INVESTIGATION OF THE ANTIBACTERIAL AND ANTIOXIDANT ACTIVITY OF COCCINIA GRANDIS HYDROALCOHOLIC EXTRACT AND IXORA COCCINEA METHANOLIC EXTRACT.

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Abstract

In the current study, methanolic extract of Ixora coccinea leaves and hydro-alcoholic extract of coccinia grandis, both of which are said to have anti-oxidant and anti-bacterial properties, are prepared and analyzed. When compared to the industry standard, the methanolic extract of Ixora coccinea leaves and hydro-alcoholic extract of Coccinia grandis (ICCG) demonstrated significant antibacterial and antioxidant test activities. The inhibition zone diameters were determined to assess the antibacterial activity against Staphylococcus aureus and E. coli using the traditional cup-plate method. In contrast to other formulations, it is clear that ICCG5, which contained clove oil (0.7%), Ixora coccinea (7%), and Coccinia grandis (8%), displayed a greater zone of inhibition. Hence, ICCG5 formulation was found to have better antibacterial and antioxidant activities than other formulations. Results of all other ICCG5 evaluation parameters were all acceptable for all formulations.

Keywords: Ixora coccinea and Coccinia grandis, Formulation, Antioxidant and Antibacterial activity.

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Introduction:

In Ayurveda, *Ixora coccinea Linn*, a member of the Rubiaceae family, is referred as vetchi, flame of the woods, or jungle of geranium. This popular flowering shrub is Asian in origin. Its name is a corruption of an Indian god. It has long been utilised for its hepatoprotective, chemotherapy-protective, antibacterial, antioxidant, anti-nociceptive, anti-mitotic, and anti-inflammatory properties [1]. The primary mechanism underlying a lot of human neurologic and other illnesses today appears to be free radicals or oxidative damage. For instance, it has been shown that diabetes is accompanied by amplified oxidative stress and declined anti-oxidant status [2]. In light of the foregoing, we created a formulation and assessed its antioxidant capability and antibacterial impact using *Ixora coccinea* and *Coccinia grandis.*Lupeol and taraxerone are the active components in the hydro alcoholic extract of the coccinia grandis, which is primarily used for wound healing and has anti-inflammatory properties [3-4].

Materials and Methods:

Materials

Clove oil was extracted at Department of Pharmacognosy. The other compounds, such as liquid paraffin, white soft paraffin, and emulsifying wax, were of analytical quality and utilised without additional purification. Sandip Institute of Pharmaceutical Sciences' Department of Pharmacognosy supplied bacterial cultures of Staphylococcus aureus and Escherichia coli.

Preparation of Crude Extract:

250 ml of methanol were thoroughly used to extract the dehydrated leaves for 6 hours in a Soxhlet equipment. The extract was then dried by air after being concentrated by methanol evaporation.

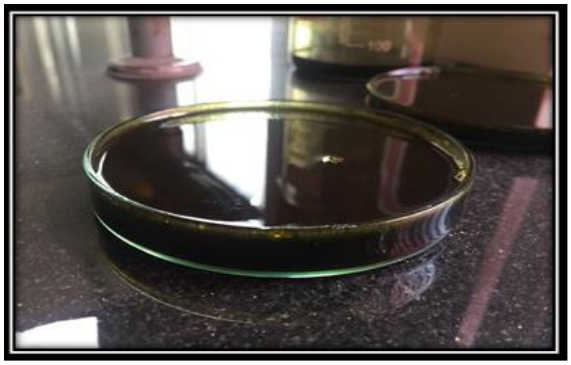

Fig No.1. Crude extract obtained

Method for preparation of ointment [5]

Combine clove oil, Ixora cocconia, and coccinia grandis extracts. With constant stirring and heat to 70–75°C, pour this mixture into the ointment base, melting it completely. Then it was stirred until the clove oil and/or Ixora coccinea were dissolved, and it was chilled to room temperature. Acacia and tragacanth should be distributed in distilled water in a beaker. With the "doubling" method, incorporate the dispersed phase into the mixture with the preservative. Table 1 lists the ingredients of the emulsifying ointment base; Table 2 lists the ingredients of various ointment formulations.

Table No.1. Emulsifying ointment base's chemical makeup

Sr. No.	Ingredients	Quantity
1	Ixora Coccinea Extract	7%
2	Hydro alcoholic extract of coccinia grandis	8%
3	Clove oil	0.7%
4	Accacia	1gm
5	Tragacanth	2gm
6	Benzoic Acid	0.5gm
7	Ointment Base	20gm

Table No.2. Composition of different ointment formulations

Item	Material name	Quantity (%)						
		ICCG 1	ICCG 2	ICCG 3	ICCG 4	ICCG 5	ICCG 6	ICCG 7
1	Clove oil	0.7	0.7	0.7	--	0.7	--	--
2	*Ixora coccinea*	5	6	4	3	7	1	2
3	*coccinia grandis*	5	4	3	6	8	2	1
4	Benzoic acid, Accacia& Tragacanth	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.
5	ointment base	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.	q.s.

Table No. 3. Phytochemical screening details

Sr. No	Phytochemical analysis of *Ixora coccinea* methanol extracts with Hydro alcoholic extract of coccinia grandis	
1.	Alkaloids	+ +
2.	Steroids	++
3.	Flavonoids	+++
4.	Tannins	++
5.	Saponins	+
6.	Glycosides	-

+: Present in low range; ++: Present in moderate range; +++: Present in high range;-: Absent

Pharmacological Evaluation:

Antibacterial activity

Escherichia coli (NCTC 10418) and Staphylococcus aureus (NCTC 6571), two bacteria that are harmful to humans, were used as in-vitro test subjects for the compounds' antibacterial activity.

a) Method: Cup-plate agar diffusion using Nutrient agar.

b) Preparation of test solutions:

Each test substance's 5 mg was mixed in 5 mL of dimethy formamide to create a stock solution with a 100 mcg/mL concentration. Following that, 0.1 mL of this solution was tested.

c) Preparation of standard solution:

Norfloxacin, a antibactial medication, was employed at concentration of 100 mcg/m.

d) Method of testing:

Testing was performed as per the ref [3-4].

Table No. 4. Inhibition zone diameters of different formulations

Sr.No.	Compd.	Zone of inhibition	
		E.coli	***S. aureus***
1	ICCG -1	30.0	29.9
2	ICCG -2	30.8	30.2
3	ICCG -3	29.5	30.5
4	ICCG -4	24.3	21.8
5	ICCG -5	33.2	34.6
6	ICCG -6	19.7	21.1
7	ICCG -7	19.4	22.0
Standard	**Norfloxacin**	**34**	**35.1**

* ZoI- Zone of inhibition. Values are average of three determinations.

Ferric reducing power assay:

Using Oyaizu's approach [8], the reducing antioxidant strength of the plant extracts was assessed. After 30 minutes, the absorbance at 700 nm was measured using a UV-Vis spectrophotometer in comparison to a blank. The reaction mixture's increased absorbance is a sign of increased reducing power. In terms of mg of gallic acid equivalents per g of plant material, the redcuing power of the plant material was calculated. [6] The ability of the formulation of IC to reduce ferric is shown in Table No. 5.

Table No.5. Ferric reducing power assay

Plant parts	Solvents	Amount (mg of gallic acid equivalents/g of plant material)
Ixora coccinea Leaves	Methanol	11.9
Fruit coccinia grandis	Hydro alcoholic	12.1

Hydrogen peroxide radical scavenging activity:

In this method, the decay or loss of hydrogen peroxide can be detected spectrophotometrically at 230 nm after a scavenger has been incubated with hydrogen peroxide. As per the aforementioned references, we carried out this task. [7-9] presented in table no. 6. The data was expressed as % inhibition. Hydrogen peroxide scavenging activity (%) = [A0– A1/ A1] × 100

Table No.6. Hydrogen peroxide radical scavenging activity

Sr.No.	Formulation	Standard (Ascorbic acid)	IC
1	ICCG -1	36.21 ± 0.23	24.12 ± 0.52
2	ICCG -2	36.21 ± 0.23	41.12 ± 0.52
3	ICCG -3	48.64 ± 0.13	28.22 ± 0.24
4	ICCG -4	46.21 ± 0.23	30.12 ± 0.52
5	ICCG -5	89.27 ± 0.12	60.31 ± 0.11
6	ICCG -6	58.64 ± 0.13	51.22 ± 0.24
7	ICCG -7	70.44 ± 0.15	45.41 ± 0.32

Conclusion:

The current investigation comes to the conclusion that Ixora coccinea and coccinia grandis formulations possess high levels of antioxidant and antibacterial properties. We also found the existence of phytochemicals such steroids, alkaloids, tannins, saponins, glycosides, and flavonoids.

Spreadability, viscosity, extrudability, antibacterial activity, and antioxidant activity were all assessed for the formulations. The findings make it abundantly clear that all formulations displayed good extrudability, viscosity, and spreadability. According to the statistics, formulation ICCG5 with clove oil (0.7%), Ixora coccinea (7%) and coccinia grandis
(8%) shown a bigger zone of inhibition than other formulations. According to the statistics, formulation ICCG5 with clove oil (0.7%), Ixora coccinea (7%) and coccinia grandis (8%) shown a bigger zone of inhibition than other formulations. Hence, ICCG5 formulation was found to have better antibacterial and antioxidant activities than other formulations. Results of all other ICCG5 evaluation parameters were all acceptable for all formulations.

References:

1. A.Elumalai, Chinna Eswaraiah, Yetcharla Venkatesh, Burle Shiva kumar and Chava Narendar Phytochemical and pharmacological profile of Ixora coccinea Linn. International Journal of Pharmacy & Life Sciences 2012, 3(3):1563-1567
2. Moni Rani Saha, Md. Ashraful Alam,Raushanara Akter and Rumana Jahangir. In-vitro free radical scavenging activity of Ixora coccinea L. Bangladesh J Pharmacol, 2008,3, 90-96.
3. Sushil D Patil, Rishikesh S Bachhav Phenolic extract of Ixoracoccinea and Hydro-alcoholic extract of Coccinia grandis fruit evaluated for antibacterial & antioxidant activity in combined formulation International Journal of Botany Studies, 2021:6(6):1390-1392
4. Sushil D.Patil Novel herbal formulation with combination of chloroform extract of lagerstroemiaspeciosa and hydro alcoholic extract coccinia grandis for treatment of mouth ulcerInternational Journal of Botany Studies, 2022, 7(5):143-145
5. G. Sandhyarani and K. Praveen Kumar Formulation and Evaluation of Neomycin Sulphate Ointment Containing Natural Wound Healing Agent Couroupita Guianensis, International Journal of Pharmacology Research 2014, 4(3):124-127.
6. S. Rohini, M. Shalini, Nithya Narayanaswamy & K. P. Balakrishnan Application of Natural Products In Cosmetics: A Study of Ixora Coccinea Extracts For Their Antityrosinase And Antioxidant Activities International Journal of Research in Cosmetic Science 2012;2 (1) :1-7
7. Manimegalai S., Sridharan T. B., Rameshpathy M. and Devi Rajeswari V. Antioxidant, phytochemical screening and antimicrobial activity of Couroupita Guianensis flower extract Der Pharmacia Lettre, 2014, 6 (6):251-256
8. Oyaizu M Studies on products of browning reaction prepared from glucoseamine. Jpn J Nutr 1986, 44,307-314.
9. Sushil D.Patil ,Laxmikant B.Borse ,Amit R.Gaikwad ,Ganesh B. Shevalkar,Hemant U. Chikhale, Evaluation Antioxidant and Antimicrobial Activity from Tablet Formulation of Chloroform Extract of Lagerstroemiaspeciosa Andhydro Alcoholic Extract Cocciniagrandis Journal of Pharmaceutical Negative Results , 2023,14(Special Issue1): 1190-1192