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Comparative Hair Growth Evaluating Activity of Marketed Formulation with Isolated Herbal Active Constituent.

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ABSTRACT:

Background: oxidative harm has been observed in various kinds of hair loss. As a polyphenolic phytoalexin Resveratrol, Anogeissus latifolia is known to be an antioxidant, Anti-inflammatory, and anti-apoptotic agent. anogeissus latifolia is known as an antioxidant, Anti-inflammatory, and anti- apoptotic agent.

Aim: Thus, we aim to examine the effects of resveratrol and Anogeissus latifolia (nanoformulation) on hair growth.

Objective: In vivo C57BL/6 mice were used to evaluate the effects of resveratrol and Anogeissus latifolia on hair cycle, hair length, skin thickness, hair follicle diameter, hair cycle score, and the percentage of hair cycle stage. Topical application of resveratrol and Anogeisus laifolia significantly promoted and stimulated the hair growth. This present experiment showed that the combination of resveratrol and Anogeissus latifolia encourages hair growth and may be a potential candidate for treating hair loss.

Keywords: Resveratrol, Anogeissus latifolia, Hair growth, Nano formulation, Antio-oxidant, Vernier caliper.

Introduction:

Resveratrol has antioxidant activity, which also reduces inflammation, and hair follicles are stimulated, this will also improve blood circulation which increases the oxygen supply to the newly growing hair. Resveratrol belongs to the resorcinol class and consists of stilbesterol and polyphenols. Resveratrol chemical name: (3,5,4'-trihydroxystilbene) which is a polyphenolic. Resveratrol can be found in the skin of grapes, dark chocolate, plants, and peanuts, blueberries, raspberries, cranberries, mulberries, pistachios, groundnuts, and purple grapes.

Anogeissus latifolia gum ghatti or Indian gum is a complex non-starch polysaccharide. Anogeissus latifolia is a plant that belongs to the combretaceace family. The bark of the Anogeissus latifolia used in mice's hair growth activity is smooth and pale to dark grey in color1. Polysaccharides, lectins, peptides, flavonoids, and tannins are the secondary metabolites of Anogeissus latifolia. Gallotannins, 3-beta-hydroxyl-28-acetyl taraxen, terpinoids,,leucocyanidine, etc..are the phytoconstituents present.

Phytoconstituents

Resveratrol (3, 5, 4 - trihydroxystilbene) is a nonflavonoid polyphenol that takes vicinity as phytoalexin. Resveratrol includes phenols or polyphenolics, phytoalexin, Trans-resveratrol, Cis- resveratrol, Vitisin A (A compound derived from resveratrol that is visible in red wines. It additionally has antioxidant properties) glycosides, Piceid (another glycosylated form of resveratrol, found in numerous plants, which contributes to the general resveratrol content material in dietary sources)[1]

Anogeissus latofolia plant contains various secondary metabolites such as Anthraquinone, Alkaloids, phenols, essential oils, tannins, flavonoids, terpenoids, saponin, xanthones, glycosides, uronic acid galactose, pentonic acid, L-arabinose and aldobionic acid on hydrolysis with sulphuric acid. These various phytochemicals are responsible for various pharmacological activities.[2]

Pharmacological activities: Anogeissus latifolia

Anogeissus latifolia is one of the crucial medicinal plants considered Ayurveda in cardiac disease. The plant is beneficial in UTI infections, pores, skin sicknesses, liver proceedings, fever, epilepsy, and many others. The plant is wealthy in pharmacologically active phenolic phytoconstituentellagic acid. It possesses recuperation capability, microbicidal activities, antiulcer capacity, hypolipidemic activities, and hepatoprotective capacity[3]. A. latifolia extract

has effective antioxidant activity. Additionally, to examine the possible purpose for the effective activity, the percentage of gallic acid became anticipated and was observed to be 0.95%, which will be one of the motives for strong antioxidant activity exhibited utilizing the plant[4]. This Anogeissus latofolia bark extract can also be used for antiulcer activity and Antibacterial activity which has already been proven. This activity is due to the presence of gallic acid (0.95%, w/w) and ellagic acid (0.25%, w/w) in the plant[5]. Due to the presence of flavonoids and phenolic compounds, it can also be used for the analgesic, Antipyretic, and anti-inflammatory activity[6].

Resveratrol

Resveratrol can be used as an antioxidant and anti-inflammatory activity in cardiovascular disease due to the presence of polyphenols in them. Resveratrol can result in primary anti-oxidant enzymes like glutathione peroxidase, heme oxygenase, and superoxide dismutase in both vascular and cardiac cells. Used in Antidiabetic and obesity effects, lipid metabolism, Anticancer activity, neurodegenerative disorders, genotoxicity effects, etc...[7] ***Alopecia:***

Hair loss, or alopecia, is a situation wherein hair in a few regions of the scalp is lost or now not regrowth.1 there are numerous forms of hair loss, along with androgenetic alopecia (AGA) and alopecia areata (AA). Hair loss is due to genetic factors and external elements specifically prominent in current society, including work pressure, emotional change, and hormone secretion disturbance. Its occurrence tends to be some of the young and it can bring about disturbed selfperception and mental struggle. in recent times, it has turned into a worldwide difficulty for all ages[8].

Material and Methods

Collection and authentication

The plant material (Anogeissus latifolia) was accrued from Srisailam hills and a specimen was dropped inside the herbarium and the leaves were authenticated by using Professor Dr.Madhavachetty, S. V. University, Tirupathi.

Preparation of extract

The leaves parts were accrued, and shadow dried after which the size decreased to small particles manually. The dried material became coarsely powdered earlier than extraction. The extracts were prepared using 2000ml soxhlet equipment non-stop and extracted with ethanol for four days. The temperature was maintained at 40-60ºC. After the extraction technique, the crude extract was collected. Resveratrol is collected from the market(resveratrol reserve) and produced by nature's answer products. It is in liquid form.

Experimental Animals:

30 healthy Wistar rats (150- 200g) were procured from ICAR- National Institute of Animal Nutrition and Physiology Adugodi, Bengaluru. The animals were acclimatized and housed in a wellventilated animal house. Laboratory conditions were maintained for 7 days previous to the experiment at a controlled temperature (22 ÃÂ± 1Ã¢ÂÂ). Relative humidity (55 ÃÂ± 10%), with 12 hours of light and dark cycle with food, water, and libitum as per the guidelines of the Committee for the Motive of Control and Supervision of Experiments on Animals (CPCSEA). Then the animals were divided into distinctive groups before initiation of the experiment. The Institutional Animals Ethics Committee (IAEC), accredited the experiment protocol employing the No. 03/1511/CCSEA 2021- 22.

Preparation of drug formulation:

At normal temperature, 0.15mg of Anogeissus latifolia extract is weighed and dissolved in 10mL of ethanol and 4ml of propylene the mixture is uniformly mixed via magnetic stirring to form an organic section. At normal temperature, 1 ml of marketed resveratrol, and 500mg of carbochol are weighed and dissolved in 10mL of water, and the mixture is uniformly mixed by magnetic stirring to form an aqueous phase. The organic phase was added into the aqueous phase with continuous stirring using a magnetic stirrer to obtain a stable and uniform emulsion with 2 drops of triethanolamine. the emulsion obtained by mixing the water phase and the oil phase shows good uniformity and stability in a state of maintaining stirring, and no obvious phase separation phenomenon occurs.

Experimental Design and Protocol

SL NO	GROUP	TREATMENT
1	CONTROL	normal saline solution 0.9% of NaCl
2	TEST	Extract of Anogeissus latifolia and resveratrol
3	STANDARD	Minoxidil Topical solution USP 5% w/v

Animals are grouped into three groups that are control, test, and standard. A total number of 18 animals were used and grouped. Each group consists of 6 animals. The Treatment period was 14 days. Wistar rats from each group were placed in separate three metabolic cages for 14 days.

Food and water are available during experimentation in cages.

Methods:

The dorsal area of 2cm into a 2cm square area is marked and shaved using a razor that can be carefully used for shaving all 18 rats' hairs. The rats were kept in an air-conditioned room. Hair growth rates were observed daily. When adequate hair lines appeared in the 18 rats, hair was cut from the hair lines and shaved dorsal areas of the rats, 20mm thick. 6 shaved rats from the control group applied the saline solution of 0.9% NaCl, 6 from the standard group applied a Minoxidil topical solution of 5%, and 6 from the test group used a combination of Anogeissus latifolia and Resveratrol extract. Growth Rate of hair was observed daily for 28 days. When adequate hair growth appeared, photographs were taken for documentation purposes.

% growth= Mean hair length(mm) or weight(gm) in week 1/ Mean hair length(mm) or weight (gm) in week 2 or 4 * 100

PERSL.N O	MEAN HAIR LENGTH	MEAN HAIR LENGTH	MEAN HAIR LENGTH	MEAN HAIR LENGTH	PERCENTAGE MEAN GROWTH%	
	IN FIRST WEEK	IN SECOND OR FOURTH WEEK	IN FIRST WEEK	IN SECOND OR FOURTH WEEK		
	(QUERCETIN)	(QUERCETI N	(MINOXIDIL)	(MINOXIDIL)		
1	1.35	1.8	1.8	2.8	SAMPL E	STANDARD
2	1.4	1.7	2.1	3.1	72%	56%
3	0.75	1.5	2.2	3.3	79%	78.50%
4	0.85	1.6	2.4	3.1	62%	62.50%
5	1.4	1.9	2.6	4.1	78%	77%
6	1.7	1.7	2.8	4.6	63.10%	62.10%
					76%	74%

Results and Conclusion:

Figure 1: hair growth in first week

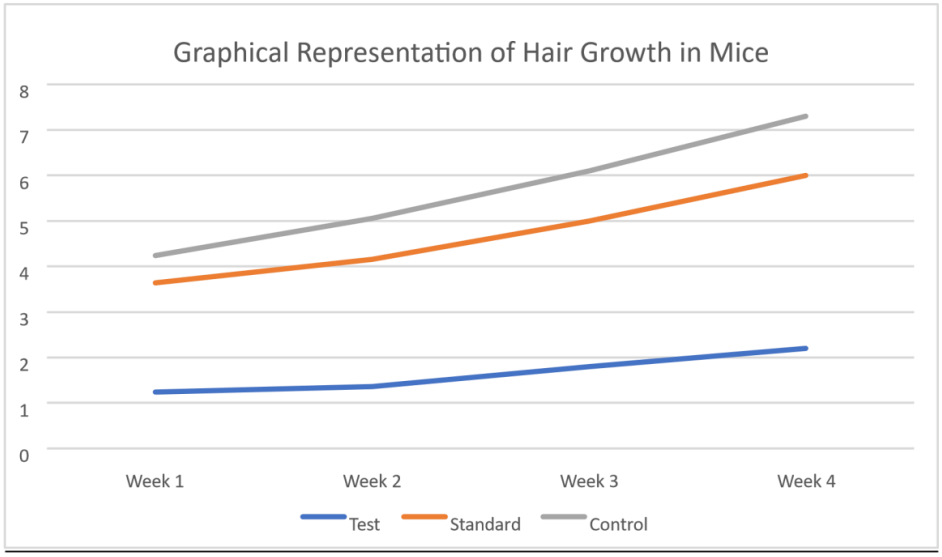
Figure 2: hair growth in last week

Among the various solutions of Anogeissus latifolia and resveratrol nanogel. The following sample was selected for the evaluation of hair regrowth activity the animals weighing in the range of 90 to 200 grams were chosen and applied with 0.5ml of a formulation consisting of 10mg/kg of active pharmaceutical ingredients and ingredients like oil phase, water phase and was applied for 28 days and was evaluated for hair regrowth which is compared with the marketed product Minoxidil 5% as a standard which came out to give a positive result. The result of percentage hair growth was calculated using a formula and the length of hair was measured with vernier caliper. Further, the histopathology study will be performed according to OECD guidelines with the permission of the animal ethical committee. For the above experiment, the nanoformulation was prepared and evaluated for the results of hair regrowth that have shown a positive result for higher hair tensile strength and length. The length of the hair gown was measured using the vernier caliper apparatus

Conclusion:

In conclusion, we found that the sample has promoting effects on hair growth and prolongs hair length. A low concentration of resveratrol can also mediate the proliferation. Hence, the sample containing Anogeissus latifolia leaf extract and resveratrol is expected to be a potential drug for the prevention and treatment of hair loss. The result results revealed that nanoformulation is a potential ingredient for hair growth stimulation.

X Axis-1 Unit-1Week
-1 Unit-0.5mm



X Axis-Duration

Y Axis

Y Axis- Length Reference

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