# MORPHOLOGICAL DESCRIPTION OF SEA BUCKTHORN "STAR" MALE PLANT - FEMALE PLANTS POLLINATION ELEMENT (POLLINATORS)

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# INTRODUCTION

*Hippöphae rhamnoides* L., (2n = 24) called sea buckthorn is part of the Eleagnales Order, fam. Eleagnaceae.

It is a species that grows in the spontaneous flora and is presented in the form of a shrub with strong thornes, with unsexed deciduous flowers, with female plants of the floral formula: K4-2CoA4G1 and male plants as pollinators.

It has a high port ranging from 1.5 to 4 m, and under favorable conditions it can grow 8-10 m height and a trunk diameter of over 15 cm. Distributed grouped cultivars of the same sex have the same hereditary attributes as those originally created from the primary one, from which they have been vegetative propagated via root suckers. The area occupied by a clonal group is small and its extension made in a centrifuged manner. Shading favors the loss of vegetative buds, a phenomenon called divestment, the area being overwhelmed by other species (Rați I. V., Rați Luminița, 2001).

The center of the sea buckthorn area is better prepared to withstand the action of the unfavorable factor, so the sea buckthorn zone is convex. The clonal group represents the total of the descendants coming from a vegetative subject (same sex). Concrete clonal group - is one part of the clonal group that has common phenotypic properties (force, productivity). The biometric characteristics of a clonal group are: • the peripheral area which is the young area, 1-3 years old and 1 m high;

• middle area of 4-10 years, with a height of 1-2 m, very productive and

• central area older than 10 years, with heights greater than 2 m.

In order to fructify sea buckthorn plants, there is a need a very good pollination that is usually done with the help of wind and insects from male plants.

Female plants (produce the fruit) and male plants (they only act as pollinators) have different traits. There are several ways of placing male plants near female plants, so pollination of female plants can be achieved in a 100% proportion. Male plants are more vigorous than female plants. They have longer, thicker and darker annual branches, also larger, elongated buds, well grouped on branches, and the bud individually has the appearance of a grape. Fructex Bacau Company has approved sea buckthorn cultivars of female plants for which male plants have been recommended. For this purpose, a good pollinator was created within the company mainly for the specific cultivars. This was created as a result of the researches undertaken to obtain the Star cultivar (Rați I. V., et al., 2013). This cultivar has an erect growth; it develops more on vertical, which allows maintaining the planting distance between the female plants and the male ones. It loads very well with buds that generate large amounts of pollen (Fig.1) ensuring good pollination.



Fig. 1 STAR plant cultivar male with pollen buds

# MATERIAL AND METHODS

Observations and determinations were made in the experimental field located at Fructex Bacau in the northern part of the town, on the Bistrita river meadow.

The pedoclimatic characteristics of the experimental field are: molluscular alluvial soil, carbonless without horizon B, sometimes with ballast spots. It has moderately alkaline pH 8.5, medium content of carbonate (7.2%) and low content of humus (1.2%).

The climate of the area is temperate continental with aridity caused by continental invasion, which can be frequently excessively cold in winter and excessively hot and dry in the summer. The average multianual temperature for the studied area is  $9.2 \degree$  C. The coldest month of the year is January with an average of - 4.2 C and the warmer the month is July the average value is of 20.5 C.

The average rainfall is below 30 mm between December and March and more than 40 mm for the April-October period. Most rainfall between 87.7-90.3 mm was recorded in June.

The multi-annual average does not exceed 540 mm. The relative humidity of the air is generally high, due to the presence of Siret and Bistrita courses and the accumulation of existing water. The major meadows in the studied area provide shelter against Western air masses. In their descent on the western slope of the Pietricica Peak, the masses of air suffer sewage processes along the Bistrita and Siret Valleys, where the experimental cultures are located (Rați I. V., Rați Luminița, 2001)

On this area, there are maintained, homologated and patented by Fructex Bacau seven female cultivars Auras, Serpenta, Victoria, Ovidiu, Silvia, Diana and Tiberiu (Rați I. V., Rați Luminița, 2003).

These cultivars are used as "BASE" plants as well as mother plantations/main and they have been used in experiments for the productivity test. This test involves determining the plant production (yield) quantities in kg of fruit per plant produced by female plants and the ability to pollinate of the Star cultivar. Research has been done on several male plants, recommending a male plant with a temporary name- STAR, a cultivar that aims to be the unique pollinator for the seven female cultivars. The application for certification was filed with the number 278 / 22.08.2014, the surveillance research ISTIS and OSIM had been carried on for a period of over four years.

The agro technological conditions used in the experimental field are 3 m between rows and 1.5 m per row. On a row of male plants correspond to seven rows of female plants. For these planting distances corresponds to a density of 2211 male and female plants per hectare.

They are conducted in free form towards the direction of the row, ensuring that the female plants benefit a proper balance between the vegetative area (growth of the shoots) and the generative fruit growth.

### **Biological Material**

In order to ensure a good pollination of the cultivars, seven cultivars of female plants obtained at Fructex Bacau company: Auras, Victoria, Serpenta, Ovidiu, Tiberiu, Silvia and Diana varieties were used.

The male biotype proposed for certification was selected as a result of a previous research that was considered the best pollinator for female cultivars created within the company.

### Materials and Methods of Research

The characterization was carried out according to the UPOV- International Union for the Protection of Vegetable Creations issued by OSIM - STATE OFFICE FOR INVENTIONS AND TRADE MARKS and ISTIS - STATE INSTITUTE FOR TESTING AND REGISTRATION OF VARIETIES which focused on/followed the expression of the characters, the type/reference of cultivar and the mark.

The harvesting of the pollen was made from sea buckthorn shoots with pollen-filled buds which were brought to the laboratory (Fig. 2).



Fig. 2. Obtaining pollen from buds using test shingle, temporary storage of pollen in Petri dishes

The anthers were removed from the filament by rubbing the buds on a sieve, and by pulling them off with the fingers or tweezers, the pollen was dried off by letting it cool in Petri dishes (Fig. 2).

Preservation was carried out in ethyl alcohol until cytogenetic preparations and kept cold.

The obtained pollen was used for describing and determining the germination capacity (according to V. Cociu research method). Observations for pollen description were made using the Euromex microscope. The pollen used for the observations was colored with Orchool solution and Lugol solution

The UPOV-TG / 240/1 and CPVO TP 240/1 protocols were used to perform the Test for Distinctness, Uniformity and Stability Test (DUS) for male sea buckthorn. The productivity test was carried out in a fruit production plantation/orchard with seven female cultivars, the results being presented in another scientific study.

# **RESULTS AND DISCUSSION**

# Description of the STAR cultivar - male pollinator for female sea buckthorn plants.

The morphological and phenotypic

characteristics highlighted by UPOV have been used in the ISTIS and OSIM inspections (see table 1).

The STAR cultivar is a male plant (Fig. 3) which ensures a very good pollination for female plants.

The description of the cultivar was compared with other male varieties (Pollmix 1). Bibliography of the reference cultivar of the world's range is Pollmix.

For the male plants, the corresponding mark is 2, Nr. UPOV 1 \* Table 1.

A male plant is recommended for every seven female plants.

Table 1. Results obtained at SC Fructex Bacau 2008. Description after UPOV
of the STAR cultivar (sea buckthorn pollinator)

No UPOV	Characteristics	State of Expression	Note
1. *	Plant: sex	Male plants	2
2. *	Plant: type of growth	type of tree	1
3. *	Plant: the position of the branches	semierect	2
4.	Plant: vigour	strong	7
5.	Plant: the density of the branches	dense	7
6.	Plant: the position of the inflorescence	-	-
7.	One year branch : thickness	medium	5
8. *	Branch : number of thorns (from the middle to the top)	absent or very small	1
9.	Branch : thorns' lenght	long	7
10. *	The leaf: shape	really narrow, eliptical	1
11. *	The leaf: size	big	7
12.	The leaf: margin undulation	absent	1
13. *	The leaf: the colour of upper side	silver	2
14.	The leaf:: intensity of green colour of the upper side	medium	2
15. *	The leaf: pubescence of the inferior side	weak	3
16. *	The fruit: size	-	-
17. *	The fruit:shape	-	-
18. *	The fruit:skin colour	-	-
19. *	The fruit: pubescence	-	-
20.	The fruit:peduncle lenght	-	-
21.	The beginning of the blossoming	-	-
22.	Fruit Maturity	-	-



Fig. 4. STAR cultivar male plants' pollen maker buds

**The type of growth** - the STAR biotype (Fig. 5) has the shape of a tree similar to the reference varieties in the worldwide series Maslichnaya, Novost, Altaya, Slovan, Vitaminaya. (bibliography).

The advantage of the STAR biotype is that having a high, erect type of growth, it can ensure a good pollination for all varieties studied by spreading the pollen.

The male STAR buds are large, folded, reddish-brown with yellowish hues and lots of pollen.

Tree-shaped plants are marked with  $1 \cdot (No. UPOV 2 * Table 1)$  (The rating conferred to this character is 1).



Fig. 5. STAR cultivar. Tree shaped plant type

**The position of the branches -** in the STAR cultivar (Fig. 6) are numerous and semi-erect, that resemble the German assortments Ascola and Frugana from the wideworld range.

Plants with erect branching correspond grade 2 (UPOV No. 3 \* Table 1).



Fig. 6. STAR cultivar - semierect branching

**The vigor of the STAR plant** (Fig. 7) is strong, which recommends it for higher densities (more plants per hectare) exploiting intensive and superintensive culture technologies with densities over 2200 plants on Ha.



Fig. 7. STAR cultivar- strong vigour

The vigour also allows a decrease in manual labor, **because** the need for fewer cuts.

The world-wide cultivar reference is Bojan and Hergo. This character is marked with 7 (UPOV No. 4 Table 1).

The density of the branches - in the STAR cultivar (Fig.8) the branches are dense, being similar to the Maslichnaya, Slovan varieties of the world assortment, thus granting the grade 7. (UPOV No. 5 tab.1)



Fig. 8. STAR cultivar-branches density

One-year branch - in the STAR cultivar (Fig.9), the expression of the character fits in the middle class as the Bojan and Slovan reference varieties from the wide-world range. The rating conferred to this character is 5 (UPOV No. 7 Table 1).



Fig. 9. STAR cultivar- one year branch detail

**Spin length** - STAR cultivar (Fig. 10). At the base of the two-year shoots and on thicker wood there are spines that are long, being appreciated by grade 7. This length provides for this plant a lack of aggressiveness because the thorns are flexible and soft. This trait is also found in the Auras and Victoria varieties (No. UPOV 9 Table 1).



Fig.10. STAR cultivar- thornes length

The number of thorns from the middle to the top - in the STAR cultivar (Fig.11). It is absent or very small, being marked with 1 point as for the Obilnaya and Yantarnaya varieties of the wide-world range (No. UPOV 8 \* Table 1).



Fig.11. STAR cultivar- the number of thorns from the middle to the top not present

**The shape of the leaf** - in the STAR cultivar (Fig.12) It is very narrow, elliptical as well as the Slovan reference cultivar according to the UPOV description,mark 1 (No. UPOV 10 \* Table 1).

**The size of the leaf** - in the STAR cultivar (Fig. 13) is large as that of the reference varieties Bojan, Leikora, Vitaminaya marked with 1 7 (No. UPOV 11 \* Table 1).

**The leaf undulation/bend** - in the STAR cultivar (Fig. 14) according to the expression of character is absent as in the reference variety Frugana and is marked with 1 (UPOV No. 12 Table 1).

**The colour of the top of the leaf** - in the STAR cultivar (Fig.15) is silver as well as the reference varieties Dorana, Leikora, Pollmix 1 and it was assigned the mark 2. (UPOV No. 13 \* Table 1).

The intensity of the green color of the upper face of the leaf in the STAR cultivar (Fig. 15) is medium as well as the Leikora reference variety, awarding Note 2. (UPOV No. 14 Table 1).

**Pubescence of the inferior leaves of the leaf** in the STAR cultivar (Fig. 16) is weak as the reference Silvia variety is given note 3. (UPOV No. 15 \* Table 1).



Fig. 12. STAR cultivar- the shape of the leaf is very narrow, elliptical



Fig. 13. STAR cultivar- leaf size - large



Fig. 14. STAR cultivar-absent edge undulation of the leaf



Fig. 15. STAR cultivar- the color of the upper face – silver; intensity of green colour- medium

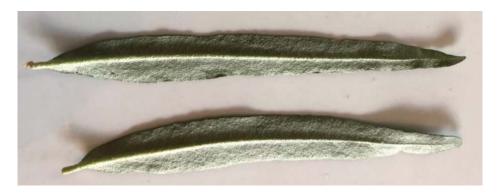


Fig. 16. STAR cultivar- pubescence of the inferior side of the leaf

### **Pollen description**

The pollen granule forms/takes shape in anthers, where it stays until full maturity when it is eliminated through its apertures. The content of each pollen granule is covered with sporopollenin.

The inner layer of the sporopollenin/ sporoderme, intine, is thinner than the cellulose type, whereas the outer layer (exine) is made of pollenine, with a high resistance to decomposition and the action of some chemical substances or agents, which offers the pollen granule a good stability for a long time.

The morphological features of pollen granules are reflected/deriving in the hybrid qualities of some plants, in the derivation of spontaneous plant culture forms, as well as (in) their genetic interrelations. Pollen was harvested using V. Cociu's method

Pollen is obtained by rubbing the buds on a sieve. Storage was done in Petri dishes in a dry and cold place. Observation of the pollen was done using the Euromex microscope.

**Results on STAR cultivar examined pollen** When pollen was examined in the STAR cultivar pollinating male (Fig. 17), several characters and attributes were noted, which we briefly list:

- the harvesting pollen presents itself under the form of irregularly shaped grains,
- the colour of the pollen is yellow,
- the taste is pleasant, slightly fragrant,
- humidity of pollen at harvest time- it is hygroscopic, attracts moisture, therefore it is a very good environment for the development of microorganisms and because of this it has to be processed very quickly,
- the size of pollen grains is medium,
- the shape of the pollen granule it is slightly triangular, rounded, spheroidal.

In culture environment, pollen from the STAR cultivar has a very good germination capacity with both colour variants (Fig. 18).

This confirms the productivity tests that recommended it as the best pollinator for the studied cultivars (Auras, Serpenta, Victoria, Ovidiu, Silvia, Diana and Tiberiu).

Popularization and marketing of sea buckthorn plants to households

The author of male and female sea buckthorn varieties, Rați Ioan Viorel, wishes to use/ to turn to account them under the protection of a trademark to be secured by OSIM and to allow the transfer of knowledge about the complexity of this plant on its importance within households.

Being a dioecious plant, the population does not have the necessary knowledge to use it, thus by applying this trademark, it can be explained what the gender diversity means to a species, what is the relationship between male and female plants, what is the final result in sea buckthorn culture.

Sea buckthorn has a great tradition in our geographic area that has created a large number of local populations and has allowed a very good selection in the early stages of creating genetic bases in order to improve the species and obtain new cultivars.

The ratio of one male plant to seven female plants allows for the first time to issue the concept of the **family of sea buckthorn seven plus one plants** (Fig. 19). The STAR male cultivar can form a family with each female plant cultivar obtained in the experimental field at Fructex.

Seven female varieties of STAR cultivar can form seven families of sea buckthorn. Each family of sea buckthorn can be grown in every household, finding a place to decorate the garden, use it as a herb, or to improve the degraded lands in the proximity of the houses. All these arguments confirm the need to launch a trademark /brand for this cultivar.

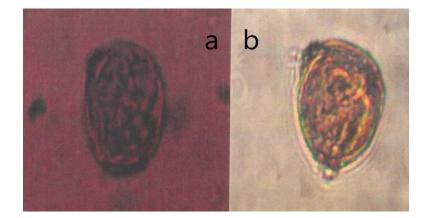


Fig. 17. The form of pollen grains in the STAR cultivar in those two dyes Orceina(a) and Lugol (b)

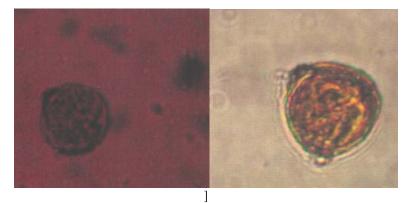


Fig. 18. The pollen grain germinated in the STAR cultivar in the two dyes Orceina (a) and Lugol (b).



Fig. 19. Trademark registered by OSIM

# CONCLUSIONS

STAR Cultivar is a male plant of the species *Hippophae rhamnoides* that provides a very good pollination for female plants in culture.

The STAR cultivar has the shape of a tree, a high erect growth ensuring a good pollination by spreading the pollen, the branches are erect allowing a vertical development.

The male / female recommended plant ratio is 1 to 7 (one male plant for seven female plants) and it is the best ratio as a result of more than over 20 years of research conducted by Professor Rati Ioan Viorel.

STAR cultivar harvested pollen is in the form of grains, with irregular contour, yellow color, pleasant taste, slightly fragrant, the shape of the pollen granule is slightly triangular, rounded, spheroidal..

The commercial exploitation of sea buckthorn plants and will be achieved under the protection of a trademark protected by OSIM under the name " FAMILIA DE CATINA ALBA 7+1 FIRE "- SEA BUCKTHORN FAMILY SEVEN PLUS ONE PLANTS.

### ABSTRACT

STAR cultivar is a male plant of the species Hippophae rhamnoides which provides a very good pollination for female plants in the *sea buckthorn* culture.

The ratio of male and female recommended plants is one male plant to seven female plants.

The STAR cultivar has a very good germination capacity. This confirms the productivity tests that recommend it as the best pollinator for the studied female cultivars: Auras, Serpenta, Victoria, Ovidiu, Silvia, Diana and Tiberiu.

The STAR cultivar has a high erect growth ensuring a good pollination by spreading the pollen for all the studied cultivars.

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