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RESEARCH ARTICLE

CHARACTERIZATION OF SEA BUCKTHORN FRUITS AND COPSES IN TERMS OF SEROTONIN AND MICROELEMENTS

I. BRAD [1], G.A.VLĂSCEANU [2], I.L. BRAD [3], ŞT. MANEA [2]

The Academy of Agricultural and Forestry Sciences, Bucharest, Romania[1]
61 Mărăşti Bvd., District, Zip Code 707183

Hofigal Export-Import S.A., Bucharest, Romania [2]
2nd Intrarea Serelor, District 4, Zip Code 041124

Janssen-Cilag Bucharest, Romania [3]
8 Sipoutel Fântânilor Str., District 1, Zip Code 707183

Abstract

The content of microelements in sea buckthorn fruits, leaves and copses was determined through neutron activation. Also, the content from fruits and leaves was analyzed by atomic absorption, for 11 sea buckthorn bio-types.

The content in the dry matter was analyzed from the ashes of the sea buckthorn, depending on the harvesting time. The variation in serotonin content was analyzed depending on the origin of the fruits (three geographic regions). Estimations were made regarding:

- ways of processing different plant organs;
- biological effects of sea buckthorn;
- perspective for serotonin as an immune inductor in different diseases

The effects of sea buckthorn are the consequece of a large number of active physiologic substances, like hydro- and lyposoluble vitamins, hormones and phytohormones, amino-acids (including the essentials), provitamins A, carotenoides, serotonin, melatonin.

Most of our study was done with research centers: industrials, medical and production units.

Serotonin's effects are well known: immune-inductor, energizing, anti-depressive, chemical mediator, involved in disabilities like insufficiency in transmitting the information through the nervous system to the organs.

The serotonin alongside with the other substances from sea buckthorn is recommended for sickness or discomfort, too, mostly concerning the severe conditions that affect the 21st century: cancer, HIV, depression, anxiety, suicide tendencies, insomnia, alcohol abuse, schizophrenia, any disease generated by a physical, chemical or biological agent.

Key words: *sea buckthorn, serotonin, phytotherapy, immunity, metabolism*

The content of micro- and infra-micro elements from sea buckthorn fruits and copses was determined by the activation of neutrons. The content of Zn, Cu. Mn, Fe, Ba, Mo, determined by fotometric of atomic absorbtion in 11 seabuckthorn bio-types, was also analysed. *(*Brad *et al.*1976*)*.

In dry material has been analysed the content of macro, micro and inframicro-elements in ashes of sea buckthorn, depending on the time of harvesting. The variation in serotonin content depending on the origin of the fruits (samples taken from three geografic regions) has been also been analysed *(*Cojocaru and Brad, 1984; Brad *et al.*1997*)*. Estimations were made regarding the following items:

- ways of processing different plant organs;
- biological effects of sea buckthorn;
- serotonin perspective as an immune inductor in different diseases.

The effects of seabuckthorn are the consequece of a large number of active physiologic substances, like hydro- and lypo-soluble vitamins, hormones and phytohormones, organic acids playing an important part in metabolism, aminoacids (including the essentials), flavonoides, pro-vitaminsA, carotenoides, serotonin, melatonin *(*Mathiev and Azizov, 1981; Uluitu and Brad, 1984*)*.

The use of seabuckthorn products (single or in association), act as immune-inductor and also have other benefits, due to the fact that they act simultaneously, synergic and harmonic, determing effects of many active ingredients from sea buckthorn fruits, leaves and copses *(*Brad and Medeşan, 1987*)*. Most of our researches were made in cooperation with research centers: industrials, hospitals and production units. *(*Uluitu *et al.* 1997; Brad et al. 2002*)*.

Today, micro-elements are considered to be ”mineral” vitamins because they meet all the requirements of what vitamins are.

The vitamins are not synthesized by the human organism; they are co-enzymes or co-factors of numerous enzymes that act in all the human chains, tracks and metabolic cycles. Macro-, micro- and inframicro-elements have different roles in the metabolic chain, being at the same time stimulants and inhibitors, synergic or antagonists, being found in the structure of some substances, carrying different physiological rols compared to vitamins. These „mineral” vitamins are function inducers and structure stabilizers or play a part in catalyst processes or governs enzymatic activities. For some micro-elements the intimate, subtle process is unknown, while their presence even in small amounts has positive effects (ppm*).(*Bazarova, 1978; Talichova, 1998; Puhalskaia, 2000; Brad *et al.,* 2007*)*

Materials and methods

Sea buckthorn fruits, leaves, offshoots and offshoot bark were used as biologic materials. Micro and inframicro-elements were determined by **the activation of neutrons** from the Atomic Physics Institute *(*Cojocaru C.,1973, Salajan M., 1983*)* and through atomic absorbtion spectrophotometry by Pedologic Institute *(ASAS)*.

Plain serotonin was isolated by using extraction methods from the concentrated acid solutions through *reverse osmosis*. Knowing its chemical proprieties, serotonin may also be associated with a precipitation element or co-precipitation.

The serotonin content was determined by means of a **fluorescent method** by researchers in the Institute of Normal and Pathologic Physiology *(*Uluitu M., Chis M., 1995*)*. The hydroponic (watery) extract was obtained by heating and boiling dry leaves and fruits in carbonated water in proportion of 1:4 vegetal material:acid solution. After filtration the determination was done and the results were expressed in micrograms, serotonin/gram of fruit or dry leaf.

Results and discusions

Microelements were detected and it could be seen how impressively quantities might vary by breed and soil/weather conditions. Content in Zn, Cu, Mn, Fe, Br, Mo was determined by neuronal activation in 11 biotypes and this is depicted in *Tables 1, 2, 4 and 7.*

Table 1. *The concentration of elements in the white seabuckthorn copses and fruits ashes (g% - analysis by neutron thermo activation)*

Element g%	Copse	Fruit
Al	0.55	0.21
Au	0.000014	0.000006
Ba	0.075	-
Ca	14.40	4.20
Co	0.0005	0.0004
Cr	0,0038	0.0032
Fe	1.09	1.10
La	0.00096	0.00040
Mn	0.106	0.034
K	25.40	31.80
Rb	0.014	0.031
Ru	-	0.0007
Sm	0.00004	0.00003
Se	0.00025	0.00014
Na	1.48	1.29
Sb	0.0004	0.0001
Th	0.00025	0.00016
Zn	0.074	0.140

A low number of micro-elements was determined by V. Ţigănuş (*Table 2*) from the Pedology Institute at ASAS, through atomic absorption spectrophotometry.

In the dry fruits and ashes there are to be found elements enumerated in alphabetical order, by us: Al, As, Au, Ba, Ca, Ce, Co, Cr, Cs, Fe, Hf, K, La, Rb, Mg, Mn, Mo, Na, Sb, Se, Se, Sm, Sr, Th, U, V, Zn, Yb. In different chains and cycles (according to the nowadays knowledge about the microelements) it was revealed that they participate or constrain in more than 80-100 metabolic sequences (*Tables no. 3, 5*). The results regarding the content of microelements in dry substance, ashes of white seabuchthorn fruits are presented in *Table 6*. The content of serotonin in leaves, offshoots, offshoot bark was also determined by using the same samples. Great differences could be seen between serotonin content from dry leaves and fruits (the offshoots contain almost the same quantity of serotonin as the dry fruits and the offshoot's bark contain 6-7 times more serotonin than the offshoot and 4 times more than the leaves, on average, the quantity of serotonin amounted between 30-100 micrograms).

Table 2. *Analytical results regarding the content of dry material, ashes and microelements in white sea buckthorn fruit samples (Station of Trees Research Bacău)*

Type	Ash (450°C)	Zn	Cu	Mn	Fe	B	Mo
		ppm/dry material in air					
Sf. Gheorghe 4	4.19	8.0	9.25	6.8	63	13.5	3.15
Sf. Gheorghe 5	3.68	10.5	5.25	9.5	280	11.0	3.10
Sf. Gheorghe 6	3.41	9.8	5.50	8.0	53	12.7	3.40
Sf. Gheorghe 9	3.65	12.5	4.50	10.8	118	19.0	2.57
Sf. Gheorghe 10	3.66	12.3	5.00	8.8	78	16.4	5.07
Delta 60 M	3.57	14.3	6.75	9.0	183	17.5	3.10
Without thorns	3.82	11.8	6.75	10.0	95	11.0	2.62
Şerbăneşti l	3.75	16.0	7.00	11.3	428	3.5	3.15
Şerpeni 1 1	3.18	12.8	4.50	9.3	323	19.0	1.43
Şerbăneşti 4	3.33	15.3	3.00	10.5	83	16.0	2.02
Ciumaşi	3.91	9.3	6.00	12.5	88	16.4	1.75

Table 3. *Analytical results regarding the content of microelements in dry substance and ash (ppm) of white sea buckthorn* ***leaves*** *samples (Station of Trees Research Bacău)*

Type	Ash (450°C)	Zn	Cu	Mn	Fe	B	Mo
		ppm/dry material in air					
Sf. Gheorghe 4	11.05	14.8	6.25	48.8	278	153.0	6.24
Sf. Gheorghe 5	7.83	18.5	6.75	56.8	300	101.0	2.76
Sf. Gheorghe 6	6.99	17.3	4.00	80.8	213	87.0	3.00
Sf. Gheorghe 9	6.35	10.5	13.70	42.5	135	43.5	2.42
Sf. Gheorghe 10	7.68	11.0	4.50	40.5	203	58.5	1.95
Delta 60 M	10.02	13.8	5.25	41.8	235	77.5	3.90
Without thorns	7.31	23.3	5.00	74.3	340	66.5	2.28
Şerbăneşti l	7.82	14.5	5.00	56.8	395	80.5	0.65
Şerpeni 11	7.73	15.5	5.00	31.8	315	66.5	2.60
Şerbăneşti 4	7.86	11.5	3.00	89.3	245	60.0	0.83
Ciumaşi	6.66	11.5	5.23	46.3	245	65.0	1.08

Table 4. *The content of macro-, micro-, semimicro-, and inframicro-elements in dry substance of white sea buckthorn fruits (Şerbăneşti biotype); analysis through neutron thermo activation: neutron flux 2 x 10^{12} $n/cm^2.s$*

Element	Date 1- 4. X Media ± s	Date 2 -27. XI Media ± s
Ca%	19.832 ± 1.188	8.013 ± 0.561
K%	8.45 ± 0.42	22.06 ± 1.10
Na%	4.095 ± 0,082	2.117 ± 0.042
Fe%	1.261 ± 9.063	0.635 ± 0.032
Zn ppm	401 ± 28	895 ± 36
Ba ppm	421 ± 70	156 ± 37
Rb ppm	105 ± 11	213 ± 24
Br ppm	127 ± 2	66 ± 2
Cr ppm	36 ± 3	18 ± 2
Ce ppm	20 ± 2	11 ± 2
As ppm	15 ± 1	4.8 ± 0.5
La ppm	12 ± 1	5.1 ± 0.2
Co ppm	6.4 ±0.6	3.9 ± 0.4
Th ppm	2.8 ± 0.3	1.2 ± 0.2
Se ppm	2.8 ± 0.1	1.5 ± 0.1
Cs ppm	2.3 ± 0.6	1.3 ± 0.3
Sm ppm	1.83 ± 0.08	0.9 ± 0.05
Yb ppm	1.4 ± 0.4	0.9 ± 0.3
Hf ppm	1.5 ± 0.3	0.5 ± 0.2
Sb ppm	1.3 ± 0.1	0.9 ± 0.1
Au ppm	260 ± 10	94 ± 6

Table 5. *The content in micro- elements and inframicro-elements in the white seabuckthorn fruit ashes (Şerbăneşti l biotype) established by neutron activation*

Microelement	Date 1-4. X Media ± s	Date 2 -27. XI Media ± s
Al %	0.263 ± 0.003	0.60 ± 0.011
As ppm	0.4 ± 0.2	0.9 ± 0.3
Au ppb	509 ± 15	55 ± 8
Br ppm	27 ± 1	50 ± 1
Ca %	5.12 ± 0.50	4.55 ± 0.45
Co ppm	5.7 ± 0.4	5.3 ± 0.8
Cr ppm	73 ± 4	21 ± 2
Fe %	0.588 ± 0.024	0.474 ± 0.019
K %	17,22 ± 0,69	24.38 ± 0.98
La ppm	1.7 ± 0.1	1.4 ± 0.1
Mg %	3.24 ± 0.22	4.89 ± 0.34
Mn %	270 ± 8	480 ± 12
Mo ppm	50 ± 5	88 ± 6
Na %	0.6986 ± 0.070	0.6256 ± 0,063
Rb ppm	43 ± 4	31 ± 3
Sb ppm	0,76 ± 0.11	0.57 ± 0.09
Se ppm	0.39 ± 0.01	0.45 ± 0.01
Se ppm	5.7 ± 0.4	3.5 ± 0.3
Sm ppm	0.21 ± 0.02	0.31 ± 0.03
Sr ppm	0.21 ± 0.02	279 ± 28
U ppm	sub 1	sub 1
V ppm	4.3 ± 0.5	5.3 ± 0.8
Zn ppm	10.89 ± 25	7.26 ± 3.5

Table 6. *The result of the biochemical analysis done on some seabuckthom biotypes (Ciumaşi, Sf. Gheorghe 4, 5, 6, 9, 10, Şerbăneşti 1, 4, Şerpeni 11, Without thorns, Delta 40 M) selected at Bacău*

Mentions	**Media ± s**	**The biotype limits**
Average Production fruits (1991-1994) Kg /plant t / ha	18.09 ± 3.40 18.197 ± 3.425	5.13 - 87.97 8.747 - 27.939
Hydrosolubile Substance (%)	12.9 ± 3.7	11.5 -15.0
Ascorbic acid (mg%)	149.06 ± 82.90	22.21 - 260.52
Total Acidity (% d.s. Malic acid)	7.6170 ± 2.0624	4.1059 -11.5487
Average Production (1991-1994) green substance t/ha dry substance t/ha	18.179 ± 7.34 3.33 ± 1.25	8.547 - 27.939 1.666 - 5.231
Oil g%t/ha	14.4671 ± 4.3319 480 ± 303	8.8800 - 21.9402 168 -1147
α-, β-, γ-caroten mg/100 g g/ha	24.12 ± 12.30 135 ± 145	6.27-42.72 28-473
Serotonin in fruits µg/g g/ha	27.28 ± 7.81 95.4 ± 53.4	16.72 - 41.59 26.80 -18.11
Serotonin in leaves µg/g g/ha	37.48 ± 8.07 143.7 ± 104.3	26.39 - 48.48 48.9 - 403.8
Microelements in leaves		
Mentions	**Media ± s**	**The biotype limits**
Zn ppm g/ha	14.7 ± 3.9 49 ± 20	10.5 - 23.3 13 - 79
Cu ppm g/ha	5.8 ± 2,8 21 ± 16	3,00 - 13,75 3.5 - 64.0
Mn ppm g/ha	55.4 ± 18.5 170 ± 54	31.8 - 89.3 85 - 244
Fe ppm g/ha	164 ± 73 853 ± 371	135 - 340 286 – 1648
B ppm g/ha	84 ± 27 267 ± 142	58.5 - 153.0 70 - 543
Mo ppm g/ha	2.52 ± 1.58 10 ± 6	0.65 - 6.24 1 - 22
Microelements in fruits		
Zn ppm g/ha	12.1 ± 2.5 49 ± 17	8 - 16 18 - 67
Cu ppm g/ha	5.77 ± 1.66 19 ± 8	3.00 - 9.25 3.4 - 33.0
Mn ppm g/ha	9.95 ± 2.18 33 ± 7	6.8 - 15.5 12 - 72
Fe ppm g/ha	173 ± 130 576 ± 487	53 - 428 97 - 1690
B ppm g/ha	14,2 ± 6.6 50.2 ± 30	3.5 - 19.0 5.2 - 99.0
Mo ppm g/ha	2.65 ± 0.98 9 ± 4	1.43 - 5.07 2 - 7

Table 7. *The variation in content of serotonin from hydrophonic (watery) extract from dry leaves and fruits, gathered from 11 types of seabuckthorn populations selected at S.C. Fructex S.A. Bacău*

Population	**Dry Fruits (µg/g)**	**Dry Leaves (µg/g)**
Without thorns	16.72	27.27
Sf. Gheorghe 5	25.30	40.00
Sf. Gheorghe 6	29.99	49.37
Şerbăneşti 1	17.90	45.27
Ciumaşi	41.59	43.29
Şerbăneşti 4	27.33	42.01
Sf. Gheorghe 9	39.12	26.39
Delta 60 M	26.36	33.05
Sf.Gheorghe 10	27.44	35.88
Şerpeni 11	21.31	48.48
Sf. Gheorghe 4	29.69	26.63
Media ± s	**22.23 ± 7.73**	**27.97 ± 14.42**

Table 8. *The variation in content of serotonin from dry seabuckthorn fruits, leaves, offshoots and offshoot barks depending on the origin.*

Fruits types	**µg/g serotonin**	**Types**	**µg/g serotonin**
Plafar Bucharest	31	Leaves	49
Plafar Craiova	30	Offshoots	31
Plafar Buzău	32	Offshoots barks	190

The serotonin analyzed was from seabuckthorn fruits obtained from different sources (Bucuresti, Buzau and Craiova). In *Table 8*, there could be seen a slightly smaller difference expressed in micrograms/gram of serotonin, the quantity was necessary and sufficient to have some effects from 8-10 g of dry substance daily, that corresponds to 40-50 g of fruits.

Conclusions

1. The sea buckthorn powder represents a true accumulator of microelements among which boron, iron, zinc, manganese, cobalt, molybdenum. In the case of the sea buckthorn, the microelements are combined as complex chelatic derivates. That is why, the sea buckthorn microelements are bioavailable.

Remarkable results have been obtained in the treatment of the sea buckthorn oil in different dilutions and associated with 1% gentian violet of the wounds. The healing was rapid and with no secondary infections.

Good results have been obtained as well in the treatment of the infectious pod dermatitis with preparations based on sea buckthorn oil, associated in different dilutions with substances whose effects produce an indirect synergy.

The results obtained with respect to applying the sea buckthorn preparations as a medicinal remedy have lead to the conclusion that this remedy provides the maximum efficiency in the treatment of some diseases.

2. Serotonin's effects are well known: immune-inductor, energizing, anti-depressive, chemical mediator, bearing implications on such disabilities as insufficiency in transmitting the information through the nervous system to the organs.

The serotonin alongside with the other substances from sea buckthorn are recommended for sickness or discomfort, too, mostly concerning the severe conditions that affect the 21st century such as cancer, HIV, depression, anxiety, suicide tendencies, insomnia, alcohol abuse, schizophrenia, any disease generated by a physical, chemical or biological agent. *(*Koslowski, 1970; Morichi, 1977; Takasahi, 1983; Muster, 1984*)*.

References

Bazarova A. (1978) *„The seabuckthorn - a drug remedium into the tibethan medicine”,* Răstit. Resursi, XIV

Brad I.L., Silva F., Cojocariu O., Jokl E., Marcu Z., Martinovski G., Voicu E. (1976) *„The dinamic's of some active principles from the fruits of Hippophae rhamnoides”,* Analele ICCPT vol.XII; p.1-9, 65 pag.

Brad L, Medeşan C. (1987), *„Research regarding the immune-inductor effect of some white seabuckthorn breeds”,* Symposium "Oltenia medicală", May 30-31, Craiova

Brad I., Ţigănuş V., Brad I.L., Raţi L., Raţi V., Acatrinei F.(1997), *„A comparative research of the ash content (total mineralization) and micro-elements at 11 white seabuckthorn breeds, selected at S.C. FRUCTEX S.A. Bacău*", Symposium "50 years since the Inauguration of The Agronomic Institute Timişoara", June 1-2, Timişoara

Brad I., Brad I.L., Radu F.(2002), *"White Seabuckthorn - a pharmacy into a plant"* , Bucharest, Editura Tehnică, p. 114,115,116; 178 pag.

Brad I., Brad I.L., Vlăsceanu G.A., Manea Şt., Tamaş V., (2007), „*Usage of lipidic extract from dried sea buckthorn fruit (Hippophae rhamnoides L.) in burns and wounds of the members*", ISA 2007, Quebec, Canada.

Cojocaru V., Brad L.(1984), *"Establishing the content of*

micro-elements in the white seabuckthorn through neutron activation", Symposium "Oltenia medicală", June 2-3, Craiova

Koslowski I. (1970) *„The Phisiopathology and the general treatment of the grave burn"*, Surgery, p. 9, 385-390, 410 pag.

Mathiev N.K.,Azizov F.S.(1981)*„Biological active agents of the fruit of the sea buckthorn*", Izv. Akad.Nauk, 6, p.118-123, 280 pag.

Morichi A.(1977) *„The vitel prognostic of burns",* Anesthesia, Analgesia, Reanimation, nr. 34, p.1293-1302, 1560 pag.

Munster A.(1984) *„The immunological answer to traumas and burns",* American Journal of Medicine, nr.76 ;3 A; p.142-145, 251 pag.

Puhalskaia E., (2000*) „The seabuckthorn extracts effect on the thumoral transplants at the animals",.* Bull.Exlt. Biol.Med., 45, p.363-364, 470 pag.

Talichova L.(1998) *„Morphological change in the rat liver under sea buckthorn oil administration against a background of alcohol poisoning"*, Experimental Pathology Pehm, 3, p.177-184, 784 pag.

Uluitu M., Brad L.(1984) *"Establishing the content of serotonin in the organs of white seabuckthorn",* Symposium "Oltenia medicală", June 2-3, Craiova

Uluitu M., Brad L, Chiş R., Brad I.L., Rați I.V., Rați L., Acatrinei F.(1997), *"Variation in seabuckthorn population (Hippophae rhamnoides) selected at S.C. Fructex S.A. Bacău".* Symposium „50 years since the inauguration of The Agronomic Institute Timişoara", June 1-3, Timişoara

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