Studies on quality assessment of ashwagandha root (Withania somnifera) powder

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Abstract
Ashwagandha root having a tremendous importance in Ayurveda and medical system. Based on these phytochemical screenings were carried out for various extracts of ashwagandha root powder. The preliminary phytochemical screening showed the presence of various phytochemicals viz., Alkaloid, Flavonoid, Tanin, Saponin, Carbohydrates and Glycosides. Various efforts have been made to evaluate functional and reconstitute properties of powder. Further ashwagandha root powder was analyzed for its physical, functional, reconstitutions and for proximate composition. Our study shows that ashwagandha root powder is great source of nutraceuticals and phytochemicals.

Keywords: functional properties, reconstitution properties phytochemical screening, proximate composition

1. Introduction
Ashwagandha is one of the most revered plants in traditional Ayurvedic medicine in India. It is an erect, greyish, subshrub with inconspicuous yellow or greenish flowers followed by small, spherical, orangish-red berries containing yellow, kidney shaped seeds. It grows three to five feet tall, mainly on waste land, but is cultivated widely as the whole plant. Most commonly the root and leaf are used medicinally (Engels and Brinckmann, 2013) [5].
Ashwagandha is a reputed health food and herbal tonic and used for cardiovascular diseases in ethno medicine. It is available for human use either as a single herb or an ingredient of polyherbal or herbomineral formulations. The human doses of Ashwagandha are generally in the range of 4-6 g/day and expected to be safe and non-toxic. Withania contains active ingredients like steroidal alkaloids and lactones known “Withanolides”. Withaferin A and withanolide D are the two main withanolides that contribute to most of the biological actions of withania (Matsuda et al., 2001; Sharma et al., 2011) [10, 14].
Survey of literature shows that most of the researches are on the medicinal, clinical properties of ashwagandha root powder but very rare information on the physicochemical properties of ashwagandha root powder. So the prime role of this investigation with objectives to evaluate functional, physical, chemical and overall other quality attributed of ashwagandha root powder.

2. Materials and Methods
2.1 Collection of Materials
The required materials for the present investigation were collected from the local markets of the Parbhani.

2.2 Preparation of Powder
The dried roots of ashwagandha root were grinded in disc mill. The obtained powder is then allowed to sieve from rotary sieve shaker containing sieves of different mesh no. viz. 30, 60 and 100.

2.3 Preparation of ethanolic, aceton and aqueous extracts of ARP (Ashwagandha Root Powder)
The powdered Ashwagandha root samples (50 g/250 mL) were extracted successively with methanol, acetone and water using soxhlet apparatus at 55-85°C for 8-10 h in order to extract the polar and non-polar compounds (Elgorashi and Staden, 2004) [14].
2.4 Proximate Composition
Ashwagandha root powder was analyzed for fat, protein, crude fibre according to AACC (2000) [1], Carbohydrate by difference method and moisture, ash as per methods of AOAC (1990) [2].

2.5 Phytochemical Screening of extracts
1. Test for Alkaloid
Wagner’s test: About ten mg of extract was taken and few drops of Wagner’s reagent (Dissolve 2 g of iodine and 6 g of KI in 100 cm³ of water) was added and the formation of a reddish brown precipitate indicates the presence of alkaloids.

2. Test for Flavonoid
Lead acetate test: Ten mg of extract was taken and few drops of 0.1% lead acetate solution was added. Appearance of yellow colour precipitate indicates the presence of flavonoids.

3. Test for Tannin
Ferric Chloride Test: To 5 ml of the sample, a few drops of 0.1% ferric chloride were added. The presence of a brownish green or blue black colour indicated that the material possessed tannins.

4. Test for Saponin
Foam test: 0.5 mg of extract was diluted with 20 ml distilled water and shaken well in a graduated cylinder for 15 min. The formation of foam to a length of 1 cm indicated the presence of saponins.

5. Test for Carbohydrates
Fehling’s test: Five ml of Fehling’s solution was added to 0.5 mg of extract and boiled in a water bath. The formation of yellow or red precipitate indicates the presence of reducing power.

6. Test for Glycosides
Glycoside test: 0.5 mg of extract was dissolved in 1 ml of water and then aqueous NaOH solution was added. Formation of yellow colour indicates the presence of glycosides.

2.6 Physical, Functional and Reconstitutional Properties of Ashwagandha Root Powder
2.6.1 Physical Properties
1. True Density
True density was calculated by filling the approximate 1 g of ground sample in a burette containing toluene. Then raised in toluene level was measured and an average of two reading of true density was calculated as,

\[ \text{True Density (g/ml)} = \frac{\text{Weight of ground sample}}{\text{Rise in toluene level}} \]

2. Bulk Density
Sample was kept in 100 ml cylinder and tapped for 25 to 30 times to allow uniform compacting of grain, then recorded the volume and weighed the sample in the cylinder. Bulk density was calculated by,

\[ \text{Bulk Density (g/ml)} = \frac{\text{Weight of sample}}{\text{Volume of sample}} \]

3. Angle of Repose
It was determined by heap method reported by Martin et al. (1991) [9]. Briefly, the powder was poured through a glass funnel from a definite distance to the smooth horizontal surface until a heap of maximum height was formed in a conical form. The diameter and the height of the heap were determined and the tangent of the angle was determined by following expression,

\[ \text{Angle of Repose (°)} = \tan^{-1}\left(\frac{h}{r}\right) \]

Where, h - the height of heap and 
\( r \) - the radius of heap made by powder

4. Carr Index
From Carr (1965) [3], Carr Index (Carr I) was calculated as the difference between the tapped and bulk densities divided by the tapped density as shown below,

\[ \text{Carr Index (%)} = \frac{\text{True Density} - \text{Bulk Density}}{\text{True Density}} \times 100 \]

5. Hausner ratio
The Hausner ratio (HR) from (Hausner, 1967) [6] was calculated as tapped density divided by bulk density,

\[ \text{Hausner ratio (HR)} = \frac{\text{True Density}}{\text{Bulk Density}} \]

2.6.2 Functional Properties
1. Water Holding Capacity
Water Holding Capacity was determined using the method of Sowbhagya et al. (2007) [15].

2. Water Absorption Capacity (WAC)
Water Absorption Capacity was determined using the method of Sowbhagya et al. (2007) [15].

3. Oil Absorption Capacity (OAC)
Oil Absorption Capacity was determined as outlined by Sangnark and Noomhorm (2004) [13]

4. Swelling Capacity
Swelling Capacity was measured as described by Sowbhagya et al. (2007) [15].

2.6.3 Reconstitution Properties
1. Solubility
One gram of powder was mixed with 100 ml distilled water and blended in a hand blender. The solution was transferred to 50 ml centrifuge tubes and centrifuged at 3000 rpm for 5 min. It was allowed to settle for 30 min and 25 ml of the supernatant was transferred to pre-weighed petri plates which was oven dried at 105°C for 5 h. The solubility (%) was calculated as the weight difference.

2. Dispersibility
Distilled water (10 mL at 25°C) was taken in a 50 ml beaker and 1 g sample was added. The sample was stirred vigorously for 15 s making 25 complete movements back and forth across the whole diameter of the beaker. The reconstituted powder was poured through a 212 μm sieve into a pre-weighed aluminium pan. The pan with sieved and sample was dried at 105°C temperature for 4 h. The dispersibility was calculated according to the formula given by Jinapong et al. (2008) [7].
Dispersibility (%) = \frac{(10 + a) \times \%TS}{a \times (10 - b) / 100}

Where, \( a \) - amount of powder (g) taken
\( b \) - moisture content in the powder and
\%TS - dry matter in percentage in the reconstituted powder after it has been passed through the sieve

3. Wettability
Wettability of sample was evaluated according to the method described by Jinapong surface to completely submerge in 400 mL of distilled water at 25°C temperature.

3. Results and Discussion
3.1 Physical properties of Ashwagandha root Powder

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Parameters</th>
<th>Mean value</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>True Density (g/ml)</td>
<td>0.55</td>
</tr>
<tr>
<td>2</td>
<td>Bulk Density(g/ml)</td>
<td>0.41</td>
</tr>
<tr>
<td>3</td>
<td>Angle of Repose (º)</td>
<td>27.80</td>
</tr>
<tr>
<td>4</td>
<td>Carr Index (%)</td>
<td>25.45</td>
</tr>
<tr>
<td>5</td>
<td>Hausner Ratio</td>
<td>1.34</td>
</tr>
<tr>
<td>6</td>
<td>Colour</td>
<td></td>
</tr>
<tr>
<td></td>
<td>L*</td>
<td>65.99</td>
</tr>
<tr>
<td></td>
<td>a*</td>
<td>3.07</td>
</tr>
<tr>
<td></td>
<td>b*</td>
<td>15.85</td>
</tr>
</tbody>
</table>

*Each values is average of three determination

3.2 Functional Properties of Ashwagandha root powder

True density is the density of the solid material excluding the volume of any open and closed pores. The true density of powders often differs from that of the bulk material because the process of comminution or grinding will change the crystal structure near the surface of each particle and therefore the density of each particle in a powder. The true density of ashwagandha root powder was found to be 0.55 g/ml.

Angle of repose of ARP was 27.80. Values for angles of repose ≤ 30º generally indicate a free flowing material and angles ≥ 40º suggest a poorly flowing material (Yüksel et al., 2007) [11].

The compressibility indices (Carr’s index and Hausner ratio) provide the flow properties and compressibility of powders. Carr’s index (CI) and Hausner’s ratio (HR) (a measure of the interparticulate friction) are useful tools in the development of new formulation. The carr (%) and Hausner ratio was 25.45, 1.34 respectively. Lower Carr Index or lower Hausner ratios of a material indicates better flow properties than higher ones. A Carr’s CI of <15% or HR of <1.11 is considered ‘excellent’ flow whereas CI>38% or HR>1.60 is considered ‘very very poor’ flow Carr, 1965 and Hausner, 1967 [1, 6]. Good flow of powder helps to avoid the extensive costs and time involved in unloading powders that will not flow out of storage containers. As well as helps to achieve the best formulation and improve the quality and consistency of the product.

3.3 Reconstitution properties of ashwagandha root powder

For the consumers, food powder dissolution will have a direct impact on their perception of the overall product quality. Various efforts have been made to develop methods quantifying the reconstitution properties of food powder. Reconstitution properties of powder have various impacts on the quality and its overall acceptability among the consumers. In view of that various efforts have been made to develop methods of quantifying reconstitution properties of ashwagandha root powder.

Many factors affect the water solubility of powdered products, including processing conditions, composition, particle size, density, pH and storage conditions (Mirhosseini and Amid, 2013) [11]. The per cent solubility of ashwagandha root powder was found to be 78%.

Kim and Bhowmik (1990) [8] stated that the wettability and dispersability depend on the particle size, density, porosity, surface charge and surface area, the presence of amphiphatic substances and surface activity of the particles. Is was found that ashwagandha root powder wet within 175 sec. and having dispersability of 16.40%

3.4 Qualitative screening of Phytochemical Constituents of ashwagandha root Powder

Preliminary qualitative phytocheical analysis was carried out to identify the secondary metabolites present in the ashwagandha root powder extracts of acetone, etahanol and aqueous. The following results had been made.

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Phytochemical Constituents</th>
<th>Ethanol</th>
<th>Acetone</th>
<th>Aqueous</th>
<th>Name of the Test</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Alkaloid</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>Wagner’s Test</td>
</tr>
<tr>
<td>2</td>
<td>Flavonoid</td>
<td>+</td>
<td>+</td>
<td>-</td>
<td>Lead acetate test</td>
</tr>
<tr>
<td>3</td>
<td>Tannin</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>Ferric Chloride Test</td>
</tr>
<tr>
<td>4</td>
<td>Saponin</td>
<td>+</td>
<td>-</td>
<td>+</td>
<td>Foam test</td>
</tr>
<tr>
<td>6</td>
<td>Carbohydrate</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>Fehling’s test</td>
</tr>
<tr>
<td>7</td>
<td>Glycoside</td>
<td>+</td>
<td>+</td>
<td>+</td>
<td>Glycoside test</td>
</tr>
</tbody>
</table>

Where, += present and - = absent
From the phytochemical tests it was observed that various extracts viz., ethanolic, acetone and aqueous extracts showed the presence and absence of phytochemical constituents. Ethanol extract found to contain presence of alkaloid, flavonoids, tannins, saponins, carbohydrates, glycosides. Acetone also possesses all those phytochemical constituents excepting saponins. Aqueous extract contains alkaloid, tannins, saponins, carbohydrates. Presence of such phytochemical constituents of crude powder extracts are of great medicinal values.

The similar findings were detected by Viswesvarai et al., (2013) who reported that aqueous extracts of ashwagandha root powder contains alkaloid, tannins, saponins, carbohydrates and glycosides.

Each of these phytochemicals is known for various protective and therapeutic effects. For instance, phenol is known to be an erythrocyte membrane modifier. Alkaloids protect against chronic diseases; saponins protect against hypercholesterolemia.

3.5. Proximate Composition of ashwagandha root powder

<table>
<thead>
<tr>
<th>Sr. No.</th>
<th>Proximate Constituents</th>
<th>Values (%)</th>
</tr>
</thead>
<tbody>
<tr>
<td>1</td>
<td>Moisture</td>
<td>7.2</td>
</tr>
<tr>
<td>2</td>
<td>Ash</td>
<td>4.2</td>
</tr>
<tr>
<td>3</td>
<td>Fat</td>
<td>0.9</td>
</tr>
<tr>
<td>4</td>
<td>Protein</td>
<td>3.3</td>
</tr>
<tr>
<td>5</td>
<td>Carbohydrate</td>
<td>51.1</td>
</tr>
<tr>
<td>6</td>
<td>Crude Fibre</td>
<td>33.3</td>
</tr>
</tbody>
</table>

*Each values is average of three determination

From the table of proximate evaluation it was revealed that ashwagandha root powder had nutrient composition viz., moisture 7.2%, ash 4.2%, fat 0.9%, 51% carbohydrates, 3.3% protein and 33% crude fibre. From this it was revealed that ashwagandha root powder is rich source of carbohydrates followed by crude fibre.

4. Conclusion
It can be concluded from the study that root powders of ashwagandha is rich source of various secondary metabolites that helps to combat from the various diseases. Different extracts of ashwagandha root powders contains many bioactive compounds including alkaloid, flavonoids, glycosides, saponins and tannins. Further investigation reveals the ashwagandha root powder possesses good functional, reconstitution and chemical properties which aids in food processing. The present study can be used for the formulation of these bio ingredients in the food products against various diseases.

5. References