DEVELOPMENT OF DRYING PROTOCOL FOR WITHANIA SOMNIFERA ROOTS

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INTRODUCTION

Ashwagandha (Withania somnifera), also known as Indian ginseng, is an important ancient plant, and the roots of which have been employed in Indian traditional systems of Medicine, Ayurveda, and Unani. Ashwagandha belongs to family Solanaceae, and it is a green shrub [1] found throughout the drier parts of India, Baluchistan, Pakistan, Afghanistan, Sri Lanka, Congo, South Africa, Egypt, Morocco, and Jordan. In India, it is widely grown in the provinces of Madhya Pradesh, Uttar Pradesh, plains of Punjab, and northwestern parts of India such as Gujarat and Rajasthan [2].

Many pharmacological studies have been carried out to describe multiple biological properties of W. somnifera. These studies have shown that the plant preparation has antitumor, immunomodulatory, antispasmodic, tuberculosiis, rheumatism, diuretic, antibiotic, adaptogenic, antioxidant activities, digestive disorders, and common cold [3].

 Constituents

It is cultivated specifically for its root, which has medicinal and commercial value due to the presence of the steroidal chemicals, withanine and somniferine; therefore, it is considered the equivalent of Chinese ginseng (Kumar et al. 2008). The major constituents of W. somnifera are steroidal alkaloids and lactones, a class of constituents together known as withanolides (steroidal lactones with ergostane skeleton) [4]. So far 12 alkaloids, 35 withanolides, and several sitoindosides have been isolated, and their structures have been elucidated [5]. The various alkaloids include withanine, somniferine, somnine, somnifermine, withanine, pseudo-withanine, tropine, pseudo-tropine, 3-α-gloyloxytropane, choline, cuscohygrine, isopelletirine, anafoline, and anahydrine [6].

It is all about the plant and the present article discusses the preservation technique/drying technique for roots of W. somnifera.

METHODS

An experiment was conducted to standardize the postharvest technology drying for W. somnifera during the year 2009–2010 at National Medicinal Plants Board Project, in B.R. Nahata College of Pharmacy, Mandsaur, M.P. The drying studies were conducted using three different methods: Sun drying, shade drying, and hot air oven drying. Fresh ashwagandha roots were brought from the B.R. Nahata College of Pharmacy, Mandsaur (M.P.). The initial moisture content (60.4%) of the Ashwagandha roots were brought from the B.R. Nahata College of Pharmacy, Mandsaur (M.P.). The initial moisture content (60.4%) of the Ashwagandha roots was obtained that is mentioned in literature (10–5%).

Result: The result revealed that hot air oven drying of roots took the lowest time (12 h), while sun drying method took 24 h for drying. The alkaloid content, namely, Withanolide A was significantly influenced by different methods of drying. The highest alkaloid content (0.010%) was observed in sun drying and hot air oven drying with 0.02 Lab%6 as compared to shade drying with 0.009% Withanolide A.

Conclusion: It may be concluded that sun drying and hot air oven drying are suitable methods for drying W. somnifera roots.

Keywords: Withania somnifera, Shade drying, Moisture content, Withanolide A.

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Drying

Drying is an essential process in the preservation of agricultural products. Drying consists of the removal of sufficient moisture content of crude drug so as to improve its quality and make it resistant to the growth of microorganisms. When medicinal plant materials are prepared for use in dry form, the moisture content of the material should be kept as low as possible to reduce damage from mold and other microbial infestation. Information on the appropriate moisture content for particular medicinal plant materials may be available from pharmacopeias or other authoritative monographs. A definition of drying that shall sharply differentiate it from evaporation is difficult to formulate. The term drying usually infers the removal of relatively small amounts of water from solid or nearly solid material, and the term evaporation is limited to the removal of relatively large amounts of water from solutions [4]. Drying inhibits partially enzymatic reactions. Drying facilitates pulverizing or grinding of a crude drug [7].
kept open to the sun from 8.00 am to 5.00 pm. Moreover, the moisture content of the dried material was taken every day using IR moisture balance. The temperature during sun drying ranged from 32°C to 42°C and during the night hours, and the samples were kept in plastic covers to prevent reabsorption of moisture. Likewise in mechanical/hot air oven drying method, the root pieces (2–4 inches) were spread uniformly in the hot air oven at temperature 50°C, 60°C, and 70°C, respectively. Moreover, in shade drying method, the root pieces (2–4 inches) were spread in a plastic tray and kept in shade, well-ventilated place. The temperature during shade drying ranged from 25.5°C to 37°C and during the night hours, and samples were kept in plastic covers to prevent reabsorption of moisture. The samples were weighed at regular intervals until a moisture content of nearly 6–5% was obtained. Table 1 shows that 6 days’ sun drying is sufficient for the roots of ashwagandha. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6% [10].

Table 2 shows that 15 days’ shade drying is sufficient for roots of ashwagandha. The moisture content limit for dried ashwagandha root is 6% or below 6%. Moreover, the experimental moisture content for dried roots of ashwagandha was 5.8% recorded.

Table 3 showing that when roots were kept in hot air oven for 12 h at 50°C and 60°C was sufficient for rapid drying of roots of Ashwagandha because it attained 6.2% and 5.8% moisture content at this time and temperature. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6%. Table 4 showing that hot air oven for 12 h at 50°C is sufficient for rapid drying of roots of Ashwagandha because it attains 6.0% moisture content at this time and temperature. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6%.

Table 5 shows that hot air oven drying for 12 h at 60°C is sufficient for rapid drying of roots of Ashwagandha because it attains 5.8% moisture content at this time and temperature. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6%.

Table 1: Sun drying of ashwagandha roots

| S. No. | Drying time (days) | Temperature (°C) | RH (%) | Weight of roots (g) | Moisture content (%) |
|--------|--------------------|------------------|--------|---------------------|---------------------|
| 1.     | 0                  | 32.5             | 67     | 2000                | 60.4                |
| 2.     | 1                  | 34.0–39.5        | 43–25  | 1500                | 50.4                |
| 3.     | 2                  | 39.5–40.0        | 30–26  | 900.0               | 30.8                |
| 4.     | 3                  | 36.0–39.5        | 43–25  | 800.5               | 24.4                |
| 5.     | 4                  | 40.0–40.5        | 40–34  | 700.0               | 17.2                |
| 6.     | 5                  | 38.5–39.0        | 34–25  | 600.5               | 10.4                |
| 7.     | 6                  | 38.5–40.0        | 38–26  | 500.0               | 5.8                 |

Table 2: Shade drying of ashwagandha roots

| S. No. | Drying time (days) | Temperature (°C) | RH (%) | Weight of dried roots of WS (g) | Moisture content (%) |
|--------|--------------------|------------------|--------|---------------------------------|---------------------|
| 1.     | 0                  | 36.5             | 67     | 2000.0                          | 60.4                |
| 2.     | 1                  | 36.0–34.0        | 43–38  | 1700.0                          | 55.0                |
| 3.     | 2                  | 37.0–36.0        | 37–34  | 1500.0                          | 50.4                |
| 4.     | 3                  | 36.5–34.0        | 46–38  | 1370.0                          | 43.8                |
| 5.     | 4                  | 37.5–36.0        | 49–37  | 1250.0                          | 39.4                |
| 6.     | 5                  | 36.5–35.5        | 43–38  | 1130.0                          | 35.8                |
| 7.     | 6                  | 37.5–36.0        | 40–35  | 1050.0                          | 32.4                |
| 8.     | 7                  | 37.0–36.5        | 39–30  | 985.0                           | 28.2                |
| 9.     | 8                  | 36.0–34.5        | 35–30  | 947.0                           | 22.6                |
| 10.    | 9                  | 37.5–36.0        | 36–34  | 939.0                           | 20.2                |
| 11.    | 10                 | 36.5–37.0        | 34–30  | 923.0                           | 16.2                |
| 12.    | 11                 | 36.0–37.5        | 40–36  | 905.0                           | 11.6                |
| 13.    | 12                 | 36.5–37.0        | 43–38  | 892.0                           | 10.0                |
| 14.    | 13                 | 34.5–36.0        | 57–37  | 885.0                           | 9.8                 |
| 15.    | 14                 | 35.5–37.0        | 46–38  | 860.0                           | 8.6                 |
| 16.    | 15                 | 36.0–37.5        | 45–35  | 545.0                           | 5.8                 |

Table 3: Hot air oven drying of ashwagandha roots at constant time (12 h)

| S. No. | Drying time (h) | Temperature (°C) | RH (%) | Weight of dried roots of WS (g) | Moisture content (%) |
|--------|-----------------|------------------|--------|---------------------------------|---------------------|
| 1.     | Fresh           | 37               | 40     | 1000.0                          | 60.4                |
| 2.     | 6               | 50               | 37     | 480.5                           | 6.2                 |
| 3.     | 12              | 50               | 21     | 430.6                           | 5.8                 |
| 4.     | 14              | 50               | 41     | 410.0                           | 5.2                 |

Drying time: Was kept constant (12 h). Temperatures: 50°C, 60°C, and 70°C of hot air oven, RH%: In hot air oven

Table 4: Hot air oven drying at constant temperature (50°C)

| S. No. | Drying time (h) | Temperature (°C) | RH (%) | Weight of dried roots of WS (g) | Moisture content (%) |
|--------|-----------------|------------------|--------|---------------------------------|---------------------|
| 1.     | Fresh           | 37               | 58     | 1000.0                          | 60.4                |
| 2.     | 6               | 50               | 78     | 700.5                           | 19.4                |
| 3.     | 8               | 50               | 70     | 580.0                           | 14.2                |
| 4.     | 10              | 50               | 58     | 480.5                           | 6.8                 |
| 5.     | 12              | 50               | 50     | 460.0                           | 6.0                 |
| 6.     | 14              | 50               | 47     | 455.6                           | 5.2                 |
showing that in hot air oven 10 h at 70°C is sufficient for rapid drying of roots of Ashwagandha because it was attaining 5.2% moisture content at that time and temperature. And moisture content limit for dried Ashwagandha root is 6% or below 6% [10].

Chemo profiling of dried roots of Ashwagandha

High-performance thin-layer chromatography (HPTLC) was performed for checking the chemical breakdown of ashwagandha roots during drying in which roots were dried by three different methods of drying. HPTLC method used as an analytical tool on the basis, of which the best method of drying for Ashwagandha roots was selected.

Specifications

CAMAG TLC scanner, stationary phase E. MERCK KGaA HPTLC plates silica gel 60F254; dimensions 20.0 cm x 10.0 cm; drying device Oven temperature 120°C for 20 min (for plate); calibration mode was single level, and evaluation mode was peak area. Sample applicator was CAMAG Automatic TLC Sampler 4. The number of tracks was 14 in the form of bands (in which number of standard levels 1 and number of samples 13), band length was 8.0 mm, and the distance between tracks was 12.3 mm. Sample volume for application was 5.0 µl for pure standard and 10 µl for samples.

Development of TLC plates was done in Twin Trough Chamber 20 cm x 10 cm and saturated for 30 min with filter paper. The mobile phase was chloroform: methanol; drying device CAMAG TLC Plate Heater III temperature 60°C for 5 min.

Derivatization post-chromatographic derivatization was done by Vanillin-Sulfuric acid reagent at CAMAG TLC plate heater III; temperature 120°C for 20 min.

RESULTS AND DISCUSSION

Chromatographic studies were done with HPTLC to reveal the concentration of withanolide A, and the results suggest that fresh sample of ashwagandha root extracts contains the highest concentration of Withanolide A as compared to shade drying, sun drying, and hot air oven drying, and in these three drying methods, sun drying is much better than shade drying. However, hot air oven drying at 50°C for 12 h shows the highest yield of Withanolide A than other two (sun and shade) drying methods. Therefore, hot air drying may be the best method of drying for ashwagandha roots.

These results show that 6 days' sun drying is sufficient for roots of ashwagandha. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6% [10].

These results show that 15 days' shade drying is sufficient for the roots of ashwagandha. The moisture content limit for dried ashwagandha root is 6% or below 6%. Moreover, the experimental moisture content for dried roots of Ashwagandha was 5.8% recorded.

| S. No. | Drying Time (h) | Temperature (°C) | RH (%) | Weight of dried roots of WS (g) | Moisture content (%) |
|-------|----------------|------------------|--------|--------------------------|---------------------|
| 1.    | Fresh          | 37               | 58     | 1000.0                   | 60.4                |
| 2.    | 6              | 60               | 76     | 680.7                    | 17.4                |
| 3.    | 8              | 60               | 75     | 560.8                    | 12.2                |
| 4.    | 10             | 60               | 50     | 450.0                    | 6.4                 |
| 5.    | 12             | 60               | 48     | 440.0                    | 5.8                 |
| 6.    | 14             | 60               | 45     | 435.0                    | 4.4                 |

Table 6: Hot air oven drying at constant temperature (70°C)

| S. No. | Drying Time (h) | Temperature (°C) | RH (%) | Weight of dried roots of WS (g) | Moisture content (%) |
|-------|----------------|------------------|--------|--------------------------|---------------------|
| 1.    | Fresh          | 37               | 58     | 1000.0                   | 60.4                |
| 2.    | 6              | 70               | 76     | 640.7                    | 15.4                |
| 3.    | 8              | 70               | 67     | 516.8                    | 13.0                |
| 4.    | 10             | 70               | 52     | 410.0                    | 5.8                 |
| 5.    | 12             | 70               | 46     | 400.0                    | 5.2                 |
| 6.    | 14             | 70               | 43     | 398.0                    | 4.0                 |
Fig. 3: Withanolide A conc. In sample 1

Fig. 4: Withanolide A conc. In sample 2

Fig. 5: Withanolide A conc. In sample 3

Fig. 6: Withanolide A conc. In sample 4

Fig. 7: Withanolide A conc. In sample 5

Fig. 8: Withanolide A conc. In standard 1
Table 7: Comparison between standard and samples for the concentration of Withanolide A (Result through area)

| S. No. | Sample ID | Sample name                          | Area     | Withanolide A (%) |
|--------|-----------|--------------------------------------|----------|-------------------|
| 1.     | Sample 1  | Fresh root extract of WS             | 1751.72  | 0.03              |
| 2.     | Sample 2  | Shade-dried root extract of WS       | 1556.70  | 0.009             |
| 3.     | Sample 3  | Sun dried root extract of WS         | 1305.60  | 0.010             |
| 4.     | Sample 4  | 10 h hot air oven dried at 50°C       | 1927.95  | 0.014             |
| 5.     | Sample 5  | 12 h hot air oven dried at 50°C       | 2943.25  | 0.021             |
| 6.     | Standard 1| Withanolide A                        | 2714.84  |                   |
| 7.     | Sample 6  | 14 h hot air oven dried at 50°C       | 1387.94  | 0.010             |
| 8.     | Sample 7  | 10 h hot air oven dried at 60°C       | 1336.16  | 0.013             |
| 9.     | Sample 8  | 12 h hot air oven dried at 60°C       | 1942.27  | 0.016             |
| 10.    | Sample 9  | 14 h hot air oven dried at 60°C       | 2193.86  | 0.012             |
| 11.    | Sample 10 | 10 h hot air oven dried at 70°C       | 1222.68  | 0.008             |
| 12.    | Sample 11 | 12 h hot air oven dried at 70°C       | 1089.47  | 0.011             |
| 13.    | Sample 12 | 14 h hot air oven dried at 70°C       | 1141.99  | 0.0031            |

Fig. 9: Withanolide A conc. In sample 6

Fig. 10: Withanolide A conc. In sample 7

Fig. 11: Withanolide A conc. In sample 8

Fig. 12: Withanolide A conc. In sample 9
When roots were kept in hot air oven for 12 h at 50°C and 60°C, it was sufficient for rapid drying of roots of Ashwagandha because it attained 6.2% and 5.8% moisture content at this time and temperature. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6% [10].

In hot air oven, 12 h at 70°C is sufficient for rapid drying of roots of Ashwagandha because it attains 5.2% moisture content at this time and temperature. Moreover, moisture content limit for dried ashwagandha root is 6% or below 6% [10].

Table 7 shows that chromatographic studies were done with HPTLC to reveal the concentration of Withanolide A, and the results suggest that fresh sample of WS root extracts contains the highest concentration of Withanolide A as compared to shade drying, sun drying and hot air oven drying and in these three drying methods. Sun drying is much better than shade drying. However, hot air oven drying at 50°C for 12 h shows the highest yield of Withanolide A than the other two (sun and shade drying) drying methods. Therefore, hot air oven drying may be the best method for drying of WS roots.

**CONCLUSION**

On a comparative account, it can be suggested that WS roots may be dried by sun drying or hot air oven drying method. There may be little loss of withanolide A on drying by these two methods. Hence, these two methods of drying may be the safe method for the drying of ashwagandha roots.

**CONFLICTS OF INTEREST**

The authors have no conflicts of interest.

**AUTHORS’ CONTRIBUTIONS**

All authors contributed to the design and implementation of the research, to the analysis of the results, and to the writing of the manuscript.

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