

Formulation And Evaluation of an Herbal Ointment containing Neem and Turmeric Extracts

¹Harshkumar Brahmhatt, ²Naseena. P, ³Nazia Malik, ⁴Anasuya Patil*, ⁵Yogesh
Dagadu Pawar, ⁶Ankur Patel, ⁷Rajeev Ranjan, ⁸Ashish Suttee,

¹Department of Pharmacy, Sumandeep Vidyapeeth Deemed To Be University At & Po. Pipariya,
Taluka: Waghodiya, Vadodara, Gujarat 391760.

²National College of Pharmacy Kozhikode Kerala 673602.

³TRR College of Pharmacy Pragathi Colony, Meerpet Hyderabad Telangana

⁴Department of Pharmaceutics, KLE College of Pharmacy, II Block Rajajinagar, Bengaluru-560010

⁵Shri Gulabrao Deokar College of Pharmacy Shirsoli Road ,Jalgaon, Maharashtra, India 425002.

⁶Department of Pharmaceutics, Sardar Patel College Of Pharmacy,Vidyanagar-Vadtal
Road,Bakrol,388315

⁷University Department of Chemistry, DSPM University, Ranchi 834008.

⁸School of Pharmaceutical Sciences, LPU, Punjab, India 144411

Corresponding Author

***Anasuya Patil,**

Department of Pharmaceutics, KLE College of Pharmacy, II Block Rajajinagar, Bengaluru-560010.

Abstract

Interest in usage of herbal remedies have significantly increased in recent years, even in areas where access to modern treatment is available. Phytochemicals and herbal remedies have gained popularity recently a lot of attention because medicinal plants are the primary source of bioactive molecules utilized in both conventional and medicine of the modern age. The purpose of this study is to create and test an ointment containing Neem (*Azadirachta indica*) and Turmeric (*Curcuma longa*) extracts. The maceration procedure was used to prepare the ethanolic extracts. The extract was blended with the base using the levigation procedure to generate the ointment after the ointment base was prepared. When it was finished, the formulation's physicochemical properties—such as color, fragrance, pH, spreadability, extrudability, consistency, solubility, and washability—were evaluated. After it was ready, the formulation was assessed for physicochemical characteristics such color, smell, pH, spreadability, extrudability, consistency, solubility, and washability. Further stability testing of the formulation at various temperatures indicated no alteration in irritancy, spreadability, or diffusion. As a result, it might become a medium for efficiently and readily utilizing the therapeutic properties of Neem and Turmeric in a simple dose form.

Keywords: Physicochemical parameters, Neem, Turmeric, antifungal activity.

1. Introduction

Medicinal or pharmaceutical chemistry is a branch of chemistry and pharmacology concerned with the design, synthesis, and development of pharmaceutical medications. The identification, production, and development of novel chemical entities appropriate for therapeutic use is the focus of medicinal chemistry [1-2]. It also involves the investigation of already available pharmaceuticals, their biological

features, and their Structure-activity correlations in quantitative terms (QSAR). Pharmaceutical chemistry focuses on the effectiveness of medications and the suitability of medical equipment for the uses for which it is intended[3].

Medicinal plants' importance in drug discovery

synthetic chemistry, combinatorial chemistry, molecular modeling, and separation from plants and other natural sources of novel chemical entities have all been used to obtain molecules for drug development [4]. About a quarter of top-selling medications globally in 2001 and 2002 were either natural ingredients themselves or were derived from them. [5]. For lead development, lead optimization, and clinical studies, the quantities of isolated natural compounds are frequently insufficient. To assess if synthesis or semi-synthesis would be feasible, cooperation with medicinal and synthetic chemists is required. [6-7].

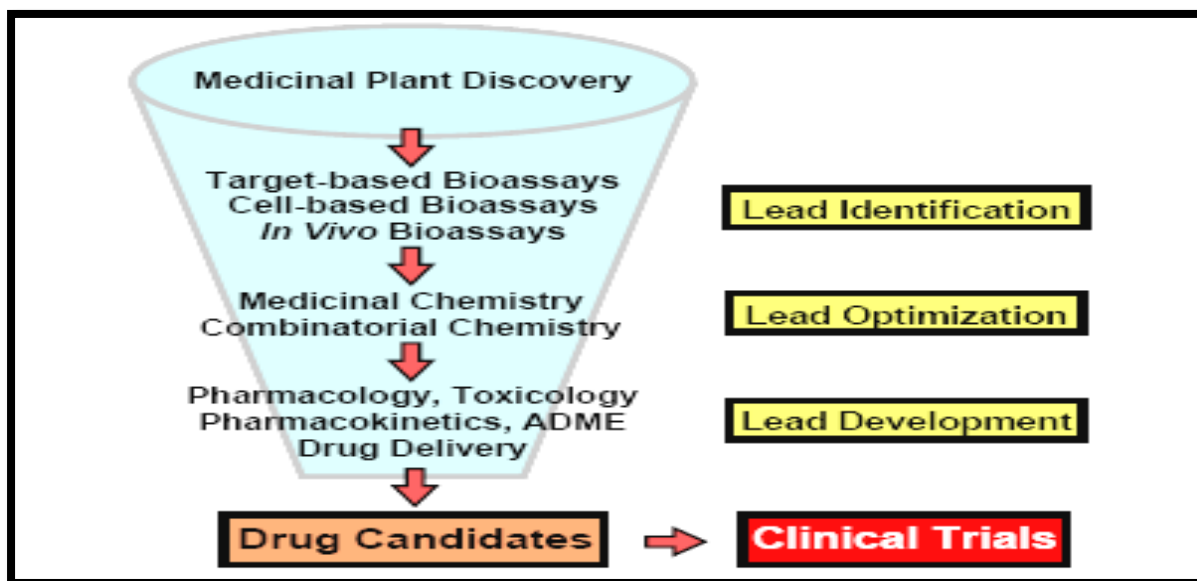


Fig.1 Comprehensive presentation of medicinal plant discoveries [8]

Medical plants have positive effects on human health [9]

- (I) Many contemporary medications, like aspirin, are made inadvertently from therapeutic plants.
- (II) Many cultures around the world, including Chinese medicine and Indian medicine, directly use plants as remedies.
- (III) Many food crops, like garlic, have therapeutic properties. New medications can be derived from medicinal plants. More than 250 000 different species of flowering plants are thought to exist.
- (IV) Studying therapeutic plants enables one to comprehend plant toxicity and safeguard both people and animals against natural toxins.
- (V) The cultivation and preservation of therapeutic plants, for instance, protects plant metabolic engineering.

2. Fundamentals of phytochemistry

In the strictest sense, phytochemistry is the study of phytochemicals. These substances are made from plants. The phrases are frequently used in a more restricted sense to refer to the numerous secondary metabolic products that are present in plants. Numerous of them are well known for offering defense against insect assaults and plant illnesses [10]. For human consumers, they also provide a number of protective activities. The separation, identification, and determination of the structure of physiologically active chemicals have become simpler thanks to the ongoing creation of spectroscopic and chromatographic analytical techniques[11]. The publication *Phytochemistry letters* promotes research in all fields involving natural products, including structural elucidation, biotechnology, pharmacology, ethnobotany and traditional use, genetics, analytical evaluation of herbal remedies. Research medicinal herbs and plants, provide training in phytochemical examination, and advise on sample recognition, processing, and production of excellent herbal products. [12-13].

The Phytochemistry Unit, which has expertise in the following areas [14], plays a crucial role in the gathering of plants that HMRC finds interesting [14].

- Gathering and preparing plant samples.
- Plant sample preparation for screening and bioassay research on plant compounds.
- Utilizing methods like Flash Chromatography, Preparative TLC, Preparative HPLC, GC-MS, HPLC-UV, HPLC-Diode Array, and LC-MS analysis, bioactive chemicals can be fractionated and isolated.
- Preparing and using HPLC to analyze standardized extracts.

Herbal medicine has been used for millennia and is being studied in some European and Asian nations. A great deal of effort has been made that is beyond the comprehension and ability of the average person [15]. The best thing about using herbal medication is that it can be used by people of any age group and has no side effects or ineffective cures. Polyherbal formulations are defined as those that contain two or more herbs. Numerous studies using turmeric rhizomes (*Curcuma longa* Family-Zingiberaceae) and neem leaves (*Azadirachta indica* Family-Meliaceae) extracts as well as several other herbal medicines have been carried out [16-17]. Ointments, a semisolid preparation used topically for a variety of reasons, including protectants, antiseptics, emollients, antipruritics, keratolytics, and astringents, are also available as dosage forms for herbal medications. Neem is made up of *Azadirachta indica* (Meliaceae) leaves and other aerial elements. Neem leaves and neem oil are being tested for their effectiveness in treating AIDS and have several qualities, including antiseptics, insecticides, antifertility, and antiviral effects [18-19]. Turmeric is made from the dried and fresh rhizomes of the Zingiberaceae plant species, *Curcuma longa* [20]. It is employed as an antibacterial, expectorant, seasoning, or condiment. Due to its high antioxidant content, studies have shown that turmeric can be used to treat conditions like arthritis, liver disease, Alzheimer and depression [21].

3. A Method and Material of plant extract

Phytoplanetlibrary

Neem leaves were gathered from the neighborhood around the city, while dried turmeric rhizomes were bought at a nearby market.

Preparation of neem extract [22]

After being carefully washed with distilled water, the plant leaves were picked and allowed to dry for 10 days in the shade. The powdered dry leaves were created.. After being ingested for three hours with 350 ml of 90% ethanol, 100 g of powder was transferred to a percolator, and 150 ml of 90% ethanol was added. The powder was macerated for seven days while being stirred occasionally. After collecting

and concentrating the ethanolic extract, a blackish-green residue was obtained. The extract was kept in a cool, dry location in an airtight container.

Turmeric extraction and processing [23]

Dried rhizomes of turmeric was ground into a powder, and the extraction procedure was the same as it was for neem leaf extract. plant extract with a ruby red hue was achieve and kept in an airtight container in a cool, dark location.

4. Developing herbal ointment [24]

Table 1: manufacturing the ointment base

S. No.	Content of ointment base	Take quantity
01	Wool fat	0.5gm
02	Cetostearyl alcohol	0.5gm
03	Hard paraffin	0.5gm
04	Yellow soft paraffin	8.5gm

Table 2: manufacturing of herbal ointment

S. No.	Name of plant extract	Take quantity
01	Extracted neem prepared	0.06gm
02	Extracted turmeric prepared	0.06gm
03	Ointment base q.s.	10 gm

The steps involve in formation of herbal ointment [25]

Topical ointment bases-simple ointment BP, emulsifying ointment BP, and aqueous cream BP-with varied degrees of aqueous or anhydrous character were made using the fusion process. Initial preparation of the ointment foundation involved carefully grated hard paraffin weight, which was then placed in an evaporating dish. Neem and turmeric extracts were accurately measured and added to the herbal ointment foundation using the Using a levigation method, a smooth paste that was two or three times as heavy as the base was produced. The ointment was subsequently placed into a suitable container after adding additional base gradually until it was homogeneous.



Fig.2 Dried ethanolic turmeric and neem extract to evaluate Physicochemical studies

6. Standards of evaluation for herbal ointments [26]

A visual examination was used to check physical characteristics, including color and smell.

Consistency

Smooth consistency and no indications of greed are present.

Solubility

Ether, alcohol, and chloroform are all soluble in and miscible with boiling water.

Washability

After the combination had been applied to the skin, the ease of water washing was evaluated.

pH

With the use of a digital PH meter, the PH of the herbal ointment was determined. 100 ml of distilled water was used to make the ointment solution, which was then left to sit for two hours. The PH of the solution was assessed three times, with the average reading being calculated.

Spreadability

The spreadability was evaluated by sandwiching excess sample between two slides that had been uniformly crushed by employing a predetermined weight for a particular period of time. The amount of time required to separate the two slides was used to calculate spreadability. The result of separating two slides faster is better spreadability. Spreadability was determined using the formula below.

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

Where,

S = Spreadability

M denotes the weight of the upper slide. L = Glass slide length

T is the duration needed to separate the slides.

Extrudability

A tube-shaped container was used to store the mixture. Extrudability was calculated as the weight of cream needed to extrude 0.5 cm of cream ribbon in 10 seconds.

Diffusion analysis

The diffusion research was carried out using the agar nutritional medium. A board with a hole in the middle was filled with ointment. It was apparent how long the ointment took to diffuse. (After 60 seconds)

Stability study

A four-week physical stability test on the herbal cream was conducted at different temperatures, including 2°C, 25°C, and 37°C. It was found that the herbal ointment was physical steadiness across a range of temperatures, including 2°C, 25°C, and 37°C, four weeks soon.

Table 3: Analysing a herbal ointment's physical-chemical composition [27]

S.No.	Physicochemical parameters	Observation
01	Colour	Yellow
02	Odour	Characteristic
03	Consistency	Smooth
04	PH	5.4
05	Spreadability (seconds)	7
06	Extrudability	0.4 gm
07	Diffusion study (after 60 min)	0.7 cm
08	Loss on drying	30%
09	Solubility	Soluble in boiling water, miscible with alcohol, ether, chloroform
10	Washability	Good
11	Non irritancy	Non irritant
12	Stability study (20°C, 25°C, 37°C)	Stable

7. The antifungal characteristics of the herbal ointment [28-30]

The herbal ointment's antifungal effectiveness was examined during the current investigation. The antifungal experiment included *C. albicans*, *M. audouinii*, *A. niger*, and *T. mentagrophytes* are four typical bacteria was obtained antifungal activity of the compounds was assessed using the disc-diffusion method.

Materials Used

- sterile Petri dishes
- Clean graduated pipettes
- Sterilised beakers, glass rods, conical flasks, and watch glasses.
- A 6mm cork borer that has been sterilised.
- A growing culture that is 18–24 hours old and is in nutrient broth.
- Forceps with fine, sterilised points.
- Syringes used to treat tuberculosis.
- Sterile cotton swabs and cotton wool.

Building nutrition agar media

The following items were used to make the nutritional agar media.

Peptone : 20 g.

Beef extract : 05 g.

Sodium chloride : 05 g.

Agar : 20 g.

Distilled water up to : 1000 ml.

Peptone and beef extract were weighed out in portions that were gently warmed in distilled water before being heated on a boiling water bath to dissolve the necessary amount of agar. Sodium chloride is then used to adjust the PH of the aforementioned solution, and the resulting product is then diluted with distilled water to make a volume of 1000 ml. The healthy agar medium is next sterilized by autoclaving it at 120 °C under 15 lbs/in 2 pressure for 20 minutes.

The creation of test solution

10 ml of DMSO and 10 mg of the plant extract were used to dissolve it. The 10 ml of solution was then diluted with DMSO to create 100 ml. Currently, the test substance's concentration is 100 gm per ml. These test-tube preparations for sample solutions were labelled and sanitized.

The creation of standard solution

The usual drug utilised throughout testing is fluconazole. The dosage of this drug is altered to contain 100 g per ml because it is water soluble.

Approach to testing

The previously created nutritious agar media is cooled to 45°C while being gently agitated to guarantee consistent cooling. This was aseptically supplemented with a culture that was 18 to 24 hours old, properly mixed, and gently shaken. This was put in the large petridishes (20 to 25 ml each) and let to sit for an hour. The cups were formed by scooping out the pierced agar after the set agar had been punctured with a clean cork borer. The circumference of each cup is 48 mm. These cups received 100 cc of the plant extract that had been prepared in DMSO. The drug solution was next added, and it

was allowed to diffuse for about 45 minutes at room temperature. The plates were then kept at 37 °C for the following 24 hours in an incubator. After 24 hours, the inhibition's diameter or area in millimeters

were

measured.

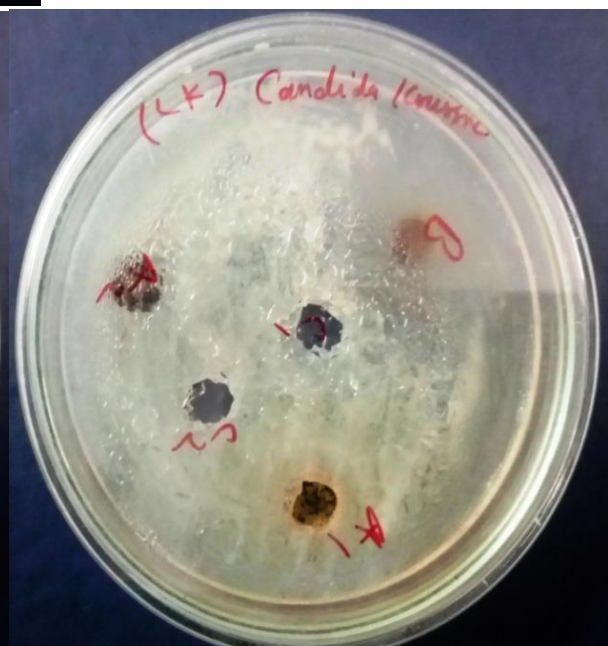
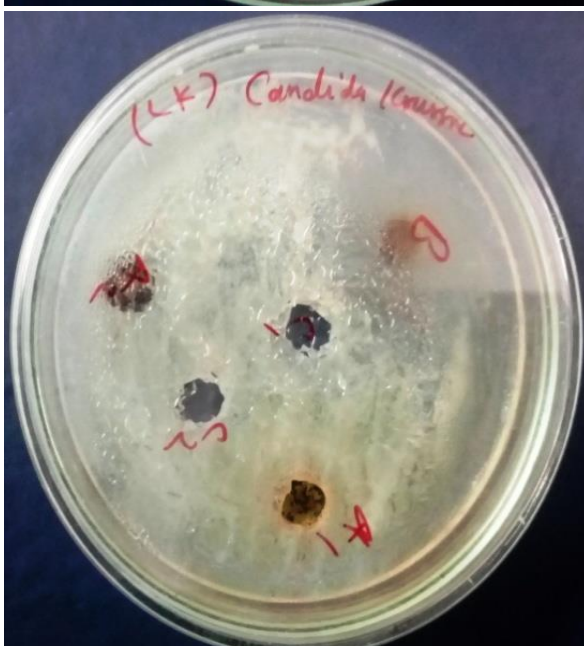
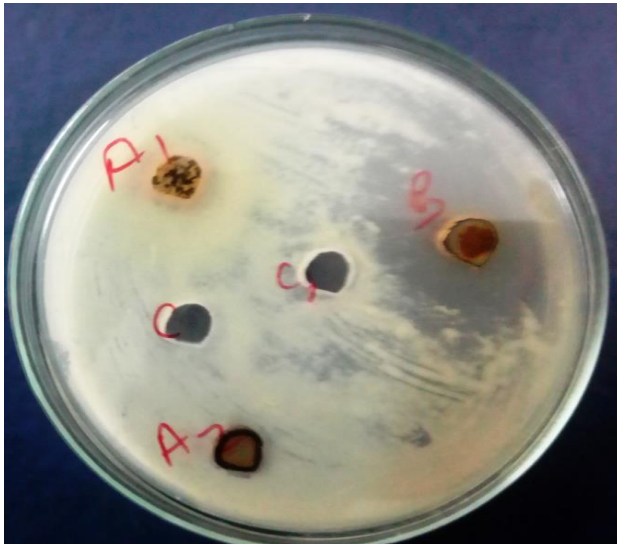
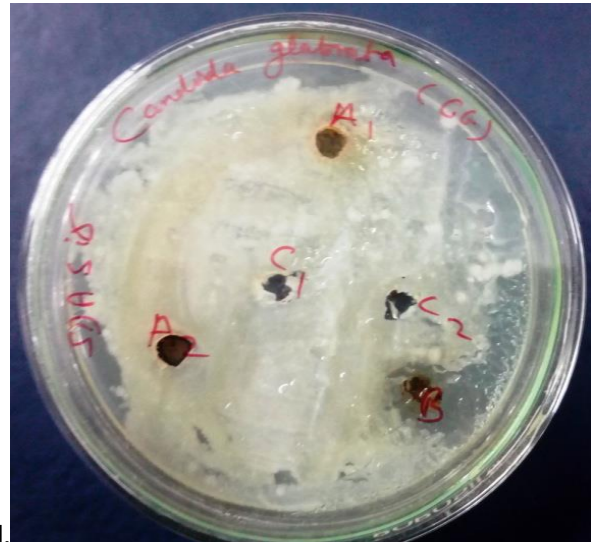


Fig.3 Diagrammatic presentation of inhibition zone area showing antifungal activity

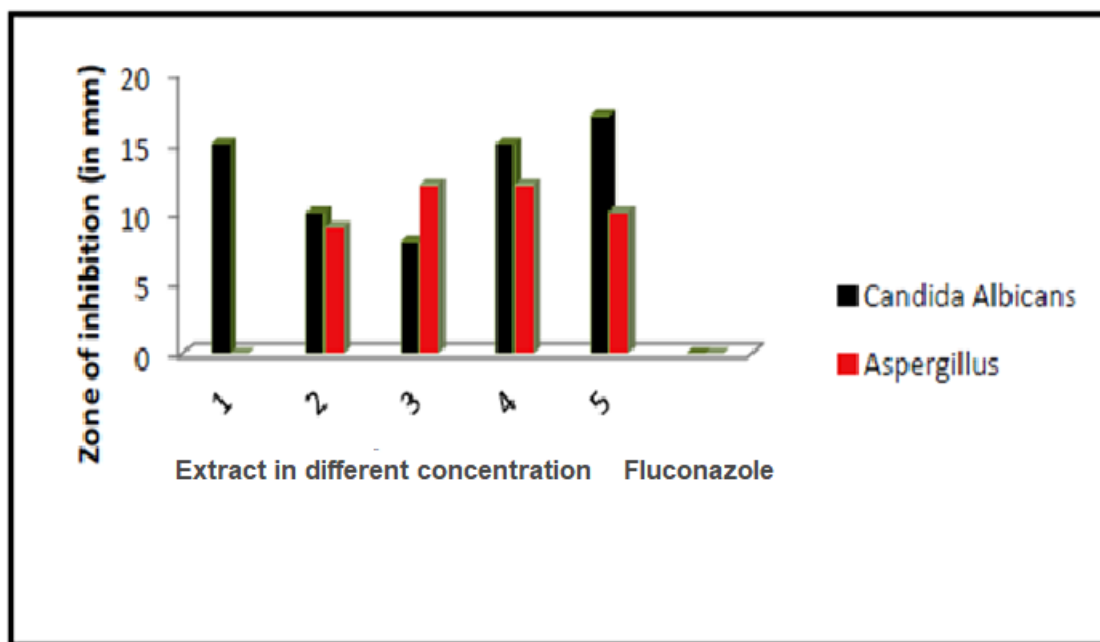


Fig.4-Antifungal activity of herbal ointment

8. Result and Discussion

The current investigation's objectives were to create and assess herbal ointment. To acquire a decent extract yield for this without damaging the chemical components or their action, Basic maceration was the technique employed to create the botanical extracts. In order to ensure that the herbal extract and ointment base were blended equally and remained stable throughout storage, the levigation technique was utilized to make the ointment.

Investigations on the physicochemical properties produced positive results for spreadability, extrudability, washability, solubility, loss on drying, and other factors.

In addition, the formulation was exposed to a variety of temperatures over the course of four weeks, including 2°C, 25°C, and 37°C, to investigate its stability. There were no changes discovered in terms of spreading potential, diffusion analysis, or annoying effect. The ethanolic extract and fractions of Neem and Turmeric were potential antifungal agents because both had MIC values of less than 1000 g/mL. With MIC values of 50–100 g/mL, the ethylacetate fraction of neem and turmeric showed mild to moderate antifungal activity, against *C. albicans* and *A. niger* species, having the maximum antifungal activity (MIC 12.5 g/mL)

Reference

1. X. Chen, H. Zhu, X. Liu, Q. Li and M. Dong, Design and synthesis of novel GluN2A NMDAR positive allosteric modulators via scaffold hopping strategy as anti-stroke therapeutic agents, *Bioorganic & Medicinal Chemistry* 83 (2023) 117236.
2. H. Sato, A. Chandela and Y. Ueno, Synthesis and characterization of novel (S)-5'-C-aminopropyl-2'-fluorouridine modified oligonucleotides as therapeutic siRNAs, *Bioorganic & Medicinal Chemistry* 87 (2023) 117317.
3. H. N. B. Nguyen and M. Y. Patuwo, Quantitative Structure-Activity Relationship (QSAR) modelling of the activity of anti-colorectal cancer agents featuring quantum chemical predictors and interaction terms, *Results in Chemistry* 5 (2023) 100888.
4. J. Almaliti and W. H. Gerwick, Methods in marine natural product drug discovery: what's new?, *Expert Opinion on Drug Discovery* 18 (2023) 687–691.
5. I. W.-J. Tian and X.-J. Wang, Broad-Spectrum Antivirals Derived from Natural Products, *Viruses* 15 (2023) 1100.
6. R. Lu, H. Akasaka and K.-H. Ruan, Design, synthesis and characterization of lead compounds as anti-inflammatory drugs targeting mPGES-1 via enzymelink screening, *Future Medicinal Chemistry* 15 (2023) 757–767.
7. M. P. Badart and B. C. Hawkins, Synthetic Strategies to Access Heteroatomic Spirocentres Embedded in Natural Products, *Synthesis* 53 (2021) 1683–1705.
8. M. Feher and J. M. Schmidt, Property Distributions: Differences Between Drugs, Natural Products, and Molecules from Combinatorial Chemistry., *ChemInform* 34 (2003).
9. A. K. Mandal, A. Pandey, P. Pant, S. Sapkota, P. Yadav and D. P. Bhandari, Formulation of Herbal Tea from Nepalese Medicinal Plants: Phenolic Assay, Proximate Composition and In-vivo Toxicity Profiling of Medicinal Plants with Nutritive Benefits, *Journal of Plant Resources* 20 (2022) 139–149.
10. I. S. Jethi, S. K. Mishra and K. B. Satapathy, Study on Ethnomedicinal Plants used against diabetes by the tribes of Gajapati district of Odisha: Ethnomedicinal plants used against diabetes, *Plant Science Today* (2023).
11. A. A. Grigoriev and I. A. Avdeenko, Regenerative ability of grafted cuttings in the callusing room by using solutions of physiologically active substances, *Agrarian science* (2023) 125–129.
12. I. Perception and use of Medicinal Plants and Chemical Products in the Traditional Treatment of Cutaneous Leishmaniasis in Morocco, *Tropical Journal of Natural Product Research* 7 (2023) 2946–2951.
13. N. Cicero, S. Gangemi and A. Allegra, Natural products and oxidative stress: potential agents against multiple myeloma, *Natural Product Research* 37 (2022) 687–690.
14. F. Barbier, F. Fichtner and C. Beveridge, The strigolactone pathway plays a crucial role in integrating metabolic and nutritional signals in plants, *Nature Plants* (2023).
15. This abstract has been withdrawn, *Heart, Lung and Circulation* 32 (2023) S228.
16. Assessment of Neem (*Azadirachta Indica*) and Lemongrass (*Cymbopogon citratus*) Extracts on *Meloidogyne incognita*, *Indian Journal of Ecology* (2023).
17. Z. El Ouafi, S. Pirro, N. Al Idrissi and H. Ghazal, The Complete Genome Sequence of *Curcuma longa* (Zingiberaceae, Zingiberales), Turmeric, *Biodiversity Genomes* (2023).
18. R. A. Chachar, Individual and Combined Effects of Moringa and Neem Leaves on Immune Response and Gut Microflora in Japanese Quails, *Pakistan Journal of Zoology* (2023).
19. K. Thangamani and Shiwani, Effect of magnetic water and neem leaves ash on strength properties, *Materials Today: Proceedings* 79 (2023) 406–410.
20. I. Gałazka-Czarnecka, Modification of color in turmeric rhizomes (*Curcuma longa* L.) with pulsed electric field, *PRZEGLĄD ELEKTROTECHNICZNY* 1 (2023) 125–127.
21. T. Asselah, What is the Path Forward to Treat Hepatitis Delta Virus?, *Clinics in Liver Disease* (2023).
22. Dr. M. Sakthivel, Dr. S. M. Halith, R. Karthikeyan, M. Kaviya, M. Kiruthika, S. Kowsalya and R. Krishnapriya, Formulation and Evaluation of Herbal Ointment Containing Neem and Turmeric Extract, *International Journal of Pharmaceutical Sciences Review and Research* 78 (2023).

23. O. Bayraktar, M. E. Uslu and G. Öder, Preparation of Encapsulated Turmeric Extract for Food Fortification, *Current Nutraceuticals* 04 (2023).
24. Dr. M. Sakthivel, Dr. S. M. Halith, R. Karthikeyan, M. Kaviya, M. Kiruthika, S. Kowsalya and R. Krishnapriya, Formulation and Evaluation of Herbal Ointment Containing Neem and Turmeric Extract, *International Journal of Pharmaceutical Sciences Review and Research* 78 (2023).
25. S. Biswas, D. Dasar, K. Khandare and A. Wanjari, Efficacy of Novel Herbal Formulation in Efficacy of Novel Herbal Formulation in Conservative Management of Mutrashmari (Urolithiasis) - A Case Report, *International Journal of Ayurvedic Medicine* 13 (2023) 1078–1082.
26. Miss. Shivani Prakash Kale, Miss. Snehal P. Patil, Sakshi T. Kale, Mr. Sachin S. Navale and Dr. Amol A. Patil, Formation and Evaluation of Herbal Ointment by using Curcuma Longa (Turmeric), *International Journal of Advanced Research in Science, Communication and Technology* (2023) 721–725.
27. M. Shelke, P. Sonawane, G. Bharskar and S. D. Mankar, Formulation and Evaluation of Antimicrobial Herbal Ointment containing Colocasia esculenta, *Research Journal of Pharmacognosy and Phytochemistry* (2022) 277–280.
28. F. A. da S. Sá, T. C. Silva, W. M. Andrade, R. I. de Ávila, M. C. Valadares, C. R. Costa, A. S. Santos, V. A. Q. Feitas, J. R. de Paula and M. do R. R. Silva, Antifungal activity of the ethanolic extract and flavonoid avicularin from *Myrcia tomentosa* (Aubl.) DC. on virulence factors of *Candida* species, *Journal of Herbal Medicine* 38 (2023) 100643.
29. Dr. M. Sakthivel, Dr. S. M. Halith, R. Karthikeyan, M. Kaviya, M. Kiruthika, S. Kowsalya and R. Krishnapriya, Formulation and Evaluation of Herbal Ointment Containing Neem and Turmeric Extract, *International Journal of Pharmaceutical Sciences Review and Research* 78 (2023).
30. A. Shah, Development and characterization of herbal ointment by using psoralia corylifolia and aloevera for the treatment of vitiligo, *International Journal of Biology, Pharmacy and Allied Sciences* 12 (2023).