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## Comparison of gravimetric method with the colorimetric method for saponin content in safed musli [*Chlorophytum borivilianum*]

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**Abstract**

Safed musli (*Chlorophytum borivilianum*) is a rare herb from India used in traditional systems of medicine including Ayurveda and Unani. It is generally used for diabetes, arthritis, cancer, improving sexual performance, boosting vitality and many others. Safed Musli is the best known source of saponins. Saponins have many health benefits. Saponins are glucosides with foaming characteristics. Sugars are combined with sapogenins to form the saponins and it is used in the commercial preparation of steroidal hormones. The dry powder of safed musli root was taken for analysis of saponin content. The saponin content was analyzed by two different methods - Gravimetric method and method developed by National Research Centre on Medicinal and Aromatic Plants, Boriavi, Anand i.e. Spectrophotometric method. The result showed that saponin content was on slightly higher side when estimated by spectrophotometric method.

**Keywords:** Safed musli (*Chlorophytum borivilianum*), saponin, gravimetric method, spectrophotometric method

**Introduction**

There are seventeen species of *Chlorophytum* present in India [1]. Among all of them, *C. borivilianum* produces the highest yield of roots and has the highest saponin content [2]. *Chlorophytum Borivilianum* (Safed Musli) is a rare herb used in Ayurveda. It was usually procured in jungles but due to its medicinal quality, it has since begun to be cultivated in farm. In India, *C. borivilianum* is generally found in Rajasthan, Gujarat, and some region of Madhya Pradesh [3]. It is also known as “White Gold” or “Herbal Viagra” [4]. The roots of *Chlorophytum borivilianum* are used as tonic and important ingredient of 20 ayurvedic and unani preparations [5]. Safed Musli is an aphrodisiac [6] as well as a tonic for diabetes, inflammation, and also used to increase body immunity [7]. It contains high amount of saponin and polysaccharide, and the water extract appears to be the most active extraction. The overall composition of the roots consists of saponins (17–20%), carbohydrates (42%), proteins (5–10%) and fiber (3–4%) [4]. Although saponin contents as low as 13.58% have been noted [8]. Rhizomes are often short and inconspicuous while roots are usually thicker or slightly fleshy.

Saponins are a group of naturally occurring plant glycosides. They are characterized by their strong foam forming properties in aqueous solution. Due to the great variability of their structures, saponins always show anti-tumorigenic effects through many antitumor pathways [9]. The presence of saponins has been reported in more than 100 families of plants out of which at least 90 kinds of natural saponins have been found to possess significant anti-cancer properties. There are more than 11 different classes of saponins including oleananes, taraxasteranes, dammaranes, tirucallanes, lupanes, lanostanes, cucurbitanes, hopanes, ursanes, cycloartanes and steroids [10]. Chemically all these can be classified as triterpenoid and steroidal glycosides. Saponins consist of non-polar aglycones (triterpene or steroid) and one or more glycone (monosaccharide) moieties [11]. When saponins are hydrolyzed they yield an aglycone or non-saccharide portion (sapogenin). Saponins have diverse properties like molluscicidal activity [12] antimicrobial and insecticidal activities [13] and reduce the harmful LDL-cholesterol [14]. Saponins are excellent foaming agents and emulsifiers because of the presence of both hydrophobic and hydrophilic moieties and this property of the saponins helps in lowering the serum cholesterol in human body [15].

At present the saponin content of fasciculated roots of Safed musli is being analysed by the gravimetric method given by Birk *et al.* (1963) [16]. A new spectrophotometric method has been developed by National Research Centre on Medicinal and Aromatic Plants, Boriavi Anand, Gujrat.

### Materials and Methods

Thirty samples of fasciculated roots of Safed musli were analysed for its saponin content by gravimetric as well as by colourimetric methods. Samples of Safed musli were taken from the fields of Nagarjun Medicinal Plants Garden, Dr PDKV, Akola (MS).

The procedures are detailed as under:

#### Method of estimation of saponins by gravimetric method:

Safed musli dry root powder 5.0 g was taken in soxhlet thimble. It was extracted with 90% V/V Ethanol by refluxing for half an hour. The process of refluxing was repeated three times and distilled off the solvent. The soft extract left over after distillation of ethanol, was treated with Petroleum ether (60-80<sup>o</sup> C) by refluxing for half an hour. The content was cooled and removed off the solvent by decantation. The same soft extract was treated similarly with Chloroform and Ethyl acetate. The solvents was poured off after cooling, keeping the soft extract in the same flask. The soft extract (after three extractions cited above) was dissolved in 25 ml methanol. This methanolic extract was filtered and concentrated to 5 ml. The methanolic extract was added drop by drop with constant stirring to 25 ml Acetone to precipitate saponins. The ppt is filtered, collected and dried to a constant weight at 105 °C. The percent saponin in given sample was calculated by given formula:

$$\text{Total Saponins (\%)} = \frac{\text{Weight of residue} \times 100}{\text{Weight of sample taken}}$$

#### Method of estimation of saponins by Spectrophotometric method [18]:

Safed musli dry root powder 1.0 g was taken and extracted thrice in (100 ml each) 85% ethanol. Partitioned with *n*-butanol and evaporated to dryness. The residue was dissolved in 100 ml distilled water and diluted 10 times as working standards. Aliquot of 0.2, 0.4, 0.6, 0.8 and 1.0 ml standard saponins solution was used for calibration. To this solution 5 ml freshly prepared vanillin reagent under ice cold condition was added. The contents were mixed in vortex and kept at 60 °C ± 1 °C on water bath for one hour. The reaction was stopped by changing the tubes to ice box for 10 minutes. The absorbance was taken at 473 nm against blank and readings were noted. Samples of safed musli root extracts were analysed as per standard calibration curve by appropriate dilution.

### Results and Discussion

The results are presented in table-1. It has been well established that the values obtained by colorimetric methods are most of the times higher than values obtained by gravimetric method [17]. The saponin content estimated by gravimetric method was in the range of 5.3-6.9% with the mean value of 6.06%. The saponin content was on slightly higher side when estimated by spectrophotometric method with the mean value 6.34%. The spectrophotometric method was less time consuming and accurate (Standard deviation ± 0.40).

**Table 1:** Saponin content in fasciculated roots of Safed musli estimated by gravimetric and spectrophotometric methods.

Saponin content (%)		
Sample Nos.	Gravimetric method	Spectrophotometric method
1	6.2	6.5
2	5.8	6.1
3	6.6	6.8
4	5.4	5.8
5	6.2	6.4
6	5.8	6.1
7	6.8	6.8
8	6.7	6.9
9	6.3	6.6
10	5.6	6.1
11	5.8	6.2
12	6.5	6.6
13	5.6	6.2
14	6.3	6.5
15	5.7	5.9
16	5.3	5.6
17	6.4	6.5
18	6.3	6.8
19	6.9	6.9
20	5.7	5.8
21	5.3	5.6
22	5.7	6.0
23	5.8	5.9
24	6.5	6.8
25	6.2	6.3
26	5.8	6.2
27	6.2	6.4
28	6.9	7.1
29	5.4	6.6
30	6.2	6.4
Mean	6.06	6.34
SD ±	0.48	0.40
CV %	8.15	6.49

### Conclusion

Compared to gravimetric method for the estimation of saponin in safed musli, the spectrophotometric method was found to be accurate, less time consuming, sensitive and reproducible.

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