

Standardization of *Valeriana officinalis* L. rhizome and roots cultivating in different regions of Armenia (Rhizomata cum *Radicibus valerianae officinalis* L.).

Hovhannisyan VG*, Chichoyan NB

Department of Pharmacognosy, Yervan State Medical University after M. Heratsi, Yerevan, Armenia

Abstract

Valerian is the most popular plant in medicine. It has been cultivated from ancient times and established itself as a profitable culture. The underground part of *valerian* is the source of commonly used sedatives, especially in chronic diseases. This plant is still the subject of considerable research aimed at establishing the chemical and pharmacological basis of the activity which has been clearly shown in a number of animal and clinical studies. The purpose of the study was to standardize the raw material of *Valeriana officinalis* L. cultivated in different regions of Armenia (Aparan, Ashotavan). As a material served rhizome with roots, obtained from the *Valeriana officinalis* L., cultivated in different regions of Armenia (Aparan, Ashotavan) in the second half of September in 2015. The commodity analysis of the raw material (volatile oil, extract, moisture, total ash) was carried out according to the requirements of WHO. The microscopic analysis was carried out with triocular electronic "Micros" microscope (10 × 40) and Olympus Digital C-3000 Zoom camera. The qualitative analysis of essential oil was carried out by GC-MS method. The research was carried out on gas chromatograph with a mass-selective spectrometer "BrukerDaltonik" (EM-640S). Unlike the pre-mountainous zone conditions of Aparan, the latter proves that in the mountainous zone conditions of Ashotavan, the plant has a developed mechanical tissue, which is displayed with the formation of sclereids. The existence of druses is also typical to a preparation, which is an anatomical differentiative feature for the raw material harvested from Ashotavan. In the result of the research confined the quality indices of *valerian* cultivated in Ashotavan and Aparan (volatile oil, the total of extracts, moisture, and total ash). The analysis showed that in volatile oil of *Valerian* climatized in Ashotavan and in Aparan the prevailing was bornyl acetate, the quantities of which were 55.2% and 90.7%.

Keywords: Aparan, Ashotavan, *Valeriana officinalis* L., Microscopical analysis, Volatile oil, Gas chromatography mass spectroscopy method, Bornyl acetate.

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Introduction

Valerian is the most popular plant in medicine. It has been cultivated from ancient times and established itself as a profitable culture. The underground part of *valerian* is the source of commonly used sedatives, especially in chronic diseases. Herbal remedy is considered as an alternative and complementary treatment of insomnia which has been reflected in many ethnobotanical investigations [1-9]. In the 2nd century AD, for example, the prominent ancient Greek physician Galen prescribed *valerian* for insomnia [10-16].

Valerian is a nutraceutical preparation from the roots of *Valeriana officinalis* L. and related species that is recommended for relief of tension, anxiety and insomnia. The greatest biological efficacy of *valerian* has been correlated with freshly harvested and carefully dried root preparations and with the iridoid alkaloid and sesquiterpene content of these preparations [17-22]. *Valeriana officinalis* L. is the species

most commonly used in northern Europe and still retains its official pharmacopoeial status although it is most commonly encountered as an ingredient of herbal medicines. This plant is still the subject of considerable research aimed at establishing the chemical and pharmacological basis of the activity has been clearly shown in a number of animal and clinical studies [23-26].

The purpose of the study was to standardize the raw material of *Valeriana officinalis* L. cultivated in different regions of Armenia (Aparan, Ashotavan).

Materials and Methods

As a material served rhizoma cum radicibus (ERE 193404), obtained from the *Valeriana officinalis* L., cultivated in different regions of Armenia (Aparan, Ashotavan) in the second half of September in 2015.

The processing of herbal raw materials was carried out in accordance with WHO guidelines governing the harvesting process [27].

The commodity analysis of the raw material (volatile oil, extracts, moisture, total ash) was carried out according to the requirements of WHO [28].

The microscopic analysis was carried out with triocular electronic "Micros" microscope (10 × 40) and Olympus Digital C-3000 Zoom camera. The qualitative analysis of essential oil was carried out by GC-MS method. The research was carried out on gas chromatograph with a mass-selective spectrometer "BrukerDaltonik" (EM-640S).

Helium was used as a gas carrier, the flow of the injector was 30 ml/min. An OPTIMA-FEAP capillary column (DF: 0.25; ID: 250) was used, the temperature gradient 40°C (1 min) to 200°C, heated up to 200°C, scanning at 30-400 m/z. The compounds were identified according to NIST (National Institute of Standard and Technology-Mass Spectrometry) data.

Results and Discussion

The results of the microscopic analysis showed that on the preparation of the raw material of *Valeriana officinalis* L. cultivated in Aparan rectangular epiblem cells with thickened walls are seen. Large parenchyma cells filled with starch granules are visible and a large number of essential oil globules of hypodermis are expressed. (Figures 1 and 2).

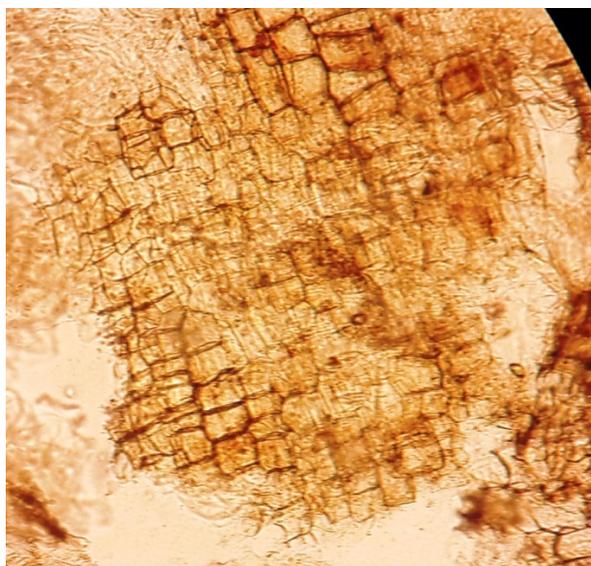


Figure 1. *Valeriana officinalis* root transverse section (Aparan). a-epiblem cells with thickened walls.

Conducting bundle with netlike vessels and parenchyma jelly starch granules are well expressed (Figure 3).

Stone cells are rare seen in the preparation of the raw material of valerian grown in Aparan (Figure 4).

The results of the preparation analysis of *Valeriana officinalis* L. cultivated in Syunik (Ashotavan) showed that in the cells of the hypoderm were well-expressed the globules of essential oil

(Figures 5a and 5b), starch granules and simple unicellular trichome (Figure 6).

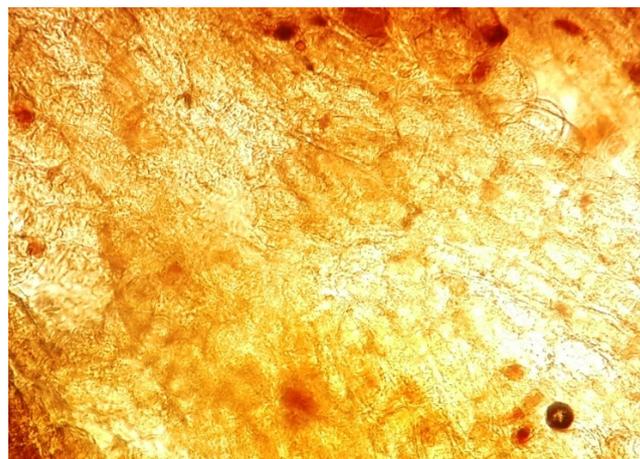


Figure 2. *Valeriana officinalis* root transverse section (Aparan). a and b-essential oil globules of hypodermis.

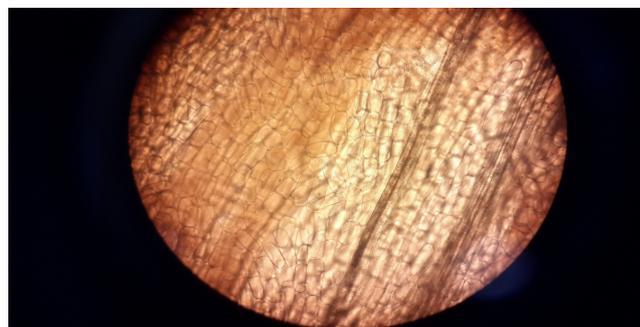


Figure 3. *Valeriana officinalis* root transverse section (Aparan). a and b-netlike vessels, c-jelly starch granules.



Figure 4. *Valeriana officinalis* rhizome transverse section (Aparan). a and b-stone cells.

Conducting system of a root is well expressed with thickened net-like vessels, which unlike the raw material of Aparan, in this case here and there are well seen thickened

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parenchymatous stone cells (Figures 7a and 7b). Unlike the pre-mountainous zone conditions of Aparan, the latter proves that in the mountainous zone conditions of Ashotavan, the plant has a developed mechanical tissue, which is displayed with the formation of sclereids.

The existence of drouses is also typical to a preparation, which is an anatomical differentiative feature for the raw material harvested from Ashotavan (Figure 8).

Recorded anatomical differentiative features prove that all these raw materials got morphological anatomical features stipulated by the influence of climatic conditions, can play a serious role in the primary standardisation process of a raw material.

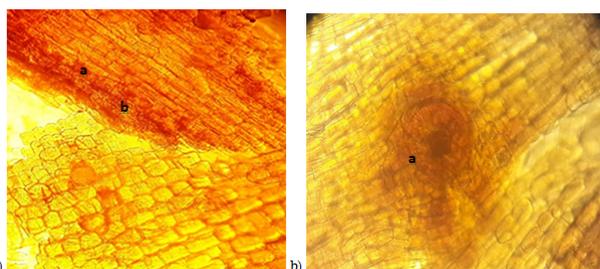


Figure 5. a) *Valeriana officinalis* rhizome transverse section (Ashotavan), a and b-essential oil globules of hypodermis. b) *Valeriana officinalis* rhizome transverse section (Ashotavan). a-essential oil globules of hypodermis.

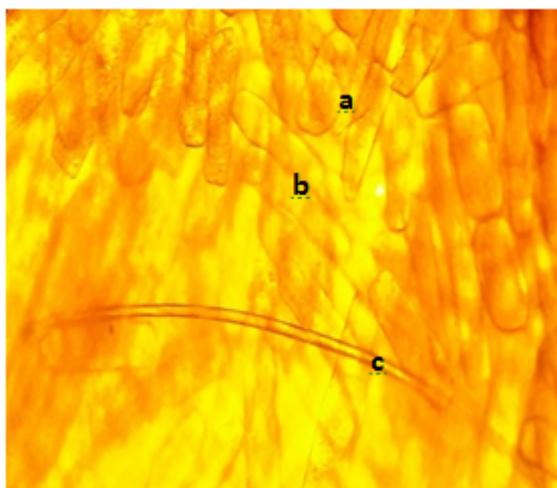


Figure 6. *Valeriana officinalis* rhizome transverse section (Ashotavan). A and b-jelly starch granules. c-simple unicellular trichome.

The outcome of the essential oil according to European pharmacopeia is considered the main criteria of good quality for the raw material of *Valeriana officinalis* L., where the amount of the essential oil mustn't be less than 5 ml/kg. According to the results of our investigation the outcome of the essential oil obtained from *Valeriana officinalis* harvested from Aparan is 1.22%. Whereas the raw material harvested from Ashotavan was 0.98%, which completely corresponds to the European pharmacopeia indices confirmed for *Valeriana officinalis* L. [29].

As the results of the research showed, the total of extracted substances was 29% in the harvested raw material of Aparan, and in the raw material of Ashotavan-27%, which also conformed the quality indices for the raw material of European pharmacopeia (not less than 25%) [29].

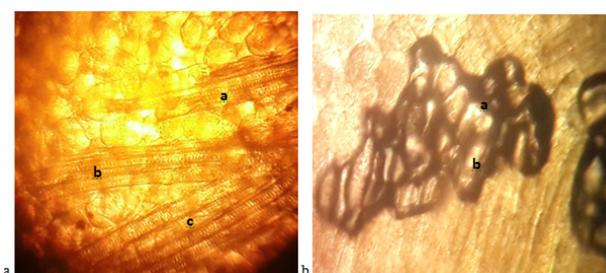


Figure 7. a) *Valeriana officinalis* rhizome with roots transverse section (Ashotavan), a-c-thickened net-like vessels, b) *Valeriana officinalis* root transverse section (Ashotavan). a and b-stone cells.

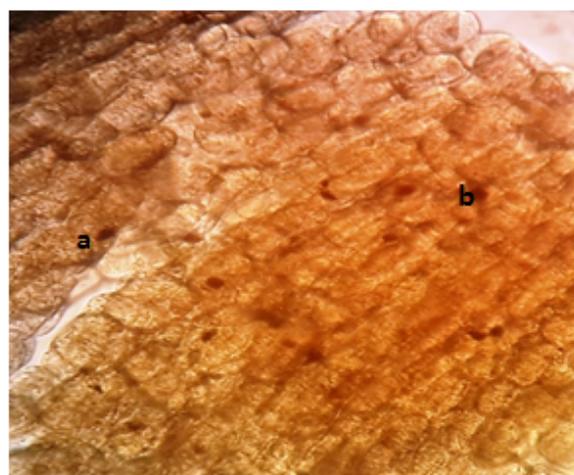


Figure 8. *Valeriana officinalis* rhizome transverse section (Ashotavan). a and b-drouses.

Table 1. The quality indices of *Valeriana officinalis* L. rhizome and roots cultivating in different regions of Armenia.

Plant name	Region	Extract (%)	Volatile oil (%)	Moisture (%)	Total ash (%)
<i>Valeriana officinalis</i>	Aparan	29	1.22	8	8
	Ashotavan	27	0.98	8.1	7.8

The total ash in the raw material harvested from Aparan was 8%, in the raw material from Ashotavan was 7.8%. These marked indices didn't exceed the European pharmacopeia (12% not over) standards. Considerably the moisture indices in the raw material cultivated from Aparan was 8%, and in the raw material cultivated in Ashotavan was 8.1% (Table 1) [29].

The qualitative and quantitative compositions of volatile oil cultivated from *Valeriana officinalis* L. climatized in Aparan and in Ashotavan were confirmed by gas chromatography-mass spectrometry method (Figures 9 and 10).

In the result of the essential oil analysis derived from the raw material of *Valeriana officinalis* L. cultivating in Aparan from invented 5 compounds, only bornyl acetate was identified, the quantity of which was 55.2% (Figure 9).

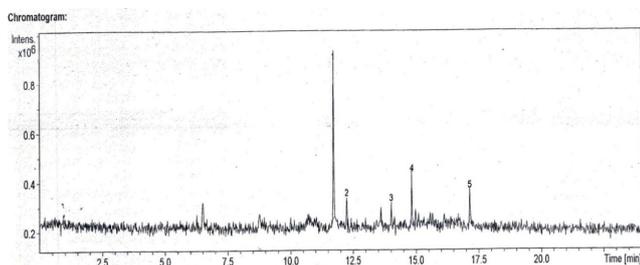


Figure 9. GC-MS analysis of volatile oil obtained from *Valeriana officinalis* rhizome with roots cultivated in Aparan. Identified substances: 1. Bornyl acetate.

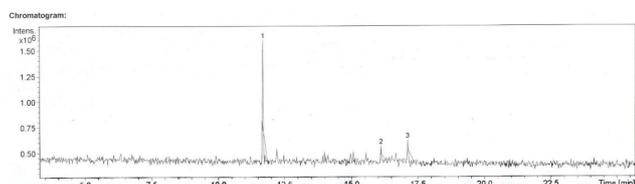


Figure 10. GC-MS analysis of volatile oil obtained from *Valeriana officinalis* rhizome with roots cultivated in Ashotavan. Identified substances: 1. Bornyl acetate.

In the essential oil of *Valeriana officinalis* L. cultivated in Ashotavan 3 compounds were invented, only bornyl acetate was identified. The quantity of which was 90.7% (Figure 10).

The analysis showed that in volatile oil of *Valeriana officinalis* L. climatized in Ashotavan and in Aparan the prevailing was bornyl acetate, the quantities of which were 55.2% and 90.7%.

Conclusion

1. Anatomical-differentiative features of *Valeriana officinalis* L. rhizomes and roots cultivated in different regions of Armenia were defined.
2. Unlike the raw material cultivated in Aparan, in the preparation of raw material in Ashotavan stone cells are well seen, which proves that in mountainous zone the mechanical tissue is well developed, as well as drouses exist (sclereids).

3. In the result of the research confined the quality indices of *Valeriana officinalis* L. cultivated in Ashotavan and Aparan (volatile oil, the total of extracts, moisture, and total ash).

4. Bornyl acetate was identified in the volatile oil obtained from the raw material of *Valeriana officinalis* L. cultivated in Ashotavan and in Aparan, which was prevailed in the raw material cultivated in Ashotavan.

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***Correspondence to**

Hovhannisyan VG

Department of Pharmacognosy

Yervan State Medical University after M. Heratsi

Armenia