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PLUM GERMPLASM RESOURCES AND BREEDING IN ROMANIA

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In Romania, work on identification, conservation and evaluation of fruit genetic resources activities was initiated in 1970 in order to limit the loss of biodiversity. There are rich sources of germplasm located in two research centres: RIFG Pitesti with 642 accessions and UCv-SCDP Vâlcea with 361 accessions, representing wild species, local populations, named cultivars, breeder's selections and rootstocks. Observations were made according to the IBPGR Prunus descriptors updated by the ECP/GR Prunus Working Group. The following genetic resources from the Piteşti and Vâlcea collections were used in a breeding programme in the development of several plum cultivars: 'Grase de Becs', 'Carpatin', 'Ialomiţa', 'Kirke', 'Wilhelmina Späth' (for resistance / tolerance to Plum pox virus); 'Vinete româneşti', 'Tuleu timpuriu', 'Anna Späth' (for late blooming), 'Tuleu gras', 'Vâlcean' (for fruit quality), 'Stanley', 'Pescăruş', 'Centenar' (for productivity), and 'Diana' (for self-fertility). The plum rootstock breeding programme used the following genotypes as sources of genes: 'Rosior vărătec', 'Brompton', 'Renclod Verde', 'Pixy', 'Saint Julien A', 'Albe mici', 'Scolduş', 'Porumbar', etc. Breeding using the germplasm in these collections resulted in the release of 40 cultivars and 11 generative and vegetative rootstocks.

Key words: *plum germplasm, breeding, genitors, cultivars, rootstocks.*

INTRODUCTION

Biodiversity is very important for humanity according to the Convention on Biological Diversity signed by more than 150 countries, at the Earth Summit in Rio de Janeiro in 1992 (Botu *et al*., 2017). Plum is considered the second most important fruit tree crop produced in temperate climates (Botu *et al*., 2012) and the most important fruit tree crop in Romania (Butac *et al*., 2011; Coman *et al*., 2012).

Romania is a country located in Southeastern Europe, which has good environmental conditions for many fruit species in the wild or under cultivation. Numerous genetic resources of plum, apple, pear, sweet and sour cherry, peach, apricot, walnut, hazelnut, sweet chestnut, and berries are present (Botu *et al*., 2017).

Over 30–40 years ago, many countries started work on identification, collection, and conservation in field collections and gene banks, for evaluation and use of genetic resources in national and international programmes. The oldest fruit tree collections were established at Teynham, Kent (England) in 1533, at Hessen, Saxony (Germany) in 1542, at Pradel (France) in 1608, at Arad (Romania) in 1877 etc. (Braniţte *et al*., 2006; Botu *et al*., 2017).

In Romania, work on identification, conservation and evaluation of fruit genetic resources was initiated in 1970 in order to limit the loss of the biodiversity due to genetic vulnerability. There are rich collections of plum germplasm for development of cultivars and rootstocks in two research centres: Research Institute for Fruit Growing (RIFG) Pitesti with 642 accessions and Fruit Growing Research and Ex-

Table 1

OVERVIEW OF *EX SITU* PLUM COLLECTIONS IN ROMANIA

No.	Centre	Type of collections	Species and interspecific hybrids	Local accessions	Foreign accessions	Other (biotypes, hybrids, mutants, etc.)	Total no. of accessions
1	RIFG Pitesti	Cultivars	7	183	320	40	550
		Rootstocks	0	82	10	0	92
	TOTAL RIFG		7	265	330	40	642
2	UCv-SCDP Vâlcea	Cultivars	27	56	125	35	216
		Rootstocks		86	21	11	118
	TOTAL ECv-SCDP		27	142	146	46	361
	GENERAL TOTAL		34	407	476	86	1003

tension Station (UCv-SCDP) Vâlcea with 361 accessions, representing wild species, local populations, named cultivars, breeder's selections and rootstocks.

The genetic resources preserved *ex situ* are very valuable and can be useful for breeding new cultivars and rootstocks. The success of any breeding programme depends on the existence of a rich source of valuable germplasm (Cociu *et al*., 1997; Butac *et al*., 2011).

MATERIAL AND METHODS

Presently, in Romania there are plum collections in two centres: RIFG Pitesti and UCv-SCDP Vâlcea. *Ex situ* conservation of accessions is done by different methods: conservation in the field collections (at RIFG Pitesti and UCv-SCDP Vâlcea), conservation in plastic containers and cryoconservation at –196 ^{o}C in liquid nitrogen (at UCv-SCDP Vâlcea) (Braniste *et al*., 2006; Botu *et al*., 2008; Butac *et al.,* 2011). At RIFG Pitesti, the plum cultivar collection established in 1997 includes 550 accessions, and the plum rootstock collection established in 2009 includes 92 accessions. At SCDP Vâlcea, the plum collections established in 1989, 1993, and 1996, include 361 accessions (species, cultivars and rootstocks). The accessions are arranged according to the ripening season. They are grafted on Myrobalan rootstock, spaced at 4 by 4 m (at RIFG Pitesti) and 5 by 4 m, respectively (at Ucv-SCDP Vâlcea). Each genotype is a variant and each variant has 2–5 replications (1 tree = 1 replication).

Between 2000–2017, in these collections of cultivars and rootstocks, observations have been made regarding phenology, productivity, vigour, fruit quality, resistance/tolerance to diseases and pests, resistance to low temperatures, resistance to frost, etc. in order to select genitors for future breeding. The observations and measurements were made according to the IBPGR Prunus descriptors updated by the ECP/GR Prunus Working Group within the Genres CT95 No. 61 project "International network on Prunus genetic resources".

The hybridisation and selection work was carried out in the plum experimental fields of the two research centres (RIFG Piteşti and UCv-SCDP Vâlcea). Current methods involved controlled cross-pollination and repeated positive selection in all development stages of hybrids.

RESULTS

In the Pitesti and Vâlcea centres there are a total of 1003 accessions (cultivars and rootstocks), of which: 34 are species and interspecific hybrids, 407 are local cultivars, 476 are foreign cultivars and rootstocks, and 86 are other genotypes (biotypes, hybrids, mutants, etc.) (Table 1).

Regarding the germplasm collections, one of the Romanian breeder's objectives is to enrich genetic resources by exchange of biological material with similar institutions, as well as by exploring the natural flora. Another objective is to identify new potential genitors for future breeding work (Table 2).

Table 2

GENITORS USED IN THE BREEDING WORK OF PLUM CULTIVARS IN ROMANIA

No.	Objectives	Genitors
1	Basic genitors	Tuleu gras, Centenar, Carpatin, Tuleu timpuriu, Tita, Alina, Record, Minerva, Piteştean, Dâmboviţa, Albatros, Pescăruş, Sarmatic, Vâlcean, Silvia, Iulia, Stanley
2	Resistance / tolerance to PPV	Oneida, Kirke, Grase de Becs, Grase de Peşteana, Agen de Sirăuţi, Boambe de Leordeni, Ontario, Wilhelmina Späth, Kirke, Jojo, Andreea
3	Fruit quality	d`Agen, Agen 707, Grand Prize, Vision, Valor, Oneida, Čačanska rodna, Grase românesti, Agen de Sirăuţi, Vâlcean, Ontario, Renclod Althan, Renclod violet, Record, Vânăt de Italia, Andreea, Agent, Romanţa, Voyageur, Hanita, Haganta, Minerva, Debreceny, Pescăruş, Ozark premier
4	Earliness	Early Rivers, Ruth Gerstetter, Diana, Ialomiţa, Čačanska lepotica, Čačanska ranna
5	Lateness	Anna Späth, Vinete româneşti, Valor, President, Jojo
6	Self-fertility	Stanley, Anna Späth, Bluefre, Ialomiţa, Diana, Romanţa, Čačanska lepotica, Andreea
7	Productivity	Stanley, Anna Späth, Bluefre, Standard, Grase de Becz, Čačanska lepotica
8	Low vigour	Stanley, Bluefre, Wilhelmina Späth

In the breeding work for cultivars, the main objectives are: improvement of the old Romanian cultivars 'Tuleu gras', 'Vinete romanesti', and 'Grase romanesti'; extension of the fruit ripening season; improvement of fruit quality for fresh market; tolerance/resistance to Plum Pox Virus (PPV); self-fertility; increased productivity; and decreased tree vigour.

Taking into account these objectives, breeding for new cultivars used the following different genitors: 'Grase de Becs', 'Carpatin', 'Ialomiža', 'Kirke', 'Wilhelmina Späth' (for resistance / tolerance to Plum pox virus), 'Vinete românești', 'Tuleu timpuriu', 'Anna Späth' (for late blooming), 'Tuleu gras', 'Vâlcean' (for fruit quality), 'Stanley', 'Pescăruș', 'Centenar' (for productivity); 'Diana', and 'Andreea' (for self-fertility) (Table 2).

The plum cultivar breeding programme began 60 years ago, using various breeding methods (controlled hybridisation, open pollination, selection from wild populations, and mutagenesis).

As a results of breeding, 40 new varieties with very good agrobiological characteristics were registered, patented or have patents pending. Conventional breeding methods (crossing) were used to modify the genetic structure of quantitative traits of new plum cultivars. Some of the phenotype characteristics were improved and others did not show positive changes. For example, regarding fruit size, most of the cultivars obtained in Romania have larger fruits than old cultivars like 'Tuleu gras' and 'Grase românești'. The productivity of some new cultivars was improved. Male sterility from 'Tuleu gras' was passed on to the whole offspring, with all cultivars obtained from this maternal parent being male sterile. Regarding the ripening time, most of the new cultivars are earlier than the old cultivars (for example, 'Centenar', 'Tuleu timpuriu', 'Carpatin', 'Tita' are earlier than 'Tuleu gras'). A very good genetic gain was achieved regarding tolerance to Plum pox virus. Several cultivars like 'Ialomiža', 'Carpatin', 'Alina', 'Roman', 'Romanža', 'Agent', 'Andreea' etc. have demonstrated tolerance to PPV (Tables 3–6).

Very good results have been obtained in the rootstock breeding programmes started 50 years ago in the Vâlcea and Pitești centres. In the breeding programmes for plum rootstocks the main objectives are: low to medium vigour;

Table 3

THE GENETIC GAIN OF NEW CULTIVARS OBTAINED FROM 'TULEU GRAS' AND HIS OFFSPRING

No.	Cultivar	Year of registration	Parents	Breeding method used	Genetic gain – trait modified
1	Tuleu timpuriu	1967	Tuleu gras × Pźche	Simple cross	Earlier and more productive than Tuleu gras
2	Superb	1968	Tuleu gras × Abbaye d'Arton	Simple cross	Earlier and bigger fruit than Tuleu gras
3	Tuleu dulce	1968	Tuleu gras × d'Agen	Simple cross	More sweet than Tuleu gras
4	Centenar	1978	Tuleu gras × Early Rivers	Simple cross	Earlier and more productive than Tuleu gras; a few later than Early Rivers
5	Albatros	1979	Tuleu gras o.p.	Open pollination	Earlier and bigger fruit than Tuleu gras
6	Dambovita	1981	Tuleu gras × Anna Späth	Simple cross	Later and bigger fruit than Tuleu gras
7	Pitestean	1982	Tuleu timpuriu × Early Rivers	Pyramid cross	Earlier and bigger fruit than Tuleu timpuriu
8	Carpatin	1982	Tuleu gras × Early Rivers	Simple cross	Earlier and bigger fruit than Tuleu gras; tolerant to PPV
9	Minerva	1984	Tuleu timpuriu × Early Rivers	Pyramid cross	Earlier than Tuleu timpuriu
10	Flora	1989	Tuleu gras × Renclod violet	Simple cross	Earlier and bigger fruit than Tuleu gras; tolerant to PPV
11	Sarmatic	1989	Tuleu timpuriu × Early Rivers	Pyramid cross	Earlier and bigger fruit than Tuleu timpuriu
12	Baragan 17	1990	Tuleu gras × Early Rivers	Simple cross	Earlier than Tuleu gras
13	Tita	1991	Tuleu gras - irradiated stones	Mutagenesis	Earlier and bigger fruit than Tuleu gras; very good fruit quality
14	Alina	1991	Tuleu gras - irradiated stones	Mutagenesis	Earlier and bigger fruit than Tuleu gras; very good fruit quality; Tolerant to PPV
15	Iulia	2002	Tuleu gras × Renclod Althan	Simple cross	Earlier and bigger fruit than Tuleu gras
16	Ivan	2003	Tuleu gras × Vănăt de Italia	Simple cross	Earlier and bigger fruit than Tuleu gras
17	Jubileu 50	2003	Tuleu gras × De Bistriža	Simple cross	Earlier, bigger fruit and more productive than Tuleu gras
18	Roman	2004	Tuleu gras × Early Rivers	Simple cross	Earlier and bigger fruit than Tuleu gras; tolerant to PPV
19	Dani	2004	Tuleu gras × Grase românești	Simple cross	Later than Tuleu gras, but earlier and bigger fruit than Grase romanesti
20	Geta	2004	Centenar × Ialomiža	Pyramid cross	Later and bigger fruit than Centenar
21	Romaner	2005	Tuleu gras × Renclod Althan	Simple cross	Earlier and bigger fruit than Tuleu gras
22	Elena	2005	Tuleu gras × Stanley	Simple cross	Later than Tuleu gras and Stanley
23	Topval	2010	Tuleu gras × Stanley	Simple cross	Earlier and bigger fruit than Tuleu gras; tolerant to PPV

Table 4

THE GENETIC GAIN OF NEW CULTIVARS OBTAINED FROM 'GRASE ROMANESTI'

No.	Cultivar	Year of registration	Parents	Breeding method used	Genetic gain – trait modified
1	Gras ameliorat	1968	Grase romanesti – self pollination	Auto pollination	Earlier, bigger and more sweet fruit; tolerant to PPV

Table 5

THE GENETIC GAIN OF NEW CULTIVARS OBTAINED FROM 'VINETE ROMANESTI'

No.	Cultivar	Year of registration	Parents	Breeding method used	Genetic gain – trait modified
1	Vinete româneşti 300	1970	Vinete româneşti selection	Selection	More productive and tolerant to PPV than Vinete româneşti

Table 6

THE GENETIC GAIN OF NEW CULTIVARS OBTAINED FROM OTHER CULTIVARS

No.	Cultivar	Year of registration	Parents	Breeding method used	Genetic gain – trait modified
1	Silvia	1979	Renclod Althan × Early Rivers	Simple cross	More productive than both parents
2	Pescarus	1979	Renclod Althan × Wilhelmina Späth	Simple cross	More productive than both parents
3	Ialomita	1981	Renclod Althan × Early Rivers	Simple cross	Self-fertile; earlier than Renclod Althan. Tolerant to PPV
4	Diana	1983	Renclod Althan ×x Early Rivers	Simple cross	Self-fertile; earlier than Renclod Althan
5	Record	1983	Renclod violet o.p.	Open pollination	Bigger fruit, more productive than Renclod violet
6	Vâlcean	1990	H 8/12 (R.C. Althan × Wilhelmina Späth) × H 5/23 (R.C. Althan x Early Rivers)	Pyramid cross	Earlier, bigger and more sweet fruit than both parents
7	Renclod de Caransebes	1991	Renclod Althan × Wilhelmina Späth	Simple cross	Earlier than both parents
8	Andreea	2000	H 27/87 OP	Open pollination	More sweet than mother parent; tolerant to PPV
9	Delia	2002	Vănăt de Italia × Anna Späth	Simple cross	Later than Vânăt de Italia; lesser vigour than both parents
10	Agent	2004	Open pollination	Selection	More sweet fruit; tolerant to PPV
11	Doina	2004	Anna Späth × Renclod Althan	Simple cross	Earlier than both parents
12	Matilda	2004	Anna Späth × d'Agen – irradiated with Co^{60}	Simple cross	More productive than Anna Späth
13	Zamfira	2005	Anna Späth × Renclod Althan	Simple cross	Later than Anna Späth; tolerant to PPV
14	Alutus	2010	(R.C. Althan × Early Rivers) × (R.C. Althan × Wilhelmina Späth) × mixed pollen	Pyramid cross	Later and bigger fruit than both parents; tolerant to PPV
15	Romanta	2012	Stanley × Vâlcean	Simple cross	Bigger fruit than Stanley More productive than Vâlcean; tolerant to PPV

tolerance to PPV and foliar diseases; easy propagation; adaptability to heavy soil; good anchorage in the soil; good compatibility with commercially propagated cultivars and positive influence on precocity, yield and fruit quality. The following genotypes have been used as sources of genes to achieve these goals over time: 'Roşior văratec', 'Brompton', 'Renclod Verde', 'Pixy', 'Saint Julien A', 'Albe mici', 'Scolduş', 'Porumbar', etc. (Table 7).

The new rootstocks obtained in Romania are easier to propagate, have lower vigour, are adapted to heavy soils and are resistant to major diseases. Among the newly developed rootstocks are: 'Oteşani 8', 'Oteşani 11', 'Miroval', 'Rival', 'Pinval', 'Corval', 'Oltval' (obtained at UCv-SCDP Vâlcea); 'Mirobolan C5', 'Mirobolan dwarf', 'Adaptabil' and 'Mirodad 1' (obtained at RIFG Piteşti) (Table 8).

DISCUSSION

Most of the Romanian accessions belong to Prunus domestica, Prunus insititia, Prunus cerasifera, Prunus spinosa and *Prunus salicina* (Botu *et al.*, 2008, 2017; Butac *et al.*, 2010).

The evaluation of accessions from collections led to the creation of a database. A large number of genotypes (292),

Table 7

GENITORS USED IN THE BREEDING WORK OF PLUM ROOTSTOCKS

No.	Species	Genitors
1	*Prunus domestica* L.	Roşior văratec, Renclod verde, Brompton, Oteşani 8, Voineşti B
2	*Prunus cerasifera* Ehrh	Corcoduş 163, Corcoduş 169, C2, C3, C4, C10, C12, C13, C16, C17, BN 4Kr
3	*Prunus insititia* Juss	Pixy, Oteşani 11, Albe mici, GF 655.2, Saint Julien A, Scolduş, Poliza de Nurcia
4	*Prunus spinosa* L.	Porumbar de Iaşi, Porumbar A, Porumbar D-PL
5	*Prunus besseyi* Bailey	Brooks, *P. besseyi* selections
6	*Prunus tomentosa* Thunb.	Orient, 473, 474

especially local varieties, have already been introduced into the European Gene Bank of *Prunus;* respectively, 181 genotypes belong to *Prunus domestica*, 38 genotypes belong to *Prunus cerasifera*, 67 belong to *Prunus insititia* and 6 belong to other species (Maggioni *et al*., 2010).

Regarding the breeding work for cultivars, the same objectives, genitors and methods were reported by Botu *et al.* in 2012. The breeding work created a large variability which was very important for future fruit growing in Romania (Butac *et al*., 2010; Butac *et al*., 2013).

Many of the cultivars obtained in the Romanian breeding programme are being cultivated in other European countries ('Tita' in the Netherlands; 'Pitestean' in Germany and Poland; 'Carpatin' in Czech Republic and Poland; 'Tuleu timpuriu' in Bulgaria, etc.) (Butac *et al*., 2011).

Botu *et al.* in 2012 reported similar results concerning the modification of the genetic structure in new plum cultivars obtained in Romania. For example, 'Andreea' and 'Agent' cvs, were described as tolerant to plum pox virus. The male sterility of the plum cultivars obtained from 'Tuleu gras' cv. was also described by Botu and collaborators in the same year.

Regarding the breeding work for rootstocks the same objectives, genitors and methods were reported by Braniste and Butac in 2006, Mazilu *et al.* in 2013. In 2013, Mazilu and his collaborators reported that the main objectives in rootstock breeding are low vigour and good compatibility with commercially cultivars, because the main rootstocks used in the past in Romania and also in Europe were 'Myrobalan' seedlings. Blazec and Pistekova in 2009 and 2012, Kaufmane *et al*. in 2007 and Sosna in 2002 wrote that 'Myrobalan' rootstock is very vigorous and has insufficient compatibility with some cultivars. The new rootstocks developed in Romania were described by Botu *et al.* in 2006, Dutu *et al*. in 1993 and Mazilu *et al.* in 2013.

Table 8

THE PLUM ROOTSTOCKS OBTAINED IN ROMANIA AND THE GENETIC GAIN ACHIEVED FOR CERTAIN CHARACTERISTICS

No.	Rootstock name	Year of registration	Parents	Breeding method used	Genetic gain – trait modified
Valcea center					
1	Otesani 8	1980	Selection in *P. domestica* population	Selection	Seedling rootstock with better grafting compatibility and induced lower vigour than Myrobalan
2	Otesani 11	1987	Selection in *P. insititia* population	Selection	Clonal rootstock with better grafting compatibility and induced lower vigour than Myrobalan
3	Miroval	1998	Selection in *P. cerasifera* population	Simple cross	Clonal rootstock with high multiplication capacity than Myrobalan
4	Rival	2003	Saint Julien H2 × *P. insititia*	Simple cross	Clonal rootstock with high multiplication capacity than Myrobalan
5	Corval	2005	Selection in *P. cerasifera* population	Simple cross	Clonal rootstock with high multiplication capacity than Myrobalan
6	Oltval	2005	(*P. besseyi* × *P. americana*) × *P. cerasifera*	Simple cross	Clonal rootstock with high multiplication capacity than Myrobalan
7	Pinval	2005	Scolduş open pollination	Simple cross	Clonal rootstock with high multiplication capacity than Myrobalan
Pitesti centre					
1	Mirobolan C5	1999	Selection in *P. cerasifera* population	Simple cross	Seedling rootstock with good compatibility with all plum cultivars
2	Mirobolan dwarf	1999	Selection in wild flora	Simple cross	Seedling rootstock which induced lower vigour than Myrobalan
3	Adaptabil (for peach and plum)	2000	*P. besseyi* × mixed pollen (*P. cerasifera, P. domestica, P. persica, P. armeniaca*)	Inter specific cross	Clonal rootstock with high multiplication capacity than Myrobalan; more resistant to heavy soils than Myrobalan
4	Mirodad 1	2017	Mirobolan dwarf × Adaptabil	Inter specific cross	Clonal rootstock which induced lower vigour than Myrobalan

CONCLUSIONS

The genetic resources preserved *ex situ* and *in situ* are very important and can be used for breeding new cultivars and rootstocks. The success of any breeding programme depends on the existence of a rich source of valuable germplasm. In Romania there are very rich plum collections. Over time different genitors were used in plum breeding activities.

As a result of breeding, 40 new varieties and 11 new rootstocks were registered. Modern orchards can be established with some of these cultivars and rootstocks, which were using different breeding methods.

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PLŪMJU ĢENĒTISKIE RESURSI UN SELEKCIJA RUMĀNIJĀ

Rumānijā augļaugu izvērtēšanas un saglabāšanas aktivitātes tika uzsāktas 1970. gadā, lai ierobežotu bioloģiskās daudzveidības samazināšanos. Ģenētiskie resursi izvietoti divās vietās: Augļkopības zinātniskajā institūtā Pitešti — 642 vienības un Valesijā — 361 vienība, kas reprezentē savvaļas sugas, vietējās populācijas, šķirnes, atlasītu selekcijas materiālu un potcelmus. No šo kolekciju ģenētiskajiem resursiem selekcijā izmantotas plūmju šķirnes 'Grase de Becs', 'Carpatin', 'Ialomita', 'Kirke', 'Wilhelmina Späth' (rezistencei/tolerancei pret *Plum pox* vīrusu); 'Vinete românești', 'Tuleu timpuriu', 'Anna Späth' (vēlai ziedēšanai), 'Tuleu gras', 'Vâlcean' (augļu kvalitātei), 'Stanley', 'Pescâruș', 'Centenar' (ražībai) un 'Diana' (pašauglībai). Plūmju potcelmu selekcijas programmai izmantoti: 'Rosior văratec', 'Brompton', 'Renclod Verde', 'Pixy', 'Saint Julien A', 'Albe mici', 'Scolduș', 'Porumbar' u.c. Selekcijas aktivitātes, kas balstītas uz ģenētisko resursu izmantošanu, rezultējušās ar 40 šķirnēm un 11 ģeneratīvi un veģetatīvi vairojamiem potcelmiem.