THE INVESTIGATION ON SUPERCRITICAL FLUID EXTRACTION AND SEPARATION OF BIOLOGICALLY ACTIVE SUBSTANCES OF *HIPPOPHAE RHAMNOIDES* AND GRAPE SEEDS

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INTRODUCTION.

Hippophae rhamnoides and grape seeds take up more noticeable place among the objects of photochemical investigations. This is connected with discovery a whole number of biologically active substances (BAS) in them. They are better saturated by essential and fat oils and represent a great interest for the pharmaceutical industry. They aquire a great value due to medicinal properties of their supercritical (SC) CO_2 extracts. In this connection it is expedient to carry out the investigation of the SC fluids extracts for hippophae pulp and grape seeds. In the work a large consideration is given to study of grape seeds. Being waste materials of wine industry they have a great pharmacological value. The aim of the work is a definition of optimal extraction pressures for given plants at a temperature up to 31^0C; a determination of effective thermodynamic parameters for the extraction of concrete BAS; to reveal the dependence of extraction efficiency on the pressure change; to analyse the extracts; comparative investigation of lipofile complexes of grape seeds obtained by extraction organic solvents and SC CO_2.

MATERIALS AND METHODS.

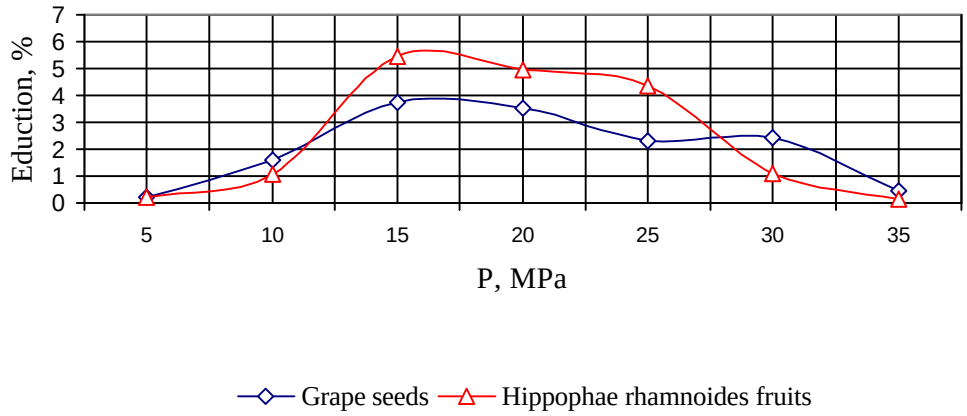
Experimental investigations are carried out with a special device allowing to observe the SC extraction processes visually.

To solute this question has been worked out an experimental device [1] allowing to make instantaneous processing and calculation of all thermodynamic parameters and with an opportunity of visual observation through the phenomena in a cell in near-critical and over critical regions, solubility and extraction process. The optimal extraction parameters are different for each kind of plants, and the working temperature of the extraction process must be relative to a natural. This allows to avoid the degeneration of thermolabile compounds and preserve the natural balance of extraction. The device allows to research phase transitions of multicomponent systems in a wide range of state parameters. A main part of the experimental device is the measuring cell of synthetic sapphire, which is placed in special flanges. To support tight seal in the measuring cell between sapphire tube and flanges can be placed fluorplastic or carbon-graphitic packings. To measure a temperature in the measuring cell in the upper flange is founded a cover from stainless steel, in where is transferred a thermocouple of chromium-alumel type. It allows to carry out the measurement on the height of the measuring cell. A plate of any material (for precise definition of interphase tension) can be put into the measuring cell for a measurement of the interphase tension and edging angles

of wettability in static conditions by the method of meniscus visual observation appearing on liquid phase boundary.

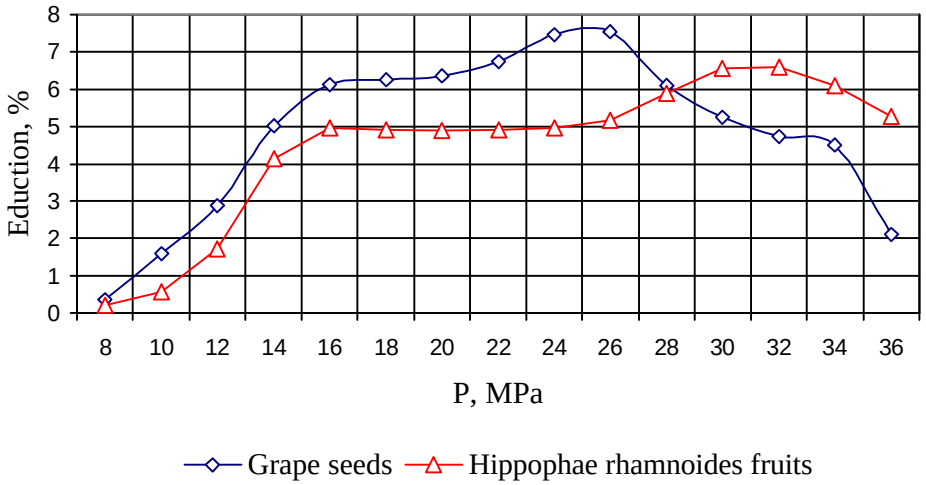
In the optical cell one observes to CO_2 solubility of extracted substance and determined a maximum result. The extraction is made at isothermic and isobaric process with constant time of extraction. An influence of pressure change upon the extraction is defined by the consecutive stepped increase of pressure completing the process with full exhaustion of raw materials for each isochor point at given thermodynanic parameters (Diagram 1).

Diagram 1. Isotherm of eduction dependence of extractive substances from raw materials "for exhaustion" upon the pressure change.



After each fixed point the obtained extract is removed out of the separator, where it passes after the extractor. One researches a raw material, at which a new portion of raw material is used for all points of isochor. (Diagram 2)

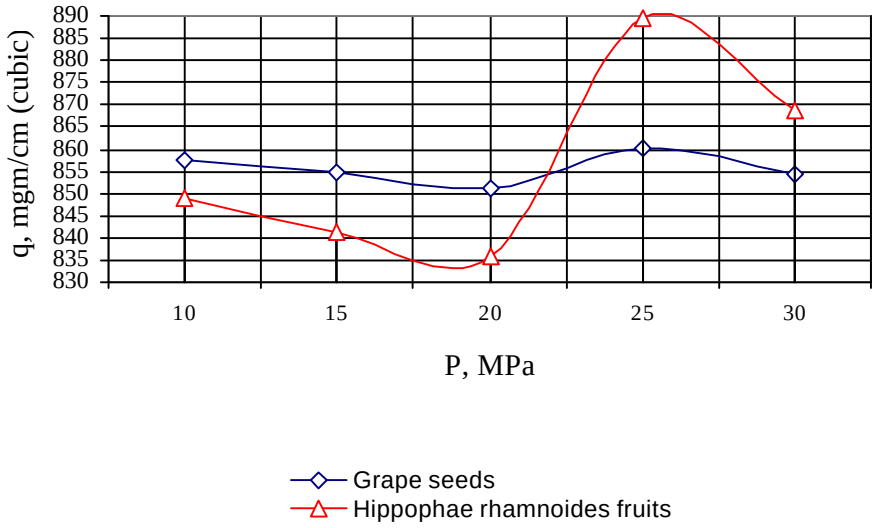
Diagram 2. Isotherm of eduction dependence of extractive substances upon the pressure change.



Extracts are investigated for a biologically valuable substances by liquidous gas chromatographs, ultra-violet spectrophotometer, atom-adsorptive spectroscope. Due to these investigations are obtained thermodynamic parameters of effective extraction of concrete valuable BAS. The qualitative and quantitative analysis of extracts obtained at different thermodynamic parameters are done and confirmed the change of chemical composition of the extract owing to change of T and P.

Observing SC fluid extraction it may be noted that sometimes a maximum result of SC fluid extraction exists in a region of SC state, where theoretical calculations show a decrease of the extraction. It is explained by the complex chemical composition of extract which can change the effective, in technological respect, region of SC extraction, authors [2, 3] point out at that fact. A comparison of Diagram 1 and Diagram 2 shows, that sum output of extract (Diagram 1) for each raw material is little larger than in (Diagram 2), where they must correspond to each other. In the first case the SC CO_2 passes larger through the raw material than in the second one, and this causes more full extraction. To our opinion this is explained by the ability of CO_2 for adsorption of restricted quantity of extract only. The extracts obtained from the same raw material at different thermodynamic parameters are differ from each other by the chemical composition. It is connected with change of extraction depth at the change of pressure. Diagram 3 shows a density change of extracted substances with change of extraction pressure at temperature 31^0C- const.

Diagram 3. Dependence of extract density upon pressure of extraction processing



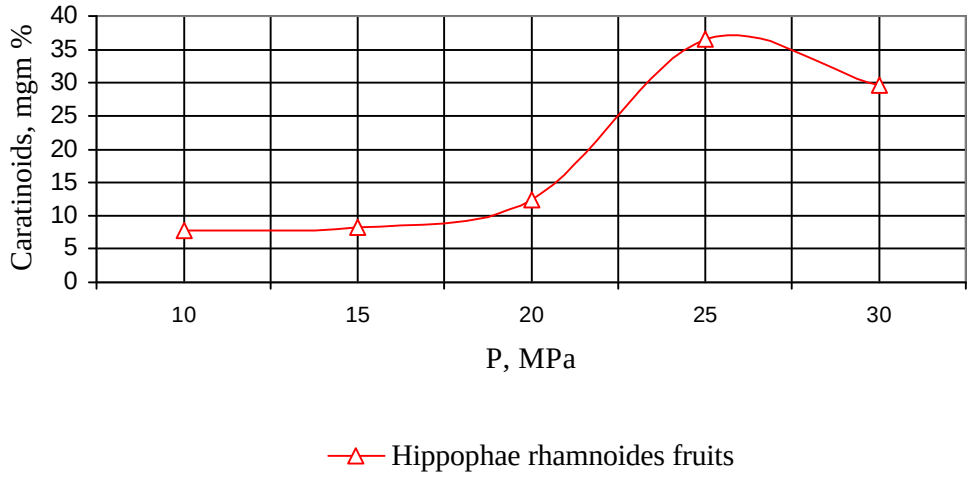
To compare numerical exponents of SC CO_2 extract of grape seeds were obtained extracts by petrolane ether, hexane, chlodone-11. Hexane and petrolane extracts are obtained by stepped irrigation of mixed processed material at optimum corelation of raw material and extragent 1:20, extraction temperature 30^0C, extraction time 2,5 hours. Chladone-11 is removed from the extract under the vacuum. Numerical exponents and chemical composition of lipofile extracts of grape seeds are give in Table 1.

Table 1.

Number exponents and chemical compound.	Extrogent			
	Petroleum ether	Hexane	Chladone-11	SC CO_2 - extract
Density, g/cm^3	0,7545	0,7537	0,7546	0,8552
Refraction exponent	1,4770	1,4763	1,4700	1,4732
Acid number	2,13	2,17	2,37	10,40
Saponification number	189,10	185,30	253,72	450,57
Ether number	186,97	183,13	251,35	440,17
Iodine number	148,31	142,37	97,47	116,10
Reichter- Meiss number	1,36	1,37	1,87	1,52
General- technological eduction	12,03	13,21	13,40	14,7
Palmitic acid, %	5,64	5,24	5,34	5,16
Stearic acid, %	5,37	5,09	2,14	1,54
Oleic acid, %	19,62	20,58	12,80	13,89
Linoleic acid, %	69,37	69,09	80,12	79,81
Caratinoid sum, mgm %	3,90	4,10	4,20	4,0
Tocopheroll sum, mgm %	285,39	276,43	270,51	269,64
Chlorophyll sum, mgm %	5,32	5,28	2,91	2,78

It is studied an influence of pressure upon the extraction of concrete BAS. On example of Hippophae rhamnoides. Diagram 4 shows the dependence of caratinoid eduction upon the change of extraction pressure at temperature 31^{0}C- const.

Diagram 4. Dependence of caratinoid eduction upon pressure of extraction processing.



CONCLUSION.

The supercritical extraction of substances by CO_2 has a number of advantages. Firstly, both extract and technological process are very high ecologic. Secondly, SC-extractions only provide practically absolute microbiologic purity. Thirdly, at SC-extractions are obtained natural, saving structure and properties, components, that confirms by the chemical analysis. And finally SC-extractions only can more completely educe the compounds from plant raw materials. Exactly supercritical parameters allow to save in the extract such concentration of components which can not be offered by and existing techniques achieved a level of industry production.

REFERENCES :

[1]. A.M. Aliev, Proc. of the 8th Meeting on Supercritical Fluids “Chemical Reactivity and Material Processing in Supercritical Fluids ”, Bordeaux (France), T.2, p.891.

[2]. I.M. Abdulagatov, Kh.S. Abdulkadyrova, M.N. Dadashev. Thermophysics of High Temperature 1993, V. 31, ? 5, p. 830

[3]. F. M. Gumerov, A.N. Sabirzyanov, G.I. Gumerov. Sub- and Supercritical Fluids in the Polymers Processing. Kazan:2000, p. 7.