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Original Research Article

Formulation and Evaluation of Polyherbal Ointment Using Carica Papaya and Leptadenia Pyrotechnica

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Abstract

The foundation of the herbal medication business is the ethnomedical values of plants. In order to create herbal medications with minimal side effects, India has contributed its expertise in traditional system medicines (Ayurveda and Siddha). These plants have historically been utilized by tribal communities in India as extracts, powders, or pastes to cure conditions including anti- microbial activity, cough and colds, fever, stomach, kidney, and liver diseases, pain, inflammation, and wounds. According to WHO data, plant extracts are used to treat more than 80% of the world's population for fundamental ailments. Natural remedies are popular in Asian nations provides a full overview of past human plant connections. Traditional medicinal plants have a diverse spectrum of bio-active compounds that are used to treat both chronic and infectious ailments.

The objective of the present study is to formulate and evaluate a polyherbal ointment. Ointment was formulated using ethanolic extracts of *Carica Papaya* and methanolic extract of *Leptadenia Pyrotechnica*. The total phenolic content (TPC) was determined by Folin-Ciocalteu method and total flavonoids content (TFC) were determined using aluminum chloride method. Formulations were prepared using different ointment bases with different concentrations of the extracts such as 1% and 2% w/w. Formulations were tested for its physicochemical properties like pH, spreadability, extrudability and viscosity. The TPC in *Carica Papaya and Leptadenia Pyrotechnica* extract were found to be 105.5 mg/gm of extract and 104.5 mg/gm of extract respectively. TFC in *Carica Papaya and Leptadenia Pyrotechnica* extract were found to be 20.5 mg QE/gm of extract and 22.5 mg QE/gm of flavonoid in dry extract respectively. Ointment was formulated using simple ointment base with 2 %w/w of dry extracts was found to most stable in its physicochemical properties. The ointment consisting of 2% extract of *Carica Papaya and Leptadenia Pyrotechnica* shows satisfactory physical properties and stability.

Keywords: Wound healing, Total Phenolic Content, polyherbal, Carica Papaya, *Leptadenia Pyrotechnica*, antioxidant.

Introduction

According to World Health Organization data, over 80% of people worldwide rely on plant extracts as their primary medical therapy. The usage of herbal treatments in Asian countries exemplifies the long history of interactions between people and plants. Not just in India but all throughout the world, the usage of herbal medicines made from traditional medicinal plants has grown significantly. Native knowledge of Indian medicine has been passed down through the years, primarily within certain areas or tribal tribes. This age-old knowledge comes from conventional Indian medical systems like Ayurveda and Siddha, and it is also gaining popularity in the West. Due to their low cost and seldom side effects, ethnomedicines and herbal medications have become more popular. The World Health Organisation (WHO) has recently recognised the value of traditional medicine in the healthcare sector. In the Ayurvedic and Siddha medical systems, remedies are made from certain plant components and used to cure various diseases. Numerous traditional medicinal plants used in Siddha and Ayurveda medical systems have been the subject of scientific study for almost 30 years. The scientific

evaluation of traditional medicinal plants provides alternative medicines that are supported by science, laying the groundwork for the herbal drug business and identifying therapeutic targets in the pharmaceutical sector. Recently there has been a shift in universal trend from synthetic to herbal medicines, which we can say 'Return to Nature'. Medicinal plants have been known for millennia and are highly esteemed all over the world as a rich source of therapeutic agents for the prevention of diseases and aliments. Nature has bestowed our country with an enormous wealth of medicinal plants; therefore, India has after been referred to as the medicinal garden of the world. The emphasis on use of medicinal plants had either to be placed on the treatment rather than prevention of diseases. Over 90% of traditional medicine remedies contain medicinal plants, the medicinal plants that have been implicated with preventive measures in diseases control strategies.

Historically, medicinal plants have been the primary source of new antimicrobial medications. These ancient medicinal plants have significantly improved human health. Plants are also known to be a major source of secondary metabolites and essential oils. The goal of this study was to determine the antioxidant activity of Carica Papaya leaf extracts in ethanol and *Leptadenia Pyrotechnica* whole plant in methanol.

Material and Methods

The fresh leave of *Carica papaya* and *Leptadenia Pyrotechnica* collected from the swami Keshwanand Rajasthan Agriculture University, Bikaner, Rajasthan. The sample was identified and authenticated by Department of Botany, Government College Kolayat, Bikaner, Rajasthan.

Preparation of the extracts

The method of extraction was conducted according to Vuong *et al*. (2013) with slight modification. Initially, the leaves were washed with tap water and dried in the oven at 40°C. Afterward, the dried leaves were ground using blender (Model: CB15V, India) to obtain a fine powder. *Carica Papaya* leaves and whole plant of *Leptadenia Pyrotechnica* were subjected to a soxhlet extraction system for 24 hrs (60 °C) using ethanol and methanol as solvent respectively. Extracts were filtered and concentrated using a rotary vacuum evaporator at 60°C. The dried extracts were stored until further use.

Determination of Total Phenolic Content (TPC)

The TPC of the *Carica papaya and Leptadenia Pyrotechnica* extracts were determined spectrophotometrically according to the Folin-Ciocalteu colorimetric method with slight modifications. 5 mg of gallic acid was dissolve into 10 ml ethanol and make up volume to 100 ml with distilled water, so the concentration of the solution 50 mcg/ml then standard series was performed in order to prepare different concentrated solution. From the standard gallic acid stock solution prepare serial dilution as different concentrated solution as 1 ml, 2 ml, 3 ml, 5 ml, 7 ml and 10 ml of six test tubes each containing gallic acid of 50 mcg, 100mcg, 150mcg, 250mcg, 350mcg, 500mcg respectively in 50 ml standard volumetric flask. Take 1ml, 2ml, 3ml, 5ml, 7ml and 10ml of plant extract sample solution is taken in separate volumetric flasks. Add 5 ml of FC reagent and 5 ml of 7.5 % sodium carbonate solution in all volumetric flasks. The test tubes were incubated for 20 minutes at room temperature to complete the reaction. Blue color is observed. Pipette out 2 ml of solutions separately from each volumetric flask and transfer to new test tubes marked as the extract series (S1 to S6) and test (T1 and T2). Make up volume of each test tube with distilled water up to 5 ml. All experiments were made in triplicates and the TPC was determined using the standard gallic acid calibration curve.

Determination of Total Flavonoid Content (TFC)

From stock solution of quercetin is diluted serially to make various concentrations of 0.25 mcg/ml, 0.5 mcg/ml, 0.75 mcg/ml, 1.0 mcg/ml and 1.25 mcg/ml solutions. 1 ml quercetin of each concentration was added to five separate test tubes containing 4 ml distilled water. 2 sample extract of 1 ml each was added to the test tube containing 4 ml of distilled water. At the same time, 0.5 ml of 5 % NaNo2 was added to test tube and 0.5 ml of 10 % AlCl3 after 5 minutes. Then 2 ml of 1M NaOH was added to the mixture after 6 minutes, the volume of mixture was made 10 ml by distilled water. Rest the test tubes for 15 minutes to incubate at room temperature. Absorbance was measured by spectrophotometer at 510 nm. The total flavonoid content was expressed as quercetin equivalents using the linear equation based on calibration curve.

Preparation of polyherbal ointment

Polyherbal ointments were prepared using different ointment bases like water soluble base, water miscible base, oleaginous base, o/w (oil-in-water) emulsion base, w/o (water- in-oil) emulsion base [8]. Formulation with code F1 – F2 was prepared using an oleaginous base with 1 and 2 % w/w extract.

Wool fat, hard paraffin, cetyl alcohol, yellow soft paraffin, methyl paraben and propyl paraben were used to prepare ointments with 1 and 2 % w/w

extract. F3 – F4 was prepared by using water miscible bases. Cetyl alcohol, white beeswax, span 60, tween 80, purified water, Methyl paraben and propyl paraben was used for the preparation of ointments with 1 and 2 % w/w extract. F5 – F6 Ointments were prepared by using water soluble base. The ointments were prepared using PEG 4000, PEG 400, methyl paraben and propyl paraben with1 and 2 % w/w extract. Formulations F7 – F8 was prepared using o/w emulsion base. White bees wax, cetyl alcohol, propylene glycol, tween 80, water, methyl paraben and propyl paraben was used for preparation of ointments with 1 and 2 % w/w extract. Formulation F9 – F10 was prepared by using w/o emulsion base. Liquid paraffin, white beeswax, wool fat, borax, water, methyl paraben and propyl paraben was using for the preparation of ointments with 1 and 2 % w/w extract. Wherever, oil phase and aqueous phase were to be added, precaution was taken that both the phases have the same temperature, 70°C. This was done to avoid the separation of aqueous phase during the mixing of content and its cooling. All formulations were prepared by using *Carica Papaya* and *Leptadenia Pyrotechnica* as an active ingredient in 1:1 ratio.

Evaluation of Polyherbal Ointment Appearance and homogeneity

Physical appearances, as well as homogeneity of all developed ointments, were tested by visual observation

Color and Odor

Sensory organs were used to evaluate color and smell. Color and smell were evaluated before and after the time frame.

Consistency

Small quantity of prepared ointment was taken. The ointment was rubbed between the thumb and index finger and the observation was noted.

pH

All the ointments were subjected to pH determination. A pH meter expressing the values digitally was utilized to determine the pH. To determine this, 1% solution of ointment was prepared. Dilution of 1 gm ointment was done with distilled water to make up the volume to 100 ml. The solutions were kept aside for 2 hours.

Diffusibility

Agar nutrition media was made and added to a Petri dish for the diffusion research. To place the prepared ointment, a hole was made in the center of the agar media. After 60 minutes, the diffusibility was tested.

Viscosity

The Brookfield viscometer was utilized to determine the viscosity of the formulation. Sample was taken in a beaker. Standard operating procedure of viscometer was followed while determining viscosity. The spindles no. 1 to 4 was used. The test sample viscosity was determined by using spindle numbers 1, 2, 3 and 4. Each spindle was fixed and rotated at speeds of 0.3, 0.6, 1.5, 3, 6, 12, 30 and 60 rpm respectively. Readings were noted only when the dial reading was above 10. This reading was considered for the calculation of viscosity.

Sensitivity

Eight volunteers had their forearms treated with some of the ointment. After 20 minutes, we looked for any toxicity (Draize *et al*; 1994).

Washability

A portion of ointment was applied on the skin and allowed to flow under the flow of tap water till its complete removal from the skin. The time of complete removal of ointment from the skin was noted.

Loss on Drying

This analytical technique was utilized to determine weight loss, placing a required quantity of ointment inside a petri-dish which in turn was placed on a water bath. The sample was dried up at 105ºC

Loss on drying expressed in percentage = $W_i - W_f / W_i * 100$

Where, W_i symbolizes the first sample weight taken, W_f symbolizes the ultimate sample weight after heating

Spreadability

To measure the spreadability parameter the sample (1 g) was sandwiched between two glass slides and weight (50 g) was applied to the slide for 5 minutes to press it to a uniform thickness. The spread ability of the ointment is measured in seconds by the time it takes to separate the two sides (Schwarz, *et al*; 2001). The formula to calculate the spreadability is as below:

$S = M \times L / T$

Where, S =Spreadability, M=Weight placed on upper slide, L = Length of Glass Slides, T=Time required to separate the slides.

Extrudability

Extrudability studies are dependent on the amount of ointment extruded when finger pressure is applied. The ointments were packed in 5 gm collapsible aluminum tubes that are lacquered. These collapsible tubes have a nasal neck tip with an opening of 5 mm. With the help of a finger, the pressure was applied to the collapsible tubes. The quantity of formulation that is extruded out from these lacquered tubes in 10 seconds was measured.

Results and Discussion

TPC

The total phenol content was determined by Folin Ciocalteu method and reported as gallic acid equivalents (GAE) concerning the standard curve.

The concentration of TPC presents in the leaf extract of *Carica papaya* in methanol and *Leptadenia Pyrotechnica* in ethanol was 105.5 mg/gm of extract and 104.5 mg/gm of extract respectively.

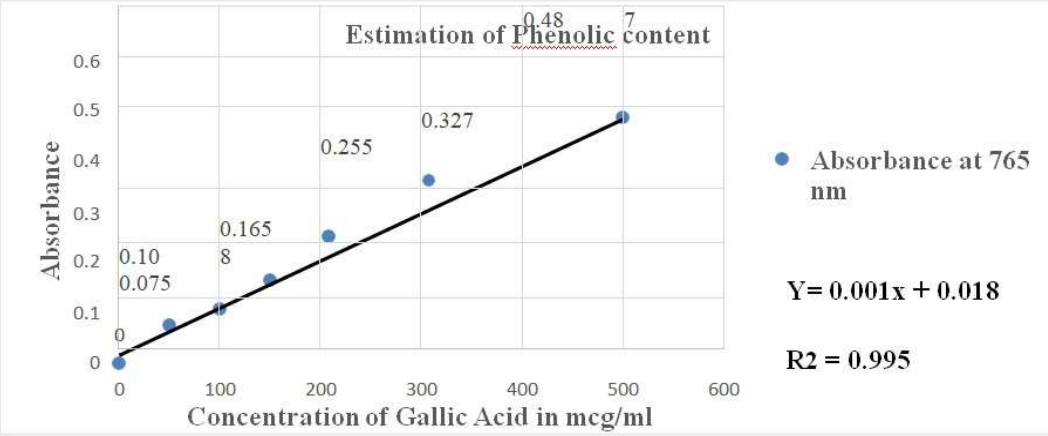

Figure 1: Standard gallic acid calibration curve. (Values are expressed as mean ± SE mg of gallic acid equivalent per gram of dry weight, that is, GAE/g of the extract triplicates of each sample extract was recorded)

TFC

The TFC was determined by aluminum chloride method and represented as quercetin equivalents (QAE) concerning the standard curve. The concentration of sample extract was evaluated by comparing it to the standard Graph 2. The concentration of TPC presents in the leaf extract of *Carica papaya* in methanol and *Leptadenia Pyrotechnica* in ethanol was 20.5 mg QE/gm of extract and 22.5 mg QE/gm of flavonoid in dry extract respectively.

$y = 0.7358x + 0.1605$, $R^2 = 0.9141$

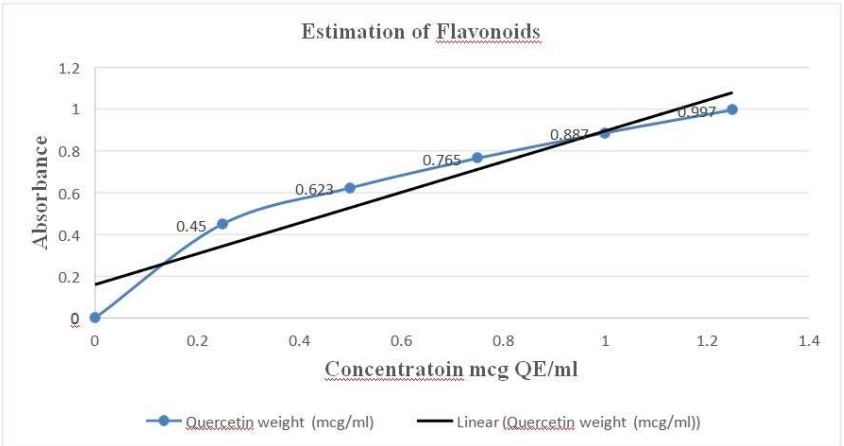

Figure 2: Standard quercetin calibration curve. (Values are expressed as mean ± SE mg of quercetin equivalent per gram of dry weight, that is, QAE/g of the extract triplicates ofeach sample extract was recorded)with a straight-line equation

Table 1: Evaluatory parameters of color, odor, consistency, pH, LOD and viscosity in formulations

Formulation	Parameters					
	Color	Odor	Consistency	pH	LOD (%)	Viscosity(cp)
F1	Yellowish	Characteristic	Irregular	7.1	29	256
F2	Yellowish	Characteristic	Smooth	7.2	27	248
F3	Yellowish	Characteristic	Irregular	6.9	28	248
F4	Yellowish	Characteristic	Irregular	6.5	25	249
F5	Yellowish	Characteristic	Smooth	7.2	26	260
F6	Yellowish	Characteristic	Smooth	5.9	28	28
F7	Yellowish	Characteristic	Irregular	6.9	26	245
F8	Yellowish	Characteristic	Smooth	6.8	32	242
F9	Yellowish	Characteristic	Smooth	7.5	28	236
F10	Yellowish	Characteristic	Irregular	7.1	32	235

Table 2: Evaluatory parameters of Spreadability, Extrudability and Washability informulations

Formulation	Parameters		
	Spreadability (sec)	Extrudability (gm)	Washability
F1	5 ± 0.02	0.44 ± 0.03	Ease
F2	4 ± 0.03	0.5 ± 0.02	Ease
F3	4 ± 0.04	0.9 ± 0.03	Ease
F4	5 ± 0.02	0.35 ± 0.02	Slightly greasy
F5	4± 0.02	0.46±0.03	Ease
F6	5± 0.03	0.39±0.02	Slightly greasy
F7	5 ± 0.03	0.44 ± 0.02	Ease
F8	6 ± 0.02	0.5 ± 0.03	Ease with soap
F9	6 ± 0.05	0.45 ± 0.04	Greasy
F10	5 ± 0.02	0.44 ± 0.03	Greasy

Discussion

Medicinal plants constitutes as an important natural wealth of the country by playing asignificant role in primary health of mankind. They importantly serve as raw material for manufacturing medicines as therapeutic drugs. *Carica papaya and Leptadenia Pyrotechnica* is used as a natural medicinal plant, recognized for its antimicrobial and wound healing activity.

The present study was done to formulate and evaluate polyherbal ointment. For this the herbal extracts were prepared by using soxhlet process to obtain a good yield of extract and there was no any harm to the chemical constituents and their activity.

The formulations (F1-F10) were prepared using different ointment bases so that uniform mixing of the herbal extract with the ointmentbase was occurred which was stable during the storage. The physicochemical properties were studied which shows satisfactory results in formulation F2 for spreadability, extrudability, washability, solubility, loss on drying and others.

References

1. Agar, O.T., Dikmen, M., Ozturk, N., Yilmaz, M.A., Temel, H. and Turkmenoglu, F.P. (2015). Comparative Studies on Phenolic Composition, Antioxidant, Wound Healing and Cytotoxic Activities of Selected *Achillea* L. Species Growing in Turkey. *Molecules,* 20(10), 17976-18000. https:// doi.org/10.3390/molecules201017976
2. Kandikonda Maneesh, Kanakam Vijayabhaskar, Heena Firdouse, Pingali Srinivasa Rao, Mothukuri Prajwitha, Sreeramula swetha. Evaluation of Antimicrobial of P. vesicularis, Streptococcus faecalis, Aeromonas hydrophilia, Salmonela typhae, Stphylococcus cohni, Serratia ficaria and E. coli. Of crude and n-butanol fraction fruit latex of Carica papaya L. (Caricaceae). Asian Journal of Pharmaceutical Research. 2021; 11(2):92-4.
3. Anand D.A.V., Arulmoli, R. and Parasuraman, S. (2016). Overviews of Biological Importance of Quercetin: A Bioactive Flavonoid. *Pharmacognosy reviews,* 10 (20), 84-89.
4. Asghar, N., Naqvi, S.A., Hussain, Z., Rasool, N., Khan, Z.A., Shahzad, S.A., Sherazi, T.A, Janjua, M.R.,Nagra, S.A., Zia-Ul-Haq, M. and

Jaafar, H.Z. (2016).Compositional difference in antioxidant and antibacterial activity of all parts of the *Carica papaya* using different solvents. *Chemistry Central Journal,* 10(5), 1-11.

5. Canini, A., Alesiani, D., D'Arcangelo, G. and Tagliatesta, P. (2007). Gas chromatography mass spectrometry analysis of phenolic compounds from *Carica papaya* L. leaf. *Journal of Food Composition and Analysis*, 20(7), 584-590.
6. Prashant Purohit and Sahil, A review on wound healing activity of medicinal plants, international journal of pharmaceutics & drug analysis, 6 (7), 2018; 545 – 547.
7. Ismail, H.F., Hashim, Z., Soon, W.T., Rahman, N.S.A., Zainudin, A.N. and Majid, F.A. (2017). Comparative study of herbal plants on the phenolic and flavonoid content, antioxidant activities and toxicity on cells and zebra fish embryo. *Journal of Traditional and Complementary Medicine*, 7(4), 452-465.
8. Kurahashi, T. and Fujii, J. (2015). Roles of Antioxidative Enzymes in Wound Healing. *Journal of developmental biology,* 3(2015), 57-70.
9. Lodhi, S. and Singhai, A.K. (2013). Wound healing effect of flavonoid rich fraction and luteolin isolated from *Martynia annua* Linn. on streptozotocin induced diabetic rats. *Asian Pacific Journal of Tropical Medicine*, 2013, 253-259.
10. Mohini Shelke, Avesh Tamboli, Pankaj Sonawane, Priyanka Sadaphal, S.D. Mankar. A Review on Pharmacognosy and Pharmacological Activity of Carica papaya Leaf. Research Journal of Pharmacognosy and Phytochemistry. 2021; 13(4):200-4.
11. Esther Lydia, Sheila John, Riyazudin Mohammed, Thiyagarajan Sivapriya. Investigation on the Phytochemicals present in the Fruit peel of Carica papaya and evaluation of its Antioxidant and Antimicrobial property. Res. J. Pharmacognosy and Phytochem. 2016; 8(4): 217-222.
12. Prashant Purohit and Himisha Gaur, "Lipid Based Nanoparticles in Cancer Therapy" Indo American Journal of Pharmaceutical Research.2023:13(07).
13. Mrunali Potbhare, Deepak Khobragade. In Vitro Evaluation of Antioxidant Potential of Ayurvedic Preparations Lauha Bhasma and Mandura Bhasma. Asian J. Pharm. Res.2017; 7(2): 63-66.
14. Sanju, S.K. Singh, Prashant Purohit,Tejaram and Kashi Ram , Preparation and Characterization of Isoniazid Loaded Nanoparticles Using Natural Polymers.(2024).Int J Pharm Sci.15(2), p31-39.
15. Selvakumar K., Madhan R., Srinivasan G. , Baskar V. Antioxidant Assays in Pharmacological Research. Asian J. Pharm. Tech. 1(4): Oct. - Dec. 2011; Page 99-103.
16. Vidya. M, Bhavya. S. V, Sunitha. P. S, Rashmi P. Benefits of Papaya. Int. J. of Advancesin Nur. Management. 2019; 7(2):165-166.
17. Tejaram, Rajesh Sharma, Prashant Purohit, Sanju, Kashi Ram, Formulation and Evaluation of Cefixime Loaded Grafted Gellan Gum Tablet, International Journal of Pharmacy and Biological Sciences (2023) 13 (3): 89-98
18. Mohan Kalyan R Konduri, Kiran B Uppuluri, Ramesh Chintha, Shaik Mulla, Rakesh Peruri. In-vitro Antimicrobial Activity of Four Indigenous Medicinal Plants Belonging to Bapatla, A.P. Research J. Pharm. and Tech. 3(2): April- June 2010; Page 461-465.
19. Wajid Azam, Vicky Rai, Rehan Jawed, Kalyani Jaywardhane, Vishal Thakur, Vishal Jain, Parwez Alam. Comparison of Antifungal and Antimicrobial activities in different parts of Carica papaya Linn. Asian Journal of Pharmaceutical Research. 2023; 13(4):213-8.
20. Kokate, C., Purohit, A., Gokhale, S., Text Book of Pharmacognosy, 13th ed. Nirali Prakashan, Pune, pp. 74-84 (1999).
21. Khare CP, 2004. Encyclopedia of Indian Medicinal Plants, Springer, Germany, pp. 23-25,333-336,432-433.