PREPARATION OF HERBELLO- AN HERBAL ANTIDANDRUFF SHAMPOO

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ABSTRACT
Malassezia furfur is reported as most common etiological agents causing Dandruff - a common skin condition that affects the scalp and hair. Several herbs are reported to be effective in controlling such fungi. In our laboratory an antidandruff herbal shampoo-Herbello was prepared and characterized. Certain improvements, specially with respect to viscosity, conditioning ability and antidandruff activity were needed. Therefore, aim of the present work is to improve the prepared shampoo. The active ingredients of various herbs were extracted and checked for their efficacy against Malassezia furfur. Most effective herbs were chosen and used in different combinations for preparation of shampoo in order to improve antidandruff activity. Various formulation of antidandruff herbal shampoo were tried out and best combination was chosen for stability testing. After stability test Herbello was provided to volunteers as samples and feedback obtained were analyzed.

KEY WORDS
Herbal shampoo, Lawsonia alba (Henna), Malassezia furfur, Paper disc-diffusion method, Phyllanthus emblica (Amla), Sapindus trifoliatus (Shikakai)

INTRODUCTION
Hair is an important part of overall appeal of human body, historically been associated with beauty and social distinction. Innumerable instances from all art forms can be cited supporting the special prominence accorded to hair by people of virtually all times and cultures[1]. There are many hair problems like hair loss, dandruff, unruly hair, thinning of hair, lack of hair volume, conditioning, immature graying, dullness, etc which have observed in some point of time by most of the individuals. Dandruff is reported to be a common problem in most of the population[1]. It is a harmless, chronic condition that occurs when scalp becomes dry or greasy and produces white flakes of dead skin that appear in hair or on shoulders. People most often think of it as anything that produces a flaky scalp. Although it is harmless, dandruff can be embarrassing for those who have it. It usually starts between the ages of 10 and 20 and affects up to 40% of people over the age of 30[1].

Microorganisms causing dandruff are fungus belonging to genus Malassezia. Genus Malassezia has been extensively studied and enlarged to include seven species comprising of taxa Malassezia furfur, Malassezia pachydermatis, Malassezia sympodiiali, Malassezia globosa, Malassezia obtusa, Malassezia restricta and Malassezia slooffiae[2].

Dandruff can be reduced by controlling growth of these dandruff causing organisms. Varieties of hair care products, including shampoos, are available in market and used frequently. A Shampoo may be described as a cosmetic preparation meant for washing of hair and scalp, packed in a form convenient for use. Its primary function is to clean the accumulated sebum,
scalp debris, oil, etc present on hair. The added functions of shampoo include lubrication, conditioning, hair building, prevention of static charge build up, medication etc. [3].

There are synthetic as well as herbal shampoos (containing ingredients obtained from plants) available commercially and used extensively for the same. The need and awareness for cosmetics with herbs is on rise, primarily because it is believed that these products are safe and free from side effects. However, herbal cosmetics in general and antidandruff shampoos in particular are considered to be less effective as compared to synthetic ones. There are large numbers of plants which are reported to have beneficial effects for controlling dandruff [3]. In our laboratory comparison of various commercially available shampoos was done with reference to their ability to control dandruff [4]. Based on these observations, an attempt was made to prepare an herbal antidandruff shampoo which is as effective as synthetic shampoos available commercially [5]. Prepared shampoo (Herbello) was analyzed and compared for it’s various physical, chemical and biological characteristics with commercially available shampoos. Samples were distributed to volunteers and feedback obtained suggested that improvement was needed in viscosity, conditioning and cleaning ability of the shampoo. Antidandruff activity was also found to be average and needs to be improved. However, the project was found to be viable by performing feasibility analysis, which can give an opportunity to the individual to be an entrepreneur and a job provider rather than job seeker. Therefore, the main aim of the present study is to improvise the shampoo Herbello to make it compatible with other herbal shampoos available commercially.

Objectives

- To compare the organisms isolated from the scalp with Malassezia furfur (MTCC no - 1374)
- To check effect of individual component of shampoo the on growth of dandruff causing microorganisms
- Sub-culturing and maintenance of cultures of dandruff causing microorganisms
- To select herbs which are effective against dandruff
- To standardize extraction procedure for active ingredient of herbs
- To check the effect of herb extracts against Malassezia furfur
- Preparation of shampoo using effective herb(s)
- Determining shampoo’s inhibitory effects on dandruff causing microorganisms
- Sample distribution and analysis of Feedback obtained
- Data Analysis and report writing
MATERIALS AND METHODS

Selection of Herbs

Table: I

<table>
<thead>
<tr>
<th>Scientific Names</th>
<th>Common Names</th>
<th>Uses or effects</th>
<th>Parts used</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Lawsonia alba</strong> or</td>
<td></td>
<td>Conditioning agent, provide body and bounce to hair, makes hair manageable</td>
<td>Leaves in dried powdered form.</td>
</tr>
<tr>
<td><strong>Lawsonia inermis</strong></td>
<td>Henna</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Citrus limon</strong></td>
<td>Lemon</td>
<td>Used in shampoo for antidandruff property, act as natural cleanser, used in herbal mixtures as decoction for normal to oily hair.</td>
<td>Fruit-fresh, juice form.</td>
</tr>
<tr>
<td><strong>Phyllanthus emblica.</strong></td>
<td>Amla</td>
<td>Promote hair growth, prevent premature graying and control dandruff</td>
<td>Fruit in dried powdered form.</td>
</tr>
<tr>
<td><strong>Azadirachta indica</strong></td>
<td>Neem</td>
<td>Used in shampoos as antimicrobial agent</td>
<td>Leaves in dried powdered form.</td>
</tr>
<tr>
<td><strong>Sapindus indica</strong></td>
<td>Soap nut</td>
<td>Used in shampoos, act as antidandruff agent, hair growth regulator and effective cleanser.</td>
<td>Fruit in dried powdered form.</td>
</tr>
<tr>
<td></td>
<td>(Shikakai)</td>
<td></td>
<td></td>
</tr>
<tr>
<td><strong>Sapindus trifoliatus</strong></td>
<td>Arishtak (reetha)</td>
<td>It is used in washing hair to get rid of lice.</td>
<td>Fruit in dried powdered form.</td>
</tr>
<tr>
<td><strong>Aloe vera</strong></td>
<td>Aloe</td>
<td>Used as an active ingredient in treating hair problems such as baldness, dandruff, dryness etc. It is good for hair and scalp</td>
<td>Leaf–fleshy part in gel form.</td>
</tr>
</tbody>
</table>

Preparation of Herb extract

Selected powdered herbs were weighed individually and suspended in distilled water /50% methanol / absolute ethanol (1:10, W/V). The mixture was allowed to boil up to 75% of the volume for water extract or allowed to evaporate methanol or ethanol. The residues were reconstituted in 5ml of distilled water and filtered with muslin cloth. The filtrate was centrifuged at 5000 rpm for 10-15 min and supernatant was used for further analysis.

Analysis of herb extracts was carried out in terms of their ability to inhibit growth of *Malassezia furfur* using paper disc-diffusion method.

Preparation of Shampoo

Basic ingredients, SLES (30%), Glycerine (18%), **Aloe vera** (12%) and Cocamidopropyl Betain (CAPB- 6%) were mixed one by one with constant stirring. Mixture of herbal extract (1:1:1,10%) was added prior to adjusting final volume using distilled water. To this Sodium Benzoate (0.5%) as preservative and NaCl (upto 1.5%) was added in parts with constant stirring to obtain viscous system. Tea tree oil was added in quantity sufficient for fragrance.
Procedure:

Analysis of Improvised Herbello

**Determination of antidandruff activity:**
Inhibitory effect of shampoo on *Malassezia furfur* was studied by paper disc-diffusion method.

**Paper disc-diffusion method**
To determine the inhibitory effect of various components / herbs /shampoo on *Malassezia species*, paper disc-diffusion method was carried out using dextrose peptone agar (Hi-Media, M-649) as medium (\(^8\)).

**Stability Test:**
Primary stability test for the shampoo was performed by The Kelkar Education Trust’s, Post Graduate Diploma in Perfumery and Cosmetics Management (PGDPCM) institution. Test for shampoo’s stability in pH value, viscosity in cps at 12rpm Spindle 64 of Brookefield Viscometer, Colour, Odour and separation were carried out for 15 days.

**Feedback analysis:**
Feedback form for the shampoo was designed and distributed to 50 volunteers (willing to try the formulation for couple of weeks and give unbiased feedback) and collected after a week for analyzing the acceptability of shampoo by the end users of new product.

SLES, Glycerine, Aloe vera, CAPB and Herb extract was added one by one in an wide mouth container in the required quantity with a...

Final volume was made with Distilled water and sodium benzoate was added with constant mixing.

NaCl was added in parts while mixing it simultaneously till a viscous system is...

Tea Tree oil (QS) was added.
RESULTS AND DISCUSSION

In our laboratory an anti-dandruff herbal shampoo named Herbello was prepared\(^5\) and tested for it’s efficacy against \textit{Malassezia furfur}-dandruff causing microorganisms. It’s physical and chemical properties\(^4\) were analysed before distributing the samples. Feedback analysis suggested that it’s viscosity needed to be improved, antidandruff activity was average and it’s conditioning and foam formation property was less as compared to commercial shampoos. However, the simple procedure and low cost makes it a potential project for a small scale start up in today’s competitive age. Keeping this in mind, the main aim of the present study is to address the short comings of Herbello. Earlier prepared Herbello showed antidandruff activity against \textit{Malassezia furfur} and other isolated \textit{Malassezia} species\(^6\). To confirm whether the inhibition activity was due to herbs or basic components of shampoo, their efficacy for inhibiting growth of \textit{Malassezia furfur} was studied by paper disc-diffusion method.

Plate 1 indicates ability of one of the components of shampoo (CAPB) to inhibit growth of dandruff causing microorganisms. Figure 1 indicates that to some extent the various components play a role in inhibiting the growth of organisms along with herbs. However, response of various cultures was different for each of the components studied (p< 0.0149) . This indicates that isolated organisms are members of genus \textit{Malassezia}, but belongs to different species. Keeping this in mind, all further experiments were designed using \textit{M. furfur}.

Based on the Literature survey, herbs selected in the present study include \textit{Aloe vera}, Reetha, Shikakai, Amla, Neem, Lemon and Henna. Water extracts of the selected herbs were prepared to determine their efficacy on \textit{Malassezia furfur} using paper disc-diffusion method. Figure 2 (a) represents size of inhibition zone produced by water extracts of different herbs on \textit{Malassezia furfur}. Water extracts of \textit{Aloe vera} and Shikakai did not show any inhibition. While extracting active ingredients of herbs in water, it was found that extracts were getting contaminated with fungi. Even after boiling it, contamination problem was persistent. To avoid contamination organic solvents like methanol and ethanol were used.

Plate 1 : A,B,C and D represent zone of inhibition by CAPB on \textit{Malassezia furfur}, Culture1, Culture2 and Culture3, respectively.
Size of inhibition zone obtained using methanolic extracts is represented in Figure 2 (b). Henna showed maximum zone of inhibition followed by Amla, Reetha and Neem. Whereas, like water extracts, methanolic extracts of Aloe and Shikakai did not show any effect against growth of *Malassezia furfur*, suggesting no role of Aloe and Shikakai in inhibiting growth of dandruff causing organisms. Methanolic extracts of herb showed similar results to that of water extract (p<0.1). However, for the purpose of cosmetics, ethanol is a preferred organic solvent with the characteristics similar to that of methanol. Thus for subsequent experiments ethanolic extracts were used.

There are many reports in the literature suggesting synergistic effect of various components (9). To study synergistic effect between different herbs, shampoo with various combinations of herbs were prepared and studied by paper disc-diffusion method. Henna was found to be very effective against dandruff causing organisms; therefore, Henna was added in combination with different herbs to check their synergistic effect. Figure 3 represents the effect shampoo prepared using different combination of ethanolic herb extracts on growth of *Malassezia furfur*. The most effective combination of herbs (Henna + Shikakai) was used further for the preparation of shampoo. Though, Shikakai was not showing any antidandruff activity, its combination with Heena was found to be very effective in controlling dandruff. Shikakai might have helped in removing build up of oil from hair and scalp to make the micro environment unsuitable for the growth of organisms.
Based on the results obtained by paper disc-diffusion method and feedback obtained regarding shampoo, ethanolic extracts of Henna, Shikakai and Amla worked the best and hence, were included in the final formulation. Addition of Amla was significantly improving conditioning ability of the product. Tea tree oil was added to improve the fragrance of shampoo. The shampoo thus prepared (Improved Herbello) was tested for its anti-microbial activity on *Malassezia furfur* by paper disc-diffusion method and showed excellent zone of inhibition (40.0 mm, Plate 2), therefore, it was prepared in bulk and given away as samples to many volunteer to obtain their view about quality and efficacy of the shampoo.

Plate 2 represents zone of inhibition produced by improvised Herbello with mixture of ethanolic extract of Henna, Shikakai and Amla on *Malassezia furfur*.

Feedback forms were designed and distributed along with the shampoo sample to 50 volunteers. Couple of weeks later the forms were collected back from 33 volunteers who had used the product and were ready for provide unbiased feedback. Analysis of the response is represented in Figure 4 where foam formation and cleaning ability of shampoo were rated excellent, viscosity, and fragrance of shampoo was rated very good and conditioning of shampoo was rated as good.
However, before commercializing the shampoo it is important to know the shelf life of the product. To determine stability of improvised Herbello, an analysis for 15 days was carried out at The Kelkar Education Trust’s - Post Graduate Diploma in Perfumery and Cosmetics Management Institution. The product was found to be stable in all the parameters apart from viscosity. The viscosity was found to be reducing at 45°C. Thus it was concluded that the product has less shelf life and needed to be improve with respect to stability at high temperature. However the product was found to be stable at room temperature as well as at lower temperature (4°C).

Feasibility study for the improvised product was performed and it was found to be a viable project to set up small scale industry. In the present era of ever increasing population and recent economic slowdown, getting a job is becoming increasingly difficult even for an educated person. Our society needs some of the educated people to come up as job providers rather than becoming job seekers. The small scale industry project such as the one described above does not require much of the technical qualification or facilities and therefore any graduate can easily become an entrepreneur and provide job opportunity to approximately 30-40 workers.

**FUTURE PROSPECTUS**

- Stability testing for longer period and improving self life
- Characterization and detailed component analysis of ethanolic extracts of Henna, Shikakai and Amla
- To popularize the scientifically prepared shampoo which can be used by general public
- To start a small scale business

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