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Antibacterial Efficiency Of *Clitoria Ternatea* Flower Aqueous Extract And It's Phytochemical Analysis

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Abstract
Clitoria ternatea flower is one of the medicinal plant also called as butterfly pea flower. It is mainly distributed in tropical Asia including India. In the present study, mainly used the blue color flower as sample extract for various analysis. Extraction was prepared by using aqueous solution then it was used for various analysis. We examined the phytochemical of *C. ternatea* flower. Different tests were conducted for phytochemical analysis, explained briefly in this paper. Extract showed positive result on flavonoids, glycoside, phenols, saponins, tannins, carbohydrates, proteins and terpenoids. Antimicrobial activity of *Clitoria ternatea* flower is exhibiting the best result. Here we used *Bacillus cereus, Klebsiella pneumonia , Staphylococcus aureus, and Morganella morgani,* four infectious microorganisms. Treatment against these organisms resulted in the best inhibition. This analysis mainly conducted by the agar well diffusion method. The aqueous flower extract of *C. ternatea* has well medicinal properties and antimicrobial activity which can treat the Urinary tract Infection(UTI) and also known to prevent diabetics.

Key words: *Clitoria ternatea* , Phytochemical, Antimicrobial activity.

Introduction
Plants are the good source of medicine and it is inherited and used as the important compound in the health care system universally. India is called as the botanical garden of the world as it produces large quantity of valuable medicinal plants. Plant medicines are used to treat infectious diseases as well as chronic diseases. (Tandon and Yadav, 2017) Drugs are being developed all the time since a growing number of people develop various acute and chronic infections, diseases, cancer, heart problems, diabetes etc., Now, it has become the major threat to human race because the consequences are devastating. Any prescribed drug itself could lead to various other minor and major health issues. (Zhao et al., 2018) Therefore, people still prefer plant-based natural medicines which has lesser or no impact. More than 80,000 plant species are been used for the treatment of various diseases as traditional medicines across the different regions of India. India is a well-known country to use medicinal plants for treating the diseases. Medicinal plants and aromatic plants are being used as raw materials for the manufacture of drugs.
Clitoria ternatea Linn. Is an attractive perennial Climber with conspicuous blue or white Flowers. It belongs to the family Fabaceae and Commonly known as "butterfly pea" and "shankhapuspi". It is traditionally used

to treat Various ailments (Sivarajan and Balachandran 1994, Kokate 1999). The plant is native to South-east Asia and distributed in tropical Asia Including India, the Philippines and Madagascar (Anonymous 1998). There are several reported Ayurvedic 'medha' drugs which include *Clitoria ternatea , Celastrus panniculatus, Acorus calamus, Centella asiatica, and Withania somnifera* (Sivaranjan and Balachandran, 1994). With the advancement of Ayurvedic tradition and its scientific exploration, several classes of plant species have been studied in order to evaluate their therapeutic potentials and to isolate the lead compounds. Clitoria ternatea has witnessed a pharmacological and toxicological evaluation of these claims pointing to some important therapeutic benefits of this traditional drug which are highlighted in this review. CT has been used as an ingredient in 'Medhya Rasayana' a rejuvenating recipe used for treatment of neurological disorders and considered to strengthen a person's intellect (Sharma and Bhagwan, 1988). CT is a potential candidate for enhancing learning and memory (Taranalli and Cheeramkuczhi, 2000, Rai et al., 2001, Rai et al., 2002, Rai et al., 2005). In traditional systems of medicine transmitted orally or in writing (esp. Ayurveda) various therapeutic effects have been attributed to roots, leaves and seeds of CT. A number of bioactive secondary metabolites and pharmacological activities of the plant have been reported. Hence, this review is a critical assessment of the currently available information on ethnobotanical and ethnomedical uses, pharmacognosy, and medicinal uses as recorded in traditional systems of medicine transmitted orally or in writing. It also reviews secondary metabolites, pharmacological and toxicological studies of this useful plant. Roots, seeds And leaves of *C. ternatea* are commonly used in The Ayurvedic system of medicine. Extracts of This plant have been used as an ingredient in the Ayurvedic 'Medhya Rasayana' as a Rejuvenating recipe used for treatment of Neurological disorders and are considered to Enhance the intellect (Sharma and Dash 1988). The whole plant and seed extracts are used for Stomatitis, piles, sterility in females, Hematemesis, insomnia, epilepsy, psychosis, Leucorrhea and polyurea (Yoganarasimhan2000). The roots are bitter, refrigerant, laxative, Intellect-promoting, diuretic, anthelmintic, Tonic and are useful in dementia, hemicrania, Burning sensations, leprosy, inflammation, Leucoderma, bronchitis, asthma, pulmonary Tuberculosis, ascites, fever, otalgia, Hepatopathy and as a cathartic (Nadkarni 1976). The root, stem and flower are also used for the treatment of snake bite and scorpion sting (Morris 1999). *C. ternatea* has been shown to have number of pharmacological activities such1.Introduction as possessing anxiolytic, antidepressant, anticonvulsant, antistress (Jain et al. 2003), sedative (Kulkarni et al. 1988), antipyretic, anti-inflammatory, analgesic (Devi et al. 2003, Gomez and Kalamani 2003), Anthelmintic (Salhan et al., 2011) and anti-microbial activities (Kamilla et al., 2009). The extract of *C. ternatea* Has been shown to improve learning ability, Enhance memory, increase apical and basal Dendritic branches, and increase acetylcholine Content and acetyl cholinesterase activity in rats (Rai et al. 2001). The plant contains several Secondary metabolites such as kaempferol and Its glucoside–clitorin, taraxerol and a lactone Aparajitin (Barik et al. 2007). Seeds contain –Sistosterol, hexacosanal, and anthoxanthin (Yoganarasimhan 2000).

Material and methods

Dry *Clitoria ternatea L.* Flower was used as a sample for various analysis. The sample of dry *Clitoria ternatea L.*(butterfly blue pea) flower was collected from Department of Applied Genetics , Karnataka University Dharwad in the month of January. The dried flower can be stored in normal atmospheric condition.

Reagents

All analytical grade chemicals, acids and solvents, media and other chemicals used in the present study were purchased from different sources. Mercury (ll) chloride (SRL), Potassium iodide (Fisher), Iodine (Fisher),Picric acid(SRL), Magnesium ribbon, Concentrated hydrochloric acid (SRL), Sodium hydroxide (Fisher), Dilute hydrochloric acid (SRL), Carbon tetrachloride (SRL) Ammonia(SRL), Glacial acetic acid(SRL), Lead acetate (SRL), Ferric chloride (Fisher), Concentrated Sulfuric acid(SRL), alcoholic alpha naphthol (SRL), Feelings A & B solution (SRL), Sodium hydroxide (SRL), copper sulphate (Fisher), Nutrient agar(SRL), Dimethyl sulfoxide(DMSO) solution (SRL)

Extract preparation

The Dry blue pea flower extract were prepared by using aquasious treatment by Taking a some amount of dry *Clitoria ternatea* and cutting the flower into small pieces using scissor, weigh 7.5 grams of chopped pieces and then pour into the conical flask add 120 ml of distill water allow the water bath for 30 minutes at 70°C then cool within the water bath for an hour then take outside the conical flask mix well we observe the

dark colour appearance after that filter the solvent using muslin cloth and again filter through the what's man number one filter paper and the collect the extract and stored at 4^{o}C.

Phytochemical analysis

During phytochemical analysis it shows positive result on flavonoids, glycosidase, phenolic compounds, tannins, carbohydrates, and proteins.

Test for Alkaloids:

Alkaloids test is mainly done by several methods, we use only three methods That is mayer's test, Wagner's test, Hager's test. In mayers test- take about 3ml of sample in test tube Add few drops of mayers reagent the treated solution was observed with the brownish precipitations it shows the positive result of the alkaloids test. In Wagner's test- take about 3ml of sample in a test tube add few drops of Wagner's reagent, the treated solution was observed the brownish precipitations, it gives a positive results of the alkaloids test. In Hanger's test- take about 2ml of extract add few drops of hangers reagents in a test tube, the treated solutions was observed the appearance of yellowish precipitation, it gives a positive results of alkaloids test.

Test for flavonoids:

In flavonoid test we use only two methods that is shinoda test, and Alkaline reagent test. In shinoda test- to take about some amount of sample in a test tubes add few fragments of magnesium ribbon add concentrated hydrochloric acid drop wise the treated solutions was observed pink scarlet, crimson red or occasionally green to blue color appears after few minutes, this indicates the presence of the flavonoids. The Alkaline reagent test: to take about some amount of samples in a test tubes add a few drops of sodium hydroxide solutions the treated solutions was observed with the formation of an intense yellow color which turns to colorless an addition of few drops of the diluted acid, it indicates the positive results of the flavonoids.

Test for saponins:

Saponins test is done by frothing test method, In frothing test- take about 3 ml of solution mixed with 10ml of distilled water in a test tube, the test tube was stoppered and shaken vigorously for about 5 min it was allowed to stand for 30 min the treated solution was observed in the formation of honey comb fourth it shows positive result of the saponins.

Test for glycosides

In glycosides test we use only two methods i.e. Borntrager's test and keller Killiani test. In Borntrager's test- take about 1ml of sample add 5 to 10 ml of dilute HCL then boil on water both for 10 minutes and filter the solution then extract of filtrate with a CCL4 or benzene is added to it after that add equal amount of ammonia solutions to filtrate and finally shake well the treated solution was observed by the appearance of pink to red color it indicates the presence of the glycosides. In Keller Killiani test- to take about 0.5 ml of solution of glacial acidic acid add 2-3 drops of Fecl3 and add 2 ml of extract add 1 ml of concentrated H2SO4 along the wall the treated solution was observed in the formation of deep blue color at the junction of 2 liquids it indicates the presence of the third glycosides.

Test for phenolic compounds:
Phenolic compounds test is done by using ferric chloride method to take about test to solution had few drops of neutral 5% of ferric chloride solution that treated solution was observed In the formation of dark green color this shows the positive result of presence of phenolic compounds.

Test for tannins:
Tannins test is done by using two methods i.e. ferric chloride methods and they lead acetate methods. In ferric chloride test- to take about test solution add few drops of neutral 5% of ferric chloride solution that treated solution was observed in the formation of dark green color this shows the positive result of presence tannins. In lead acetate test- to take about some amount of test solution add few drops of 10% of lead acetate solution the treated solution was observed in formation of white PPT it indicates the presence of tannins.

Test for carbohydrates:
In carbohydrates test we use only two methods i.e. Molisch's test and Fehling's test. In Molisch's test- take one ml of test solution with the few drops of alcoholic alpha napthol add 0.2 ml of concentrated H2SO4 along this side of test solution that treated solution was observed in formation of purple to violet color ring appears at the junction it shows the positive result of presence of the carbohydrates. In Fehling's test- to take equal volume of Fehling's A and Fehling's B solution add some amount of test solution in a test tubes appearance of yellow green PPT indicates the presence of little or less amount of reducing sugar.

Test for proteins:
Biuret test- to take about test solution add 4% NaoH solution and few drops of 1% cuso4 solution in a test tube appearance of green or blueish color indicates the presence of protein.

Test for Terpenoids:
Salkowski test- to take about 5ml of extract was mixed with 2 ml of chloroforms and 3 ml of concentrated sulfuric acid was added to forms a layer, in observations a radish brown coloration of the interface was formed It indicates the presence of the Terpenoids.

Antimicrobial activity determination
Agar well diffusion method: The Bacterial strains were obtained from the Department of Applied Genetics at Karnataka University, Dharwad and incubated in nutrient broth at 37ºC for 24 hours. Kanamycin was used as the positive control. The Nutrient Agar media is prepared and microwells were made on cultural media in 6mm diameter with the help of gel puncture machine(Sawan S et al.2014) The microwells were filled with10µl, 50µl, 100µl, 200µl of plant extract the petridishes used for antibacterial screening were incubated at 37^{o}C for 24-48 hours, In a current study, the effectiveness of *Clitoria ternatea* as an antibacterial property which is compared with the antibiotic kanamycin with four different bacteria :*Bacillus cereus, Klebsiella pneumonia , Staphylococcus aureus, and Morganella morgani.* the activity was measured in terms of zone of inhibition (mm) appearing around the microwells.(Sawan S et al.2014).

Result and discussion
Phytochemical analysis:
Phytochemicals are rich in bioactive Compounds it has a medicinal property in plants to prevent various disease like microbial activity and oxidative stress and provides various health benefits. Today food Industries are very interested in using the plant extracts having Good total phenolic content.(Sawan S et al., 2014). The phytochemical extraction of *Clitoria ternatea* flower is very important in Identifying new sources of therapeutical and industrial application. The present study gives the valuable information of bioactive compounds presence in *Clitoria ternatea* flower. Phytochemical analysis is done by alkaloids Tannins glycosides saponins flavonoids and phenols carbohydrates proteins and the terpenoids. In a present study alkaloid shows negative result in that *Clitoria ternatea* flower(aq. extract) it changes color but it to do not form the precipitation so this experiments result shows the absence of alkaloids, Alkaloids are produced by large variety of organisms including bacteria, fungi, plants and animals, and Some alkaloids have a bitter taste While many are toxic to other organisms . In their study, alkaloids are present in shoot, flower and seed

extracts, and absent in leaf and shoot extract (P.Manjula et al., 2013). In flavonoids it shows positive result it means the pink color scarlet is observed by the shinoda test and Alkaline test shows color changes to blackish grayish resulting in the presence of flavonoids. Flavonoids were reported in root extract and in shoot and flower extract and seed extract, flavonoids have been reported to possess many useful properties, including anti-inflammatory, oestrogenic, enzyme inhibition, antimicrobial, anti-allergic, antioxidant, vascular and cytotoxic antitumor activity(P.Manjula et al., 2013).similar that the study of glycoside shows the positive results which means that the deep brown color at the junction of two liquids by the Keller Kliliani test so the result shows the presence of the glycosides, Glycoside compounds were present in *Clitoria ternatea* leaf, shoot, flower and seed positive results however absent in root extracts.(P.Manjula et al., 2013). in the study of saponins it shows positive results to observe honey comb fourth by the frothing test. saponins were reported in the root of *Clitoria ternatea,* saponins were presently absent in shoot and seed extract, traditionally saponins were extensively used as detergent, pesticides, and molluscicides, in addition to their industrial application as forming and surface active agents they also have beneficial health effects (P.Manjula et al., 2013).in the given experiment similar to the study of phenolic compound shows the formation of dark green color by the ferric chloride test it indicates the presence of phenolic compounds. Phenolic compounds are of great importance as cellular support material because they form the integral part of cell wall structure by polymeric phenolic(P.Manjula et al., 2013).in the study of tannins the formation of white PPT is observed conducted by the lead acetate test it shows the presence of the tannins. Tannins and Resins reported in the present study in shoots, leaves, flowers and seeds, it was attributed that tannins contributed the property of astringency leading to faster healing of wounds and inflamed mucos membrane(P.Manjula et al., 2013). similar that the study of carbohydrate shows the formation of purple to violet coloring at the junction by Molisch's test and the appearance of yellow or greenish PPT by the Fehling's test it shows the presence of the carbohydrates. The extracts almost have a same amount of carbohydrates but as calculated seed methanol showed (78.22) a high content (sriyeta c et al., 2017). in the study of proteins the greenish or the blueish color is appears indicates the presence of the proteins. in the study of terpenoids formation of reddish brown color of the interface was formed it shows the presence of Terpenoids. *Clitoria ternatea* showed the presence of active component like saponins, tannins, flavonoids, terpenoids, glycosides, amino groups (partibha s et al., 2023). Crude ethonolic extract of the fresh *C. ternatea* flowers are used for phytochemical screening, were sterols, terpenoids, flavonoids, Alkaloids, saponins, reducing sugar and tannins shows an Positive result. The presence of various phytochemical constituents contributes to the biological activities of the plant (Rosalinda Torres c et al.2022).

Results and Discussion Flower morphometric and phytochemical analysis was performed and following results were obtained. 3.2. Morphomatric Variation in Flowers of *C. ternatea* There were three different colour producing *C. ternatea* plants were observed and visual examination revels that the three different number of petal produced by *C. ternatea* observed and categorized as 1A: Blue colour more petals; 1B: Blue colour single petal; 2A: white colour more petals; 2B: white colour single petal, and 3A: purple colour single petal (H.D.K.Buddik et al., 2021).

Phytochemicals	Result
Alkaloids	-ve result
Flavonoids	+ve result
Glycosides	+ve result
Phenols	+ve result
Saponins	+ve result
Tannins	+ve result
Carbohydrates	+ve result
Proteins	+ve result
Terpenoids	+ve result

+ve :presence -ve : Absences

Table 1 phytochemical analysis of *Clitoria ternatea* flower

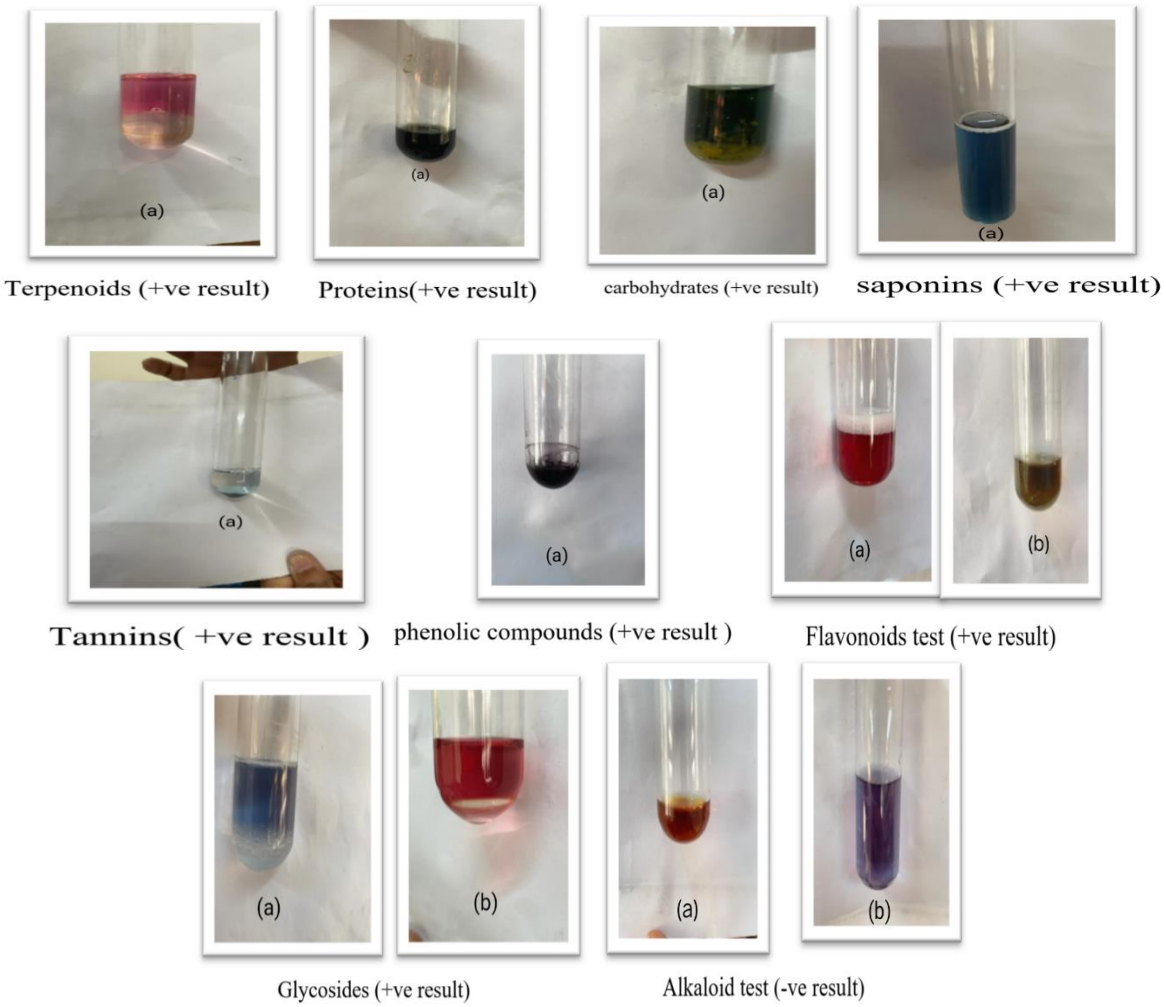

Figure 1 : phytochemical analysis -Alkaloids shows negative result (a)-Wagner's test (b)-Hanger's test both are shows negative result. Flavonoids shows positive result, (a)-shinoda test (b)- Alkaline reagent test. glycosides shows positive result, (a)-Keller killiani test (b)- Borntrager's test. Phenolic compounds shows positive result,(a)-ferric chloride method. saponins shows positive result, (a)- frothing test method. Tannins shows positive result,(a)-ferric chloride methods. Carbohydrates shows positive result,(a)-Fehling's test. Protein shows positive result,(a)-Biuret test. Terpenoids shows positive result, (a)-Salkowski test.

Antimicrobial activity determination

Bacillus cereus, Klebsiella pneumonia , Staphylococcus aureus, and Morganella morgani were used to screen antimicrobial activity of the dry *Clitoria ternatea* flower extract. Agar well diffusion method as used to assess the activity against the bacteria by measuring the zone of inhibition. Inhibiting concentration used for the sample here is 2mg/ml of DMSO (Sawan S et al.2014). Antibacterial growth was determined by measuring the diameter of the inhibition zone(SD +/-mean) anti bacterial property of *Clitoria ternatea* was investigated by agar diffusion method. The aqueous solvent extract from the flower of *Clitoria ternatea* were a tested against kanamycin.(Rajesh K et al., 2017). The antimicrobial activity of *Clitoria ternatea* flower shows good inhibition activity at 200 µg/mL against bacterial strains.

Name of the bacteria	Zone of inhibition (diameter in mm)				
	mean ± standard error				
	Kanamycin	*Plant Extract*			
		10 µg/mL	50 µg/mL	100 µg/mL	200 µg/mL
B. cereus	25.76 ± 0.2514	13.84 ± 0.0606	15.99 ± 1.0955	14.53 ± 0.1311	17.07 ± 0.6258
K. pneumonia	27.24 ± 0.4071	13.05 ± 0.3093	14.63 ± 0.7578	13.35 ± 0.4030	14.21 ± 0.1460
M. morganii	22.85 ± 1.1200	13.58 ± 0.5963	12.95 ± 0.6829	17.57 ± 1.1484	19.07 ± 0.7785
S. aureus	28.30 ± 0.6905	10.50 ± 0.2535	15.27 ± 0.5302	14.20 ± 0.6270	13.58 ± 0.1103

Table 2 – Antimicrobial activity determination

Aqueous extract of dry *Clitoria ternatea* flower shows good inhibiting activity against two (gram +ve)bacterial species that is *M.morganii* and *B.cereus*.(table 2) (Sawan S et al., 2014).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.(Ethel jeyaseela J et al., 2021).The leaf and seed of plant included in the present study were found to be active on at least one of the selected microbial strains tested. In general, among the tested microbial strains, bacteria were found to be more sensitive to the test extracts than fungi. The preliminary disk diffusion assay of methanol *C. ternatea* extracts against microbes showed that the leaf and seed extracts were more favourably compared to the rest of the extracts (sriyeta c et al., 2017).

Additionally, a research was done to investigate antimicrobial activity under minimum inhibitory concentration on *C. ternatea* flower using disc diffusion. Extractions of aqueous, methanol, petroleum ether, hexane and chloroform were used against Escherichia coli, *K. pneumoniae*, and *Pseudomonas aeruginosa* and a positive control Amikacin was used. The study indicated that the inhibitory zone of methanol extract was between 16 mm to 26 mm, and in chloroform extract between 14 mm to 18 mm. In the aqueous extract, a zone of inhibition with a diameter of 12 mm was obtained while petroleum ether and hexane extract did not exhibit any antimicrobial properties. The results show that the methanol extracts possess high antimicrobial activity as compared to chloroform and hexane extracts(Nadzirah jamil1 et al., 2018).

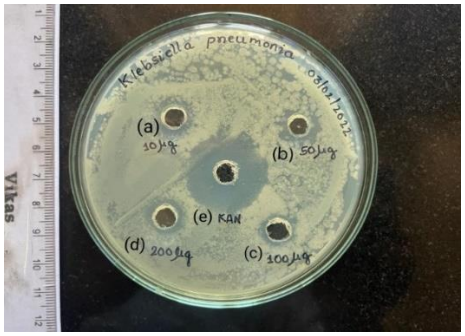

Figure ; *-Klebsiella pneumonia* Here this image shows the inhibition of *K .pneumonia* strain against aqueous extract of *Clitoria ternatea* flower. Were 200µg/mL shows good result compared with the standard strain kanamycin. Here (a)= 10µg, (b) = 50µg, (c) = 100µg, (d) = 200µg, (e) =Kanamycin.

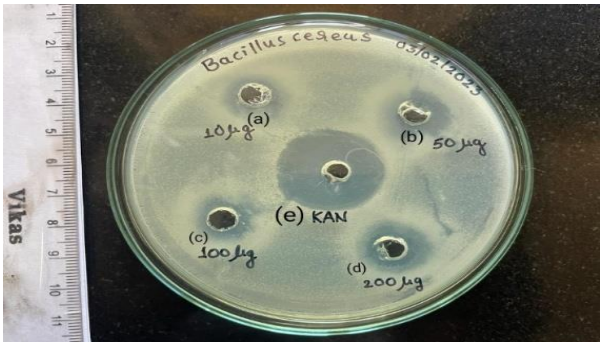

Figure ; ***Bacillus cereus*** **-**Here this image shows the inhibition of *B.cereus* strain against aqueous extract of *Clitoria ternatea* flower. Were 200µg/mL shows good result compared with the standard strain kanamycin. Here (a)= 10µg, (b) = 50µg, (c) = 100µg, (d) = 200µg, (e) =Kanamycin.

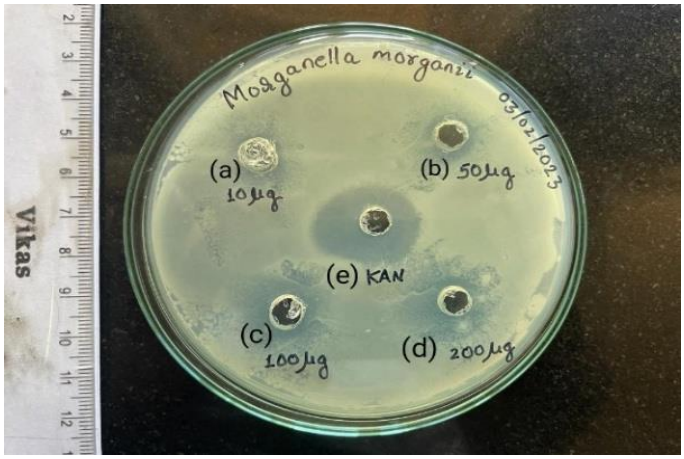

Figure ; ***Morganella morganii*** **-**Here this image shows the inhibition of *M .morganii* strain against aqueous extract of *Clitoria ternatea* flower. Were 200µg/mL shows good result compared with the standard strain kanamycin. Here (a)= 10µg, (b) = 50µg, (c) = 100µg, (d) = 200µg, (e)= Kanamycin.

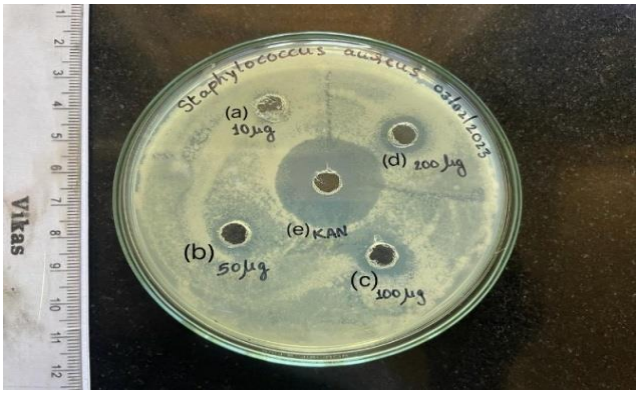

Figure ; ***Staphylococcus aureus*** **-**Here this image shows the inhibition of *S .aureus* strain against aqueous extract of *Clitoria ternatea* flower. Were 200µg/mL shows good result compared with the standard strain kanamycin. Here (a)= 10µg, (b) = 50µg, (c) = 100µg, (d) = 200µg, (e)= Kanamycin

Conclusion

It is concluded that *Clitoria ternatea* is a plant with a variety of ethnic medicinal uses. The qualitative analysis of butterfly pea flower show the presence of bioactive compounds such as flavonoids, glycosides, tannins, carbohydrates, proteins saponins, Terpenoids. It also shows antibacterial activity which inhibit the growth of microorganisms like *Bacillus cereus, klebsiellosis pneumonia, Marganella morganii, Staphylococcus aureus.* The aqueous flower extract of C. ternatea has well medicinal properties and antimicrobial activity which can treat the Urinary tract Infection(UTI) and also known to prevent diabetics.

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Reference

1. Sawan Sharma, Gouri Satpathy, Rajinder K. Gupta* Nutritional, phytochemical, antioxidant and antimicrobial activity of Prunus armenicusJournal of Pharmacognosy and Phytochemistry2014; 3 (3): 23-28.
2. P. Manjula, CH. Mohan, D. Sreekanth, B. Keerthi and B. Prathibha Devi Phytochemical analysis of Clitoria ternatea Linn., A Valuable Medicinal Plant, Vol. 92 (3&4) 2013 : 173-17.
3. Nadzirah Jamil1 and Furzani Pa'ee Antimicrobial Activity from Leaf, Flower, Stem, and Root of Clitoria ternatea – A Review. AIP Conf.Proc.2002, 0200441-1-020044-5;
4. CH.N.Durga Maha Lakshmi1, B.Deva Prasad Raju2, T.Madhavi1 and N.John Sushma1* Identification of Bioactive Compounds by FTIR Analysis and In vitriol Antioxidant Activity of Cltoria ternatea Leaf and Flower Extracts Indo American Journal of Pharmaceutical Research, 2014 ISSN NO: 2231-6876.
5. Narasimhamurthy Konappa, Arakere C. Udayashankar, Soumya Krishnamurthy, Chamanalli Kyathegowd Pradeep, Srinivas Chowdappa & Sudisha Jogaiah, GC–MS analysis of phytoconstituents from Amomum nilgiricum and molecular docking interactions of bioactive serverogenin acetate with target proteins, Published: 02 October 2020.
6. Rosalinda C. Torres1*, Ma. Rachel V. Parcon1, Harvy Jay N. Esmundo1, Danielle Camille P. Canillo1,And Cyril C. Ramil1 Antioxidant activity and phytochemical constituents of Philippine Clitoria ternatea flowers as a potential therapeutic agent against infectious diseases, Issues in Biological Sciences and Pharmaceutical Research Vol.10 (2),pp.12-18, May 2022.
7. Partibha Siddham, Randive Sonali* and Jagtap MN Phytochemical Analysis and Antimicrobial Screening of Clitoria ternatea L,Acta Scientific Microbial(ISSN: 2581-3226) Volume 6 Issue 4 April 2023.
8. Rajesh Kumar T, Santhosh Kumar R and Anju VS Phytochemical and antibacterial activities of crude leaf and root extracts of Clitoria ternatea varieties (Fabaceae) Journal of Pharmacognosy and Phytochemistry 2017; 6(6): 1104-1108.
9. Ethel Jeyaseela Jeyaraj, 1 , 2 ,† Sheila Nathan, 3 ,† Yau Yan Lim,‡ 1 ,† and Wee Sim Choocorresponding author 1 ,*†Antibiofilm properties of Clitoria ternatea flower anthocyanin-rich fraction towards Pseudomonas aeruginosa Access Microbiol. 2022; 4(4): 000343. Published online 2022 Apr 26.
10. Anupsingh Vijaysingh Thakur1, Sonu Ambwani1*, Tanuj Kumar Ambwani2, A. H. Ahmad3 and Dharmendra Singh Rawat4 Evaluation of phytochemicals in the leaf extract of Clitoria ternatea Willd. through GC-MS analysis August 2018Tropical Plant Research 5(2):200-206.
11. 1B Narmadha,2Dr. M. Job Gopinath, 3J. Crashing Prema KUMARI GC-MS Analysis of Phytochemicals in Methanolic Leaf Extracts of Commonly Available Herbal Plants Invellore Tamil Nadu, INDIA. Volume 10, Issue 10 October 2022| ISSN: 2320-2882
12. R. Kavitha Fluorescence, FT-IR and GC-MS Determination of Bioactive constituents of leaf extract of Clitoria ternatea Int J Pharm Bio Sci 2017 Apr ; 8(2): (P) 299-307.Sriyeta C, Souvagyalaxmi S, Anjana B, Sangita D (2017). Studies on antimicrobial activity, phytochemical screening tests, biochemical evaluation of clitoreaternatealinn. Plant extracts. International Journal of Research Granthaalayah, 5(10):197-208.
13. Tandon, N., Yadav, S.S., 2017. Contributions of Indian Council of Medical Research (ICMR) in the area of Medicinal plants/Traditional medicine. J. Ethnopharmacol. 197, 39–45. https://doi.org/10.1016/j.jep.2016.07.064.
14. Zhao, X., Chen, L., Lu, J., 2018. A similarity-based method for prediction of drug side effects with heterogeneous Information. Math. Biosci. 306, 136–144.https://doi.org/10.1016/j.mbs.2018.09.010
15. Mittal, A.K., Chisti, Y., Banerjee, U.C., 2013. Synthesis of metallic nanoparticles using plant extracts. Biotechnol. Adv.31, 346–356. https://doi.org/10.1016/j.biotechadv.2013.01.003
16. R. GovindarajanM. Vijayakumar, P. PushpangadanAntioxidant approach to disease management and the role of 'Rasayana' herbs of Ayurveda Journal of Ethnopharmacology, 3 June 2005 Pages 165-178.
17. Neeti N. Jain, C.C. Ohal, S.K. Shroff, R.H. Bhutada, R.S. Somani, V.S. Kasture, S.B. Kasture*Clitoria ternatea and the CNSPharmacology, Biochemistry and Behavior 75 (2003) 529–536 Pharmacology, Biochemistry and Behavior 75 (2003) 529–536.

18.Osborn RW, De Samblanx GW, Thevissen K, Goderis I, Torrekens S, Van Leuven F,et al. Isolation and characterization of plant defensins from seeds of Asteraceae, Fabaceae, Hippocastanaceae and Saxifragaceae. FEBS Lett 1995;368:257–62.
19.Anonymous. Indian medicinal plants, vol. 2. Madras: Orient Longman; 1995. p. 129–32.
20.Taranalli AD, Cheeramkuczhi TC. Influence of Clitoria ternatea on mem- ory and central cholinergic activity in rats. Pharm Biol 2000;38:51–6.
21.Lin, W. S., He, P. H., Chau, C. F., Liou, B. K., Li, S., and Pan, M. H. (2018). The feasibility study of natural pigments as food colorants and seasonings pigments safety on dried tofu coloring. Food Sci. Hum. Wellness 7, 220–228. Doi: 10.1016/j.fshw.2018.09.002
22.Dilrukshi, P. G. T., Munasinghe, H., Silva, A. B. G., and De Silva, P. G. S. M. (2019). Identification of synthetic food colours in selected confectioneries and beverages in jaffna District, Sri Lanka. J. Food Qual. 2019:453169. doi: 10.1155/2019/7453169
23.FDA (2018). Q3C–Tables and List Guidance for Industry. Available online at: www.fda.gov/regulatory-information/search-fda-guidance-documents/q3c-tables-and-list-rev-4 (accessed September 8, 2021)
24.Asadnejad, S., Nabizadeh, R., Nazarinia, A., Jahed, G. R., and Alimohammadi, M. (2018). Data on prevalence of additive colors in local food and beverage products, Tehran, Iran. Data Brief 19, 2104–2108. Doi: 10.1016/j.dib.2018.07.001
25.Gayan Chandrajith Vidana Gamage. Yau Yan Lim. Wee Sim Choo.Anthocyanins From Clitoria ternatea Flower: Biosynthesis, Extraction, Stability, Antioxidant Activity, and Applications 17 December 2021Volume 12 – 2021 |
26.Sen, T., Barrow, C. J., and Deshmukh, S. K. (2019). Microbial pigments in the food industry—challenges and the way forward. Front. Nutr. 6:7. Doi: 10.3389/fnut.2019.00007
27.Feketea, G., and Tsabouri, S. (2017). Common food colorants and allergic reactions in children: myth or reality? Food Chem. 230, 578–588. Doi: 10.1016/j.foodchem.2017.03.043
28.Choo, W. S. (2019). “Fruit pigment changes during ripening,” in The Encyclopedia of Food Chemistry, eds L. Melton, F. Shahidi, and P. Varelis (Elsevier: The Netherlands), 17–123.
29.Landim Neves, M. I., Silva, E. K., and Meireles, M. A. A. (2021). Natural blue food colorants: consumer acceptance, current alternatives, trends, challenges, and future strategies. Trends Food Sci. Technol. 112, 163–173. Doi: 10.1016/j.tifs.2021.03.023
30.Gustiningtyas, A., Setyaningsih, I., Hardiningtyas, S. D., and Susila, A. A. R. (2020). Improvement stability of phycocyanin from Spirulina platensis encapsulated by water soluble chitosan nanoparticles. IOP Conf. Ser. 414:012005. Doi: 10.1088/1755-1315/414/1/012005
31.Buchweitz, M. (2016). “Natural solutions for blue colors in food,” in The Handbook on Natural Pigments in Food and Beverages, eds R. Carle and R. M. Schweiggert (Oxford: Woodhead Publishing), 355–384. Doi: 10.1016/B978-0-08-100371-8.00017-8
32.Jing, P., and Giusti, M. M. (2007). Effects of extraction conditions on improving the yield and quality of an anthocyanin-rich purple corn (Zea mays L.) color extract. J. Food Sci. 72, C363–C368. Doi: 10.1111/j.1750-3841.2007.00441.x
33.Salehi, B., Sharifi-Rad, J., Cappellini, F., Reiner, Ž., Zorzan, D., Imran, M., et al. (2020). The therapeutic potential of anthocyanins: current approaches based on their molecular mechanism of action. Front. Pharmacol. 11:1300. Doi: 10.3389/fphar.2020.01300
34.Khoo, H. E., Azlan, A., Tang, S. T., and Lim, S. M. (2017). Anthocyanidins and anthocyanins: colored pigments as food, pharmaceutical ingredients, and the potential health benefits. Food Nutr. Res. 61:1361779. Doi: 10.1080/16546628.2017.1361779
35.Garcia, C., and Blesso, C. N. (2021). Antioxidant properties of anthocyanins and their mechanism of action in atherosclerosis. Free Radic. Biol. Med. 172, 152–166. Doi: 10.1016/j.freeradbiomed.2021.05.040
36.Nair, V., Bang, W. Y., Schreckinger, E., Andarwulan, N., and Cisneros-Zevallos, L. (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. Doi: 10.1021/acs.jafc.5b00928
37.Nadzirah Jamil1 and Furzani Pa’ee2, 3, a)Antimicrobial Activity from Leaf, Flower, Stem, and Root of Clitoria ternatea – A Review AUGUST 15 2018

38. Citation: Buddhika, H.D.K.; Dharmadasa, R.M.; Menuka Arawwawala, L.D.A.; Pakeerathan, K. Phytochemical Properties of Clitoria ternatea L. (Fabaceae)—A Distinct Flower Morphometric Plants Available in Sri Lanka. Proceedings 2021, 68, x. https:// doi.org/10.3390/xxxxx