International Journal of Pharmaceutical and Bio-Medical Science

ISSN(print): 2767-827X, ISSN(online): 2767-830X

Volume 04 Issue 04 April 2024

Page No: 370-377

DOI: https://doi.org/10.47191/ijpbms/v4-i4-18, Impact Factor: 7.792

Preliminary Phytochemicals Investigation in Begonia Cucullata

Ramesh Londonkar[1], Hemalatha S.A.[2]

[1]Professsor, Department of Biotechnology, Gulbarga University, Kalaburagi, (Karnataka), India.

[2]Ph. D Scholar, Department of Biotechnology, Gulbarga University, Kalaburagi, (Karnataka), India

ABSTRACT

People specifically or by implication profited of plants. *Begonia cucullata*, a perpetual blooming plant taxonomically considered to the family of Begoniaceae. *Begonia cucullata* is local to the all-South American nations. In India found in Assam, Meghalaya and Manipur. The shade dried plant powder was taken for extraction through organic solvents like petroleum ether, chloroform, methanol and water extract. The Biochemicals investigation is performed by taking after standard methods. Primary phytoconstituents investigation of the plant extract indicated the existence of carbohydrates, glycosides, flavonoids, phytosterols, and saponins in methanol and watery extricate. The plant extricate of methanol and refined water have appeared most extreme positive tests for Phytochemicals, which clearly shows that the existence of a few dynamic standards within the plant that are having vital restorative esteem.

KEYWORDS: Phytochemicals anayysis, Begonia cacullata, Soxhlet extraction, flavonoids,active principles.

ARTICLE DETAILS

Published On:
22 April 2024

Available on:
https://ijpbms.com/

INTRODUCTION

The plants are necessarily portion of nature and valuable for human. The herbal medicines have been used since past several years for treatment of various diseases. Plant origin biochemicals are devoid of nutrition such chemicals act as curative agents (Suresh.S.N *et al., 2011*). In the medical management of Global South and Global North nations, the phytomedicines highlight over miracle drugs. The WHO report explains majority of the countryside population in the Global South and Global North nations of world continue to use phytomedicines as their well ness. As phytomedicines are affordable, more efficient with less or no aftereffect in contrast to modern prescription (Jayaprakash.K *et al.,* 2011). In any conventional framework of pharmaceutical, cleaned and dried plant parts are ordinarily utilized in making phytomedicines. Rectify information of such drugs is exceptionally critical viewpoint in planning and security of phytomedicines (Gokhale.S.B *et al.,* 1995). Phytochemicals play a crucial part in treatment of number of maladies such as asthama, joint pain, malignancy etc.Begonia plant is reported as a repository of anthocyanin. Carl Ludwig Willdenow described it in 1805. It is a succulent herb and it can reach a highest point of 0.6 m . The leaf toothed with the teeth tipped by a hair otherwise the margins are strongly wavy. The stems have a glossy pale green to reddish brown appearance. The petals are rose pink with a white-tinge or pink and the leaves are pale to dark green with 9cm length and 7.3cm width. Begonia moreover known as Wax begonia as these are well known bedding plants with erect stems and reflexive circular takes off, regularly green or bronze in color. The blooms are up to 6 cm and many in number, showing up in changing shades of ruddy, pink and white. They are ordinarily delivered from seed and developed as annuals, with proper care and by growing indoors in winters can prolong their life.

Leaves are pale to dark green with 9cm length and 7.3cm width. Begonia moreover known as Wax begonia as these are well known bedding plants with erect stems and reflexive circular takes off, regularly green or bronze in color. The blooms are up to 6 cm and many in number, showing up in changing shades of ruddy, pink and white. They are ordinarily delivered from seed and developed as annuals, with proper care and by growing indoors in winters can prolong their life.

Table1. Illustrating Classification of Begonia cucullata

Kingdom	Plantae
Division	Tracheophyta
Class	Magnoliopsida
Order	Cucurbitales
Family	Begoniaceae
Genus	Begonia
Species	*Begonia cucullata* Willd

Begonia could be a very diverse genus that includes more than 1800 species, with maximum levels of micro endemics and diversity hotspots within the South Asia and Andes (Huges *et al.*, 2018). The primary live plant in Genus Begonia was presented of Europe in the nineteenth century, and later more than 400 common varieties were presented as the gardening plants (Tebbitt, 2005). Begonias are among the foremost well-known decorative plants within the world (Sakhanokho *et al.*, 2013; Twyford *et al.*, 2014). Begonias were utilized for nursery purpose; pruned plant along with leaf in salads in numerous Countries, the underground parts and rhizome of a few varieties were detailed to posses germicidal properties so were utilized in curing different sicknesses (Sakhanokho *et al.*, 2013).

Begonias are highlighted for their pain relieving, anti-inflammatory, spasmolytic, constringent and atomachic properties, used as plaster and to treat vision related problems (Mlcek *et al.*, 2011). The radiantly colored Begonia infloresence exhibit delightful, slight vinegary flavour with fresh surface (Lim 2014). Servings of mixed greens, sandwiches and garnishes are prepared using their petals (Creasy 1999). Flowers of ornamental plants used for their flavors, colors and pleasing shapes. KFDA acknowledges *Begonia cucullata* flowers as edible and can be used as food ingredients (Jung - Hua kwon *et al.*, 2019).

In the preparation of fish and meat begonia leaves used as flavoring agent. The studies show that, Begonia species is a repository of Vitamin C. It is used in medication and food. In the Country Nepal, a few varieties of Begonia were utilized for nourishment and in medication. Within the West Indies, tea prepared from Begonia leaves are consumed for curing commom cold. Begonia's underground part and rhizome were utilized in laxative preparation. It is reported that the diseases such as respiratory tract diseases, diarrhea, blood cancer, and skin maladies might be cured by Begonia leaves. Begonia leaves were used against HIV infections (Kiritkar and Basu ,1975; Wu *et al.*,2004). Reports revealed the details of Begonia class containing about 57 varieties recorded in India that are disseminated as 7 areas; most varieties are found in Himalayas- "The Land of Snow'' , South Indian and the country Shri Lanka (Uddin, A 2007).

It is reported as positive test when tested for the several phytoconstituents and negative test for alkaloids in the plant extracts of *Begonia malabarica* (Ramesh N *et al*., 2002); the Biochemical tests of *Begonia floccifera* in organic flower extracts were carried out showed positive tests for secondary metabolites (Jeeva S, 2012). Several reports indicate the biochemical compositions of flowers of Begonia species both native and decorative species are showing the presence of secondary metabolites, isotiocyanates, phenolics, probiotics phytooestrogens, fibres, indispensable amino acids and vitamins. (Kaisoon *et al*., 2011, Cavaivolu *et al*., 2013, Lim 2014, Koike *et al*., 2015). The biochemical tests of *Begonia picta* indicated positive for various secondary metabolites (Shrestha N *et al.*, 2015); *Begonia nelumbiifolia* confirmed the presence of organic acids, carotenoids, flavonoids in both leaf and stalk extracts (Nemesio villa *et al.*, 2017); *Begonia nelumbiifolia*, a traditional edible plant of Puebla, Mexico. The Phytochemicals analysis of *Begonia roxburghii* in hexane extract tested positive for secondary metabolites (Horne Mubarak *et al.*, 2018). The study of *Begonia grandis* by histochemical method showed presence of secondary metabolites in the leaf exudates (Karpova *et al*., 2019). The Phytochemicals investigation carried out on *Begonia versicolar* leaves indicated positive for phytoconstituents (Abriyani Ermi *et al.*, 2020); the biochemical testing of few Begonia species, namely *Begonia megaptera*, *Begonia vanithina* and *Begonia palmata* reported positive for phytoconstituents and absence of few secondary metabolites, mainly alkaloids (Bhattarai.B. *et al* ., 2020) ; the biochemical studies indicate positive for secondary metabolites in *Begonia medicinalis* in Phytochemicals screening carried out by Ultrasound Assisted Extraction method (Prihardina B *et al*., 2021) .

The research studies on phytoconstituents attain academic interest in today's world. The knowledge of existence of phytoconstituents in plants material definitely makes way to isolate and identify final compounds in the selected plant. From the available literature it was learnt that there is very less work carried out on Phytochemicals analysis of *Begonia cucullata.* The literature review revealed the use of Begonia species in food and medicine, in the treatment of diseases, presence of bioactive and nutraceutical compounds in the plant. This attracted attention to select *Begonia cucullata* plant for Phytochemicals analysis.

MATERIALS AND METHODS

The Department of Biotechnology, Gulbarga University, Kalaburagi was selected place to perform laboratory work, and work carried out from Feb 2022 to Aug 2022.

Distribution and Collection of Plant material: This plant Begonia is extensively cultivated in the nurseries of Pune, Maharashtra. For current investigation the plant was procured from local nurseries of Pune, Maharashtra during Feb 2022. And authenticated by Prof Rajasamarsen Modi, Associate Professor and Head Department of Studies and Research in Botany, Autonomous Government College, Kalaburagi. A Herbarium specimen was preserved bearing Voucher No HGUG.17.

PLANT EXTRACT PREPARATION

Entire collected plant was cleaned using tap water and allowed to dehydrate for several days. The powdered plant material was utilized for the process of extraction with various organic solvents and distilled waterThe powdered plant material was subjected for the process of extraction with organic chemiclas namely Petroleum ether in first , Chloroform in second extraction and followed by methanol making use of 1000 ml capacity Soxhlet apparatus and aqueous extraction by boiling in distilled water (Azwanida N N 2015). The plant powder was placed in a "thimble". Filter paper can be used to make thimble. In the round bottom flask the respective organic solvent is placed where the dissolved compounds are drawn out into the solvent.

50 g of plant material was immersed in 800 ml respective solvent and extraction was performed for 10 to 12 hrs at temperature 60 to 65˚C in heating mantle for organic solvents. Later solvent with extract was allowed to evaporate and the plant extract was used for further analysis .The aqueous extraction was performed by boiling powered plant material in 800 ml volume of distilled water kept for 2hrs and cooled for some time and the dried filtrate was collected. The percentage of yield for petroleum ether, chloroform, methanol and aqueous extraction was 1.62g, 1.1g, 5.34g and 2.32g respectively.

To calculate the yield in percent the following formula is applied:

$$\text{Yield (\%)} = \frac{\text{Dried quantity of extract}}{\text{Dried quantity of plant material}} \times 100$$

Phytochemicals Screening of the plant extracts: Preliminary Phytochemicals tests were performed to detect phytoconstituents. The procedures for the Phytochemicals tests were followed as per scientific recommendation. The plant extract dissolved in respective solvent (50g/50ml) and taken into different test tubes. The Phytochemicals analysis was done using plant extract taken in three organic solvents and one in water extract in four labeled test tubes. The below mentioned tests were performed for Phytochemicals testing.

Tests for carbohydrates:In respective solvent the plant extract dispensed into labelled test tubes and the following carbohydrate tests were performed (Raaman N, 2006).

Molisch's test: In a labelled test tube take in 2 ml of plant extracts add few drops of α-naphthal prepared in alcohol is added. With the help of dropper slowly drop concentrated sulphuric acid on one side of test tube keeping the tube in inclined position. The purple ring appears at the interface of two layers interprets carbohydrate positive test.

Benedict's test: Take in 2 ml of Plant extracts and drop equal volume of Benedict's reagent and keep for boiling in water bath. The test indicates positive for reducing sugars on appearance of orange and slight red precipitate.

Fehling's test: Take in 2 ml of plant extract and 1ml each of A- Fehling's solution and B- Fehlings solution in a test tube and kept in water bath for boiling. The tests interpreted positive for reducing sugars with occurrence of red precipitate.

Anthrone test

Take 2 ml **of** plant extracts, 2ml water and 2ml anthrone reagent was added, the mixture were mixed properly and warmed gently. Change in colour to green or blue shows the test is positive for carbohydrates.

Alkaloids test

Mayer's test: Take 2ml of plant extracts and from the sides of test tube add Mayer's reagent dropwise 2 to 3 drops slowly. The test is confirmed as positive for alkaloids with the appearance of white creamy precipitate. (Evans .W.C, 1997).

Wagner's test

Take in 2ml of plant extracts in the test tube and from one side of test tube add 8 to 10 drops Wagner's reagent. The test indicates positive for alkaloids with the appearance of brownish or reddish precipitate (Wagner, 1993).

Test for amino acids

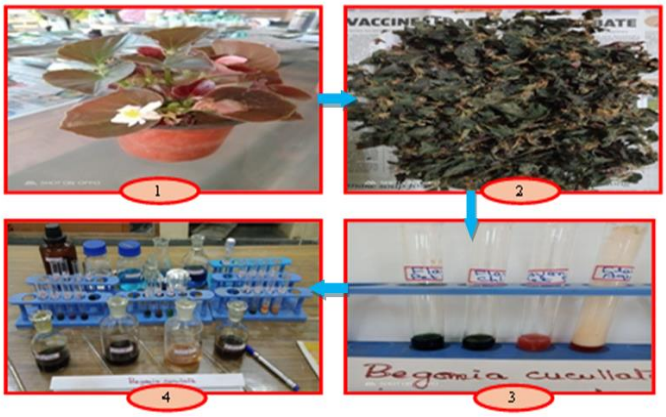
Ninhydrin test: Take 2 ml of plant extracts and mix with Ninhydrin solution and boil the mixture for two minutes. If there is change to purple colour in the solution this confirms for amino acids. (Yasuma *et al.*, 1953).

To test fixed oils and fats

Spot test: 2-3 drops of 0.5 N alcoholic solution of potassium hydroxide with a drop of phenolphthalein were added to 50 to 100 ml of plant extracts in a small conical flask and cover the mouth of conical flask with aluminum foil, keep the conical flask with mixture on boiling water bath for two hrs. It is better to take more quantity of reaction mixture to prevent evaporation. If soap formation or partial neutralization of alkali is observed, this shows the presence of fixed oils and fats (Raaman N, 2006).Test for glycosides

Take 2 ml of plant extracts and add equal volume of glacial acetic acid and add ferric chloride and from one side

of test tube add 2 ml of concentrated sulphuric acid. Appearance of brownring at the intersection of 2 layers shows the nearness of glycosides (Harborne, J.B.1973).



1. *Begonia cucullata* plant 2. *Begonia cucullata plant* kept for drying
3. Test for flavonoids 4. Phytochemicals tests

Fig.1. Phytochemicals tests of *Begonia cucullata* plant

Concentrated sulphuric acid test

To 2 ml of plant extracts add equal volume of Concentrated H_2SO_4 into a clean tube and the mixture were left undisturbed for few minutes. The test indicates positive for glycosides with appearance of red colour.

To test phenolic compounds and tannins

Phenol test: Put in 3-4 drops of freshly prepared 5% ferric chloride and add 2 ml of plant extracts. colour change to blue black or dark green indicates the test is positive for phenols (Tiwari P *et al*., 2011) .

Ellagic test: To 2 ml of plant extract add 3 to 4 drops of 15% freshly prepared glacial acetic acid and few drops of sodium nitrite. Appearance of brown precipitate confirms test positive for phenol.

Ferric chloride test: Take 2 ml of plant extracts and mix with 3 drops of 5% solution of ferric chloride . colour change to dim greenish demonstrates the test is positive for phenolic compounds (Raaman N, 2006).

Lead acetate test: Take 5ml of The plant extracts in a clean glass tube and add 3 ml of 10% solution of lead acetate and mix thoroughly. Appearance of white precipitate shows the test is positive for phenolic compounds (Raaman N, 2006).

Gelatin test:

To 2 ml of plant extracts add 1 ml of gelatin solution, include 1 ml of 10% Nacl . Appearance of white precipitate shows the test is positive for phenolic compounds and tannins (Evans .W.C, 1997).

Flavonoid test

Flavonoid test: Take 2 ml of plant extracts and add few drops of concentrated sulphuric acid and magnesium turnings. Pink or magenta colour shows the test is positive for flavonoids (Raaman N 2006).

Magnesium and hydrochloric acid reduction:

Take 20 mg plant extracts in 2 ml alcohol to this mixture adds magnesium turnings and concentrated HCl. Pink colour to crimson demonstrates the test is positive for flavonol glycosides (Raaman N, 2006).

Ferric chloride test: To 2 ml of plant extricate add few ml of 5% solution of ferric chloride . Appearance of green precipitate demonstrates the nearness of flavonoids (Audu .S.A *et al.*, 2007) .

Lead acetate test: 2ml of plant extract were mixed with 0.5 ml of 10 % lead acetate. Colour change to yellowish demonstrates the test is positive for flavonoids (Tiwari P *et al*., 2011).

Liebermann –Bur chard's test: Put in 2.5 ml of acetic anhydride and add 50 mg of plant extract . To the mixture from one side of tube slowly drop concentrated sulphuric acid. A cluster of Color appearance in the solution indicates the test is positive for phytosterols (Raaman N 2006).

Salkowaski's test: Dissolve plant extract in chloroform and separate the filtrate. To the filtrate add 3 to 4 drops of acetic anhydride and leave the tube in hot water bath for few sometime. On cooling the solution occurrence of brown zone at the intersection point confirms the test is positive for phytosterols (Tiwari P *et al.*, 2011).

Test for proteins

The aqueous and organic extracts were used for protein test. Experiment protocol of Raaman N(2006) was followed:

Biuret test: Take in few drops of 2% copper sulphate and mix with 2ml of plant extract, put in the tube 95% of ethyl alcohol half the volume of plant extract along with 1 or 2 pellets of potassium hydroxide. Colour change demonstrates

the positive test for proteins with appearance of pink. (Raaman N 2006).

Test for saponins:

Take on 50 mg of plant extracts and mix with pure water. Transfer the above solution of 20 ml volume to a measuring cylinder and thoroughly shake for 20 minutes. A layer of foam formation shows the test is positive for saponins (Banu KS *et al.*, 2015)Results And Discussion On the literature available it can be said that only few researchers have studied the Phytochemicals analysis of *Begonia cucullata* . The Phytochemicals tests carried out answered positive for of carbohydrates, glycosides, flavonoids, phytosterols and saponins, and the tests answered negative for alkaloids, phenolic compounds, fixed oils and fats, amino acids , proteins and tannins in the solvent extracts of *Begonia cucullata* (Table. 2). The biochemical tests with methanol and aqueous extract of *Begonia cucullata* have shown positive for carbohydrates, flavonoids, phytosterols, saponins and glycosides; and the tests with petroleum ether extract and chloroform extract of *Begonia cucullata* have shown positive for carbohydrates. The maximum metabolites were soluble in methanol and distilled water and few are soluble in petroleum ether and chloroform. results revealed the presence of carbohydrates and several phytoconstituents in the solvent extract of *Begonia cucullata* (Table. 2). The extract of *Begonia cucullata* have shown negative test for proteins and secondary metabolites but evidence the presence of sugars and several phytoconstituents in methanol and aqueous extract also revealed the presence of sugars in extricate of petroleum ether, and chloroform. The maximum metabolites were soluble in methanol and distilled water and few are soluble in petroleum ether and chloroform.

The Phytochemicals investigation for secondary metabolites from plants is attaining scientific interest due to health benefits of phytoconstituents as nutraceuticals, medicines, food ingredients and drugs. The results of Phytochemicals examination comprehensively approve the nearness of remedially imperative and important Phytochemicals such flavonoids, phytosterols, saponins and glycosides etc. etc. The natural world has been a origin of therapeutic specialist for considerably long time with a noteworthy of numerous advanced drugs have been purified from characteristic biological sources (Thoudam.B. *et al*., 2011) . *Begonia cucullata* having a place to the family Begoniaceae are broadly developing plant all through India.

Studies from the reports show that the biochemicals in the Begonia species are the secondary metabolites with curative properties (Suresh *et al*., 2009). Phytochemicals analysis are carried out with several organic solvents distilled water to investigate biochemicals solubility in different extraction solvents, influenced by the polarity of organic solvents. Hence it is necessary to find out the Phytochemicals present in the plant extract and check for their bioactive properties. The biochemical testing conducted by Jeeva *et al.*, and Ariharan *et a*l., (2012) with extricate of *Begonia malabarica* and *Begonia floccifera* indicates existence of several secondary metabolites specially Vitamin C.

The similar results with respect to flavonoids were shown in the following studies of Karpova *et al*., the flavonoids test were positive in many Begonia species and cultivars of Begonia genus, tests performed by Karpova *et al*., (2009). The Phytochemicals studies of *Begonia roxburghii* in hexane extract showed positive for the flavonoid tests ,glycosides, tannins, alkaloids ,carbohydrates and saponins (Hosne Mobarak *et al*., 2018). The biochemical tests carried out using the methanol flower extracts of *B.floccifera* indicates positive for maximum phytoconstituents (Jeeva *et al.*, 2012). Reports studied, Preliminary Phytochemicals screening of methanolic extract of *B. floccifera* flower (Jeeva *et al*., 2012) and of *Begonia picta* (Shrestha N *et al.*, 2016) proved to be positive for many phytoconstituents and and negative for alkaloids in both cases.

For the Phytochemicals present in methanol extract, the present study is similar to previous studies. It may be because of Phytochemicals similarity in the genus Begonia. Studies reported the biochemical tests positive for flavonoids, carotenoids and organic acid in the leaf and stalk preparation of *Begonia nelumbiifolia* (Nemesio villa *et al.*, 2017) .Studies revealed positive for flavonoids, phenolics, terpenoids, steroids, alkaloids, saponins, carbohydrates and reducing sugars in the Phytochemicals screening of methanol extract of *Begonia versicolar* (Abriyani ermi *et al.*, 2020). The biochemical testing of organic preparation of *Begonia xanthina* tested positive for flavonoids, phenolic compounds, glycosides, saponins, carbohydrates etc and negative for alkaloids (Bhattarai.B. *et al*., 2020) .The findings of present biochemical testings with the methanol extract of *Begonia cucullata* also shows positive for phenolic compounds, flavonoids and negative for alkaloids , resembling the earlier testing outcome. . The high flavonoid content in the distilled water extract of *Begonia medicinalis* reveals the potential of anticancer abilities (Priharidina *et al.*, 2021), in present study with aqueous extract of *Begonia cucullata* the flavoniod is tested positive. Plants are important source of chemotherapeutic agents. From the reports studied, a high content of secondary metabolites were present in the flowers of *Begonia cucullata*, NMR and MS studies have indicated positive for anthocyanins this also shows polyophenols and flavonoids to be present in Begonia. Few flavonoids were isolated from the organic extricate of *Begonia cucullata* inflorescence, by TLC separation. Further with NMR and HPLC studies , the four flavonoids were identified as quercetin, kaempferol, astragalin and isoquercetin, experimented by Jung - Hua kwon *et al.*, (2019).

Table 2.Illustrating the Phytochemical analysis of *Begonia cucullata.*

Sl No.	Solvent	Petroleum ether	Chloroform	Methanol	Distilled water
1.	**Alkaloids**				
	Mayers test	-	-	-	-
	Wagners test	-	-	-	-
	Hagers test	-	-	-	-
2.	**Amino acids**				
	Ninhydrin test	-	-	-	-
3.	**Carbohydrates**				
s	Molischs test	+	+	+	+
	Benedicts test	-	-	-	-
	Fehlings test	-	-	-	-
	Anthrone test	-	-	-	-
4.	**Fixed oil and fats**				
	Spot test	-	-	-	-
	Saponification	-	-	-	-
5.	**Glycosides**				
	Killer killani test	-	-	+	+
	Conc. sulphuric acid test	-	-	+	+
6.	**Phenolic compounds and tannins**				
	Phenol test	-	-	-	-
	Ellagic test	-	-	-	-
	Ferric chloride test	-	-	-	-
	Lead acetate test	-	-	-	-
	Gelatin test	-	-	-	-
7.	**Flavonoids**				
	Flavonoids test	-	-	+	-
	Magnesium and hydrochloric acid reduction	-	-	+	-
	Ferric chloride test	-	-	-	-
	Lead acetate test	-	-	+	+
8.	**Phytosterols**				
	Liberman -Burchard's test	-	-	+	+
	Salkowaski test	-	-	+	+
9.	**Proteins**	-	-	-	-
10.	**Saponins**	-	-	+	+

(+ indicates presence; - indicates absence)

CONCLUSIONS

It is well known fact that the Phytochemicals present in the plant are having immense medicinal importance. In the present investigation the preliminary testing of phytoconstituents in the plant *Begonia cucullata*, tested positive for several secondary metabolites.. This proved that the plant is reserve of Phytochemicals which exhibit a significant role in treating various ailments and repository of pharmaceuticals.

Future Scope: The present study indicates the existence of phytoconstituents applicable as therapeutics, in the plant extract. In this regard further studies need to be carried out to explore *Begonia cucullata* for its potential and specific isolation of compound

responsible in preventing and treating various diseases. Thus the current research investigations help in the preparation of novel drug candidate.

ACKNOWLEDGEMENTS

To the Department of Biotechnology, Gulbarga University, Kalaburagi both the authors are thankful for providing the necessary infrastructure to carry out the research work.

CONFLICT OF INTEREST

All the authors declare that there is no conflict of interest.

REFERENCE

I. Abriyani, E., & Fikayuniar, L. (2020). Screening Phytochemicals, antioxidant activity and vitamin c assay from bungo Perak-perak (*Begonia versicolar irmsch)* leaves. Asian journal of pharmaceutical research, 10(3), 183-187.

II. Ariharan, V.N., Meena Devi, V.N., Rajakokhila, M., & Nagendra Prasad, P. (2012). A new natural source for vitamin C. In International Journal of plant, Animal and environmental sciences, 2(3), 92-94.

III. Audu, S.A., Mohammad, I., & Kaita, H. (2007). Phytochemicals screening of the leaves of Lophira lanceolata (Ochanaeae). Life science Journal, 4(4), 75-79.

IV. Azwanida, N.N. (2015). A review on the extraction methods use in medicinal plants, principle, strength and limitation. Medicinal and aromatic plants, 4(3), 1-6.

V. Banu, K.S., & Cathrine, L. (2015). General techniques involved in Phytochemicals analysis. International journal of advanced research in chemical science, 2(4), 25-32.

VI. Bhattarai, B., & Rana, M. (2020). Diversified morphological and Phytochemicals screening of wild Begonia of Sikkim Himalaya. Eco Env Cons, 26, S129-S138.

VII. Cavaiuolo, M., Cocetta, G., & Ferrante, A. (2013). The antioxidant changes in ornamental flowers during development a senescence. Antioxidants, 2, 132-155.

VIII. Creasy, R. (1999). The edible flower garden. Periplus, Singapore.

IX. Evans, W.C. (1997). Trease and Evans Pharmacognosy. Harcourt Brace and company. Asia Pvt Ltd. Singapore, 33.Wagner, "Pharmazeutische Biologie", 5th Edition, AUFI.15 BN 3-437-20 498-X, 1993.

X. Gokhale, S.B., Kokate, C.K., & Purohit, A.P. (1995). A textbook of pharmacognosy, Pun, India; Nirali Prakashan.

XI. Harborne, J.B. (1973). Phytochemicals Methods. Chapman and Hall Ltd., London, 49-188.S.

XII. Huges, M., Peng, C., Lin, C.W., Rubite, R.R., Blanc, P., & Chung, K.F. (2018). Chloroplast and nuclear DNA exchanges among Begonia sect. Baryandra species (Begoniaceae) from Palawan Island, Philipines and descriptions of five new species. In Plos One, 13(5), eo194877. httos://doi.org/10.1371/journal.pone.019487.

XIII. Jayaprakash, K., Ayyanar, M., Geetha, K.N., & Sekar, T. (2011). Traditional use of medicinal plant among the tribal people in Theni District (Western Ghats). Southern India Asian Pac J Trop Biomed, 1(1), S20-S25.

XIV. Jeeva, S., & Marimuthu, J. (2012). Anti–bacterial and phytochemical studies on methanolic extracts of Begonia floccifera Bedd. flower. Asian Pacific Journal of Tropical Biomedicine, 2(1), S151-S154.

XV. Jung-Hwa Kwon et al. (2019). Pharmacological activity and quantitative analysis of flavinoids isolated from flowers of *Begonia semperflorens* Link et Otto. Appl Biol Chem. 62, 11.

XVI. Karpova, E.A., Krasnikov, A.A., Fershalova, T.D., Baikova, E.V., Petruk, A.A., & Yakimova, Y.L. (2019).Phenoliccompounds and antimicrobial properties of Begonia grandis Dryand. subsp. grandis leaves. Botanical Pacifica: a journal of plant science and conservation, 8(2), 51-61.

XVII. Kaisoon, O., Siriamornpun, S., Weeapreeyakul, N., & Meeso, N. (2011). Phenol compounds and antioxidant activities of edible flowers from Thailand. J. Funct. Foods, 3, 88-99.

XVIII. Karpova, E.A., Kchramova, E.P., & Fershalovas, T.D. (2009). Flavonoids and ascorbic acid in the representatives of the genus *Begonia* L. In Himija Rastitel'nogoSyr'ja (Chemistry of the Plant Raw Material), 2, 105-110.

XIX. Kiritkar, K.R., & Basu, B.D. (1975). Indian Medicinal Plants, Indological and Oriental Publishers, Delhi, 2nd Edition, 215.

XX. Wu, P.L., Lin, F.W., Wu, T.S., Kuoh, C.S., Lee, K.H., & Lee, S.J. (2004). Cytotoxic and anti- HIV principles from the rhizomes of Begonia nantoensis. Chemical and pharmaceutical bulletin, 52(3), 345-9.

XXI. Koike, A., Barreira, J.C.M., Barros, L., Santos-Buelga, C., Villavicencio, A.L.C.H., & Ferreira, I.C.F.R. (2015). Edible flowers of *Viola tricolour L*, as a new functional food: Antioxidant activity,

individual phenolics and effects of gamma and electron-beam irradiation. Food chem, 179, 6-14.

XXII. Kokate, C.K. (1999). Practical Pharmacognosy 4th edition, Vallabh Prakashan Publication, New Delhi, India.

XXIII. Lim, T.K. (2014). Edible medicinal and non-medicinal plants. Flowers, Vol.7 and 8. Springer science+ Business media, Dordrecht.

XXIV. Mace, M.D. (1963). Histochemical localization of phenols in healthy and diseased tomato roots. Phytopathology, 16, 915-925.

XXV. Mlcek, J., & Rop. (2011). Fresh edible flowers of ornamental plants- A new source of neutraceutical foods. Trends in Food Science Tech, 22, 561-569.

XXVI. Mobarak, H., Meah, M.S., Sikder, N., Tareq, M., Azad, A., Khatun, R., & Reza, A.A. (2018). Investigation of preliminary phytochemicals, analgesic, anti-arthritic, thrombolytic and Cytotoxic Activities of Begonia Roxburghii (Mix.) DC. Leaves. Med One, 3(1).

XXVII. Nikam, M.B., Mundada, P.S., Kadam, D.A., Jadhav, S.N., & Aparadh, V.T. (2013). Comparative Screening of Various solvent for Phytochemicals Testing Using Some Commelinaceae Members. (IRJPAS), 3(2), 18-20.Prihardina, B., & Fatmawati, S. (2021). Cytotoxicity of *Begonia medicinalis* aqueous extract in three cancer cell line. In IOP Conference Series: Earth and Environmental Science, 913(1), 012-084. IOP Publishing.

XXVIII. Raaman, N., (2006). Phytochemicals techniques. Edn.1, New India Publishing agency, New Delhi, 19.

XXIX. Ramesh, N., Viswanathan, M.B., Saraswathy, A.,Balakrishna, K., Brindha, P.,& Lakshmanaperumalsamy, P.(2002). Phytochemicals and antimicrobial studies of *Begonia malabarica*. Journal of Ethno pharmacology.

XXX. Sakhanokho, H.F., Pounders, C.T., & Blythe, E.K. (2013). Algenate encapsulation of Begonia microshoots for short –term storage and distribution. In Scientific World Journal, 2013, 341568. http://dx.g/10.1155/0213/341568.

XXXI. Shrestha, N., Itani, R., & Khanal, D.P. (2016). Pharmacognostic, Phytochemicals, antioxidant and antibacterial activity studies on *Begonia picta*. World Journal of Pharmaceutical Research, 5(1), 979-997.

XXXII. Suresh, S.N., Sagadevan, P.S., Rathish Kumar, Rajeshwari, V. (2011). Phytochemicals analysis and antimicrobial potential of *Abitulon indicum* (Malvaceae). IJPRD, 4(2), 132-135.

XXXIII. Tibbett, M.C. (2005). Begonias: Cultivation, Identification, and Natural History. Timber Press, Portland.

XXXIV. Thoudam, B., Kirithika, T., Ramya, J., & Usha, K. (2011). Phytochemicals constituents and antioxidant activity of various extract of corn silk (Zea maysL.). Research Pharmaceutical, Biological and Chemical Sciences, 2(4), 986-993.

XXXV. Twyford, A.D., Kidner, C.A., & Ennos, R.A. (2014). Genetic differentiation and species cohesion in two widespread central American Begonia species. In Heredity (Edinb), 112(4), 382390. http://dx.doi.org/10.1038/hd y.2013.116.

XXXVI. Uddin, A. (2007). Distribution and Status of Indian Begonia species. J. Econ. Taxon. Bot, 31(3), 591-597.

XXXVII. Uddin, A., Gogoi, R., Mao, A.A., & Phukan, S. (2008). Rediscovery of Begonia adscendens, C. B. Clarke from Esii Hill, Manipur, India. Rheedea, 18(1), 53- 55.

XXXVIII. Villa-Ruano, N., Pacheco-Hernández, Y., Cruz-Durán, R., Lozoya-Gloria, E., & Betancourt-Jiménez, M.G. (2017). Seasonal variation in Phytochemicals and nutraceutical potential of *Begonia nelumbiifolia* consumed in Puebla, México. Journal l of Food Science and Technology, 54, 1484-1490.

XXXIX. Yasuma, A., & Ichikawa. (1953). Ninhydrin - Schiff and alloxan -Schiff staining methods for proteins. J .Lab clin Med, 41, 296-29