

## RESEARCH ON HIGH-POTENTIAL BIOTYPES OF SEA-BUCKTHORN (*HIPPÖPHAE RHAMNOIDES* L.) FOR PROMOTING THEM IN IMPROVEMENT WORKS AND ECOLOGICAL PLANTATIONS

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### INTRODUCTION

*Hippöphae rhamnoides* ( $2n = 24$ ), known as sea-buckthorn belongs to the order *Eleagnales* in the family *Eleagnaceae* (M. Botez, Bădescu Gh., Botar A., 1984). Sea-buckthorn is considered one of the most valuable species of fruit shrubs from the spontaneous flora. Its application areas are extremely broad, yielding spectacular effects in human and veterinary medicine, animal husbandry, cosmetics, agriculture, microbiology, food industry, forestry. In landscape architecture it is used as an ornamental plant due to its particularly nice foliage and yellow orange fruit that remain on the plant during winter, being also a plant that participates in the spiritual and cultural ties between people (Beldeanu C., 1972).

The research conducted in the country and abroad have shown that the sea-buckthorn leaves, shoots and fruits contain a number of biologically active substances with an essential role in regulating metabolism. Buckthorn fruits are regarded as natural multivitamins as they are rich in the main vitamins (A, B1, B6, C, E, F, K, P). The vitamin C content of sea-buckthorn not only surpasses that of all other indigenous fruit species, such as the black currant, but even that of citrus fruits (lemon) by over 10 times. Only dog-rose fruits may sometimes exceed the amount for 100 g of fresh matter, depending on area conditions. In the highest mountainous areas, sea-buckthorn varieties may exceed 1.500 mg / 100 g, considerably exceeding the dog-rose vitamin C content (Brad I., Brad Ioana, Radu Florica, 2002).

The high content of vitamins, associated with numerous micronutrients that are found in sea-buckthorn fruits, led to a variety of uses in the food industry, being used either alone or mixed with other fruits to prepare juices, syrups, jams, jelly, nectar, marmalade, candy fillings, liquor etc. (Bendorf Florica, Georgescu Alexandra, Marchidan Alina, 1997).

The plant contributes to increasing forest areas by fighting erosion of degraded soils (*due to its*

*exceptional rooting potential, emitting suckers up to 24 m away from the trunk*), helping soil improvement by assimilating atmospheric nitrogen directly through the roots nodosities formed in symbiosis with the *Actinomycete* fungi (Panaitescu C. and Podeanu Gh., 1966).

Its long and rigid thorns made this species to be used for impenetrable fences that may surround orchards instead of wire fences (Haralamb A. T., 1986). It is also particularly valuable as a melliferous plant. It can be successfully used in animal fodder, adding special nutrients to it. Sea-buckthorn has multiple uses and its fields of application are constantly expanding.

This study aimed to morphologically and physiologically highlight the most valuable sea-buckthorn biotypes from the collection of germplasm of Fructex Bacău, in order to conduct full description of these and identify the biotypes suitable to soil improvement works, with superior qualities needed to create new varieties, which can be subsequently used to establish organic crops.

### RESEARCH MATERIAL AND RESEARCH METHODS

The biological material used in the selection study was the collection of 61 sea-buckthorn biotypes selected from 12 counties from Romania. The sea-buckthorn germplasm collection is located in the research field of the Fructex company and covers an area of 2 ha. The plantation is located in the meadow of Bistrița River on a typically alluvial soil. The planting distances between sea-buckthorn individuals are 3 m between rows and 1.5 m between plants in a row, generating a corresponding density of 2211 plants per ha. This culture system allows mechanical work on the rows. The pruning of the plant crown is free with

orientation along the row direction, ensuring a balance between the vegetative and the generative areas.

To determine the properties of production and behaviour under circumstances of growing the biotypes from the collection of germplasm, there were carried out, in 2013, morphological measurements (colour, type of fructification) and biometric measurements (fruit weight, weight of 100 fruits, large diameter, small diameter, shape index – the ratio between the large and small diameter) of fruits, as well as physiological ones (dry matter content of fruit) according to the biotypes categories based on their ripening time.

The ripening period has been established as follows: very early ripening takes place in July; early ripening period, August 1-15; middle ripening period, August 16-30; late ripening period, September 1-15; very late ripening period, after September 15.

Biometric measurements were performed in the laboratory using digital callipers and the analytical scales, and measuring the amount of dry matter % in the sea-buckthorn fruit pulp was performed with a Zeiss refractometer.

## RESULTS AND DISCUSSIONS

The studied sea-buckthorn biotypes were classified into five ripening periods as follows: a very early biotype (*July*), two biotypes with early ripening (*1-15 August*), twenty-two biotypes with middle ripening (*16-30 August*), fourteen biotypes with late ripening (*1-15 September*) and twenty-two biotypes with very late ripening (*after September 15*) (Figure 1).

Following the interpretation of the biometric data according to the ripening period, it was established that biotype R2 / 43-45 (*Table 1*) with very early ripening produces very small fruits (*0.09 g / fruit*) with a number of 1112 fruits per 100g fruits and the shape index value of 1.778% circumscribing this biotype within the category of elongated fruits.

In the middle ripening group (*Table 2*), there were identified twenty-two biotypes with fruit weight values between 0.11-0.30 g/fruit, values corresponding to a number of 917-333 fruits per 100 g of fruits.

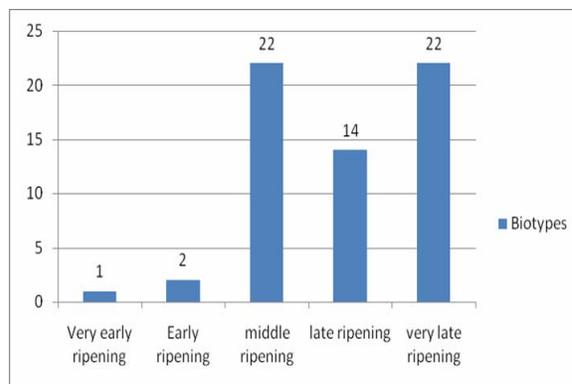


Fig. 1. Classification of sea-buckthorn biotypes according to the ripening time – 61 studied biotypes

With less than 400 fruits per 100 g there were highlighted five biotypes, respectively R1/101, R2/116, R2/118, R3/5. Eleven biotypes recorded between 400-600 fruits per 100 g, namely: R1/107-113, R2/1, R2/2-9, R2/95,98, R2/109-111, R3/1, R3/6,7, R11/4-8, R16/1, R18/2,3,5,6, R20/5,6,7. With more than 600 fruits per 100 g there were highlighted six biotypes: R2/101-108, R3/111-120, R4/67-75, R9/92-98, R10/4, 5, 7, R19/1.

The average weight below 0.20 g was found at 10 biotypes: R2/1, R2/2-9, R2/101-108, R2/109-111, R3/1, R3/111-120, R4/67-75, R9/92-98, R10/4,5,7, R19/1; the average weight above 0.20 g was found at 12 biotypes: R1/101, R1/107-113, R2/95,98, R2/116, R2/118, R3/5, R3/6,7, R11/4-8, R16/1, R18/2,3,5,6, R20/5,6,7 (*Table*).

We note the fact that no biotype with middle ripening recorded values lower than 1, and the biotypes with shape index values close to 1 have a round shape, and those with shape index values close to 2 has an elongated shape. The biotypes R20/5,6,7 - R16/1 highlight the shape index value between 1.10 and 1.76%.

From the biotypes with late ripening, there were highlighted 14 biotypes with fruit weight values between 0.09 - 0.27 g / fruit and 377-1031 fruits per 100 g (R13/1-9 – R1/98) (*Table 3*).

Table 1. Biometric measurements of fruits from the selected biotypes with very early and early ripening, Bacău, 2013

Ripening period	Biotype	Number of fruits in 100 g	Fruit weight (g)	Large Diameter (mm)	Small Diameter (mm)	Shape index
Very early ripening	R2/43-45	1112	0,09	7,40	4,16	1,78
Early ripening	R2/97,99,100	128	0,79	6,96	5,89	1,18
	R12/97-105	1539	0,07	5,95	4,98	1,20

Table 2. Biometric measurements of fruits from the selected biotypes with middle ripening, Bacău, 2013

No.	Ripening period	Biotype	Number of fruits in 100 g	Fruit weight (g)	Large Diameter (mm)	Small Diameter (mm)	Shape index
1	middle ripening	R1/101	388	0,26	9,93	5,99	1,66
2		R1/107-113	476	0,21	6,86	6,11	1,12
3		R2/1	592	0,17	8,64	5,41	1,60
4		R2/2-9	535	0,19	8,21	6,12	1,34
5		R2/95,98	469	0,21	7,55	6,36	1,19
6		R2/101-108	613	0,16	8,10	5,36	1,51
7		R2/109-111	588	0,17	8,58	5,21	1,65
8		R2/116	394	0,25	8,51	6,54	1,30
9		R2/118	323	0,31	8,53	7,46	1,14
10		R3/1	575	0,17	8,38	6,08	1,38
11		R3/5	289	0,35	8,89	7,10	1,25
12		R3/6,7	417	0,24	8,49	5,95	1,43
13		R3/111-120	667	0,15	8,56	6,54	1,31
14		R4/67-75	730	0,14	7,83	5,32	1,47
15		R9/92-98	917	0,11	6,84	5,25	1,30
16		R10/4,5,7	671	0,15	10,08	7,37	1,37
17		R11/4-8	476	0,21	9,88	6,76	1,46
18		R16/1	420	0,24	8,76	4,99	1,76
19		R18/2,3,5,6	427	0,23	8,53	5,81	1,47
20		R19/1	699	0,14	8,12	5,11	1,59
21		R20/5,6,7	476	0,21	7,45	6,79	1,10
22		Jud. Neamt	333	0,30	9,56	7,25	1,32

Table 3. Biometric measurements of fruits from the selected biotypes with late ripening, Bacău, 2013

No.	Ripening period	Biotype	Number of fruits in 100 g	Fruit weight (g)	Large Diameter (mm)	Small diameter (mm)	Shape index
1	late ripening	R1/98	377	0,27	9,13	6,52	1,40
2		R2/52-56	971	0,10	6,74	5,03	1,34
3		R3/100-109	649	0,15	8,91	4,85	1,84
4		R4/89-99	485	0,21	7,48	5,78	1,30
5		R4/109-119	649	0,15	6,74	5,19	1,30
6		R5/1-42	455	0,22	10,92	5,94	1,84
7		R5/61-69	613	0,16	6,75	5,10	1,32
8		R6/1-6	429	0,23	7,95	6,09	1,31
9		R6/114-117	538	0,19	8,13	6,31	1,29
10		R7/92-100	455	0,22	7,80	6,37	1,22
11		R9/46-53	435	0,23	7,72	6,53	1,18
12		R11/31-40	452	0,22	7,99	6,13	1,30
13		R13/1-9	1031	0,09	7,60	4,57	1,66
14		R13/110-112	391	0,26	9,15	6,12	1,50

The value of the shape index falls between 1.18 % - the biotype R9/46-53 and 1.84 % -the biotype R5/1-42, hence the fruits have an elongated shape.

Regarding the very late ripening period (Table 4), there were determined 22 biotypes with fruit weight values between 0.09 – 0.33 g/ fruit, corresponding to 303 - 1064 fruits per 100 g.

With less than 400 fruits per 100 g there were highlighted eight biotypes: R1/23-37, R1/76-78, R1/87-95, R3/97-99, R6/25-31, R9/34-35, R12/89-97, R13/93-10; with 400 – 600 fruits per 100 g, nine

biotypes: R1/6-11, R3/50-51, R3/71-82, R3/83-88, R4/40-46, R4/47-54, R9/19-26, R11/48-54, R11/92-99. Five biotypes recorded over 600 fruits per 100 g: R2/18-22, R5/113-115, R8/2-7, R9/99-107, R12/9.

Ten biotypes recorded a fruit weight below 0.20 g: R2/18-22, R3/71-82, R3/83-88, R4/40-46, R4/47-54, R5/113-115, R8/2-7, R9/19-26, R9/99-107, R12/9. Two biotypes recorded a fruit weight above 0.20 g: R1/6-11, R1/23-37, R1/76-78, R1/87-95, R3/50-51, R3/97-99, R6/25-31, R9/34-35, R11/48-54, R11/92-99, R12/89-97, R13/93-101.

Table 4. Biometric measurements of fruits from the selected biotypes with very late ripening, Bacău, 2013

No.	Ripening period	Biotype	Number of fruits in 100 g	Fruit weight (g)	Large Diameter (mm)	Small diameter (mm)	Shape index
1	very late ripening	R1/6-11	476	0,21	10,16	6,66	1,53
2		R1/23-37	303	0,33	8,99	6,10	1,47
3		R1/76-78	379	0,26	8,73	7,17	1,22
4		R1/87-95	341	0,29	8,57	6,52	1,31
5		R2/18-22	735	0,14	6,15	5,83	1,06
6		R3/50-51	488	0,21	8,10	6,27	1,29
7		R3/71-82	581	0,17	9,27	6,43	1,44
8		R3/83-88	524	0,19	7,13	5,12	1,39
9		R3/97-99	312	0,32	9,15	7,36	1,24
10		R4/40-46	559	0,18	9,70	5,38	1,80
11		R4/47-54	575	0,17	10,14	5,60	1,81
12		R5/113-115	741	0,14	7,14	6,14	1,16
13		R6/25-31	357	0,28	9,94	6,70	1,48
14		R8/2-7	667	0,15	7,86	4,48	1,76
15		R9/19-26	526	0,19	9,49	5,83	1,63
16		R9/34-35	313	0,32	7,99	7,31	1,09
17		R9/99-107	1064	0,09	6,76	4,84	1,40
18		R11/48-54	476	0,21	10,15	6,48	1,57
19		R11/92-99	417	0,24	10,08	6,46	1,56
20		R12/9	704	0,14	8,35	5,26	1,59
21		R12/89-97	323	0,31	8,65	6,81	1,27
22		R13/93-101	369	0,27	9,45	6,66	1,42

The biotypes with shape index values close to 1 have a round shape and the values close to 2 have an elongated shape. The lowest shape index value (1.06) was at the biotype R2/18-22, and the highest value (1.81) at biotype R4/47-54.

The biotype with very early ripening period (July) record a dry matter content of 12.1%, the two

biotypes with early ripening (1-15 August) a dry matter content between 12.68 and 13.88%. The biotypes with middle ripening period (16-30 August) recorded either very low values of dry matter content (9.30 – 10.52%) or very high values (17.98 – 18.76) (Table 5).

Table 5. Comparative study of the dry matter content according to the ripening period, Bacău, 2013

No	Very early ripening		Early ripening		Middle ripening		Late ripening		Very late ripening	
	Biotype	Dry matter %	Biotype	Dry matter %	Biotype	Dry matter %	Biotype	Dry matter %	Biotype	Dry matter %
1	R2/43-45	12,10	R2/97,99,100	12,68	R1/101	12,98	R1/98	14,74	R1/6-11	13,60
2			R12/97-105	13,88	R1/107-113	16,08	R2/52-56	11,76	R1/23-37	11,00
3					R2/1	15,16	R3/100-109	15,76	R1/76-78	11,42
4					R2/2-9	10,52	R4/89-99	12,52	R1/87-95	12,46
5					R2/95,98	14,86	R4/109-119	14,02	R2/18-22	13,00
6					R2/101-108	17,60	R5/1-42	13,66	R3/50-51	13,04
7					R2/109-111	17,94	R5/61-69	16,56	R3/71-82	13,96
8					R2/116	13,44	R6/1-6	13,60	R3/83-88	14,86
9					R2/118	9,30	R6/114-117	13,42	R3/97-99	12,28
10					R3/1	18,76	R7/92-100	13,06	R4/40-46	13,70
11					R3/5	12,00	R9/46-53	15,54	R4/47-54	14,52
12					R3/6,7	14,66	R11/31-40	12,48	R5/113-115	14,60
13					R3/111-120	11,08	R13/1-9	17,66	R6/25-31	12,58
14					R4/67-75	15,34	R13/110-112	13,76	R8/2-7	16,50
15					R9/92-98	16,64			R9/19-26	12,90
16					R10/4,5,7	11,98			R9/34-35	12,18
17					R11/4-8	12,58			R9/99-107	17,66
18					R16/1	11,76			R11/48-54	13,40
19					R18/2,3,5,6	15,30			R11/92-99	14,52
20					R19/1	16,30			R12/9	12,96
21					R20/5,6,7	14,02			R12/89-97	12,40
22					Jud. Neamt	14,58			R13/93-101	14,30

The biotypes with late ripening (*1-15 September*) showed low levels of dry matter content, respectively from 11.76 to 13.60%, as well as high levels, from 16.56 to 17.66%.

The biotypes with very late ripening (*after September 15*) recorded values of dry matter content between 11.00 and 16.50%. The studied biotypes were classified into five groups, according to fruit colour, as follows: 10 biotypes with yellow fruits, 14 yellow-orange biotypes, 27 orange biotypes, 4 orange-tile-coloured biotypes, five tile-coloured biotypes (Table 6, Fig. 2-6).

Table 6. The biodiversity of sea-buckthorn biotypes according to berry colour, Bacău, 2013

No	Yellow fruits	Yellow-orange fruits	Orange fruits	Orange-tile-coloured fruits	Tile-coloured fruits
1	R1/23-37	R1/6-11	R1/97-98	R1/101	R2/1
2	R4/40-46	R1/76-78	R2/18-22	R1/107-122	R2/43-45
3	R4/47-54	R1/87-95	R2/52-56	R2/2-9	R2/95,98
4	R4/67-75	R3/50-51	R2/101-108	R2/116-117	R2/97,99,100
5	R8/2-7	R3/97-99	R2/109-111		R12/98-106
6	R9/34-35	R3/100-109	R2/118		
7	R11/92-99	R5/113-115	R3/1		
8	R12/4-9	R6/25-31	R3/5		
9	R12/89-97	R9/19-26	R3/6,7		
10	R13/93-101	R9/99-107	R3/71-82		
11		R11/4-8	R3/83-88		
12		R11/31-40	R3/111-120		
13		R13/110-112	R4/89-99		
14		R19/1	R4/109-120		
15			R5/1-42		
16			R6/1-6		
17			R6/114-117		
18			R7/92-100		
19			R9/46-53		
20			R9/92-98		
21			R10/4,5,7		
22			R11/48-54		
23			R13/1-9		
24			R16/1		
25			R18/2,3,5,6		
26			R20/5,6,7		



Fig. 2 Yellow - 10 biotypes



Fig. 3 Yellow-orange fruits - 14 biotypes



Fig. 4. Orange - 27 biotypes



Fig. 5. Orange-tile-coloured fruits - 4 biotypes



Fig. 6 .Tile-coloured fruits - 5 biotypes

The production potential is directly dependent on the distribution of fruits along the shoots (the kind of fruiting). After morphological observations of sea-buckthorn, there were identified 13 biotypes with rare distribution of fruits along the shoots, 35 biotypes with dense distribution, 11 biotypes with very dense distribution and 2 biotypes with very dense distribution ( Fig. 7-10, Table 7).



Fig. 7. R2/115 Biotype with a rare distribution of fruits along the shoots - 13 biotypes



Fig. 8. R2/102-109 Biotype with a dense distribution of fruits along the shoots - 35 biotypes



Fig. 9. R2/117 Biotype with a very dense distribution of fruits along the shoots - 11 biotypes



Fig. 10. R9/46-53 Biotype with a very very dense distribution of fruits along the shoots - 2 biotypes

Table 7. Biometric observations of fruit distribution along the shoots, Bacău, 2013

No	Fruit distribution along the shoots			
	1- rare distribution	2- dense distribution	3- very dense distribution	4- very very dense distribution
1	R2/43-45	R1/23-37	R1/6-11	R3/5
2	R3/50-51	R1/87-95	R1/76-78	R3/111-120
3	R3/71-82	R1/98	R1/107-113	
4	R3/83-88	R1/101	R2/1	
5	R3/100-109	R2/52-56	R2/2-9	
6	R4/40-46	R2/95,98	R2/18-22	
7	R8/2-7	R2/97,99,100	R2/116	
8	R9/19-26	R2/101-108	R2/118	
9	R9/34-35	R2/109-111	R3/97-99	
10	R11/92-99	R3/1	R6/114-117	
11	R12/97-105	R3/6,7	R12/89-97	
12	R13/1-9	R4/47-54		
13	R13/93-101	R4/67-75		
14		R4/89-99		
15		R4/109-119		
16		R5/1-42		
17		R5/61-69		
18		R5/113-115		
19		R6/1-6		
20		R6/25-31		
21		R7/92-100		
22		R9/46-53		
23		R9/92-98		
24		R9/99-107		
25		R10/4,5,7		
26		R11/4-8		
27		R11/31-40		
28		R11/48-54		
29		R12/9		
30		R13/110-112		
31		R16/1		
32		R18/2,3,5,6		
33		R19/1		
34		R20/5,6,7		

## CONCLUSIONS

The research results have allowed the classification of sea-buckthorn biotypes from the germplasm collection into five groups based on the ripening period, six groups based on the colour of mature fruits and four groups based on the distribution of fruits along the shoots.

Of the 61 studied biotypes, there were identified, according to ripening time, the following biotypes: 1 biotype with very early ripening, 2 biotypes with early ripening, 22 biotypes with middle ripening, 14 biotypes with late ripening and 22 biotypes with very late ripening.

The biotypes studied according to their ripening period ensure spaced-out harvesting over more than 75 days, which allows good technological management.

There were highlighted 14 biotypes with more than 15% dry matter. The highest content was found at biotypes: R3/1 – 18.76%, R2/109-111 – 17.94% and R2/101-108 – 17.6%.

The biotypes studied in terms of fruit colour were classified into five groups: 10 yellow biotypes, 14 orange-yellow biotypes, 27 orange biotypes, 4 orange-tile-coloured biotypes, 5 tile-coloured biotypes.

The production potential is directly dependent on the distribution of fruits along the shoots (the type of fruiting). There were identified 13 biotypes with rare distribution of fruits along the shoots, 35 biotypes with dense distribution, 11 biotypes with very dense distribution and 2 biotypes with very very dense distribution.

The studied biotypes enable the selection of the most valuable biotypes and their promotion in improvement works to create new varieties and complete the selection in order to obtain certified biological material.

Certified seeding material allows the establishment of fruit shrubs plantations and the absorption of Community funds for organic crops.

## ABSTRACT

Sea-buckthorn, besides being considered the “Romanian ginseng” or “divine oil” due to its therapeutic properties, is also a species that can bring substantial income to those who want to develop a business in this area. This study is intended to become a tool in this respect and also a way to promote this miraculous plant, a plantation model for people interested in ecological farming who want to access European funds allotted to this type of plantations.

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