The Formulation, Preparation and the Study of Antimicrobial Activity of Triphala Churna

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ABSTRACT

Triphala contain many useful human health related contents. Herbal Products of higher plants has antimicrobial mechanism of action with minimum side effects. Triphala is a herbal formulation, consisting of equal parts of fruits of Amla (Emblica officinalis), Harda (Terminalia chebula) and Baheda (Terminalia bellerica). These are demanded for digestive track, antioxidant, antiviral, antifungal and antibacterial effects. Current study was aimed to formulate, evaluate and identify antimicrobial effect of Triphala against common bacterial isolates such as S. aureus, E. coli, were inoculated on nutrient agar media and to compare the antimicrobial effect of water & ethanol based extracts of it. Wells were made on nutrient agar media plates and herbal extract, were added into it. After 72 hours of incubation at 35 °C, zone of inhibition surrounding wells were measured and recorded. We have observed antimicrobial activity of triphala against S. aureus, E. coli organisms. It was also observed that triphala is having more antimicrobial effect for ethanolic extract as compared to water extract. We had observed difference in antimicrobial effect between water and ethanol extracts of the triphala herbal preparations.

Keywords: Triphala formulation, amla, Harda, Baheda, antimicrobial effect

1. INTRODUCTION

Triphala consist three medicinal herbs (in Sanskrit, “tri” means “three” and “phala” means “fruits”). This triphala is an antioxidant-rich herbal formulation described as a Rasayana medicine by Ayurvedic practitioners.1 Triphala are demanded for digestive track, antioxidant, antiviral, antifungal and antibacterial effects. Combining the three fruits powder is said to be triphala. Aqueous and alcoholic extracts of both Triphala were used, to evaluate antimicrobial activity. The antimicrobial agents which are used to prevent or kill or inhibit the growth of microorganism such as bacteria, fungi and algae.2 This Triphala inhibits the dose-dependent growth of Gram-positive and Gram-negative bacteria.3 This Triphala contains various phenolic and nonphenolic compounds which are active against both pathogenic and non-pathogenic bacterial strains so, it has shown broad spectrum antimicrobial activity against some resistant bacterial isolates, due to its components.4 The Triphala extract was used to evaluate the microbial test against Gram-positive and Gram-negative bacteria obtained from Enterococcus faecalis, S. aureus, P. aeruginosa, K. pneumonia, E. coli, P. mirabilis, A. baumannii with Triphalahydroalcoholic extract by using Disc diffusion test.5

2. MATERIALS

2.1. Plant Material: Sample of Triphala- Amla, Behda, Hirda, Asafoetida.
2.2. Chemicals-Solvents- Distilled water, 90% ethanol,
2.3. Nutrient Agar media- agar, peptone, sodium chloride, Beef extract, Yeast extract activated carbon.
2.4. Micro-organism: Gram positive and Gram negative (E. coli, S. aureus).

3. METHODOLOGY

3.1. Formulation of Triphalachurna-

The ingredients used in the Triphala Churna are Amla (Emblica officinalis), Behda (Terminalia bellerica), Hirda (Terminalia chebula), and Asafoetida (Ferula foetida) were purchased from local market. Drugs are cleaned, dried and kept separately and powdered. They are sieved using 100-mesh sieve and each powder weighed separately as per formula and mixed. It is then kept in air tight containers in cool and dry place.6
3.2. Triphala Evaluation-

It can be evaluated by Physical parameters: It include appearance, colour, Odour, taste, pH, Bulk Density, Tap density, Angle of repose, carr’s index, Housners ratio, Moisture content, ash values, etc.

A. Bulk and Tapped density

A quantity of 10 gm of powder was introduced in to 25 ml measuring cylinder. Note the initial volume and the cylinder was allowed to fall under its own weight on to a wooden surface from the height of 2.5 cm at next intervals. Tapping was continued till no further change in volume was noted. Bulk density(BD) and Tapped density (TD) were calculated by using these equations.

\[ \text{BD} = \frac{\text{Weight of the powder}}{\text{Untapped Volume of the packing}} \]

\[ \text{TD} = \frac{\text{Weight of the powder}}{\text{Tapped Volume of the packing}} \]

B. Carr’s Index (Compressibility index)

The Carr’s Index of the powder was determined by Carr’s compressibility index as per the given formula: Carr’s Index (%) = [(TD-BD) ×100]/BD

C. Housner’s ratio

It is the ratio between the tapped density and the bulk density.

Housner’s ratio = Tapped density/Bulk density.
D. Angle of Repose

The angle of repose of churna was determined by the funnel method. The accurately weight the powder and taken in the funnel. The height of the funnel was adjusted in such a way the tip of the funnel just touched the apex of the churna powder. The diameters of the churna cone were measured and calculate the angle of repose by using the given equation.

\[ \tan \theta = \frac{h}{r} \]

Where, \( h \) and \( r \) are the height and radius of the churna cone.

E. Ash Values

Take 3 gm of the triphalachurna in a silica crucible which was previously weighed. Then the crucible was incinerated gradually by increasing the temperature until red hot and free from carbon. The crucible was cooled and reweighed, till its constant value. Determine the percentage of the total Ash value for triphalachurna.

F. Loss on drying (LOD)

Weighed 2 gm of triphalachurna, was taken in china dish. Dried in the hot air oven at 100-105°C. Cooled in a desiccator and reweighed, till its constant value. After that record the moisture content.

3.3. Triphala extraction-

A weighed quantity of powder (5 gm.) was passed into sieve no. 100 and macerate for extraction with distilled water and 90% ethanol solvents. Kept at room temperature for 72 hrs with occasional stirring. The extract was filtered and concentrated by using water bath.

3.4. Antimicrobial activity-

The Agar plate media was prepared by heating, 28 g of nutrient agar powder with 1 liter of distilled water to fully dissolve all components. Sterilized the dissolved mixture at 121°C for 15 minutes. Once the nutrient agar has been sterilized, allowed it to cool but not solidify. Then inoculated the selected microorganism in nutrient agar medium, mixed well and poured into petri plates and set aside on the sterile surface until the agar has solidified. In this cup-plate method/agar well diffusion method, after solidification, holes about 9 mm in diameter are cut in the medium with a sterile borer. The antimicrobial agents (Triphala extracts) directly placed in the holes. The zone of inhibition is observed after incubation period, at 30 to 35ºC for 2 to 3 days. Note the diameter for zone of inhibition was measured in millimeters.

RESULT AND DISCUSSION:

Formulation of Triphala churna-

The all weighted powder ingredients of Triphalachurnai.e Amla, Behda, Hirda, Asafetida are sieved using 100-mesh sieve and mixed as per formula shown in Table no 01.

Table no 01. Formulation of Triphala churna.

<table>
<thead>
<tr>
<th>Sr. No</th>
<th>Ingredients (Powder)</th>
<th>Biological source</th>
<th>Quantity taken</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Amla fruit</td>
<td>Embellicaofficinalis</td>
<td>1 part</td>
</tr>
<tr>
<td>2</td>
<td>Behda fruit</td>
<td>Terminaliabellerica</td>
<td>1 part</td>
</tr>
<tr>
<td>3</td>
<td>Hirda Fruit</td>
<td>Terminaliachebula</td>
<td>1 part</td>
</tr>
<tr>
<td>4</td>
<td>Asafetida</td>
<td>Ferula foeitida</td>
<td>Qty. sufficient</td>
</tr>
</tbody>
</table>

Triphala Evaluation-

It can be evaluated by following Physical parameters: as shown in Table no 02.

Table no 02. Evaluation of Triphala churna.

<table>
<thead>
<tr>
<th>Evaluation parameters</th>
<th>Observation</th>
</tr>
</thead>
<tbody>
<tr>
<td>Color</td>
<td>Reddish brown</td>
</tr>
<tr>
<td>Odor</td>
<td>Pungent and spicy</td>
</tr>
<tr>
<td>Taste</td>
<td>Pungent</td>
</tr>
<tr>
<td>pH</td>
<td>6.5</td>
</tr>
<tr>
<td>Bulk Density (g/ml)</td>
<td>0.57</td>
</tr>
<tr>
<td>Tapped density (g/ml)</td>
<td>0.78</td>
</tr>
<tr>
<td>Angle of repose (°)</td>
<td>32.6</td>
</tr>
<tr>
<td>Carr's index</td>
<td>28.32</td>
</tr>
</tbody>
</table>
Hausner’s ratio 0.97
Ash value (%) 4.9
Loss on drying (%) 3.96

Study of Antimicrobial Activity—The Triphala was tested for antimicrobial activity against human pathogens namely Escherichia coli, Staphylococcus aureus by cup plate method and evaluated by measuring the diameters of the zone of inhibition in mm against the test microorganisms respectively, as shown in Table no03 and following Figure no 04.

Table no 3. Antimicrobial activity of Triphala churna.

<table>
<thead>
<tr>
<th>Organisms</th>
<th>Gram positive bacteria</th>
<th>Gram negative bacteria</th>
</tr>
</thead>
<tbody>
<tr>
<td>Solvents</td>
<td></td>
<td></td>
</tr>
<tr>
<td>Water (w)</td>
<td>S. aureus</td>
<td>E. coli</td>
</tr>
<tr>
<td>Ethanol (E)</td>
<td>22mm</td>
<td>12.5mm</td>
</tr>
<tr>
<td>Ethanol (E)</td>
<td>25.5mm</td>
<td>17.5mm</td>
</tr>
</tbody>
</table>

Figure no 04: Zone of Inhibition

CONCLUSION

The above study was concluded that the Triphala have significant anti-Microbial activity. Like Triphala, other individual herbs can also be tested for their antimicrobial effects. Triphala and its individual components have significant anti-bacterial activity against S. aureus and E. coli. There is higher antimicrobial effect of ethanolic extract of Triphala than the water extract against human pathogens namely Escherichia coli, Staphylococcus aureus by cup plate method. The results was found to be satisfactory. So we can use this triphala for digestive as well as the antimicrobial purpose that means it gives the synergistic effect.

REFERENCES

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