

OBTAINING OF CAROTENOID EXTRACT FROM *LYCIUM CHINENSE* AND CHARACTERIZATION USING SPECTROMETRICAL ANALYSIS

I. R. BUNGHEZ^{a,b*}, AVRAMESCU SORIN MARIUS^c, NEATA MARIAN^c,
RADULESCU GEORGETA^d, ION RODICA-MARIANA^{a,b}

^aNational R&D Institute for Chemistry and Petrochemistry – ICECHIM, 202 Spl. Independentei, Bucharest, Romania

^bValahia University of Targoviste, 2 Carol I Blvd., Targoviste, Romania

^cFaculty of Chemistry, University of Bucharest, 90-94 Panduri St., Bucharest, Romania

^dNational R&D Institute for Pharmaceutical Chemistry, I.C.C.F, 112 Vitan St., Bucharest, Romania

Carotenoids are known as photoprotection agents, defending the existing fotodestruction many biologically active substances in cells and tissues. Carotenoids are naturally pigments, with different colors like red, orange or yellow. Carotenoids are used as natural colorants for food and cosmetics. They are essential for plant growth and photosynthesis, and are a main dietary source of vitamin A in humans. They are thought to be associated with reduced risk of several chronic health disorders including some forms of cancer, heart disease and eye degeneration. In this study it is presented a method for obtaining a carotenoidic extract from goji berry. The obtained extract was characterized using spectrometrical analysis (UV-VIS, TGA and HPLC).

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1. Introduction

Lycium Chinense (Goji) is an important Chinese medicine used for energy restoring tonic, to cure a wide range of ailments from skin rashes and to treat and prevent diseases such as insomnia, liver dysfunction and eyesight problems to diabetes, high blood pressure, vertigo, lumbago, cancer impotence and menopausal complaints. It has various biological activities, such as enhancing immunity, protecting liver damage and reducing the side effects of chemotherapy and radiotherapy [1].



* Corresponding author: raluca_bunghez@yahoo.com

Fig. 1 Goji fruits

Research on goji berries is over the past 30 years, and a big awareness was demonstrated about its benefits of health and therapeutic benefits are growing [1]. So on international level, it has discovered the benefits of this fruit. Existing proves that Goji possess antioxidant [2], antitumor [3], anticancer, hypoglycaemic, immunological [4], neuroprotective effects [5] and enhance immunity activities [6], [7,8].

It was present in fruits of Goji a lot of carotenoids, flavonoids, polysaccharides and a group of lipid-soluble compounds with color ranging from yellow to red, has been widely used in the health food industry because of its possible role in the prevention of chronic disease like age-related degeneration [9,10]. The presences of this various functional components are responsible for these benefits effect [6]. In fact, in 1983 the Ministry of the Public Health of China approved goji berry to be marketed as a botanical medicine [11].

The more important compounds of this fruits are polysaccharides and carotenoids (beta-carotene, lutein, zeaxanthin).

Polysaccharides ($C_x(H_2O)_y$) are long-chain sugar molecules, are a distinguishing characteristic of goji berry, are a primary source of dietary fiber in the intestinal system, and once they are metabolized polysaccharides support and maintain the health of the colonic mucosal lining, lower pH and reduce colon cancer risk, enhance mineral uptake, stabilize blood glucose levels, stimulate the immune system, offer antioxidant protection [12].

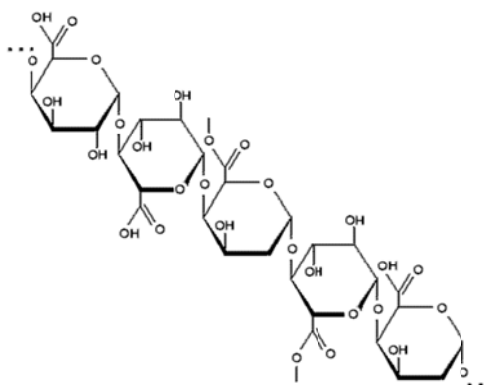


Fig.2. Pectine - a polysaccharide structure [14]

Zeaxanthin (b,b-carotene-3,30-diol) is an antioxidant in the carotenoid family. It plays a key role in our immune system support and is abundantly found in goji berry. Zeaxanthin is one of the most common carotenoid alcohols found in nature, its play a key role in our immune system support and are abundantly found in goji berry. Many researchers believe zeaxanthin and lutein, may be a potent protectant against macular degeneration 1-6, and may retard aging of the lens in preventing of forming cataract. So, vision support is another health benefit of goji berry fruits [13].

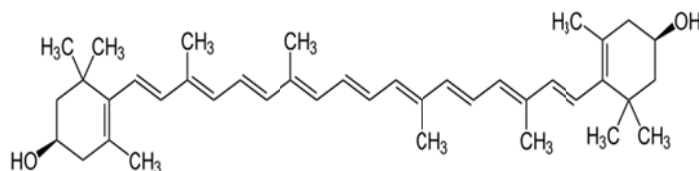


Fig. 2 Structure of zeaxanthin [14]

Beta-carotene ($C_{40}H_{56}$) is a carotenoid pigment in orange-red foods like goji berry, pumpkins, carrots, etc. It is important for the synthesis of vitamin A (a fat-soluble nutrient and

antioxidant that is essential for normal growth), vision, cell structure, bones and teeth, and healthy skin. Goji berry's beta-carotene content is among the highest for edible plants.

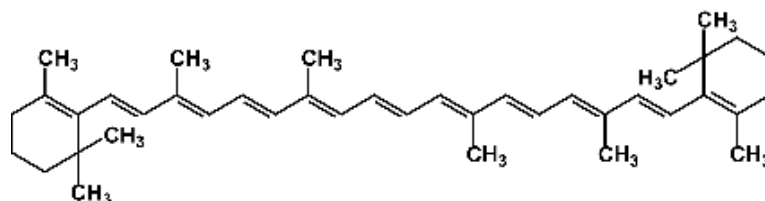


Fig. 3 Beta-carotene structure [14]

2. Experimental part

2.1. Materials

Goji (*Lycium Chinese*) fruits were available commercial. Beta-carotene standard, was from Merk. The HPLC grade solvents such as ethanol (95%), n-hexane, acetonitrile, were from Merk and Sigma Aldrich. Potassium hydroxide and anhydrous sodium sulfate were from Riedel-de Ha.

2.2 Apparatus

2.2.1. UV-VIS

A M400 Carl Zeiss Jena UV spectrophotometer with a 1 nm slit width, 1 nm step size, 0.3 nm/s average scan rate, deuterium lamp, double beam, microprocessor and quartz cell was used to measure the aqueous solution absorbance and the molar absorption spectra for each sample at 22 °C.

2.2.3. TGA

The sample was examined using a Mettler 4000 TA, TG 50 thermal analyzer system at a rate of 10°C min⁻¹ in a static air atmosphere; and a Perkin-Elmer thermoanalyzer TG S-2. The crucibles used were from Al₂O₃ and the diameter was from 70 µL. About 20 mg of sample were subjected to analysis in a temperature range 25-500°C (20°C min⁻¹). Heating speed was 10 degrees 20°C/min. Liquid air's flow rate was set around 3 l/h.

2.2.2. HPLC

HPLC spectrometer from Varian performed on a system with an Intersil ODS-3 column (150mm×4.6mm ChromSep stainless steel). The mobile phase performed on a system with acetonitrile/ethanol:70/30. The flow rate was 1ml/min, temperature 40°C. The chromatograms was monitored at λ= 450 nm, using UV detector.

2.3. Extraction of pigments from Goji

10 grams of dried goji (*Lycium Chinese*) were pulverized. The powder was collected and was stirred with mixed solution hexane/chloroform: 50/50, at room temperature for 3 hours, using an agitator. Upon was filtered the solution and washed the goji berries with hexane, a clear dark red-yellow solution was obtained.

The extracts that contain carotenoids pigments are usually submitted to saponification for the scope rejecting the carotenoids that are under esters form and to release the fats that can be saponificated. To the solution obtained was added KOH dizolvated in ethanol. The mixture was treated with a dilute acid until the pH was 7. Then, the extract was dried using Na₂SO₄ anhidre; was cooled and kept in the dark place. Next day was analyzed at UV-VIS, TGA and HPLC.

3. Results and discussions

UV-VIS analysis revealed very well the zeaxanthin carotenoid, which have three sholders at 433 nm, 448 nm and 479 nm (fig 4). This aspect is confirmed in the literature, using chloroform solvent extract (Rodriguez-Amaya, 2002).

Next thermogram (fig. 5) show very clearly melting point of carotenoids. For lutein and lycopene the melting point is between 160-170°C and for zeaxanthin between 203-205°C, more accurate.

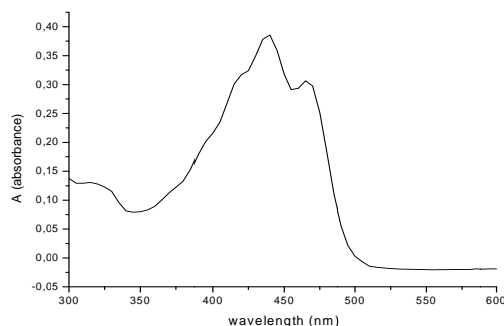


Fig. 4. UV-VIS spectra of goji extract

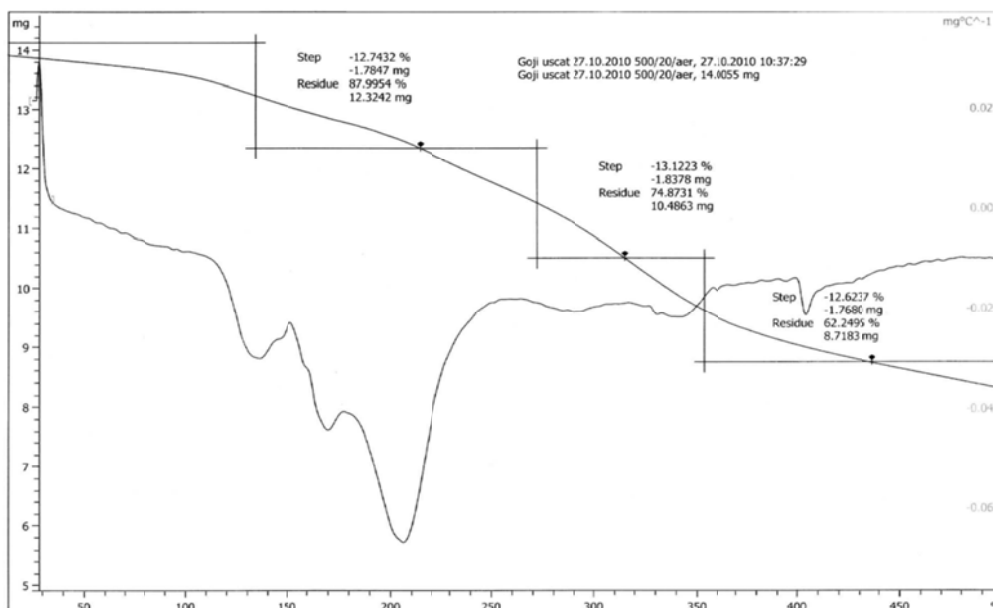
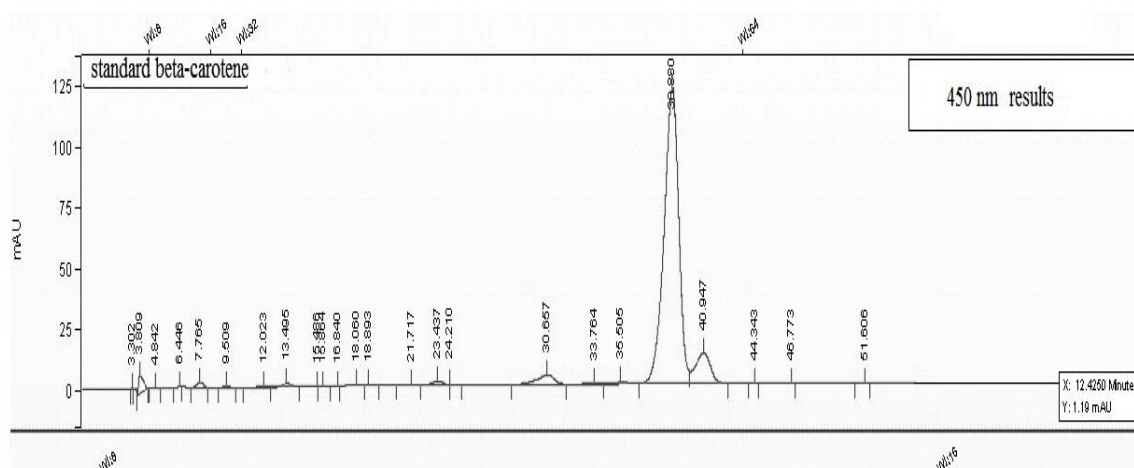
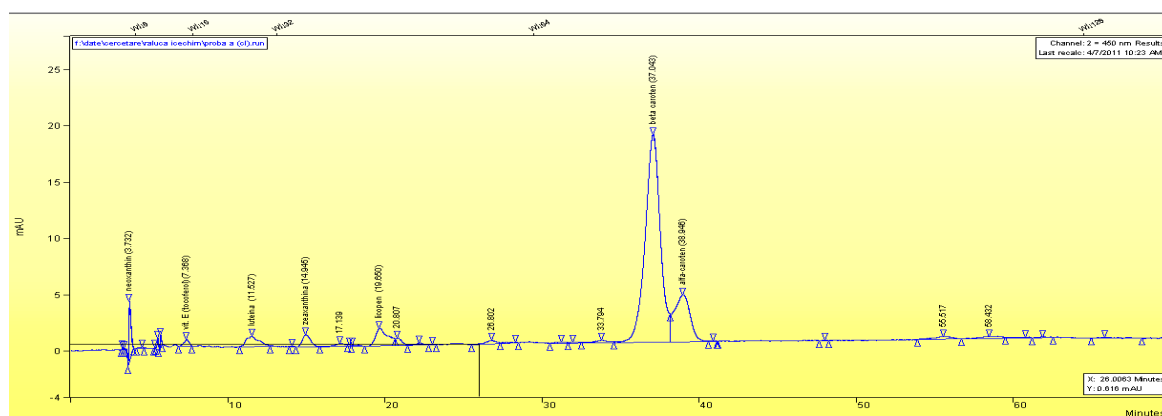


Fig. 5. TGA thermogram of goji extract



a)



b)

Fig.6 HPLC cromathogram of the a) beta-carotene standard and b) goji extract

Fig. 6. shows the HPLC chromatogram of carotenoid extracts from dried Goji fruits.. In our study, a large amount of carotenoids were still present after simultaneous extraction and saponification.

Table 1. Total carotenoids contents in saponificated extract

| Carotenoids | | Standard time retention |
|-------------------------|------------|-------------------------|
| Neoxanthin | | 3,733 |
| Vitamin E (tocopherols) | | 7,368 |
| Xantophylls | Lutein | 11,530 |
| | Zeaxanthin | 14,929 |
| Lycopene | | 19,646 |
| Beta-carotene | | 37,048 |
| Alpha-carotene | | 38,953 |

4. Conclusions

Following research findings can be drawn:

Goji berries represent a rich source of carotenoids including β -carotene predominates, α -carotene, xanthophylls and vitamin E (tocopherols).

The extraction with hexane and chloroform was more efficient, than to observe clearly peaks at UV-VIS and HPLC.

Datas of literature confirmed that goji has verified anti-cancer/immune-enhancing, antioxidant, anti-diabetic, neuroprotective, DNA protective, liver protective and anti-fatigue properties, and may help reduce hypertension.

In this research it has been demonstrated the components of this fruit. So, it is obvious that we must eat healthy for a good life.

Aknowledgements

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