

## Results regarding compatibility of some plum cultivars grafted on different rootstocks

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### ABSTRACT

In Romania, the most used plum rootstock, 'Mirobalan' does not any problems in terms of graft compatibility and that is why it is widely used. However, it was found that this generative rootstock has low compatibility with the 'Tuleu gras' cv. and with all the plum cultivars obtained from it. In this paper, we study the degree of compatibility of 5 plum cultivars ('Andreea', 'Pitestean', 'Romaņa', 'Čačanska Lepotiča', 'Jojo') with 7 Romanian rootstocks ('Adaptabil', 'Mirobolan dwarf', 'Mirodad 1', 'Mirodad 2', 'Redutabil', 'BN-4Kr' and 'B 83/8'). The evaluation of graft compatibility of these cultivar-rootstock combinations was made through two indices: morphological thickness index of the grafting area (IMÎZA - as a result of the ratio between the surfaces section of the scion, rootstock and grafting union) and anatomical similarity index of the symbionts (IMAAAtS) calculated based on the number and diameter of the woody vessels in the scion, rootstock and graft union. Following the measurements, very significant results were obtained, which can explain the degree of affinity between cultivars and rootstocks. The morphological thickness index of the grafting area had values above 0.300, which indicates good and very good graft compatibility in most of the cultivar-rootstock combinations studied, with the exception of the association with 'Redutabil' rootstock which have values below 0.300, and the thickness differences between scion, rootstock and graft union were very large, indicating a low level of compatibility between the cultivars studied and this rootstock. The anatomical similarity index of the symbionts had values close to 1, indicating good and very good compatibility. The exception was the combinations of cultivars with the 'Redutabil' rootstock, the anatomical similarity index of the symbionts having values below 1.

**Keywords:** cultivar, rootstock, compatibility, morphological thickness index, anatomical similarity index.

### INTRODUCTION

In Romania, plums are the main types of fruit crops, occupying an area of 66.730 hectares and producing 807.170 tons of fruit (Butac *et al.*, 2014; Butac *et al.*, 2015; Coman *et al.*, 2012; Data FAO, 2023).

The most popular rootstock in most European countries is 'Myrobalan' seedling (*Prunus cerasifera*), which has some disadvantages: large tree vigor, sensitivity to Plum Pox Virus, incompatibility with some cultivars (e.g. 'Tuleu gras' and its progenies), late bearing and

intensive suckering (Blažec and Pištěková, 2009, 2012; Butac *et al.*, 2016; Kaufmane *et al.*, 2007; Sosna, 2002).

According to different authors (Errea *et al.*, 2001; Mladin *et al.*, 2006; Zarrouk *et al.*, 2006; Zlati, 2009; Zlati *et al.*, 2007, 2009; Dogra *et al.*, 2018) the compatibility / incompatibility between rootstock and cultivar is a limiting factor for orchard performance and longevity. The mechanism of graft incompatibility is not yet fully understood. Much research has been carried out in this direction to understand the mechanisms of graft development. Graft incompatibility can be determined by genetic, anatomical, physiological and biochemical factors but also by external factors such as temperature, water, soil and pathogens. Symptoms of incompatibility may appear in the first year after grafting or later, after several years in the orchard and can lead to major economic losses (Asănică, 2012; Dogra *et al.*, 2018; Rasool *et al.*, 2020; Arghavan *et al.*, 2022).

Two types of incompatibility are recognized: translocated and localized graft incompatibility (Asănică, 2006, 2012; Dogra *et al.*, 2018). The first type of incompatibility is usually expressed in the nursery, in the first year after grafting as growth cessation, yellowed foliage, fragility and breakage of the graft union and has been associated with biochemical and functional alterations at the graft union due to carbohydrate blockage at the scion, above the graft union (Monig *et al.*, 1988; Asănică, 2006, 2012; Dogra *et al.*, 2018; Zamfirescu, 2022). The second type of incompatibility is characterized by anatomical irregularities at the graft union which induce mechanical weakness in the union area (interruption in vascular and cambial continuity of undifferentiated parenchyma and poor vascular connections) (Mladin *et al.*, 2006; Zlati *et al.*, 2007, 2009; Koepke and Dhingra, 2013; Dogra *et al.*, 2018). This type of incompatibility appears in the orchard after several years.

To avoid the occurrence of graft incompatibility in the orchard there are different possibilities of early detection.

The aim of this paper was to determine graft compatibility of 5 plum cultivars ('Andreea', 'Piteștean', 'Romaña', 'Čačanska Lepotiča', 'Jojo') with 7 Romanian rootstocks ('Adaptabil', 'Mirololan dwarf', 'Miroadad 1', 'Miroadad 2', 'Redutabil', 'BN-4Kr' and 'B 83/8') using anatomo-morphological indices for early detection, from the nursing phase, of incompatible cultivar-rootstock combinations to the plum species.

## MATERIALS AND METHODS

The research was carried out within the Propagation Laboratory in the second field of the nursery, between 2020-2022 periods.

5 Romanian and foreign cultivars ('Andreea', 'Piteștean', 'Romaña' – Romanian cultivars, 'Čačanska Lepotiča' – Serbian origin, 'Jojo' – German origin) grafted on 7 Romanian vegetative and generative rootstocks ('Adaptabil', 'Mirololan dwarf', 'Miroadad 1', 'Miroadad 2', 'Redutabil', 'B 83/8' – vegetative rootstock and 'BN-4Kr' – generative rootstock) were studied. The experimental plot comprises 3 trees of each combination cultivar - rootstock in 3 replications.

In order to highlight the differences of vigor between the partners, the diameter of the stem was determined 5 cm upper and under the graft union, as well as on the contact graft area between symbionts. The difference of thickness between the symbionts was determined by the calculation of the surfaces section of the scion (SSTA), rootstock (SSTPA) and the graft union (SSTŽÎ), by applying the formula:  $SST = (D/2)^2 \times 3.14$ , in which SST = surface section of stem (cm<sup>2</sup>); D = diameter of stem (cm).

After performing the three measurements, the morphological thickness index of the grafting area (IMÎZA) was calculated as the result of the ratio between the surfaces section of the scion, rootstock and graft union (Mladin *et al.*, 2006).

- IMÎZA = SSTA/SSTPA/SSTŽÎ.

When this index has values higher than 0.330, combinations cultivar-rootstock has very good compatibility – type A; when the index is between 0.300 – 0.330 the compatibility is good – type B and when the index is under 0.300 – the compatibility is poor – type C (Mladin *et al.*, 2006).

In order to carry out some anatomical and morphological evaluations from the grafted trees removed from the nursery, biological samples were taken, respectively 4-5 cm stem fragments from the graft union and then fixed in ethylic alcohol 70°.

After that, transversal and longitudinal sections were made through the graft union with the microtome. On transversal micro sections, microscope observations were made regarding: the number and diameter of woody vessels in the scion at 5 cm upper the graft union, in the rootstock at 5 cm under the graft union as well as on the joining point. Also, on longitudinal micro sections were made observations concerning the continuity and orientation of the xylem vessels through the joining point; the way of arrangement of the vessels - if they are linear or if they present involution and sinuous aspect, if xylem fascicle is continuous or interrupted in the joining point, if the parenchyma cells are differentiated or not (asymmetrical cells or their complete lack).

After these measurements and observations, the anatomical similarity index of the symbionts (IMAATS) was calculated, applying the following formula (Mladin *et al.*, 2006):

- IMAATS =  $[(N.v.a/N.v.pa + N.v.a/N.v.zî + N.v.pa/N.v.zî)/3] / [(\emptyset.v.a/\emptyset.v.pa + \emptyset.v.a/\emptyset.v.zî + \emptyset.v.pa/\emptyset.v.zî)/3]$ , in which:

- IAATS = anatomical similarity index of the symbionts;
- N.v.a. = number of woody vessels at 5 cm upper the joining point;
- N.v.pa = number of woody vessels at 5 cm under the joining point;
- N.v.zî = number of woody vessels on the joining point;
- $\emptyset.v.a$  = diameter of woody vessels at 5 cm upper the joining point;
- $\emptyset.v.pa$  = diameter of woody vessels at 5 cm under the joining point;
- $\emptyset.v.zî$  = diameter of woody vessels on the joining point.

When this index has a value 1 or close to 1, the graft compatibility is good and very good (Mladin *et al.*, 2006).

The data were statistically interpreted with the SPSS 14.0 program, which uses the Duncan test (multiple t-tests) at a 0.05 level of probability.

## RESULTS AND DISCUSSION

### Morphological thickness index of the grafting area

After the measures, very significant results have been obtained, which can explain the degree of affinity between cultivars and rootstocks. In all cultivar-rootstock combinations, it was found that the surface section of the scion at 5 cm upper the graft union was smaller than under the graft union and on the joining point. If the value of the stem section in the graft union of symbionts is farther than the values of the stem section at 5 cm upper and under the graft union of symbionts the graft compatibility of the combination is lower.

The morphological thickness index of the grafting on all 5 plum cultivars associated with the 'Redutabil' rootstock had low value (below 0.300), and the differences between the surface section of the scion and the surface section of the rootstock were very large, which indicates a low level of compatibility (Table 1).

When the plum cultivars were grafted on 'Redutabil' rootstock, the values of the morphological thickness index of grafting area varied from 0.255 to the 'Andreea/Redutabil' combination to 0.295 to the 'Romaņa/Redutabil' combination, and the thickness differences between the cultivars and rootstock were between 0.50 cm<sup>2</sup> at the 'Romaņa/Redutabil' combination and 0.79 cm<sup>2</sup> at the 'Piteștean/Redutabil' combination.

If the plum cultivars were associated with the other rootstocks, the morphological thickness index of the grafting area had values over 0.300 which indicates a good and very good compatibility (Table 1).

**Table 1.** The morphological evaluation of the grafting area at cultivar-rootstock combinations

| Cultivar          | Rootstock       | Surface section of stem (cm <sup>2</sup> ) |             |             | Thickness difference between scion and rootstock (cm <sup>2</sup> ) | IMÎZA        |
|-------------------|-----------------|--|-------------|-------------|---|--------------|
|                   |                 | SSTA                                       | SSTPA       | SSTZÎ       |   |              |
| Andreea           | Mirodad 1       | 2.17 b                                     | 1.86 b      | 2.51 c      | 0.31 b  | 0.341 b      |
|                   | Mirodad 2       | 2.07 b                                     | 1.77 bc     | 2.42 d      | 0.30 b  | 0.353 b      |
|                   | Mirobolan dwarf | 1.94 b                                     | 1.58 c      | 2.32 e      | 0.36 b  | 0.351 b      |
|                   | Adaptabil       | 2.66 a                                     | 2.35 a      | 2.92 a      | 0.31 b  | 0.303 c      |
|                   | Redutabil       | 2.05 b                                     | 1.33 d      | 2.54 c      | 0.72 a  | 0.255 d      |
|                   | BN 4 Kr         | 1.92 b                                     | 1.70 bc     | 2.21 f      | 0.22 c  | 0.401 a      |
|                   | B 83/8          | 2.68 a                                     | 2.32 a      | 2.83 b      | 0.36 b  | 0.306 c      |
|                   | <b>Average</b>  | <b>2.21</b>                                | <b>1.84</b> | <b>2.53</b> | <b>0.37</b>   | <b>0.330</b> |
| Piteștean         | Mirodad 1       | 2.49 a                                     | 2.14 a      | 2.73 a      | 0.35 d  | 0.314 e      |
|                   | Mirodad 2       | 2.40 b                                     | 2.06 b      | 2.70 b      | 0.34 d  | 0.318 e      |
|                   | Mirobolan dwarf | 1.70 f                                     | 1.35 f      | 2.00 e      | 0.35 d  | 0.397 a      |
|                   | Adaptabil       | 2.10 c                                     | 1.61 c      | 2.32 c      | 0.49 b  | 0.330 d      |
|                   | Redutabil       | 1.96 d                                     | 1.17 g      | 2.31 c      | 0.79 a  | 0.258 f      |
|                   | BN 4 Kr         | 1.89 e                                     | 1.54 d      | 2.23 d      | 0.35 d  | 0.365 b      |
|                   | B 83/8          | 1.85 e                                     | 1.43 e      | 2.17 e      | 0.42 c  | 0.356 c      |
|                   | <b>Average</b>  | <b>2.05</b>                                | <b>1.61</b> | <b>2.35</b> | <b>0.44</b>   | <b>0.334</b> |
| Romaņa            | Mirodad 1       | 1.76 a                                     | 1.43 b      | 2.06 a      | 0.33 bc   | 0.394 d      |
|                   | Mirodad 2       | 1.65 b                                     | 1.33 c      | 1.97 b      | 0.32 cd   | 0.409 b      |
|                   | Mirobolan dwarf | 1.59 c                                     | 1.29c       | 1.83 d      | 0.30 e  | 0.443 b      |
|                   | Adaptabil       | 1.80 a                                     | 1.49 a      | 2.09 a      | 0.31 de   | 0.396 cd     |
|                   | Redutabil       | 1.57 c                                     | 1.07 d      | 1.88 c      | 0.50 a  | 0.295 e      |
|                   | BN 4 Kr         | 1.34 d                                     | 1.00 d      | 1.90 c      | 0.34 b  | 0.393 d      |
|                   | B 83/8          | 1.66 b                                     | 1.33 c      | 1.98 b      | 0.33 bc   | 0.405 bc     |
|                   | <b>Average</b>  | <b>1.62</b>                                | <b>1.27</b> | <b>1.96</b> | <b>0.35</b>   | <b>0.391</b> |
| Jojo              | Mirodad 1       | 2.80 b                                     | 2.55 b      | 3.02 a      | 0.25 d  | 0.302 c      |
|                   | Mirodad 2       | 2.77 b                                     | 2.50 c      | 3.00 a      | 0.27 c  | 0.301 c      |
|                   | Mirobolan dwarf | 2.24 d                                     | 1.93 e      | 2.61 c      | 0.31 b  | 0.330 b      |
|                   | Adaptabil       | 2.86 a                                     | 2.60 a      | 3.03 a      | 0.26 cd   | 0.300 c      |
|                   | Redutabil       | 1.40 e                                     | 0.81 g      | 2.00 d      | 0.59 a  | 0.289 d      |
|                   | BN 4 Kr         | 1.32 f                                     | 1.02 f      | 1.68 e      | 0.30 b  | 0.460 a      |
|                   | B 83/8          | 2.66 c                                     | 2.35 d      | 2.94 b      | 0.31 b  | 0.301 c      |
|                   | <b>Average</b>  | <b>2.29</b>                                | <b>1.96</b> | <b>2.61</b> | <b>0.33</b>   | <b>0.326</b> |
| Čačanska Lepotiča | Mirodad 1       | 1.83 b                                     | 1.49 b      | 2.11 b      | 0.34 b  | 0.386 d      |
|                   | Mirodad 2       | 1.68 d                                     | 1.35 d      | 1.99 d      | 0.33 bc   | 0.404 c      |
|                   | Mirobolan dwarf | 1.74 c                                     | 1.43 c      | 2.04 c      | 0.31 de   | 0.403 c      |
|                   | Adaptabil       | 2.25 a                                     | 1.95 a      | 2.57 a      | 0.30 e  | 0.337 e      |
|                   | Redutabil       | 1.35 f                                     | 0.79 f      | 2.00 d      | 0.56 a  | 0.292 f      |
|                   | BN 4 Kr         | 1.26 g                                     | 0.93 e      | 1.65 f      | 0.33 bc   | 0.447 a      |
|                   | B 83/8          | 1.63 e                                     | 1.31 d      | 1.95 e      | 0.32 cd   | 0.412 b      |
|                   | <b>Average</b>  | <b>1.68</b>                                | <b>1.32</b> | <b>2.04</b> | <b>0.36</b>   | <b>0.383</b> |

\*Duncan multiple ranges test. Numbers followed by the same letter within a column are not significantly different ( $P \leq 0.05$ ).

### Anatomical similarity index of the symbionts

One of the theories regarding the causes of the incompatibility phenomenon suggests that it would be generated by the differences between the number and diameter of the woody vessels in the scion and the rootstock. That is why the anatomo-morphological analysis of the grafting union was performed, correlating the size of the xylem vessels with some anomalies that appear in cultivar-rootstock combinations with a low degree of affinity (Mladin *et al.*, 2006; Zlati, 2009).

At the incompatible combinations, discontinuities of the woody vessel, zone of undifferentiated parenchyma tissue between cultivars and rootstocks, and in some cases necrotic parenchyma tissue appear.

At the compatible combinations, the grafting union between cultivars and rootstocks has a satisfactory development, with a completely differentiated woody tissue, but with deviations from the radial structure of the location of the xylem vessels due to the grafting process.

Anatomical studies were performed under a microscope with a size unit  $7 \times 10 = 70$ , on micro sections made with microtome.

The microscopic analysis of the transversal sections at the cultivar-rootstock combinations studied at 5 cm upper the grafting union, 5 cm under the grafting union and through the grafting union, highlighted a variable number of woody vessels with different diameters from one combination to another (Fig. 1).

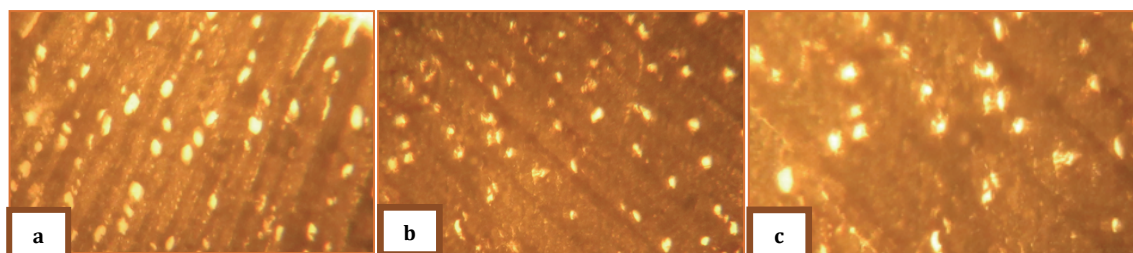


Fig. 1. Transversal section at the 'Piteștean/Adaptabil' combination  
a - upper the grafting union; b - under the grafting union; c - on the grafting union

The number and diameter of the woody vessels were higher under the grafting union than through the grafting union and upper the grafting union at all the combinations studied except the combinations with the 'Redutabil' rootstock to which the number and diameter of the woody vessels were higher through the grafting union than under and upper grafting union.

The average number of woody vessels was 50.28 vessels in scion with variations from 49.85 ('Jojo' cv.) to 49.09 ('Romaņa' cv.); 49.09 vessels in rootstock ranging from 51.42 ('Andreea' cv.) to 50.28 ('Romaņa' cv.); 49.42 vessels in the grafting area ranging from 50.14 ('Jojo' cv.) and 48.66 ('Andreea' cv.) (Table 2).

The average diameter of the woody vessels was 0.49  $\mu$  in scion, 0.52  $\mu$  in rootstocks and 0.48  $\mu$  in the grafting area, between the cultivar-rootstock combinations being significant differences. The average diameter of the woody vessels varied between 0.49  $\mu$  ('Andreea', 'Piteștean', 'Romaņa', 'Čačanska Lepotiča' cvs.) and 0.48  $\mu$  ('Jojo' cv.) in scion; 0.52  $\mu$  ('Andreea', 'Piteștean', 'Romaņa' cvs.) and 0.53  $\mu$  ('Jojo', 'Čačanska Lepotiča' cvs.) in rootstock; 0.47  $\mu$  ('Andreea' cv.) and 0.48  $\mu$  ('Piteștean', 'Romaņa', ('Jojo', 'Čačanska Lepotiča' cvs.) in grafting union (Table 2).

The anatomical similarity index of the grafting area had values close to 1, indicating good and very good compatibility. The exception made the combinations of cultivars with 'Redutabil' rootstock, at which this index had values less than 1, respectively 0.70 at

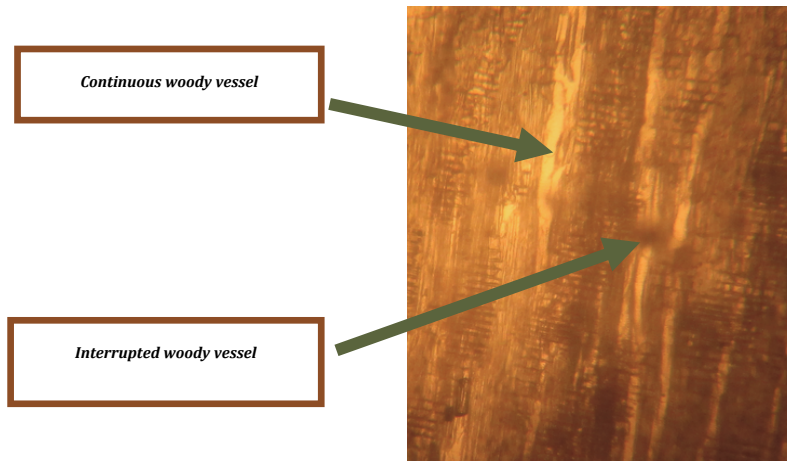
'Andreea' cv., 0.72 at 'Piteștean' cv., 0.69 at 'Romaņa' cv., 0.71 at 'Čačanska Lepotiča' cv. and 0.72 at 'Jojo' cultivar (Table 2).

**Table 2.** The anatomical evaluation of the grafting area at cultivar-rootstock combinations

| Cultivar          | Rootstock       | Number of woody vessel |              |              | Diameter of woody vessel ( $\mu$ ) |             |             | IMAAoS      |
|-------------------|-----------------|------------------------|--------------|--------------|------------------------------------|-------------|-------------|-------------|
|                   |                 | SSTA                   | SSTPA        | SSTZĪ        | SSTA                               | SSTPA       | SSTZĪ       |             |
| Andreea           | Mirodad 1       | 50.33 b                | 52.66 ab     | 45.00 c      | 0.48 b                             | 0.50 c      | 0.46 bc     | 1.05 a      |
|                   | Mirodad 2       | 51.00 ab               | 53.66 a      | 47.33 b      | 0.50 a                             | 0.52 b      | 0.49 a      | 1.04 a      |
|                   | Mirobolan dwarf | 48.00 b                | 51.00 b      | 46.00 c      | 0.51 a                             | 0.53 b      | 0.50 a      | 1.03 a      |
|                   | Adaptabil       | 50.00 b                | 53.66 a      | 48.66 b      | 0.50 a                             | 0.51 bc     | 0.49 a      | 1.04 a      |
|                   | Redutabil       | 41.66 c                | 40.00 c      | 52.33 a      | 0.46 c                             | 0.57 a      | 0.44 c      | 0.70 c      |
|                   | BN 4 Kr         | 52.33 a                | 54.00 a      | 50.00 a      | 0.48 b                             | 0.50 c      | 0.46 bc     | 1.00 b      |
|                   | B 83/8          | 53.00 a                | 55.00 a      | 51.33 a      | 0.49 b                             | 0.51 bc     | 0.47 b      | 0.99 b      |
|                   | <b>Average</b>  | <b>49.47</b>           | <b>51.42</b> | <b>48.66</b> | <b>0.49</b>                        | <b>0.52</b> | <b>0.47</b> | <b>0.98</b> |
| Piteștean         | Mirodad 1       | 51.00 ab               | 52.33 a      | 46.00 b      | 0.49 b                             | 0.51 c      | 0.47 bc     | 1.03 a      |
|                   | Mirodad 2       | 50.00 b                | 52.66 a      | 47.00 b      | 0.51 a                             | 0.53 b      | 0.50 a      | 1.04 a      |
|                   | Mirobolan dwarf | 49.00 b                | 52.00 a      | 47.00 b      | 0.50 a                             | 0.52 bc     | 0.51 a      | 1.06 a      |
|                   | Adaptabil       | 51.00 ab               | 52.66 a      | 49.33 ab     | 0.49 b                             | 0.50 c      | 0.48 b      | 1.02 a      |
|                   | Redutabil       | 40.00 c                | 39.00 b      | 50.66 a      | 0.45 c                             | 0.56 a      | 0.44 c      | 0.72 c      |
|                   | BN 4 Kr         | 51.33 a                | 53.00 a      | 52.00 a      | 0.49 b                             | 0.51 c      | 0.47 bc     | 0.96 b      |
|                   | B 83/8          | 52.00 a                | 54.00 a      | 51.00 a      | 0.50 a                             | 0.52 bc     | 0.48 b      | 0.98 b      |
|                   | <b>Average</b>  | <b>49.19</b>           | <b>50.81</b> | <b>49.00</b> | <b>0.49</b>                        | <b>0.52</b> | <b>0.48</b> | <b>0.97</b> |
| Romaņa            | Mirodad 1       | 50.33 ab               | 51.00 b      | 46.33 c      | 0.50 a                             | 0.52 c      | 0.48 b      | 1.01 a      |
|                   | Mirodad 2       | 51.00 a                | 51.66 b      | 48.00b       | 0.50 a                             | 0.51 d      | 0.49 b      | 1.02 a      |
|                   | Mirobolan dwarf | 49.33 b                | 51.66 b      | 48.33 b      | 0.51 a                             | 0.53 b      | 0.50 a      | 1.01 a      |
|                   | Adaptabil       | 50.33 ab               | 51.33 b      | 49.66 b      | 0.50 a                             | 0.52 c      | 0.49 b      | 0.98 a      |
|                   | Redutabil       | 39.00 c                | 38.33 c      | 51.33 a      | 0.40 b                             | 0.55 a      | 0.43 c      | 0.69 b      |
|                   | BN 4 Kr         | 52.00 a                | 54.33 a      | 51.00 a      | 0.50 a                             | 0.52 c      | 0.49 b      | 1.00 a      |
|                   | B 83/8          | 51.66 a                | 53.66 a      | 51.33 a      | 0.50 a                             | 0.52 c      | 0.49 b      | 0.99 a      |
|                   | <b>Average</b>  | <b>49.09</b>           | <b>50.28</b> | <b>49.42</b> | <b>0.49</b>                        | <b>0.52</b> | <b>0.48</b> | <b>0.96</b> |
| Jojo              | Mirodad 1       | 51.66 ab               | 53.00 a      | 49.00 c      | 0.50 b                             | 0.52 b      | 0.49 a      | 1.01 a      |
|                   | Mirodad 2       | 51.00 b                | 52.33 b      | 48.66 c      | 0.48 c                             | 0.51 b      | 0.47 b      | 0.99 a      |
|                   | Mirobolan dwarf | 50.33 c                | 51.66 b      | 49.33 c      | 0.50 b                             | 0.52 b      | 0.49 a      | 0.99 a      |
|                   | Adaptabil       | 51.66 ab               | 53.00 a      | 50.33 b      | 0.50 b                             | 0.53 ab     | 0.49 a      | 0.98 a      |
|                   | Redutabil       | 41.00 d                | 39.33 c      | 52.00 a      | 0.40 d                             | 0.55 a      | 0.45 c      | 0.72 b      |
|                   | BN 4 Kr         | 52.00 a                | 53.66 a      | 51.00 b      | 0.52 a                             | 0.54 a      | 0.50 a      | 0.98 a      |
|                   | B 83/8          | 51.33 b                | 53.66 a      | 50.66 b      | 0.49 b                             | 0.51 b      | 0.48 b      | 1.00 a      |
|                   | <b>Average</b>  | <b>49.85</b>           | <b>50.95</b> | <b>50.14</b> | <b>0.48</b>                        | <b>0.53</b> | <b>0.48</b> | <b>0.95</b> |
| Čačanska Lepotiča | Mirodad 1       | 51.00 a                | 52.33 b      | 48.66 b      | 0.51 a                             | 0.53 a      | 0.49 a      | 1.00 a      |
|                   | Mirodad 2       | 51.33 a                | 52.00 b      | 48.33 b      | 0.49 a                             | 0.51 b      | 0.48 a      | 1.01 a      |
|                   | Mirobolan dwarf | 50.00 ab               | 52.00 b      | 49.00 b      | 0.50 a                             | 0.52 b      | 0.49 a      | 1.00 a      |
|                   | Adaptabil       | 51.00 a                | 52.33 b      | 50.00 a      | 0.51 a                             | 0.53 a      | 0.50 a      | 0.99 a      |
|                   | Redutabil       | 40.00 b                | 38.00 c      | 51.00 a      | 0.41 b                             | 0.54 a      | 0.44 b      | 0.71 b      |
|                   | BN 4 Kr         | 51.66 a                | 53.33 a      | 51.33 a      | 0.51 a                             | 0.53 a      | 0.50 a      | 0.99 a      |
|                   | B 83/8          | 51.00 a                | 54.00 a      | 50.00 a      | 0.51 a                             | 0.52 b      | 0.49 a      | 1.01 a      |
|                   | <b>Average</b>  | <b>49.43</b>           | <b>50.57</b> | <b>49.76</b> | <b>0.49</b>                        | <b>0.53</b> | <b>0.48</b> | <b>0.96</b> |

\*Duncan multiple ranges test. Numbers followed by the same letter within a column are not significantly different ( $P \leq 0.05$ ).

Microscopic observations on longitudinal sections have shown that besides the combinations of cultivars with ‘Redutabil’ rootstock, signs of incompatibility have also appeared at the ‘Piteștean/Adaptabil’ combination manifested by discontinuities of the woody vessels, the trajectory of wood vessels being sometimes interrupted by undifferentiated parenchyma (Fig. 2). It is known that the ‘Adaptabil’ rootstock was registered mainly for peaches, but also for some plum cultivars except for the ‘Tuleu gras’ cv. and its progenies (Duțu *et al.*, 2001; Mazilu *et al.*, 2013). The ‘Piteștean’ cv. is a descendent of the ‘Tuleu gras’ cv. and maybe that is why such discontinuities of the woody vessels appear (Zamfirescu, 2022). That is why a compatibility evaluation is recommended in the orchard.



**Figure 2.** Longitudinal section on the grafting area at the ‘Piteștean/Adaptabil’ combination

## CONCLUSIONS

Early diagnosis of graft compatibility was performed by the complex morpho-anatomic analysis of the grafting area.

Evaluation of the compatibility of 5 plum cultivars grafted on 7 Romanian rootstocks was made through two indices: morphological thickness index of the grafting area (IMÎZA - as a result of the ratio between the surfaces section of the scion, rootstock and graft union) and anatomical similarity index of the symbionts (IMAAAtS) calculated based on the number and diameter of the woody vessels in the scion, rootstock and graft union.

The different growth rates of the symbionts in the grafting area recorded by the calculation of the surface section of stem at 5 cm upper and under the grafting union area, as well as on the grafting union, constituted in morphological thickness index, provided significant information for assessing the degree of compatibility. The morphological thickness index of the grafting area had values above 0.300, which indicates good and very good graft compatibility in most of the cultivar-rootstock combinations studied, with the exception of the association with ‘Redutabil’ rootstock which have values below 0.300, and the thickness differences between scion, rootstock and graft union were very large, indicating a low level of compatibility.

The number, diameter and trajectory of woody vessels in the grafting area, with different values depending on the cultivar-rootstock combination, have correlated with more or less successful welding of symbionts. The anatomical similarity index of the partners in the grafting area, calculated by an original formula, has highlighted the delayed incompatibility at all the studied cultivars grafted on the ‘Redutabil’ rootstock. Also, the ‘Piteștean/Adaptabil’ combination shows some discontinuities of the woody vessels, the trajectory of wood vessels being sometimes interrupted by undifferentiated parenchyma - a sign of incompatibility.

## REFERENCES

1. Arghavan S., Ganji-Moghadam E., Fahadan A. and Zamanipour M. (2022). Possibility of early detection of graft incompatibility in some commercial plum cultivars by phenolic compounds analysis. *J. Hort. Sci.* vol. 17 (2): 488-495.
2. Asănică A. (2012). Cireșul în plantațiile moderne – între compatibilitate și incompatibilitate. Ed. Ceres. București (in Romanian).
3. Asănică A. (2006). Cercetări anatomo-fiziologice și biochimice privind compatibilitatea la altoire a unor soiuri noi de cireș asociate cu diferiți portaltoi. Teză de doctorat (in Romanian).
4. Blažek J. and Pištěková I. (2009). Preliminary evaluation results of new plum cultivars in a dense planting. *Hort. Sci. (Prague)*. 36 (2): 45-54.
5. Blažek J. and Pištěková I. (2012). Initial results from the evaluation of plum cultivars grown in a very dense planting. *Acta Horticulturae* 968: 99-108.
6. Butac M., Chitu E., Sumedrea D. and Militaru M. (2014). Evaluation of some plum cultivars in a high density system. *Fruit Growing Research*. vol. XXX: 37-41.
7. Butac M., Chitu E., Militaru M., Sumedrea M., Sumedrea D. and Plopa C. (2015). Orchards performance of some Romanian plum cultivars grafted on two rootstocks. *Agricultural Science Procedia* 6 (2015). Elsevier: 118-123.
8. Butac M., Dutu I., Mazilu Cr., Sumedrea D., Militaru M., Cojocar M., Zamfirescu B. and Coman R. (2016). Preliminary results regarding the influence of the rootstocks to the vigour and precocity of some plum cultivars. *Fruit Growing Research*. Vol. XXXII: 71-76.
9. Coman M., Butac M., Sumedrea D., Dutu I., Iancu M., Mazilu Cr. and Plopa C. (2012). Plum culture in Romania – Current status and perspectives. *Acta Horticulturae* 968: 35-40.
10. Dogra K., Kour K., Kumar R., Bakshi P. and Kumar V. (2018). Graft-incompatibility in horticultural crops. *International Journal of Current Microbiology and Applied Sciences*. vol. 7 (2): 1805-1820.
11. Duțu I., Parnia P., Viscol I., Mazilu Cr. and Ancu S. (2001). Influența a doi portaltoi vegetativi asupra mărimii fructelor și productivității soiului de piersic Redhaven. *Lucrările Științifice ale ICDP Pitești-Mărăcineni*. Vol. XX: 75-77 (in Romanian).
12. Errea P., Garay L. and Marin J.A. (2001). Early detection of graft incompatibility in apricot (*Prunus armeniaca*) using *in vitro* techniques. *Physiologia Plantarum*. 112: 135-141.
13. Kaufman E., Rubauskis E. and Skrivele M. (2007). Influence of different rootstocks on the growth and yield of plum cultivars. *Acta Horticulturae* 734: 387-391.
14. Koepke T., Dhingra A. (2013). Rootstock scion somatogenetic interactions in perennial composite plants. *Plant Cell Rep.*, 32: 1321-1337.
15. Mazilu Cr., Duțu I., Mladin Gh., Ancu S., Coman M., Rovină A. and Plopa C. (2013). Achievements and prospects regarding vegetative rootstock breeding at the RIFG Pitesti, Romania. *Acta Horticulturae* 981 (2): 407-412.
16. Moing A. and Carde J.P. (1988). Growth, cambial activity and phloem structure in compatible and incompatible plum grafts. *Tree. Physiol.* 4 (4): 347-359.
17. Mladin Gh., Petrescu S. and Butac M. (2006). Rezultate preliminare privind unele elemente morfo-fiziologice implicate în conviețuirea simbioților soi-portaltoi la cireș. *Lucrări științifice ICDP Pitești-Mărăcineni*. Vol. XXII: 182-189 (in Romanian).
18. Rasool A., Mansoor S., Bhat K.M., Hassan G.I., Baba T.R., Alyemeni M.N., Alsahli A.A., El-Serehy H.A., Paray B.A. and Ahmad P. (2020). Mechanisms underlying graft union formation and rootstock scion interaction in horticultural plants. *Front. Plant Sci.* 11: 590847.
19. Sosna I. (2002). Growth and cropping of four plum cultivars on different rootstocks in South Western Poland. *Journal of Fruit and Ornamental Plant Research*. Vol. X: 95-103.
20. Zamfirescu B. (2022). Cercetări privind compatibilitatea la altoire a unor soiuri de prun cu diferiți portaltoi în condiții de pepinieră și livadă. Teză de doctorat (in Romanian).
21. Zarrouk O., Gogorcena Y., Moreno M.A. and Pinochet J. (2006). Graft compatibility between peach cultivars and *Prunus* rootstocks. *Amer. Soci. Horti. Sci.* 41: 1389-1394.
22. Zlati C., Cireașă V. and Grădinaru G. (2007). Researches regarding the influence of some physiological processes upon scion-rootstock association at four plum varieties. *Scientific Papers. Horticulture Series-USAMV Iasi*, 50: 609-614.
23. Zlati C. (2009). Influența unor procese fiziologice și biochimice asupra asociației altoi-portaltoi la unele specii pomicele în vederea creșterii agroproductivității. Teză de doctorat (in Romanian).
24. Zlati C., Grădinaru G. and Istrate M. (2009). Investigation of anatomical structure of graft union in sweet cherry. *Scientific Papers. Horticulture Series-USAMV Iasi*, 52: 597-602.
25. Zlati C., Grădinaru G. and Istrate M. (2009). Incompatibility aspects that appear in scion-rootstock association at some pear and plum varieties. *Scientific Papers. Horticulture Series-USAMV Iasi*, 52: 603-608.
26. \*\*\* (2023). *FAO Statistics Division*.



# The behaviour of some plum cultivars to brown rot fruit infection in northern Transylvania

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## ABSTRACT

The plum tree (*Prunus domestica* L.) is the dominant fruit tree species in Romania, according to the FAOSTAT (2021) data base. Globally, Romania ranks the second place after China at the top of major plum growing countries. This species is susceptible to various economically impactful diseases such as brown rot, produced by *Monilinia* spp. Climatic conditions have an important role in the occurrence and frequency of disease damage depending of a cultivar. Therefore, in 2023 at the FRDS Bistrita, was monitored the behaviour of brown rot damage on 18 plum cultivars with different ripening periods. During the growing stage, 12 conventional phytosanitary treatments were applied up to the harvest time. The determinations were made in the field after fruit harvesting, at the consumer's ripeness stage. Expectedly, the response to brown rot infection on fruits was different through all the cultivars studied. The results revealed low infections with *Monilinia* spp. on 'Zamfira' (6.9%), 'Anna Späth' (7.0%), and 'Doina' (7.7%), while 'Matilda' (39.5%), 'Elena' (33.1%), and 'Jubileu 50' (31.9%) expressed symptoms and a higher percentage of infected fruits. All the data obtained are statistically supported. The results are encouraging, allowing a selection of resistant or tolerant cultivars to brown rot, considering the increasing impact of climate change. Furthermore, the global trend toward organic farming requires the use of resistant cultivars to problematic pathogens for successful farming.

**Keywords:** brown rot, climatic changes, cultivar diseases, plum.

## INTRODUCTION

*Prunus domestica* L., commonly known as the plum tree, is a highly popular and valued fruit-bearing species worldwide. Due to its high adaptability to various environmental conditions and its nutritional value (vitamins and antioxidants), it is recommended to consume 2–4 fruits daily for a healthy diet (Gil *et al.*, 2002; Ortega-Vidal *et al.*, 2022). Plums are enjoyed fresh, processed (dried plums, jams, preserved) and distilled to produce alcoholic beverages (plum brandy – Botu *et al.*, 2008). Interest in plums is continuously increasing, as evidenced by the over 2000 cultivars of this species available (Sottile *et al.*, 2022). According to data base from FAOSTAT (2021), worldwide plum production ranks fifth after apples, bananas, pears and oranges. Regarding the top plum-

producing countries, China leads in global plum fruit production with 6.61 million tons and Romania ranks second with 0.8 million tons.

The trend among farmers to establish high-performance orchards, in Romania, has led to the introduction of new, largely unknown foreign cultivars, in terms of their adaptability and behavior in the face of various economically impactful diseases. In this context, both foreign and Romanian cultivars require increased attention to their susceptibility to specific pathogens, especially in the context of increasingly prevalent climate changes (Milošević and Milošević, 2023).

*Monilinia laxa*, *Monilinia fructicola* and *Monilinia fructigena* are a group of fungal diseases that cause the brown rot of the fruits in many fruit species including *Prunus* spp. They belong to the phylum Ascomycota, class Leotiomycetes, order Helioteliales, family Sclerotiniaceae, and genus *Monilinia* (Tronsmo *et al.*, 2020). All these three *Monilinia* species have resembling symptoms on tree organs including fruits, and consequently, visual observation is not enough to differentiate them. Therefore, for precise identification molecular techniques are required (Oliveira Lino *et al.*, 2016).

All of these species have been reported over all continents and the genetic similarity is extremely high: 97.5% of the DNA is identical to *M. fructigena* and *M. fructicola* and 99.1% of *Cyt b* gene is identical for *M. fructigena* and *M. laxa* (Hily *et al.*, 2011). This is the reason why many researchers including the present study referred to the above-mentioned species of *Monilinia* as *Monilinia* spp.

For growers, it is low interest to perform an accurate of the fungus identification because species of *Monilinia* have similar damaging effects on fruits and the treatments are similar. Although damages various tree organs (blossoms, twig, immature and mature fruits) the most sensitive phenological phase is represented by the blossoms and the mature fruits before and after harvesting. Brown rot is an extremely dangerous disease due to its long period of incidence, from bloom to fruit maturity and divers' climatic factors that favour disease spread (Florin and Puia, 2021).

These characteristics added up to the restrictions on pesticide use and low availability of biological control methods form an accurate picture of the difficulty of brown rot control. In these circumstances, the objective of finding resistant cultivars to fruit brown rot and breeding new cultivars with this trait is considered very important all over the world. Selecting cultivars with reduced requirements for external control methods is even greater when aiming for the ecological cultivation of plums (Moldovan *et al.*, 2022).

The behavior of different stone fruit cultivars (including plum) has been studied in many parts of the world but the results are difficult to compare because of the variability of the method used to investigate the susceptibility (Oliveira *et al.*, 2016) and maybe even more important because of the high variability of the behavior of the disease in different conditions. This is the reason for which it is very important to know how the cultivars behave in specific conditions and it is the aim of our study.

## **MATERIALS AND METHODS**

The study was performed in a young plum orchard (Fig. 1) established in 2020 at the Fruit Research and Development Station Bistrita (FRDS Bistrita). It involved a sample of 18 different plum cultivars, both Romanian ('Matilda', 'Elena', 'Gras ameliorat', 'Zamfira', 'Iulia', 'Agent', 'Flora', 'Andreea', 'Jubileu 50', 'Minerva', 'Doina', 'Centenar', 'Carpatin' and 'Diana') and foreign ('French Improved', 'Jojo', 'Anna Spath', 'Stanley') with varying ripening periods.

The cultivars were monitored throughout the entire growing season to assess their behavior in response to various pathogens. In the current study, the focus was on the

sensitivity of plum cultivars to brown rot (*Monilinia* spp.) infection. All data were collected from the same plot, under the same environmental conditions, and using the same agricultural methodology.



**Figure 1.** The plum orchard where the study was performed

During the growing season 12 conventional phytosanitary treatments were applied, distributed along the vegetative period (Table 1).

**Table 1.** Chemical fungicide treatments applied into experimental plot

|                                      |   |
|--------------------------------------|---|
| <b>I. Winter treatments</b>          | Copper (Cu) 380 g/l ( <b>3.3 l/ha</b> )                     |
| <b>II. Growing season treatments</b> |   |
| a.) Contact treatments               | Copper (Cu) 380 g/l ( <b>1.5 l/ha</b> )                     |
| b.) Systemic treatments              | Cyprodinil 500 g/kg ( <b>0.5 kg/ha</b> )                    |
|                                      | 26.7% Boscalid, 6.7% Piraclostrobin ( <b>0.5 kg/ha</b> )    |
|                                      | Cyprodinil 375 g/kg, Fludioxonil 250 g/kg ( <b>1kg/ha</b> ) |

In the current study, to establish the sensitivity of cultivars to brown rot infection, the frequency (F%) of disease on fruit was assessed. Six trees of each cultivar (3 repetitions x 2 trees) were selected for this assessment. From the total fruit yield on each tree, healthy fruits were separated from those affected by *Monilinia* spp., and then affected fruits were quantified and expressed as a percentage relative to the total number of fruits per tree. The calculation formula used to determine the frequency was:

$$F\% = \frac{\text{total number of fruits with brown rot/tree}}{\text{total number of fruits/tree}} \times 100$$

The fruit harvesting and the determination of the frequency of brown rot infection on fruits (Fig. 2) were carried out for each cultivar separately based on the fruit ripening period. Fruit inspection was conducted in the field visually, immediately after harvesting.



**Figure 2.** Brown rot symptoms on ‘Centenar’ plum cultivar