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Determination of the Amount of Water-Soluble Vitamins in the "Asbosim" Food Supplement Used in the Treatment of Hypertension Disease

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| Article History | Abstract | | | |
|-----------------------|---|--|--|--|
| | The purpose of the study is to determine the amount of water-soluble vitamins in the natural | | | |
| Received: 08 Aug 2023 | biologically active food supplement "Asbosim" used in the treatment of hypertension. The | | | |
| Revised: 29 Sept 2023 | method of high-performance liquid chromatography was used in the study. In this, LC-40 Nexera Lite high-performance liquid chromatograph (Shimadzu, Japan) and LabSolutions ver. | | | |
| Accepted: 29 Oct 2023 | 6.92 software was used. As a result, it was found that "Asbosim" a natural biologically active food additive contains water-soluble vitamins B1, B2, B9, B6, B12, C. The conclusion is that | | | |
| | "Asbosim" contains a large amount of water-soluble vitamins C, B2, and B6, which lower | | | |
| | blood pressure and improve blood circulation. That is why this natural food additive has the | | | |
| | property of lowering blood pressure. | | | |
| | | | | |
| CCLicense | Keywords: Hypertension, Asbosim, LC-40 Nexera Lite, vitamins: B1, B2, B9, B6, B12, C. | | | |
| CC-BY-NC-SA 4.0 | | | | |

1. Introduction

It is known that one of the diseases that are difficult to treat in modern medicine is an increase in blood pressure, i.e. hypertension. Hypertension is the pathology of the cardiovascular system, a complex disease characterized by an increase in arterial pressure, followed by organomorphological changes [1]. Hypertension is mainly caused by constant nervousness, depression and stress, increased amount of adrenaline (epinephrine) in the blood, alcohol consumption, obesity and other factors. This disease is one of the main diseases that lead to complications such as myocardial infarction, stroke, heart attacks, and heart and kidney failure [2]. The main symptoms of high blood pressure are headache, dizziness, noise in the ears and heart failure. High blood pressure is more common in middle-aged and elderly people.

Considering the above, we developed a new biologically active food supplement called "Asbosim" based on plants such as saffron, ajgon and mint, which has the characteristics of effective treatment of hypertension and contains natural biologically active compounds. This food supplement was approved by the Ministry of Health of the Republic of Uzbekistan and put into practice [3,4]. This article presents the results of research on determining the amount of vitamins in the natural biologically active food supplement "Asbosim" [8].

2. Materials and Methods

In this study, the amount of water-soluble vitamins B1, B2, B9, B6, B12, and C [5] contained in the food supplement "Asbosim" was determined using the method of high-performance liquid chromatography. Here, Vitamin B12 was obtained from "Rhydburg Pharmaceuticals" (Germany) [6], and vitamins B1, B2, B6, B9 and C from "DSM Nutritional Products GmbH" (Germany) [7]. HPLC-grade water, acetonitrile, chemically pure-grade acetic acid and sodium hydroxide reagents were used. The amount of water-soluble vitamins in the plant

was determined using an LC-40 Nexera Lite high-performance liquid chromatograph manufactured by Shimadzu, Japan.

2.1. Used reagents and equipment

Vitamin B12 was obtained from "Rhydburg Pharmaceuticals" (Germany), and vitamins B1, B2, B6, B9 and C from "DSM Nutritional Products GmbH" (Germany). HPLC-level pure water, acetonitrile, chemically puregrade acetic acid and sodium hydroxide reagents were used. The amount of water-soluble vitamins in the plant was measured using an LC-40 Nexera Lite high-performance liquid chromatograph manufactured by Shimadzu, Japan.

2.2. Preparation of standard solutions

Solutions of vitamins C (CAS 50–81–7), B1 (CAS 70–16–6), B6 (CAS 65–23–6) and B12 (CAS 68–19–9) (100 mg/l) of each vitamin 5 mg is prepared by dissolving in 50 ml of HPLC grade water. Standard solutions of vitamins B2 (CAS 83-88-5) and B9 (CAS 59-30-3) were prepared by dissolving 5 mg of these vitamins in 50 ml of 0.025% sodium hydroxide solution. Then, all B vitamins were mixed together and a general solution was prepared. 1.5 ml of the resulting solution was filtered using a 0.45 μ m syringe filter, poured into a vial and used for analysis.

2.3. Preparation of sample extract

For the extraction of water-soluble vitamins, 1 g of the tested sample was weighed with an accuracy of 0.01 g on a scale manufactured by OHAUS company (USA) NV222 [9], placed in a 50 ml conical flask, and 25 ml of 0.1 N HCl solution was added. The mixture was extracted in an ultrasonic bath of GT SONIC-D3 (China) [10] at a temperature of 60 °C for 20 minutes. Then the mixture was cooled, filtered and made up to 25 ml with water in a volumetric flask. 1.5 ml of the extract was filtered through a 0.45 μ m syringe filter placed in a vial and used for analysis.

2.4. Determination of vitamins contained in "Asbosim" extract

Standard solutions and sample extracts LC-40 Nexera Lite high-performance liquid chromatograph consisting of LC-40D pump, SIL-40 autosampler, SPD-M40 photo-diode array detector (PDA) [11] and LabSolutions ver. 6.92 software was analysed. Shim pack GIST C18 ($150 \times 4.6 \text{ mm}$; 5 µm, Shimadzu, Japan) [12] reverse-phase column and a gradient mobile phase consisting of acetonitrile (A) and a 0.5% solution of acetic acid in water (B) (Table 1) was used. The injection volume was set at 10 µL, the flow rate at 0.9 mL/min, and the column thermostat temperature at 35 °C. A chromatogram of the extract of the sample in 0.1 N HCl was obtained (Figure 1) and the results were processed and presented in Table 2. The analytical signal (peak area) of each vitamin in the extract was recorded at four wavelengths 361, 291, 265 and 244 nm (Figure 1).

| Table 1. Gradient program mobile phase | | | |
|--|---------------------|--------------------------|--|
| Time | Acetonitrile (A), % | 0,5 % acetic acid (B), % | |
| 0 | 0 | 100 | |
| 6,8 | 0 | 100 | |
| 15,5 | 50 | 50 | |
| 17,4 | 95 | 5 | |
| 17,5 | 0 | 100 | |



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Figure-1. The chromatograms of determination of the amount of vitamins in the "Asbosim" extract

3. Results and Discussion

It can be seen from Figure 1 above that "Asbosim" extract contains vitamins B1, B2, B9, B6, B12, C. Its amount is given in the table below. From the above table 2, it can be seen that the amount of vitamin C (136,025 mg/100g) in the food supplement "Asbosim" is significantly higher than others. It is also known from the table above that the amount of vitamins in "Asbosim" decreases in the order C>B2>B6>B9>B12>B2.

| Table 2. The amount of vitamins in "Asbosim" extract and retention times | | | | | |
|--|----------------------|---------------------|--------------------------------|--|--|
| Vitamin | Retention times, sec | Concentration, mg/l | The amount in 100 g sample, mg | | |
| Vitamin B1 | 2,062 | 0,133 | 3,325 | | |
| Vitamin B9 | 13,651 | 0,299 | 7,475 | | |
| Vitamin B2 | 15,469 | 1,662 | 41,550 | | |
| Vitamin B6 | 3,585 | 0,563 | 14,075 | | |
| Vitamin B12 | 13,628 | 0,21 | 5,250 | | |
| Vitamin C | 2,627 | 5,441 | 136,025 | | |

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It is known that American scientists proved that vitamin C is one of the substances to treat high blood pressure [14, 15]. Studies conducted in this regard showed that the effect of vitamin C on normalizing systolic and diastolic blood pressure is high. This is great scientific evidence for saying that "Asbosim" natural food supplement has a high amount of vitamin C, and it shows high biological activity in the treatment and prevention of hypertension.

4. Conclusion

In conclusion, the natural food supplement "Asbosim" that we developed based on medicinal plants contains water-soluble vitamins C, B2, B6, B9, B12, and B2. Among them, the amount of vitamin C is significantly higher than others, which gives this food supplement the property of treating and preventing hypertension. Considering the above scientific results, people suffering from hypertension are recommended to use the biologically active food supplement "Asbosim" instead of synthetic drugs.

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