



**INTERNATIONAL JOURNAL OF
PHARMACEUTICAL SCIENCES**
[ISSN: 0975-4725; CODEN(USA):IJPS00]
Journal Homepage: <https://www.ijpsjournal.com>



Research Article

Formulation Development And Evaluation Of The *Tinospora Crispa* Ointment

Savita Yadav¹, Gaurav Dubey², Aditi Omprakash Jyotishi³, Shailendra Singh Chandel^{*4}

¹IES Institute of Technology and Management, Bhopal, M.P.

²NIMS Institute of Pharmacy, NIMS University Rajasthan, Jaipur-303121, India.

³Dr. Vedprakash Patil College of Pharmacy, Sambhaji Nagar, Aurangabad, Maharashtra.

⁴NIMS Institute of Pharmacy, NIMS University Rajasthan, Jaipur-303121, India.

ARTICLE INFO

Received: 03 May 2024

Accepted: 07 May 2024

Published: 20 May 2024

Keywords:

Tinospora crispa,
Phytochemical screening,
Ointment, Formulation,
Evaluation

DOI:

10.5281/zenodo.11228397

ABSTRACT

The aim of study was to extract, screen, formulate and evaluate *Tinospora crispa* ointment via phytochemical techniques and phytochemical methods. The objectives of this work were to understand the pharmacogenetics of *Tinospora crispa*, to determine suitable procedures and solvents for the extract, to understand the phytochemical screening of the extract, to formulate an ointment of *Tinospora Crispa* extract and to evaluate ointment on the basis of various evaluation parameters. The plant *Tinospora crispa* was selected on the basis of a literature review. Then it was collected from an online store and extracted by Soxhlet extraction. The extract was subjected to phytochemical screening and after that ointment was formulated using drug extract and various excipients, after which various evaluation parameters were tested. On the basis of various literature surveys the plant *Tinospora crispa* was selected and authenticated by the Nims Institute of Pharmacy. The plant was extracted by Soxhlet extraction using methanol. Various phytochemical tests revealed the presence of saponins, tannins, steroids, carbohydrates and fixed oils. On the basis of various evaluation tests the ointment was found to be safe and effective for treating various skin problems. *Tinospora crispa* is a remarkable herb that can be used to treat many types of illnesses. Therefore, additional research is needed in addition to clinical studies to demonstrate the health advantages of this herb. The results of this study support the use of *T. crispa* ointment in the treatment of a variety of skin conditions, including acne, eczema, dark under circles, and skin allergies.

***Corresponding Author:** Savita Yadav

Address: IES Institute of Technology and Management, Bhopal, M.P.

Email ✉: ysavita929@gmail.com

Relevant conflicts of interest/financial disclosures: The authors declare that the research was conducted in the absence of any commercial or financial relationships that could be construed as a potential conflict of interest.



INTRODUCTION

Not only in the present but also in the distant past, trees and plants have been essential to human life. Early man relied on them for both his spiritual requirements, such as magic or ritualistic practices, as well as his physical needs, such as sources for food, housing, clothes, medicine, adornment, and tools. Locally grown medicinal herbs are typically accessible local and traditional treatments are nontoxic, safe, affordable, and socially and culturally acceptable for every reason¹. Numerous researchers have extensively studied the genus *Tinospora* and claim that it contains a number of phytochemicals with notable medicinal efficacy. Tropical lowland areas are home approximately 70 genera and 450 species of plants that make up the Menispermaceae plant family. These shrubs are rarely climbing or twining plants. Leaves are alternate or lobed, flowers are small chimes, and seeds are usually hooked or uniform. This family is a rich source of alkaloids and terpenes². Both the traditional medical system and Ayurveda highlight the plant's healing properties. The plant can be found in the tropical region of India from Kumaon to Assam and further north via West Bengal, Bihar, Deccan, Konkan, Karnataka, and Kerala, up to 1,200 m above sea level. It is a common shrub that grows over hedges and small trees in deciduous and dry woodlands. It enjoys a variety of soil types, from acidic to alkaline, and requires a moderate amount of soil moisture³.

Plant profile:

Kingdom: Plantae Division: Magnoliophyta Class: Magnoliopsida, Order: Ranunculaceae Family: Menispermaceae Genus: *Tinospora*

Common names⁴:

Latin:

Tinospora cordifolia (willd.) Hook.F. & Thomson

English:

Gulantha, Indian *Tinospora*

Sanskrit:

Guduchi, Madhuparni, Amrita, Chinnaruha, Vatsadaani, Tantrika, Hindi: Giloya, Guduchi

Bengal:

Gulantha

Telugu:

Tippatiga Tamil: Shindilakodi Marathi: Shindilakodi Gujarathi: Galo Kannada: Amrita balli



A



B



C

Fig. no. 1: A: Leaves of *T. Crispa* B: Flowers of *T. Crispa* C: Stem of *T. Crispa*

Botanical description:

Large, glabrous, deciduous, climbing shrub *T. crispa*. The stem structure is fibrous, and the transverse slice reveals a yellowish wood with wedge-shaped wood bundles containing large

vessels that are radially organized and spaced apart by narrow medullary rays. The stem has rosette-like lenticles, and the bark ranges in color from creamy white to gray and is deeply spiralled. The leaves are cordate and membranous in texture. Unisexual, small, yellow, and in the axillary position, the flowers have a raceme length of 2 to 9 cm and are borne on leaflet branches. Female flowers are often solitary, whereas male blooms are grouped. Curved seeds are present. Fruits have a solitary seed and are meaty. Fruits ripen in the winter, and flowers ripen in the summer⁵.

Morphological Description:

A large, widely spreading climbing deciduous shrub with many coiling branches is called *Tinospora crispa*. The following types of morphology can be observed in various *Tinospora* regions.

Stem:

This plant has a long, filiform, fleshy, climbing stem that is fairly succulent in appearance. The branches give rise to aerial roots. The bark is deeply left spirally and ranges in tint from creamy white to gray⁶.

Aerial Root:

There are aerial roots, and the fundamental structure of these aerial roots ranges from a tetra to a penta-arch. However, the cortex of the root is split into an inner parenchymatous zone and an exterior thick walled zone⁷.

Leaves:

Simple, alternate, exstipulate, round, pulvinate, heart-shaped, partially twisted, and halfway circular leaves of this plant have a length of approximately 15 petioles. Oval, 10–20 cm long, 7 nerved, profoundly cordate at the base, and membranous are the characteristics of the lamina⁸.

Flowers:

Unisexual, receptive, and greenish yellow in hue, flowers only bloom when a plant has no leaves. Female flowers are observed in single inflorescences, while male flowers are grouped.

There are two series of three sepals each, totaling six. The inner sepals are smaller than the outer sepals. Additionally, six petals are membranous, free, and smaller than the sepals. The flowering season lasts from March to June⁹.

Fruit:

These fruits are orange–red in color, fleshy, have an aggregate of one to three smooth, ovoid drupelets on a thick stem, and have a subterminal style scar. Fruits grow in the winter¹⁰.

Seed:

There have been reports of curved seeds of this species. As a result, this family is also known as the moonseed family. The embryo instantly assumed a curved shape, much like the shape of seeds. Additionally, the endocarp has different ornamentations and has crucial taxonomic characteristics.

METHODS

MATERIAL

For Formulation

- *Tinospora crispa* plant extract
- Wool fat
- Hard paraffin
- Soft paraffin
- Cetostearyl alcohol

Chemicals-

All the chemicals used for the study were of analytical grade and were purchased from R.S. Enterprises, Jaipur (Rajasthan).

Other Materials

- Beaker
- Conical flask
- Cotton
- Butter paper
- Spatula
- Forceps
- China dish
- Filter paper

Research Methodology

Selection of plants:



The plant *Tinospora crispa* was selected on the basis of various literature surveys and its future prospects in the herbal industry.

Collection of plants:

The whole plant powder of *Tinospora crispa* was collected from an online herbal store called Indianjadibooti.com in March 2023.

Authentication:

The plant species were identified and authenticated by the Department of Pharmacognosy of NIMS Institute of Pharmacy and reference no. is given as NU/Nip/2023/524.

Storage: -

The dried powder of *Tinospora crispa* plants was preserved in tightly closed airtight containers and stored in a suitable cool and dry place.

Extraction: -

- Thirty grams of dried and powdered *Tinospora crispa* plants were weighed properly.
- The powder was then extracted with 100 ml of alcoholic solvent (methanol) using Soxhlet extraction for 18 hrs.
- The extract obtained was then concentrated in a water bath and after cooling the extract was stored in a refrigerator.



Fig. no. 2: Soxhlet apparatus

Phytochemical Screening

Phytochemical examinations of the extracts were carried out according to standard methods.

Alkaloid

extracts were dissolved individually in dilute hydrochloric acid and filtered. The filtrate was used to test for the presence of alkaloids.

a Mayer's test-

Filtrates were treated with Mayer's reagent (potassium mercuric iodide). The formation of a yellow precipitate indicates the presence of alkaloids.

b Wagener's test-

filtrates were treated with Wagener's reagent (iodine in potassium iodide). The formation of a brown/reddish precipitate indicates the presence of alkaloids.

c Dragendroff's test-

filtrates were treated with dragendroff's reagent (potassium bismuth iodide). The formation of a red precipitate indicates the presence of alkaloids.

d Hager's test-

filtrates were treated with Hager's reagent (saturated picric acid solution). The presence of alkaloids was confirmed by the formation of a yellow precipitate.

Flavonoids

a Alkaline reagent test-

Extracts were treated with a few drops of sodium hydroxide solution. The formation of an intense yellow color, which becomes colorless upon the addition of dilute acid, indicates the presence of flavonoids.

b Lead acetate test-

Extracts were treated with a few drops of lead acetate solution. The formation of a yellow precipitate indicates the presence of flavonoids.

Tannins

a. Ferric chloride test-

Approximately 0.5 ml of extract was boiled in 10 ml of water in a test tube. A few drops of 0.1% ferric chloride were added and the samples were observed for brownish green or blue-black coloration. This indicates the presence of Tannins.

Phenols

a. Ferric Chloride Test-

Extracts were treated with 3-4 drops of ferric chloride solution. The formation of a bluish black color indicates the presence of phenols.

Saponins

a. Froth test

extracts were diluted with distilled water to 20 ml and shaken in a graduated cylinder for 15 minutes. The formation of a 1 cm layer of foam indicates the presence of saponins.

b. Foam test-

First, 0.5 g of extract was shaken with 2 ml of water. The persistence of foam produced for ten minutes indicates the presence of saponins.

Glycoside

a Keller- Killiani test-

First, 0.5 ml of extract was boiled with 5 ml of distilled water and 2 ml of glacial acetic acid containing 1 drop of 0.5% ferric chloride solution was added. This solution was mixed with 1 ml of concentrated sulfuric acid. The formation of a violet ring below the brown ring and a greenish ring in the acetic layer just above the brown ring and gradual spreading throughout this layer indicated the presence of cardiac glycoside.

b Molisch's reagent test-

Two to three drops of Molisch reagent were added to the extracts and mixed well. A few drops of conc. sulfuric acid were added carefully. The formation of a reddish-purple ring at the junction of the two layers indicates the presence of glycosides.

Terpenoids-

a. Chloroform test-

The extract was mixed with 2 ml of chloroform, and concentrated H₂SO₄ (3 ml) was carefully added to form a layer. A reddish brown coloration of the interface is formed to indicate the presence of terpenoids.

Carbohydrate-

a. For the Fehling's reagent test, 2 ml of the given extract was added to a clean test tube. Then 2 ml of Fehling's solution A and Fehling's solution B

were added. The solution was kept in a boiling water bath for approximately 10 minutes. If a red precipitate formed, then the presence of carbohydrates was confirmed.

b. Benedict's reagent test-

One milliliter of the extract was mixed with 2 ml of Benedict's reagent and heated in a boiling water bath for 3 to 5 minutes. The development of a brick-red precipitate of cuprous oxide confirmed the presence of carbohydrates.

c. Molisch's reagent test-

Two to three drops of Molisch's reagent must be added to a small amount of the extract in a test tube and mixed well. Now, a few drops of concentrated sulfuric acid must be added drop wise along the walls of the test tube to facilitate the formation of a layer and avoid mixing. The development of a purple ring at the layer formed by the concentrated acid is a positive indicator of carbohydrates.

Steroids-

a. Chloroform test-

The development of a greenish color when 2 ml of the extract was dissolved in 2 ml of chloroform and treated with concentrated sulfuric acid and acetic acid indicates the presence of steroids.

Fixed oils-

a. Spot test-

In this test, the given sample to be tested was rubbed between the folds of filter paper. The appearance of translucent spots confirms the presence of fats in the given sample.

Anthraquinones

a. Bontrager's test-

One gram of extract was mixed with 5-10 ml of dilute HCl and boiled in a water bath for 10 minutes. The solution was filtered, and the extract of the filtrate was treated with CCl₄ or benzene and an equal amount of ammonia solution. After shaking, the appearance of a pink to red color indicated the presence of an anthraquinone moiety.

Formulation:

1. First, all the ingredients were weighed properly as per the manufacturer's requirements.
2. Twenty-nine grams of cetosteryl alcohol and 5 g of hard paraffin were weighed and stored in a china dish.
3. Both the ingredients were melted in a water bath.
4. Five grams of wool fat and 85 g of soft white paraffin were added to the melted mixture.
5. After the ointment base was prepared, 5% plant extract was added to 95% ointment base and mixed well.
6. The ointment was then stored in airtight containers and kept in a cool and dry place.



Fig. no. 3: *Tinospora Crispa* Ointment

Evaluation

The following parameters were evaluated for the formulated ointment.

1. Color and odor-

Physical parameters such as color and odor were examined by visual examination.

2. Consistency-

Examined by applying it to the surface of the skin.

3. Homogeneity-

The homogeneity of the formulated gels was examined by visual inspection for the presence of any aggregates.

4. pH Value-

The pH values of the prepared formulations were measured by using a digital pH meter. The solution of ointment, cream, and gel was prepared by using 100 ml of distilled water and was set aside for 2 hrs. The pH of the solution was

determined in triplicate, and the average value was calculated.

5. Spreadability-

The spreadability was determined by placing an excess of sample between two slides, which were compressed to a uniform thickness by placing a definite weight for a certain time. The time required to separate the two slides was measured as spreadability. A shorter separation time for two slides results in better spreadability.

The spreadability was calculated by the following formula:

$$S=M \times L / T,$$

where

S= Spreadability

M= Weight tide to the upper slide

L= Length of glass slide

T= Time taken to separate the slides

6. Solubility-

Soluble in boiling water, miscible with alcohol, ether or chloroform.

7. Washability-

The formulations were applied to the skin, and then the extent of washing with water was checked.

8. Non-irritancy Test-

The prepared formulations were applied to the skin of humans, and their effects were observed.

9. Stability study-

Physical stability tests of the formulations were carried out for four weeks at various temperatures, such as 2 °C, 25 °C and 37 °C.

10. Viscosity-

The viscosity of the formulations was checked using a Brookfield Viscometer.

Results

Selection of Plants

The plant was selected on the basis of various literature surveys. A total of 42 articles were considered of which 12 were review articles and 30 were research articles on the basis of which the plant *Tinospora crispa* was selected.

Table no. 1: Selection of plants

Sr. No	Name of Plant	Review articles	Research articles	Total no. of articles
1.	Tinospora Crispa	12	30	42

Plant collection

Whole plant powder was collected from the online store Indianjadibooti.com. in the month of March.

Table no. 2.: Plant collection

Sr. No.	Name Of Plant	Part of Plant	Source	Date
1.	Tinospora Crispa	Whole plant powder	Indianjadibooti.com	05-03-23

Authentication-

The plant material was then authenticated by Dr. Gaurav Dubey from the Nims Institute of

Pharmacy in the month of March 2023, and the certificate with reference no. NU/Nip/2023/524 was provided on 16-03-23.

Table no. 3: Plant authenticity

Sr. No.	Name Of Plant	Authenticated by	Reference no.	Date
1.	Tinospora Crispa	Dr. Gaurav Dubey Nims Institute of Pharmacy	NU/Nip/2023/524	16-03-23

Extraction-

The whole plant powder was extracted by Soxhlet extraction using methanol as the solvent.

Table no. 4: Extraction of plant material

Sr. No.	Name of Plant	Method	Solvent
1.	Tinospora Crispa	Soxhlet extraction	Methanol

Phytochemical screening-

Various phytochemical tests were performed on the extract, and the results showed that the plant contains terpenoids, saponins, tannins, steroids, carbohydrates and fixed oils.

- A solution of the extract was made using methanol as the solvent.
- A small amount of the solution was taken to test the presence of different phytoconstituents using various chemicals and reagents.

Table no. 5: Phytochemical screening of the extract

Sr. No.	Test	Observation	Result
1.	Terpenoids Chloroform test	Reddish brown color on the interface.	+
2.	Flavonoids Alkaline reagent test	Formation of yellow color which disappear on addition of dilute acid.	-
3.	Saponins Foam test	Stable persistent froth on shaking.	+
4.	Tannins Ferric chloride test	Brown-green or blue-black Coloration.	+
5.	Alkaloid	Presence of red precipitate.	-



	Dragendroff's test		
6.	Anthraquinone Bontrager's test	Appearance of pink to red color.	-
7.	Glycosides Keller-Killani test	Formation of violet ring below the brown ring.	-
8.	Steroids Chloroform test	Development of greenish color	+
10.	Carbohydrates Molisch reagent test	Formation of purple ring at the layer	+
11.	Fixed oils Spot test	Appearance of translucent spot	+

Formulation-

An emulsion-based ointment was prepared using drug extract, hard paraffin, yellow soft paraffin,

wool fat and cetosteary alcohol as the main ingredients.

Table no. 6 : List of ingredients

Sr. No.	Ingredients	Quantity
1.	Hard paraffin	5 grams
2.	Wool fat	5 grams
3.	yellow soft paraffin	85 grams
4.	Ceto stearyl alcohol	29 grams
5.	Drug extract	25 ml

Evaluation-

Various evaluation tests were performed on the formulated ointment, which showed that

- The ointment is a light yellowish color and contains a petroleum-like odor.
- The ointment was smooth in consistency and homogenous.
- It has a pH of 6.13 and a spreadability of 7.
- The ointment is soluble in boiling water and miscible in alcohol, ether and chloroform.
- The ointment is moderately washable and is not an irritant.
- The ointment is stable at different temperatures, such as 2°C, 25°C and 37°C.
- The ointment is very viscous and has a viscosity of approximately 32.2±0.51.

Table no. 7: Evaluation parameters for ointment

Sr. No.	Parameters	Observation
1.	Color	Light yellow
2.	Odor	Petroleum like smell
3.	Consistency	Smooth
4.	Homogeneity	Homogenous
5.	PH	6.13
6.	Spreadability	7
7.	Solubility	Soluble in boiling water, miscible with alcohol, Ether, chloroform.
8.	Washability	Moderate
9.	No irritancy	Nonirritant
10.	Stability	Stable at 2°C, 25°C, 37°C



DISCUSSION

Due to its health advantages, *Tinospora crispa* (Giloy), a medicinal herb, is employed in the Indian Ayurvedic medical system. *Tinospora crispa*, also known as Amrita, Giloy, and Guduchi, is frequently used in the "Rasayanas" of the Ayurvedic medical system to boost the immune system and increase body resistance to illnesses. It is a large, glabrous, deciduous climbing shrub from the Menispermaceae family and is regarded as one of the most adaptable plants for rejuvenation in both folk and Ayurvedic medicine. Its numerous uses include osteoprotection, cardio protection, anticancer, ant diabetic, immunomodulator, ant allergic, antioxidant, and ant diabetic treatments. It has many advantages for treating skin issues such as wrinkles, wounds, acne, eczema and dark circles. An ointment containing *T. crispa* extract was created for the aforementioned ailments. The extraction process used whole-plant powder. A number of phytochemical experiments were carried out using the extract. Cetostearyl alcohol, wool fat, hard paraffin, yellow soft paraffin, and *T. crispa* extract are the main components used to formulate emulsion-based ointments. Various parameters were evaluated using the prepared formulation.

CONCLUSION

Tinospora crispa is a remarkable herb that can be used to treat many types of illnesses. The Federal Drug Administration has not approved *Tinospora crispa*, and similar to other herbal remedies and prescription drugs, it can have undesirable side effects such as constipation. Therefore, additional research is needed in addition to clinical studies to demonstrate the health advantages of this herb. Before using this medication, a person should also talk to their doctor if they have any health issues, are pregnant, or are nursing a baby. The development of a semisolid dosage form (ointment) with better formulation parameters is also covered in the current study, which offers

useful information regarding the identification and authenticity of the plant *T. crispa*. Nutraceuticals made from antioxidant-rich plants minimize oxidative stress and, as a result, slow the progression of degenerative diseases. The results of this study support the use of *T.crispa* ointment for the treatment of a variety of skin conditions, including acne, eczema, dark under eye circles, and skin allergies. Therefore, additional research might be performed to isolate and purify significant chemicals from this plant, enabling the scientific community to use it as a beneficial source for the creation of diverse herbal medications. It also exhibits a wide range of pharmacological actions. Therefore, this gives us a wide range of inquiries into potential futures.

REFERENCES :

1. Sharma R, Amin H, Prajapati PK. Antidiabetic claims of *Tinospora cordifolia* (Willd.) Miers: critical appraisal and role in therapy. *Asian Pacific Journal of Tropical Biomedicine*. 2015 Jan 1;5(1):68-78.
2. Bala M, Pratap K, Verma PK, Singh B, Padwad Y. Validation of ethnomedicinal potential of *Tinospora cordifolia* for anticancer and immunomodulatory activities and quantification of bioactive molecules by HPTLC. *Journal of ethnopharmacology*. 2015 Dec 4; 175:131-7.
3. Mishra P, Jamdar P, Desai S, Patel D, Meshram D. Phytochemical analysis and assessment of in vitro antibacterial activity of *Tinospora cordifolia*. *International Journal of Current Microbiology and Applied Sciences*. 2014;3(3):224-34.
4. Bonvicini F, Mandrone M, Antognoni F, Poli F, Angela Gentilomi G. Ethanolic extracts of *Tinospora cordifolia* and *Alstonia scholaris* show antimicrobial activity toward clinical isolates of methicillin-resistant and carbapenemase-producing bacteria. *Natural*



- product research. 2014 Sep 17;28(18):1438-45.
5. Yusoff M, Hamid H, Houghton P. Anticholinesterase inhibitory activity of quaternary alkaloids from *Tinospora crispa*. *Molecules*. 2014 Jan 20;19(1):1201-11.
 6. Li P, Yin ZQ, Li SL, Huang XJ, Ye WC, Zhang QW. Simultaneous determination of eight flavonoids and pogostone in *Pogostemon cablin* by high-performance liquid chromatography. *Journal of Liquid Chromatography & Related Technologies*. 2014 Jul 21;37(12):1771-84.
 7. Sharma A, Gupta A, Batra S.S.A. *Tinospora cordifolia* (Willd.) Hook. F. & Thomson - A plant with immense economic potential. *Journal of Chemical & Pharmaceutical Research* 2010, 2 (5): 327-33.
 8. Bairy KL, Rao Y, Kumar Das S, Kumar KB. Efficacy of *Tinospora cordifolia* on learning and memory in healthy volunteers: A double-blind, randomized, placebocontrolled study. *Iranian Journal of Pharmacology and Therapeutics*. 2004 Nov 10;3(2):57-60.
 9. Sharma A, Gupta A, Singh S, Batra A. *Tinospora cordifolia* (Willd.) Hook. F. & Thomson-A plant with immense economic potential. *J. chem. pharm. Res*. 2010;2(5):327-33.
 10. Raghunathan K. The aqueous extract of *T. cordifolia* caused reduction of blood sugar in alloxan induced hyperglycemic rats and rabbits. *J Res Ind Med*. 1969; 3:203-11.
 11. Spandana U, Ali SL, Nirmala T, Santhi M, Babu SS. A review on *Tinospora cordifolia*. *International Journal of Current Pharmaceutical Review and Research*. 2013;4(2):61-8.
 12. Kongsaktragoon B, Tamsiririrkkul R, Suvitayavat W, Nakornchai S, Wongkrajang Y. The antipyretic effect of *Tinospora crispa* Mier ex Hook. f. & Thoms. *Mahidol University Journal of Pharmaceutical Sciences*. 1984;21(1):1- 6.
 13. Dweck AC, Cavin JP. A review of *Andawali* (*Tinospora crispa*). *Personal Care Magazine*. 2006;7(1):1-7.
 14. Li S, Long C, Liu F, Lee S, Guo Q, Li R, Liu Y. Herbs for medicinal baths among the traditional Yao communities of China. *Journal of ethnopharmacology*. 2006 Nov 3;108(1):59-67.
 15. R, Ashcroft SJ. The hypoglycemic and insulin tropic activity of *Tinospora crispa*: studies with human and rat islets and HIT-T15 B cells. *Diabetologia*. 1989 Jun; 32:354-9.
 16. Roosita K, Kusharto CM, Sekiyama M, Fachrurazi Y, Ohtsuka R. Medicinal plants used by the villagers of a Sundanese community in West Java, Indonesia. *Journal of ethnopharmacology*. 2008 Jan 4;115(1):72-81.
 17. Ahmad FB, Ismail G. Medicinal plants used by Kadazandusun communities around Crocker Range. *ASEAN Review of Biodiversity and Environmental Conservation (ARBEC)*. 2003 Jan 1;1(1):1-0.
 18. Rahmatullah M, Noman A, Hossan MS, Rashid MH, Rahman T, Chowdhury MH, Jahan R. A survey of medicinal plants in two areas of Dinajpur district, Bangladesh including plants which can be used as functional foods. *American Eurasian Journal of Sustainable Agriculture*. 2009 Dec 1;3(4):862-76.
 19. Hout S, Chea A, Bun SS, Elias R, Gasquet M, Timon-David P, Balansard G, Azas N. Screening of selected indigenous plants of Cambodia for antiplasmodial activity. *Journal of Ethnopharmacology*. 2006 Aug 11;107(1):12-8.
 20. Singh SS, Pandey SC, Srivastava S, Gupta VS, Patro B. Chemistry and medicinal properties of *Tinospora cordifolia* (Guduchi).

- Indian journal of pharmacology. 2003 Mar 1;35(2):83
21. Maurya R, Handa SS. Tinocordifolin, a sesquiterpene from *Tinospora cordifolia*. *Phytochemistry*. 1998 Nov 5;49(5):1343-5.
 23. Van Kiem P, Van Minh C, Dat NT, Hang DT, Nam NH, Cuong NX, Huong HT, Van Lau T. Aporphine alkaloids, clerodane diterpenes, and other constituents from *Tinospora cordifolia*. *Fitoterapia*. 2010 Sep 1;81(6):485-9.
 24. Ghosal S, Vishwakarma RA. Tinocordiside, a new rearranged cadinane sesquiterpene glycoside from *Tinospora cordifolia*. *Journal of Natural Products*. 1997 Aug 22;60(8):839-41.
 25. Ali H, Dixit S. Extraction optimization of *Tinospora cordifolia* and assessment of the anticancer activity of its alkaloid palmatine. *The Scientific World Journal*. 2013 Jan 1;2013.
 26. Sharma U, Bala M, Kumar N, Singh B, Munshi RK, Bhalerao S. Immunomodulatory active compounds from *Tinospora cordifolia*. *Journal of ethnopharmacology*. 2012 Jun 14;141(3):918-26.
 27. Upadhyaya R, Pandey RP, Sharma V, Verma Anita K. Assessment of the multifaceted immunomodulatory potential of the aqueous extract of *Tinospora cordifolia*. *Research Journal of Chemical Sciences*.
 28. Bhawya D, Anilakumar KR. In vitro antioxidant potency of *Tinospora cordifolia* (gulancha) in sequential extracts. *International Journal of Pharmaceutical & Biological Archives*. 2010;1(5):448-56.
 29. Gupta R, Sharma V. Ameliorative effects of *Tinospora cordifolia* root extract on histopathological and biochemical changes induced by aflatoxin-B1 in mice kidney. *Toxicology international*. 2011 Jul;18(2):94.
 30. Narayanan A, Raja S, Ponmurugan K, Kandekar S, Natarajaseenivasan K, Maripandi A, Mandeel Q. Antibacterial activity of selected medicinal plants against multiple antibiotic resistant uropathogens: a study from Kolli Hills, Tamil Nadu, India. *Beneficial Microbes*. 2011 Sep 1;2(3):235-43.
 31. Shanthi V, Nelson R. Antibacterial activity of *Tinospora cordifolia* (Willd) Hook.F. Thoms on urinary tract pathogens. *Int J Curr Microbiol App Sci*. 2013;2(6):190-4.
 32. Shanish Antony A, Partha DebRoy, Vadivelan R, Jaysankar K, Vikram M, Nandini S, Sundeep M, Elango K, Suresh B; Amelioration of CNS Toxicities of L-DOPA in Experimental Models of Parkinson's disease by Concurrent Treatment with *Tinospora cordifolia*. *Hygeia J D Med*, 2010; 2(1): 28-37.
 33. Gupta R, Sharma V. Ameliorative effects of *Tinospora cordifolia* root extract on histopathological and biochemical changes induced by aflatoxin-B1 in mice kidney. *Toxicology international*. 2011 Jul;18(2):94.
 34. Sharma V, Pandey D. Protective role of *Tinospora cordifolia* against lead- induced hepatotoxicity. *Toxicology international*. 2010;17(1):12.
 35. Kirtikar K. R. and Basu, BD, *Indian Medicinal Plants II*. International Book Distributors, Dehradun. 1975.
 36. Zhao T, Wang X, Rimando AM, Che CT. Folkloric medicinal plants: *Tinospora sagittata* var. *cravaniana* and *Mahonia bealei*. *Planta Medica*. 1991 Oct;57(05):505.
 37. Nayampalli SS, Ainpure SS, Samant BD, Kudtarkar RG, Desai NK, Gupta KC. A comparative study of diuretic effects of *Tinospora cordifolia* and hydrochlorothiazide in rats and a preliminary phase I study in human volunteers. *Journal of Postgraduate Medicine (Bombay)*. 1988;34(4):233-6.

38. A. Singla, Mr Akant Priya, P. Singla, "Review of Biological Activities of *Tinospora cordifolia*", WebmedCentral Pharmaceutical Sciences, 2010; 1(9).
39. Zulkhairi A, Abdah MA, Kamal NH, Nursakinah I, Moklas MA, Hasnah B, Fazali F, Khairunnur FA, Kamilah KA, Zamree MS, Shahidan MM. Biological Properties of *Tinospora crispera* (Akar Patawali) and Its Antiproliferative Activities on Selected Human Cancer Cell Lines. Malaysian Journal of Nutrition. 2008 Sep 1;14(2).
40. Kamarazaman IS, Amom ZH, Ali RM, Akim AM, Azman KF, Arapoc DJ, Hassan MK, Shahidan M, Arshad M, Shah ZM, Kadir KK. Protective effects of *Tinospora crispera* extracts on H₂O₂-induced oxidative stress and TNF- α -induced inflammation on human umbilical vein endothelial cells (HUVECs). Journal of Medicinal Plants Research. 2012 Apr 23;6(15):3013-21.
41. Rahman NN, Furuta T, Takane K, Mohd MA. Antimalarial activity of extracts of Malaysian medicinal plants. Journal of ethnopharmacology. 1999 Mar 1;64(3):249-54.
42. Amom Z, Azman KF, Ismail NA, Shah ZM, Arshad MS. An aqueous extract of *Tinospora crispera* possesses antioxidative properties and reduces atherosclerosis in hypercholesterolemic-induced rabbits. Journal of Food Biochemistry. 2011 Aug;35(4):1083-98.
43. Almeida RN, Navarro DS, Barbosa-Filho JM. Plants with central analgesic activity. Phytomedicine. 2001 Jan 1;8(4):310-22.
44. Sulaiman MR, Zakaria ZA, Lihan R. Antinociceptive and anti-inflammatory activities of *Tinospora crispera* in various Animal models. Int J Trop Med. 2008; 3:66-9.
45. Usia T, Iwata H, Hiratsuka A, Watabe T, Kadota S, Tezuka Y. CYP3A4 and CYP2D6 inhibitory activities of Indonesian medicinal plants. Phytomedicine. 2006 Jan 5;13(1-2):67-73.
46. Usia T, Iwata H, Kadota S, Tezuka Y. Mechanism-based inhibition of CYP3A4 and CYP2D6 by Indonesian medicinal plants. Journal of ethnopharmacology. 2006 May 24;105(3):449-55.
47. Kalikar MV, Thawani VR, Varadpande UK, Sontakke SD, Singh RP, Khiyani RK. Immunomodulatory effect of *Tinospora cordifolia* extract in human immunodeficiency virus positive patients. Indian journal of pharmacology. 2008 Jun;40(3):107.
48. Akhtar S. Use of *T. cordifolia* in HIV infection. Ind J pharmacol. 2010; 42:57- 63.
49. Hawkins, E.B., Ehrlich, S.D., 2007. Herbal Medicine: Overview. Naini V, Mamidala E. An ethnobotanical study of plants used for the treatment of diabetes in the Warangal district, Andhra Pradesh, India. Biolife.2013;1(1):24-8.
50. Li P, Xu G, Li SP, Wang YT, Fan TP, Zhao QS, Zhang QW. Optimizing ultraperformance liquid chromatographic analysis of 10 diterpenoid compounds in *Salvia miltiorrhiza* using central composite design. Journal of agricultural and food chemistry. 2008 Feb 27;56(4):1164-71.
51. Yi Y, Zhang QW, Li SL, Wang Y, Ye WC, Zhao J, Wang YT. Simultaneous quantification of major flavonoids in "Bawanghua", the edible flower of *Hylocereus undatus* using pressurized liquid extraction and high-performance liquid chromatography. Food chemistry. 2012 Nov 15;135(2):528-33.
52. Zhou YQ, Zhang QW, Li SL, Yin ZQ, Zhang XQ, Ye WC. Quantitative determination of 13 flavonoids in seed of *Oroxylum indicum* by

high-performance liquid chromatography. *Curr. Pharm. Anal.* 2012;8(2):206-13.

53. Du G, Zhao H, Song Y, Zhang Q, Wang Y. Rapid simultaneous determination of isoflavones in *Radix puerariae* using high-performance liquid chromatography–triple quadrupole mass spectrometry with novel

shell-type column. *Journal of separation science.* 2011 Oct;34(19):2576-85.

HOW TO CITE: Savita Yadav, Gaurav Dubey, Aditi Omprakash Jyotishi, Shailendra Singh Chandel, Formulation Development And Evaluation Of The *Tinospora Crispa* Ointment, *Int. J. of Pharm. Sci.*, 2024, Vol 2, Issue 5, 979-991. <https://doi.org/10.5281/zenodo.11228397>