923/jas.2011.1044.1048</u>.

Azahar S.S., Raja P.B., Ibrahim M.N.M., Awang K., Zakeyuddin M.S., Hamidon T.S. and Hussin M.H. (2024). Extraction of flavonoids from Butterfly blue pea (*Clitoria ternatea*) flower as carbon steel corrosion inhibitor in CO<sub>2</sub> environment: Experimental and theoretical approaches, Journal of Molecular Liquids, (396): 124056. <u>https://doi.org/10.1016/j.molliq.2024.124056</u>

Azima, A.S., Noriham, A. and Manshoor, N. (2017). Phenolics, antioxidants and color properties of aqueous pigmented plant extracts: Ardisia colorata var. elliptica, *Clitoria ternatea*, Garcinia mangostana and Syzygium cumini. J. Funct Foods, 38: 232–241. https://doi.org/10.1016/j.jff.2017.09.018

**Bishoyi, A.K. and K.A. Geetha (2013)**. Polymorphism in flower colour and petal type in Aparajita (*Clitoria ternatea*). Open Access Journal of Medicinal and Aromatic Plants, 3(2): 12-14.

**Borikar, S.P., Kallewar, N.G. and Mahapatra, D.K. (2018)**. Dried flower powder combination of *Clitoria ternatea* and Punica granatum demonstrated analogous anti-hyperglycemic potential as compared with standard drug metformin: in vivo study in Sprague Dawley rats. J. Appl. Pharm Sci, 8:75–79. https://doi.org/10.7324/japs.2018.81111

Brune, K. and Patrignani, P. (2015). New insights into the use of currently available non-steroidal antiinflammatory drugs. J. Pain Res, 8:105. <u>https://doi.org/10.2147/jpr.s75160</u>

**Chayaratanasin, P., Barbieri, M.A. and Suanpairintr, N. (2015)**. Inhibitory effect of *Clitoria ternatea* flower petal extract on fructoseinduced protein glycation and oxidation-dependent damages to albumin in vitro. BMC Complement Altern Med 15:27. <u>https://doi.org/10.1186/s12906-015-0546-2</u>

Chayaratanasin, P., Caobi, A. and Suparpprom, C. (2019). *Clitoria ternatea* flower petal extract inhibits adipogenesis and lipid accumulation in 3T3-L1 preadipocytes by downregulating adipogenic gene expression. Molecules, 24:1894. <u>https://doi.org/10.3390/molecules24101894</u>

**Chusak, C., Thilavech, T. and Henry, C.J. (2018)**. Acute effect of *Clitoria ternatea* flower beverage on glycemic response and antioxidant capacity in healthy subjects: a randomized crossover trial. BMC Complement Altern Med, 18:1–11. <u>https://doi.org/10.1186/s12906-017-2075-7</u>

**Chusak, C., Ying, J.A.Y. and Zhien, J.L. (2019)**. Impact of *Clitoria ternatea* (butterfly pea) flower on in vitro starch digestibility,texture and sensory attributes of cooked rice using domestic cooking methods. Food Chem, 295: 646–652. <u>https://doi.org/10.1016/j.foodchem</u>

Curigliano, G., Cardinale, D. and Suter, T. (2012). Cardiovascular toxicity induced by chemotherapy, targeted agents and radiotherapy: ESMO Clinical Practice Guidelines. Ann Oncol 23:55–66. https://doi.org/10.1093/annonc/mds293

Daisy, P. and Rajathi, M. (2009). Hypoglycemic effects of *Clitoria ternatea* Linn. (Fabaceae) in alloxan

induced diabetes in rats. Trop J. Pharm Res, 8: 393-398. https://doi.org/10.4314/tjpr.v8i5.48082

**Dayal, D., A. Kumar, M.L. Swami, D. Machiwal, S. Manglassery, S.C. Vyas and H. Kunpara (2015)**. Chapter 18: Management Practices for Improved Forage Production of Butterfly Pea (*Clitoria ternatea*). p. 215-224. In: Improving Productivity of Drylands by Sustainable Resource Utilization & Management, Edition: 2016.

**Deka, M. and Chandra, K.J. (2011)**. Preliminary phytochemical analysis and acute oral toxicity study of *Clitoria ternatea* Linn roots in albino mice. International Research Journal of Pharmacy, 2(12): 139-140.

**De Souza, E.S., Burity, H.A., Oliveira, J.D.P., Figueiredo, M., and Lyra, M. (1996)**. N2-fixation and growth of the calopogonium (*Calopogonium mucunoides* Desv.) and of the clitoria (*Clitoria ternatea* L.) after sucessive cuts. Revista da Sociedade Brasileira de Zootecnia (Brazil), 25(6):1036-1048.

**European Food Safety Authority (E.F.S.A.) (2022)**. Notification of Dried Flowers of *Clitoria ternatea* L. as a Traditional Food from a Third Country Pursuant to Article; European Food Safety Authority: Parma, Italy, No. December 2021.

Fantz, P. R. (1981). A monograph of the genus *Clitoria* (Leguminosae: Glycineae).

Gamage, G.C.V., Lim, Y.Y. and Choo, W.S. (2021). Anthocyanins from *Clitoria ternatea* flower: Biosynthesis, extraction, stability, antioxidant activity, and applications. Frontiers in Plant Science, 12: 792303. <u>https://doi.org/10.3389/fpls.2021.792303</u>.

**Gengatharan, A., Dykes, G.A. and Choo, W.S. (2015)**. Betalains: natural plant pigments with potential application in functional foods. LWTFood Sci Technol 64: 645–649. https://doi.org/10.1016/j.lwt.2015.06.052

Gew, L.T., Teoh, W.J., Lein, L.L., Lim, M.W., Cognet, P. and Aroua, M.K. (2024). Glycerol-based extracts of *Clitoria ternatea* (Butterfly Pea Flower) with enhanced antioxidant potential. PeerJ Analytical Chemistry, 6:e30. <u>http://doi.org/10.7717/peerj-achem.30</u>.

Govaerts, R. (1999). World Checklist of Seed Plants 3(1, 2a & 2b): 1-1532. MIM, Deurne. [Cited as *Clitoria ternatea.*]

**Gupta, G.K., Chahal, J. and Bhatia, M. (2010)**. *Clitoria ternatea* (L.): Old and new aspects. Journal of Pharmacy Research, 3(11): 2610-2614.

Goh, S.E., Kwong, P.J., Ng, C.L., Ng, W.J. and Ee, K.Y. (2022). Antioxidant-rich *Clitoria ternatea* L. flower and its benefits in improving murine reproductive performance. Food Sci. Technol, Campinas, (42): e25921. DOI: https://doi.org/10.1590/fst.2592.

Hasanah, N.N.; Mohamad Azman, E.; Rozzamri, A.; Zainal Abedin, N.H. and Ismail-Fitry, M.R. (2023). A Systematic Review of Butterfly Pea Flower (*Clitoria ternatea* L.): Extraction and Application as a Food Freshness pH-Indicator for Polymer-Based Intelligent Packaging. Polymers, 15: 2541. https://doi.org/10.3390/polym151125411.

**Husain, S. and Devi, K.S. (1998)**. Fatty acid composition of three plant species: *Clitorea ternatea*, Mandulea suberosa and Ruta chalapensis. Journal of the Oil Technologists Association of India (30): 162-164.

**Iamsaard, S., Burawat, J. and Kanla, P. (2014)**. Antioxidant activity and protective effect of *Clitoria ternatea* flower extract on testicular damage induced by ketoconazole in rats. J. Zhejiang Univ. Sci. B., 15: 548–555. <u>https://doi.org/10.1631/jzus.b1300299</u>

Islam, M.A., Mondal, S.K., Islam, S., Akther Shorna, M.N., Biswas, S., Uddin, M.S., Zaman, S. and Saleh, M.A (2023). Antioxidant, Cytotoxicity, Antimicrobial Activity and In Silico Analysis of the Methanolic Leaf and Flower Extracts of *Clitoria ternatea*. Biochem Res Int., 2023: 1-12. https://doi.10.1155/2023/8847876.

Jeyaraj, E.J., Lim, Y.Y. and W.S. Choo (2021). Extraction methods of butterfly pea (*Clitoria ternatea*)

flower and biological activities of its phytochemicals. Journal of Food Science and Technology, 58(6): 2054-2067. <u>https://doi.org/10.1007/s13197-020-04745-3</u>.

Kamkaen, N., and Wilkinson, J.M. (2009), The antioxidant activity of *Clitoria ternatea* flower petal extracts and eye gel. Phytother Res, 23:1624–1625. <u>https://doi.org/10.1002/ptr.2832</u>

Kamilla, L., Mnsor, S.M., Ramanathan, S. and Sasidharan, S. (2009). Antimicrobial activity of *Clitoria ternatea* (L.) extracts. Pharmacologyonline, (1): 731-738.

**Kartesz, J.T. (1994)**. A synonymized checklist of the vascular flora of the United States, Canada, and Greenland. 2<sup>nd</sup> edition. 2 vols. Timber Press, Portland, OR.

Kazuma, K., Noda, N. and Suzuki, M. (2003). Flavonoid composition related to petal color in different lines of *Clitoria ternatea*. Phytochemistry 64: 1133–1139. <u>https://doi.org/10.1016/s0031-9422(03)00504-1.</u>

Kelemu, S., Cardona, C. and Segura, G. (2004). Antimicrobial and insecticidal protein isolated from seeds of *Clitoria ternatea*, a tropical forage legume. Plant Biochemistry and Physiology, (42): 867-873

Kosai, P., Sirisidthi, K., Jiraungkoorskul, K. and Jiraungkoorskul, W. (2015). Review on ethnomedicinal uses of memory boosting herb, butterfly pea, *Clitoria ternatea*. J. Nat. Remedies, 15(2): 71-76. https://doi:10.18311/jnr/2015/480.

**Kumar, B.S. and Bhat, K.I. (2011)**. In-vitro cytotoxic activity studies of *Clitoria ternatea* linn flower extracts. Int J Pharma Sci Rev Res 6:120–121

Leong, C.R., Azizi, K. and Afif, M. (2017). Anthocyanins from *Clitoria ternatea* attenuate food-borne Penicillium expansum and its potential application as food biopreservative. Nat Prod Sci 23:125–131. https://doi.org/10.20307/nps.2017.23.2.125

Lo´pez Prado, A.S., Shen, Y. and Ardoin, R. (2019). Effects of different solvents on total phenolic and total anthocyanin contents of *Clitoria ternatea* L. petal and their anti-cholesterol oxidation capabilities. Int. J. Food Sci. Technol, 54: 424–431. <u>https://doi.org/10.1111/ijfs.13953.</u>

Mahmad, N., Taha, R.M. and Othman, R. (2018). Anthocyanin as potential source for antimicrobial activity in *Clitoria ternatea* L. and Dioscorea alata L. Pigm Resin Technol, 47:490–495. https://doi.org/10.1108/prt-11-2016-0109

Macedo, M.L.R. and Xavier-Filho, J. (1992). Purification and partial characterization of trypsin inhibitors from seeds of *Clitoria ternatea*. Journal of the Science of Food and Agriculture, (58): 55-58.

Mathew, N., Anitha, M.G., Bala, T.S., Sivakumar, S.M., Narmadha, R. and Kalyanasundaram, M. (2009). Larvicidal activity of *Saraca indica*, *Nyctanthes arbortristis*, and *Clitoria ternatea* extracts against three mosquito vector species. Parasitol Res, 104(5): 1017-1025.

Mehmood, A., Ishaq, M. and Zhao, L. (2019). Impact of ultrasound and conventional extraction techniques on bioactive compounds and biological activities of blue butterfly pea flower (*Clitoria ternatea* L.). Ultrason Sonochem, 51:12–19. <u>https://doi.org/10.1016/j.ultsonch.2018.10.013</u>

**Morris, J.B. (2009)**. Characterization of butterfly pea (*Clitoria ternatea* L.) accessions for morphology, phenology, reproduction, and potential nutraceutical, pharmaceutical trait utilization. Genetic Resources and Crop Evolution, 56(3): 421-427. <u>https://doi.org/10.1007/s10722-008-9376-0</u>.

Mukherjee, P.K.; Rai, S.; Kumar, V.; Mukherjee, K.; Hylands, P.J. and Hider, R.C. (2007). Plants of Indian Origin in Drug Discovery. Expert Opin. Drug Discov. 2, 633–657. <u>https://doi.org/10</u>. <u>1517/17460441.2.5.633</u>

Mukherjee, P.K., Kumar, V., Kumar N.S. and Heinrich M. (2008). The Ayurvedic medicine *Clitoria ternatea*-From traditional use to scientific assessment. Journal of Ethnopharmacology, 120(3): 291-301. https://doi:10.1016/j.jep.2008.09.009.

Nair, V., Bang, W.Y. and Schreckinger, E. (2015). Protective role of ternatin anthocyanins and

quercetin glycosides from butterfly pea (*Clitoria ternatea* Leguminosae) blue flower petals against lipopolysaccharide (LPS)-induced inflammation in macrophage cells. J. Agric. Food Chem., 63: 6355–6365. <u>https://doi.org/10.1021/acs.jafc.5b00928</u>

Nawaz, A.H., Hussain, M., Karim, M., Khan, M., Jahan, R. and Mohammed, R. (2009). An ethnobotanical survey of Rajshahi district in Rajshahi division, Bangladesh, American-Eurasian Journal of Sustainable Agriculture, 3(2): 143-150.

Neda, G.D., Rabeta, M.S. and Ong, M.T. (2013). Chemical composition and anti-proliferative properties of flowers of *Clitoria ternatea*. Int. Food Res. J., 20: 1229–1234

Nguyen, G.K.T., Zhang, S., Nguyen, N.T.K., Nguyen, P.Q.T., Chiu, M.S., Hardjojo, A. and J.P. Tam (2011). Discovery and characterization of novel cyclotides originated from chimeric precursors consisting of albumin-1 chain a and cyclotide domains in the Fabaceae family. J. Biol. Chem., 286(27): 24275–24287.

Nithianantham, K., Ping, K.Y. and Latha, L.Y. (2013). Evaluation of hepatoprotective effect of methanolic extract of *Clitoria ternatea* (Linn.) flower against acetaminophen-induced liver damage. Asian Pac J. Trop Dis., 3:314–319. <u>https://doi.org/10.1016/s2222-1808(13)60075-4</u>

Nirmal, S.A., Bhalke, R.D., Jadhav, R.S. and Tambe, V.D. (2008). Anthelmintic activity of *Clitoria ternatea*. Pharmacologyonline, 1: 114-119.

Nishigandha, S., Alkhayer, K., and Behera, A. (2024). Efficient removal of environmental pollutants by green synthesized metal nanoparticles of *Clitoria ternatea*. Heliyonm (10) 9: e29865 <a href="https://doi.org/10.1016/j.heliyon.2024.e29865">https://doi.org/10.1016/j.heliyon.2024.e29865</a>

**Oblisami, G. (1974)**. Studies on the Rhizobium and nodulation pattern in a forage legume, *Clitoria ternatea* Linn. Proc. Ind. Nat. Sci. Acad., B, Biol. Sci., 40: 618-623.

**Oguis, G.K., Gilding, E.K., Jackson, M.A. and D.J. Craik (2019)**. "Butterfly Pea (*Clitoria ternatea*), a Cyclotide-Bearing Plant with Applications in Agriculture and Medicine." Frontiers in Plant Science, 10: 645. <u>https://doi:10.3389/fpls.2019.00645</u>.

**Perfect. J.R. (2016)**. Is there an emerging need for new antifungals? Expert Opin Emerg Drugs, 21:129131. https://doi.org/10.1517/14728214.2016.1155554

**Pengelly, B. C. and M. J. Conway (2000)**. Pastures on cropping soils: Which tropical pasture legume to use?. Tropical Grasslands, 34: 162-168. <u>https://doi.org/10.2346-3775</u>.

**Pham-Huy, L.A., He, H. and Pham-Huy, C. (2008)**. Free radicals, antioxidants in disease and health. Int. J. Biomed Sci., 4:89–96

**Phrueksanan, W., Yibchok-anun, S. and Adisakwattana, S. (2014)**. Protection of *Clitoria ternatea* flower petal extract against free radicalinduced hemolysis and oxidative damage in canine erythrocytes. Res. Vet. Sci., 97: 357-363. <u>https://doi.org/10.1016/j.rvsc.2014.08.010.</u>

**Punjanon, T. and Arpornsuwan, T. (2009)**. Studies of the mutagenic activities of synthetic hair dyes and natural hair dyes. Bull Health Sci. & Tech, 9(1-2): 33-39.

**Rai, S.S., Banik, A., Singh, A. and Singh, M. (2015)**. Evaluation of anti-ulcer activity of aqueous and ethanolic extract of whole plant of *Clitoria ternatea* in albino Wistar rats. International Journal of Pharmaceutical Sciences and Drug Research, 7(1): 33-39.

**Rajamanickam, M., Kalaivanan, P. and Sivagnanam, I. (2015)**. Evaluation of anti-oxidant and antidiabetic activity of flower extract of *Clitoria ternatea* L. J. Appl. Pharm Sci., 5:131–138. <u>https://doi.org/10.7324/japs.2015.50820</u>

**Raju, S.A.J. and Ramana, V.k. (2021)**. A study on pollinationecology of butterfly pea, *Clitoria ternatea* L. (Fabaceae). Species, 22(69): 29-35.

Ravindran, P.N. (2017). The Encyclopedia of Herbs and Spices; CAB International: Wallingford, UK,;Vol

1.

**Reid, R. and Sinclair, D. (1980)**. An evaluation of a collection of *Clitoria ternatea* for forage and grain production. Agricultural and Food Sciences.

**Ripperger, H. (1978)**. Solation of stigmast-4-ene-3,6-dione from *Hamelia patens* and *Clitoria ternatea*. Pharmazie, 33(1):82-83.

**Rugayah, R. A., Windadri, F.I. and A. Hidaya** (2004). Pengumpulan Data Taksonomi. In Rugayah, E. A. Widjaya, & Praptiwi, eds. Pedoman Pengumpulan Data Keanekaragaman Flora. Bogor: Puslit Biologi - LIPI, pp. 5-42.

Salhan, M., Kumar, B., Tiwari, P., Sharma, P., Sandhar, H.K. and Gautam, M. (2011). Comparative anthelmintic activity of aqueous and ethanolic leaf extracts of *Clitoria ternatea*. Int. J. Drug Dev. & Res., 3 (1):68-69.

Sarma, D.S.K., Kumar, D., Yamini, C., Santhalahari, C., Lahari, C., Kumar, G.C., and Lahitha M. (2023). Review on *Clitoria ternatea*. International Journal of Pharmaceutical Sciences and Medicine, 8(9): 43-58.

Shen, Y., Du, L. and Zeng, H. (2016). Butterfly pea (*Clitoria ternatea*) seed and petal extracts decreased Hep-2 carcinoma cell viability. Int. J. Food Sci. Technol., 51: 1860–1868. https://doi.org/10.1111/ijfs.13158

Shekhawat, N. and Vijayvergia, R. (2011). Anthelmintic activity of extracts of some medicinal plants. International Journal of Computational Science and Mathematics, 3(2): 183-187.

Shyamkumar, I.B. and Shwar, B. (2012). Anti-inflammatory, analgesic and phytochemical studies of *Clitoria ternatea* Linn flower extract. Int. Res. J. Pharm., 3: 208–210

Singh, N.K., Garabadu, D. and Sharma, P. (2018). Anti-allergy and antitussive activity of *Clitoria ternatea* L. in experimental animals. J. Ethnopharmacol, 224: 15–26. <u>https://doi.org/10.1016/j.jep.</u> 2018.05.026

Sinha, A. (1960). ß-Sitosterol from the seeds of *Clitoria ternatea*. Current Science, (29): 180-181.

**Srichaikul, B. (2018)**. Ultrasonication extraction, bioactivity, antioxidant activity, total flavonoid, total phenolic and antioxidant of *Clitoria ternatea* linn flower extract for anti-aging drinks. Pharmacogn Mag. 14: 322. <u>https://doi.org/10.4103/pm.pm\_206\_17</u>

**Staples**, (1992). *Clitoria ternatea* L., Record from Proseabase. Mannetje, L.'t and Jones, R.M. (Editors). PROSEA (Plant Resources of South-East Asia) Foundation, Pudoc Scientific Publishers, Wageningen, Netherlands. pp: 94-97.

Suarna, I.W. and I.M.S. Wijaya (2021). Butterfly pea (*Clitoria ternatea* L.: Fabaceae) and its morphological variations in Bali. Journal of Tropical Biodiversity and Biotechnology, 6(2): 1-12. https://doi.org/10.22146/jtbb.63013.

Surya, D., Rajamani, K., Suresh, J. and Uma, D. (2022). Morphological characterization and assessment of anthocyanin in three different genotypes of *Clitoria ternatea* L. The Pharma Innovation Journal, 11(7): 2388-2392.

Siti Azima, A.M., Noriham, A. and Manshoor, N. (2014). Anthocyanin content in relation to the antioxidant activity and colour properties of Garcinia mangostana peel, Syzigium cumini and *Clitoria ternatea* extracts. Int. Food Res. J., 21: 2369–2375

Taur, D.J. and Patil, R.Y. (2011). Antihistaminic activity of *Clitoria ternatea* L roots. J Basic Clin Pharm, 2(1): 41-44.

**Terahara, N., Toki, K. and Saito, N. (1998)**. Eight new anthocyanins, ternatins C1–C5 and D3 and preternatins A3 and C4 from young *Clitoria ternatea* flowers. J. Nat., 61: 1361–1367. <u>https://doi.org/10.1021/np980160c.</u> Tjitrosoepomo, G. (1985). Morfologi Tumbuhan, Yogyakarta: Gadjah Mada University Press.

**Uma, B., Prabhakar, K. and Rajendran, S. (2009)**. Phytochemical analysis and antimicrobial activity of *Clitorea ternatea* Linn against extended spectrum beta lactamase producing enteric and urinary pathogens. Asian J. Pharm Clin. Res., 2: 94–96

Verma, P.R., Itankar, P.R. and Arora, S.K. (2013). Evaluation of antidiabetic antihyperlipidemic and pancreatic regeneration, potential of aerial parts of *Clitoria ternatea*. Rev Bras Farmacogn; 23: 819-829.

Zakaria, N.N.A., Okello, E.J. and Howes, M.J. (2018). In vitro protective effects of an aqueous extract of *Clitoria ternatea* L. flower against hydrogen peroxide-induced cytotoxicity and UV-induced mtDNA damage in human keratinocytes. Phytother Res., (32): 1064-1072. DOI: https://doi.org/10.1002/ptr.6045.



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