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EVALUATION OF SELF-INCOMPATIBILITY LOCUS DIVERSITY OF DOMESTIC PLUM (*Prunus domestica* L.) USING DNA-BASED *S*-GENOTYPING

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*There is limited information on genetics of self-incompatibility in domestic plum (*P. domestica *L.). In comparison with other* Prunus *species, there are no S-allele specific markers for the identification of compatibility groups, and thus, genetic diversity of self-compatibility genes is not clear. Six S-locus-specific markers previously used for other* Prunus *species were used to study genetics of self-compatibility of plums, and 33 domestic plum cultivars were genotyped. The applied primer pairs showed good transferability among* Prunus *species and showed high diversity in the tested plant material (14–37 alleles per marker, average observed heterozygocity — 0.953). Applification of the tested primer pairs allowed discrimination of all plum cultivars by unique S-genotypes. Based on the obtained results, primer pairs EM-PC2consFD/ EM-PC3cons RD, PasPcons-F1/ PaC1cons-R1 and F-Box50A/ F-Box intronR are suggested as supplementary markers for characterisation and identification of plum germplasm with potential functional relevance. Although the markers used did not strictly distinguish plum cultivars according to compatibility groups (self-compatible, partly self-compatible and self-incompatible), group-unique amplification fragments were identified, which can serve as a baseline in further development of specific markers.*

Key words: Prunus*, plums, self-incompatibility, genetics, molecular markers.*

INTRODUCTION

Domestic plum (*Prunus domestica* L.) is hexaploid (2n = 6x = 48) allopolyploid species of the Rosaceae family (Botu *et al*., 2002). It is assumed that domestic plum is a naturally formed hybrid that originated about 2000 years ago from diploid (2n = 16) myrobalan (*P. cerasifera*) and tetraploid (2n = 32) *P. spinosa* L. plums (Zohary and Hopf, 2000). Thus, domestic plum has a three-component genome (D1D2C), with genetic formula D1D1D2D2CC, where part C represents the genome of diploid myrobalan plum inherited into domestic plum (Botu *et al*., 2002; Decroocq *et al*., 2004). Plants of the *Prunus* genus have a gametophytic self-incompatibility system. However, unlike other *Prunus* species, domestic plum has both sporophytic and gametophytic incompatibility systems (Botu *et al*., 2002). Sporophytic incompatibility is determined by a mutation in the locus responsible for pollen formation and cytoplasmatic sterility. The gametophytic incompatibility system is based on allele polymorphism in the *S*-RNase coding gene (*S*-gene) and determines three types of pollination: complete compatibility, partial compatibility and complete incompatibility (Botu *et al*., 2002). Due to the three component genome it is difficult to find the same allelic formula or structure for two domestic plum cultivars, since each genome has its own *S*-gene with multiple alleles. For this reason, any plum cultivar having fertile pollen can pollinate any other cultivar, but in different degrees (Selesses and Bonnet, 1994). Extensive studies on self-compatibility have been conducted on other *Prunus* species: sweet cherries (*P. avium* L.) (e.g. Lacis *et al*., 2008; Ipek *et al.*, 2011), sour cherries (*P. cerasus* L.) (e.g. Tsukamoto *et al*., 2008), apricots (*P. armeniaca* L.) (e.g. Jie *et al.*, 2005; Donoso *et al*., 2009) and on some diploid plum species, such as Japanese plum (*P. salicina* Lindl.) (e.g. Hegedüs and Halįsz, 2007; Guerra *et al.*, 2009) and myrobalan plum (*P. cerasifera* Ehrh.) (Sutherland *et al*., 2009). Domestic plum is the least-investigated species in the *Prunoideae* subfamily, which is partly due to its polyploidy and complex structure of genome (Selesses and Bonnet, 1994; Botu *et al*., 2002; Decroocq *et al*., 2004; Hegedüs and Halász, 2007; Sutherland *et al*., 2007; Tao and Iezzoni, 2010). The compatibility groups of domestic plum cultivars are not defined and there is no clear information about genetic diversity of self-incompatibility alleles.

The plum germplasm collection at the Latvia State Institute of Fruit-Growing (LSIFG) contains a wide diversity of domestic plum cultivars (Kaufmane *et al*., 2006; Kārkliņš *et al*., 2007) representing diverse geographic origin (Belarus,

Canada, Estonia, France, Germany, Italy, Latvia, Lithuania, Norway, Russia, Sweden, Ukraine, USA, etc.). Since the incompatibility problem has always been stressed by fruit growers, cytoembryological investigations on plums grown in Latvia have been performed (Kaufmane, 1991). The DNA-based research is in the first steps of implementation (Kota and Lācis, 2013) and *S*-alleles, which determine pollination compatibility, have not yet been studied in plum cultivars and their genetic diversity has not been evaluated. Lack of this information limits detailed characterisation of germplasm, breeding efforts and appropriate orchard design. The aim of the study was to perform DNA-based *S*-genotyping to evaluate genetic diversity of the *S*-locus in plum germplasm and to characterise possible *S*-allele composition and occurrence.

MATERIAL AND METHODS

Plant material. The study was performed at LSIFG on 33 domestic plum (*Prunus domestica* L.) cultivars. Eleven self-compatible, six partly self-compatible and sixteen self-incompatible cultivars were included in the study. Allocation of cultivars to a particular group of self-incompatibility was based on published cultivar descriptions (Kaufmane, 1991; Kārkliņš *et al*., 2007), fertilisation field evaluation and cytoembryology research (Kaufmane, 1991).

Isolation of genomic DNA. Total DNA was isolated from young leaves using a Genomic DNA Purification Kit (Fermentas, Lithuania). The quantification and quality evaluation of DNA was performed by a spectrophotometer NanoDrop 1000 (NanoDrop products, USA).

PCR and fragment analysis. S-genotyping of domestic plum cultivars was performed using five pairs of consensus primers spanning the first and second introns of S-RNase gene (Fig. 1). One primer pair was used to amplify the intron of the SFB gene responsible for pollen-associated self-incompatibility.

Fluorescently labelled primers PaSPons-F1 (Vaughan *et al*., 2006) and PaC1cons-R1 (Sonneveld *et al*., 2006) were used in PCR to amplify the *S*-RNase first intron and F-BOX50A and F-BOXintronR (Vaughan *et al*., 2006) to amplify the SFB intron. Touch-down PCR was employed with cycling of 95 ^{o}C for 15 min followed by 10 cycles of 94 ^{o}C for 30 s, 55 ^{o}C for 90 s with a reduction of annealing temperature by 0.5 ^{o}C per cycle, with final elongation 72 ^{o}C for 60 s, then 25 cycles of 94 ^{o}C for 30 s, 48 ^{o}C for 90 s, 72 ^{o}C for 60 s and a final elongation of 60 ^{o}C for 30 min (Vaughan *et al*., 2006).

PCR amplification products obtained using fluorescently labelled primers were collected and allele sizes determined using a ABI PRISM ® 3100 Genetic Analyzer (Applied Biosystems) and genotyping programme GeneMapper ® Software v4.0 (Applied Biosystems).

Six degenerate primers were used for amplification of the *S*-RNase second intron (Fig. 1). Three of them were EM-PC2consFD (C2), EM-PC3consRD (C3) and EM-PC5consRD (C5) (Sutherland *et al*., 2004). These EM primers were used in combinations C2 + C3 and C2 + C5. PCR conditions were as follows: initial denaturation at 94 ^{o}C for 2 min and 35 cycles of 94 ^{o}C for 10 s, 56 ^{o}C for 2 min and 68 ^{o}C for 2 min, with an increment of 10 s per cycle on the extension step after the 10th cycle of amplification (Sutherland *et al*., 2004). Another three applied primers were PruT2, PruC2 (Tao *et al*., 1999) and PCE-R (Ushijima *et al*., 1998). The primer pair combinations used for PCR amplification were PruT2 + PCE-R and PruC2 + PCE-R. PCR conditions were: an initial step of 3 min at 94 ^{o}C, 35 cycles of 1 min at 94 ^{o}C, 1 min at 56 ^{o}C and 3 min at 72 ^{o}C, and a final step of 7 min at 72 ^{o}C (Guerra *et al*., 2009). Amplification products were separated on 2% TAE agarose gel in 1X TAE buffer and stained with ethidium bromide. A 100 bp DNA ladder (O'RangeRuler 100 bp DNA Ladder, Fermentas, Lithuania) was used for fragment size determination.

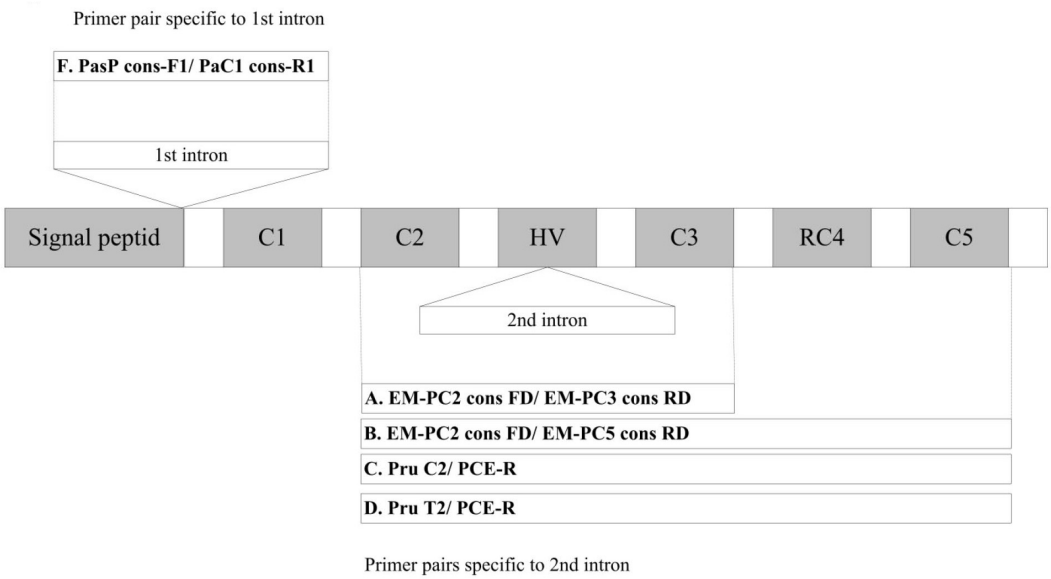

Fig. 1. Annealing sites in the *S*-RNase coding region of *S*-locus specific consensus primer pairs tested on domestic plum (*P. domestica*) cultivars. Structure of a Prunus S-RNase coding region by Ushijima *et al*., 1998 and Tao *et al*., 1999.

Table 1

CHARACTERISTICS OF *S*-RNASE AND SFB INTRON FLANKING CONSENSUS PRIMER PAIRS AND THEIR ABILITY TO DISCRIMINATE PLUM CULTIVAR SELF-COMPATIBILITY GROUPS

Praimer pair	Number of amplification fragments	Fragment length range (bp)	Observed heterozygocity	Gene diversity [PIC value]	Ability to discriminate plum cultivar self-compatibility groups according to AMOVA		
					Number of discriminative amplification fragments	Range of variability among plum cultivar self-compatibility groups, %	Mean variability among plum cultivar self-compatibility groups, %
A. EM-PC2consFD/ EM-PC3consRD	20	270–3000	1.000	0.929	9	1–35	12.9
B. EM-PC2consFD/ EM-PC5consRD	19	490–2500	0.935	0.915	9	1–42	12.4
C. PruC2/ PCE-R	14	300–1970	1.000	0.885	6	7–33	16.8
D. PruT2/ PCE-R	16	700–2600	0.909	0.911	7	2–40	18.1
E. F-Box50A/ F-Box intronR	19	196–235	0.875	0.907	8	1–12	6.1
F. PasPcons-F1/ PaC1cons-R1	31	236–422	1.000	0.942	12	1–40	12.3
Average:	19.8	-	0.953	0.915	8.5	–	13.1

Data analysis. Detected PCR amplification fragments (bands) were coded as present (1) or absent (0) in a binary matrix for statistical analyses. Genetic parameters of tested primer pairs were calculated, as well as Analysis of Molecular Variance (AMOVA) and Principal Component Analysis (PCA) were performed to estimate genetic diversity using GenAlEx 6.4.1 software (Peakall and Smouse, 2006).

RESULTS

S-allele genotyping. All six tested primer pairs exhibited successful PCR amplification and generated amplification products for 30 plum cultivars, with 14 to 31 amplification fragments identified per marker (21.2 in average) (Table 1) showing high genetic diversity. The highest number of amplification fragments was determined for primer pair PasPcons-F1/PaC1cons-R1, and in primer pair PruC2/ PCE-R (Table 1). Application of six tested primer pairs allowed discrimination of all plum cultivars by unique *S*-genotype. Amplification was not successful only for cultivars 'Ave' (using primer pair F-Box50A/F-BoxintronR), 'Gult Äggplomon' (using primer pairs EM-PC2consFD/EM-PC3consRD and EM-PC2consFD/EM-PC5consRD) and 'Zīlīšu Sarkanā Olplūme' (primer pair EM-PC2consFD/ EM-PC5consRD). Full *S*-genotypes (six amplification fragments) were obtained for four plum cultivars ('Lāse', 'Lāse clone 1', 'Melnā Renklode' and 'Ullenas Renklode') using primer pair EM-PC2consFD/ EM-PC3consRD. Partial *S*-genotypes (1–5 amplification fragments identified) were obtained for other plum cultivars, since there was no information on particular fragment copy number or genomic organisation. Heterozygosity for primer pairs ranged from 0.875 to 1.000 with an average value of 0.953 (Table 1). Gene diversity values varied from 0.885 to 0.942, with an average value of 0.915 (Table 1).

Since domestic plum is hexaploid, a maximum of six amplification fragments can be found in one sample. Consensus primer pair PasPcons-F1/ PaC1cons-R1 flanking the first intron of *S*-RNase gene amplified two or five amplification fragments (ranging from 236 to 422 bp) (Table 1). Primer pairs EM-PC2consFD/ EM-PC3consRD, EM-PC2consFD/ EM-PC5consRD, PruC2/ PCE-R and PruT2/ PCE-R ensured amplification of the second intron of the *S*-RNase gene (Fig. 1). Primer pair EM-PC2consFD/EM-PC3consRD amplified two or six putative alleles (ranging from 270 to 3000 bp), primer pair EM-PC2consFD/ EM-PC5consRD had one to five amplification fragments per domestic plum sample (ranging from 490 to 2500 bp), whereas primer pair PruC2/ PCE-R amplified two to five amplification fragments (ranging from 300 to 1970 bp) and primer pair PruT2/ PCE-R had one to six fragments, ranging from 700 to 2600 bp (Table 1). Consensus primer pairs specific for the SFB intron (F-Box50A/ F-Box intronR) amplified one or five amplification fragments (range 196 to 235 bp) (Table 1).

Marker EM-PC2consFD/ EM-PC3consRD had the highest number of amplification fragments per sample (4.0 in average), followed by PasPcons-F1/ PaC1cons-R1 (3.9). The smallest number of fragments per sample was for primer pairs PruT2/ PCE-R and F-Box50A/ F-Box intronR (2.9 in average). The most common number of amplification fragments per individual sample was three and four, and the most seldom number was one and six fragments (Fig. 2). This was observed for all studied markers and could be described by a normal distribution (Fig. 2). Primer pair Pru T2/ PCE-R had comparatively high frequency of cases with two amplification fragments per sample, whereas primer pairs EM-PC2consFD/ EM-PC3consRD and PasPcons-F1/ PaC1cons-R1 had high frequency of five fragments per sample (Fig. 2).

Plum compatibility groups. Eleven self-compatible, six partly self-compatible and sixteen self-incompatible cultivars were genotyped in the study. Significant differences were found in the total number of fragments between compatibility groups of domestic plums. The highest number of amplification fragments was found in the group of self-incompatible plums (93) and the smallest in the group

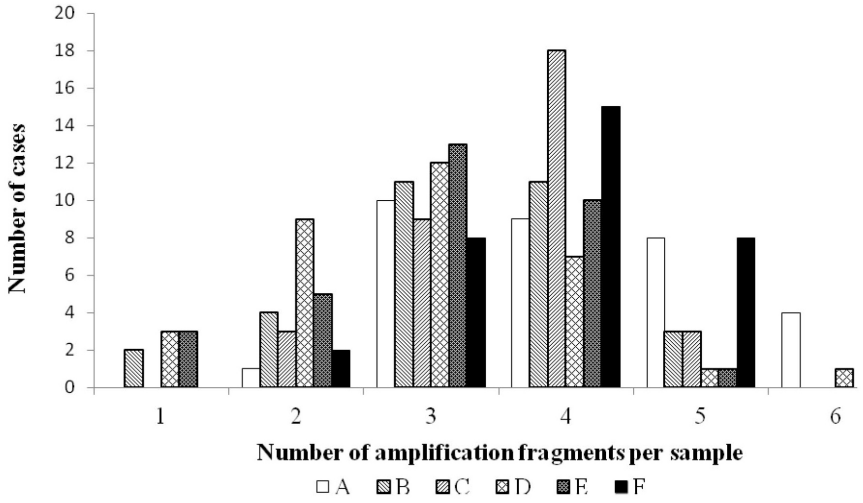

Fig. 2. Frequency of amplification fragments per sample detected in domestic plum (*P. domestica*) cultivars by *S*-locus specific consensus primer pairs.

A - EM-PC2consFD/ EM-PC3consRD

B - EM-PC2consFD/ EM-PC5consRD

C - PruC2/ PCE-R

D - PruT2/ PCE-R

E - F-Box50A/ F-Box intronR

F - PasPcons-F1/ PaC1cons-R1

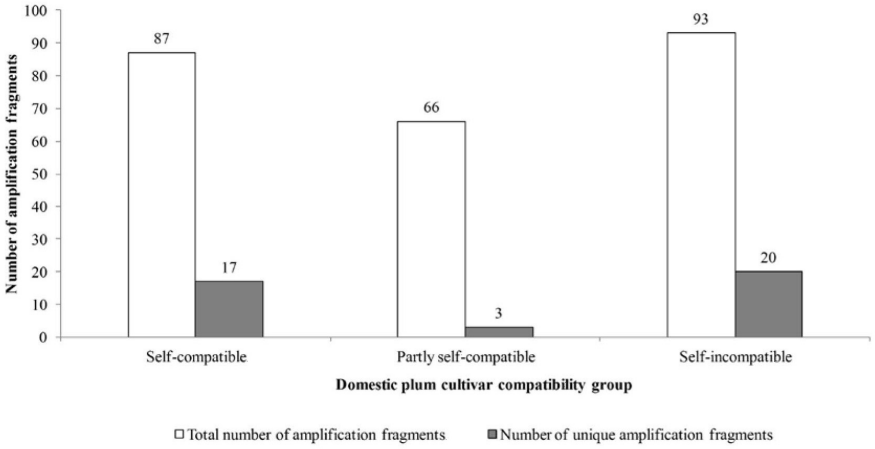

Fig. 3. Number of amplification fragments detected by *S*-locus specific consensus primer pairs depending on the domestic plum (*P. domestica*) compatibility group.

Table 2

CHARACTERISTICS OF *S*-RNASE AND SFB INTRON FLANKING CONSENSUS PRIMER PAIRS BY PLUM CULTIVAR SELF-COMPATIBILITY GROUP

	Consensus primer pair*						Average
	A	B	C	D	E	F	
Group of self-compatible domestic plum cultivars							
Observed heterozygocity	1.000	1.000	1.000	0.909	0.909	1.000	0.970
Gene diversity [PIC value]	0.885	0.889	0.861	0.873	0.914	0.927	0.892
Number of amplification fragments	15	12	11	13	16	20	14.5
Group of partly self-compatible domestic plum cultivars							
Observed heterozygocity	1.000	1.000	1.000	0.833	1.000	1.000	0.972
Gene diversity [PIC value]	0.903	0.898	0.865	0.853	0.872	0.896	0.881
Number of amplification fragments	12	11	9	9	10	12	10.5
Group of self-incompatible domestic plum cultivars							
Observed heterozygocity	1.000	0.867	1.000	0.938	0.813	1.000	0.936
Gene diversity [PIC value]	0.924	0.902	0.878	0.892	0.880	0.932	0.901
Number of amplification fragments	19	14	10	14	12	23	15.3

* A - EM-PC2consFD/ EM-PC3consRD

B - EM-PC2consFD/ EM-PC5consRD

C - PruC2/ PCE-R

D - PruT2/ PCE-R

E - F-Box50A/ F-Box intronR

F - PasPcons-F1/ PaC1cons-R1

of partly self-compatible plums (66). The group of self-incompatible plums had the highest number of unique fragments (20). Seventeen unique amplification fragments were identified in the group of self-compatible and three for partly self-compatible plums (Fig. 3). The highest number of amplification fragments was found for group of self-incompatible plums and the lowest in partly self-compatible cultivars (Table 2, Fig. 3). The highest heterozygocity was

observed in the group of partly self-compatible plum cultivars (0.972), and the highest average gene diversity for the group of self-incompatible cultivars (0.901). Observed heterozygocity for self-compatible and partly self-compatible groups was higher than the average for the whole set of tested plum cultivars, and lower in the self-incompatible group (0.936 and 0.953, respectively) (Tables 1 and 2).

Although unique amplification fragments for the selected compatibility groups were identified, AMOVA detected only 5% difference among these groups of cultivars. For each primer pair, AMOVA identified amplification fragments that were able to distinguish cultivar compatibility groups (Table 1). For the primer pair spanning the *S*-RNase first intron, twelve amplification fragments differed among the groups of cultivars with average variability of 12.3%. The primers pair spanning the *S*-RNase second intron had six to nine amplification fragments and the SFB intron primer pair had eight amplification fragments that could be used to discriminate compatibility groups (Table 1). Wide range of variability among self-compatibility groups was observed: from one to 42% of total allelic variability (Table 1). The lowest average variability among self-compatibility groups was found for the SFB intron primer pair (6.1%), and the highest for primer pair PruT2/ PCE-R (18.1%) spanning the second intron of *S*-RNase gene.

PCA differentiated plum cultivars based on the selected amplification fragments, which corresponded to self-compatibility determined by the field observations (Fig. 4). Self-compatible plum cultivars formed a close group, surrounded by cultivars representing partly self-compatible plums. Self-incompatible plums showed high diversity and formed several distant groups of cultivars (Fig. 4).

DISCUSSION

S-allele genotyping. Genotyping of domestic plum (*P. domestica* L.) cultivars was performed using five *S* locus specific consensus primer pairs that amplied the first and second intron of the the *S*-RNase coding region (Fig. 1) and one primer pair that amplified the SFB intron responsible for the pollen-associated self-compatibility. These markers were developed and used earlier to study genetics of self-compatibility in other *Prunus* species (Ushijima *et al*., 1998; Tao *et al*., 1999; Sutherland *et al*., 2004; Sonneveld *et al*., 2006; Vaughan *et al*., 2006; Guerra *et al*., 2009), with limited application in domestic plum (Sutherland *et al*., 2007). Successful amplification was detected for all primer pairs included in this study; one to six amplification fragments per cultivar were detected. The results confirmed high conservatism of *S*-gene among *Prunus* species and primer pair transferability, as already shown for primer pairs EM-PC2consFD/ EM-PC3consRD, EM-PC2consFD/ EM-PC5consRD, and Pru-C2/ PCE-R in cherries, almonds and apricots (Sutherland *et al*., 2004; Vieira *et al*., 2008). Primer pairs EM-PC2consFD/ EM-PC3consRD, EM-PC2consFD/ EM-PC5consRD, and PruT2/ PCE-R had not been used previously in domestic plum, but were found to ensure good applicability. Conservatism of the *S*-gene locus was supported also by the amplified fragment range of the same primer pairs used in different species. Similar length range of amplification fragments detected by the same primer pair was found in sweet cherries (Vaughan *et al*., 2006) and Japanese plums (Guerra *et al*., 2009).

Consensus primers showed good applicability for characterisation of plum germplasm and cultivar discrimination using a limited number of molecular markers. No identical genotypes were identified and discrimination of closely related clones was possible (e.g. six clones of cultivar 'Latvijas Dzeltenā Olplūme' collected in different locations of Latvia and from neighbouring countries). The applied primer pairs showed very high polymorphism, as 14–31 amplification fragments were detected in 33 domestic plum cultivars, with high observed heterozygocity (average 0.953) and PIC values (average 0.915) (Table 1). A similar number of amplification fragments for domestic plums was found also by Sutherland *et al.* (2007). Sutherland and colleagues (2007) pointed out that due to high polymorphism, application of primer pair PasPcons-F1/ PaC1cons-R1 spanning the first intron of *S*-RNse was sufficient for successful genotyping. Good applicability of this primer pair in the domestic plum cultivars was found earlier in previous studies

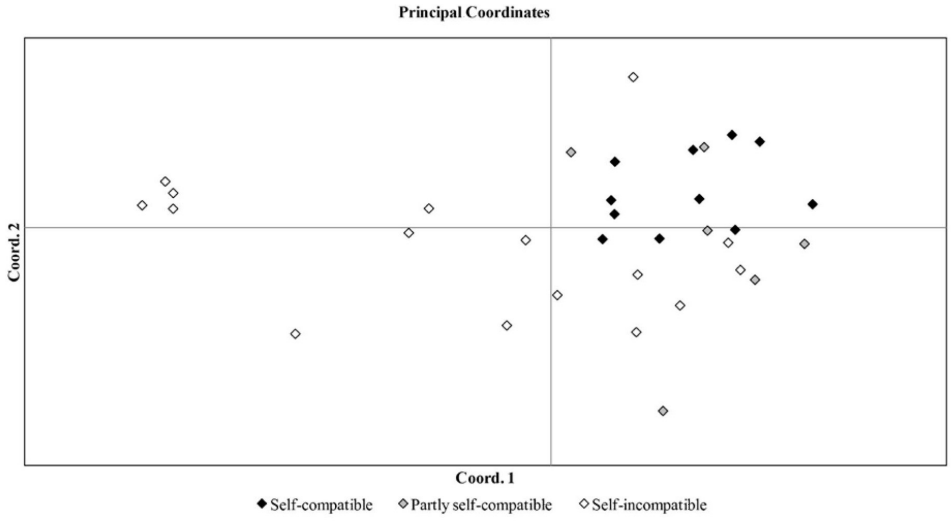

Fig. 4. PCA (principal component analysis) grouping of domestic plum (*P. domestica*) cultivars based on selected amplification fragments detected by *S*-locus specific consensus primer pairs.

on Latvian plum self- compatibility. In those studies, 92 domestic plum and seven diploid plum cultivars of diverse origin were genotyped. In total 13 amplification fragments were detected in the domestic plums, with high observed heterozygocity (1.000) and gene diversity (PIC value 0.895), ensuring good applicability for characterisation of plum germplasm and cultivar discrimination using a limited number of markers (Kota and Lācis, 2013). In this study, primer pair PasPcons-F1/ PaC1cons-R1 produced two to six amplification fragments for domestic plums (ranging from 231 to 415 bp), which is in the range detected for the same primer pair in sweet cherries (Vaughan *et al*., 2006), for which these primers were developed. In the investigation of Sutherland and colleagues (2007), primer pairs spanning the 2nd intron were not sufficiently informative due to a narrow range of amplification fragments, difficulties in scoring scoring stuttered bands and excessive number of detected fragments (more than six per sample). Thus, it was suggested that primer pairs specific for the second intron were not suitable for genotyping. However, in our investigation all primer pairs spanning the second intron of *S*-RNase gene amplified up to six fragments. The lowest distance between bands was 20 bp, and thus the bands were well detectable on agarose gel. Therefore, markers specific for the second intron of *S*-RNase gene could be used for the genotyping of domestic plums, especially primer pair EM-PC2 cons FD/ EM-PC3 cons RD that had high polymorphism (20 amplification fragments detected, fragment length range 279–3000 bp, high heterozygocity (1.000) and PIC value (0.929) (Table 1). These results concur also with previous *S*-genotyping for primer pair Pru C2 / PCE-R, which revealed a high number of amplification fragments for domestic plums (15), high observed heterozygocity (0.967) and gene diversity value (0.880) (Kota and Lācis, 2013). Detailed information on domestic plum self-compatibility can not been acquired without considering pollen-associated compatibility. Therefore, primer pair F-Box50A/ F-Box intronR needs to be employed to ensure more complete characterisation. Based on the obtained results we suggest that primer pairs EM-PC2 cons FD/ EM-PC3 cons RD, PasP cons-F1/ PaC1 cons-R1 and F-Box 50A/ F-Box intron R can be used as supplementary markers in the characterisation and identification of plum genetic resources with potential functional relevance.

Plum compatibility groups. PCA grouping of cultivars based on *S*-genotyping (Fig. 4) was not in accordance with previous cultivar compatibility characterisation conducted by cultivar evaluation and field trials. Information about compatibility of these cultivars was collected from publications and field observations and trials. One of the reasons for this might be that particular alleles do not ensure self-compatibility or self-incompatibility, and that allele combinations and their complex interactions are important. Domestic plum is a hexaploid allopolyploid species (Botu *et al*., 2002) for which inheritance of the *S*-gene has still not been elucidated. Although direct association among polyploidy and self-compatibility has not been observed (Mable, 2004), it may influence the compatibility reaction due to competition among alleles, as in sour cherries, or other mechanisms and demonstrate biases of the self-compatibility reactions. The functions of self-compatibility reactions of allopolyploid species have not been sufficiently studied (Mable, 2004). In addition, field observations and trials need to be supported by cytoembryological investigations, since pollination processes are affected by the environment, which may hide heredity or modify its external manifestation. Although the applied markers could not strictly distinguish selected plum cultivars according to compatibility groups (self-compatible, partly self-compatible and self-incompatible), group unique amplification fragments were identified (Fig. 3), which could serve as a baseline in further development of specific markers.

AMOVA was applied to evaluate the differences among plum compatibility groups and to select amplification fragments potentially applicable for discrimination of the groups. Although variability between self-compatibility groups was low (5%), six to twelve fragments that significantly differed between the groups were identified, which ensured the discrimination of self-compatibility groups (Table 1). These alleles accounted for 6.1 to 18.1% of the variability among plum cultivar groups. Selected amplification fragments were used in subsequent PCA to test their discrimination and grouping power (Fig. 4). Analysis of selected amplification fragment data showed correlation with phenotypical observations, as expected. The highest diversity was found among self-incompatible plum cultivars (Fig. 4). They formed a diffuse group with several closely related sub-groups, which could be used for further designation of domestic plum cultivar cross compatibility groups.

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MĀJAS PLŪMJU (*Prunus domestica* L.) PAŠNESADERĪBAS LOKUSA DAUDZVEIDĪBAS NOVĒRTĒJUMS, IZMANTOJOT *S*-GENOTIPĒŠANU

Joprojām ir nepietiekama informācija par pašnesaderības iedzimšanu mājas plūmēm (*P. domestica* L.). Salīdzinājumā ar citām *Prunus* sugām, tām nav izstrādāti *S*-alēlēm specifiski molekulārie marķieri, identificētas šķirņu saderības grupas, nav zināma pašnesaderības lokusa daudzveidība. Tāpēc, izmantojot sešus citām *Prunus* sugām izstrādātos *S*-lokusam specifiskos molekulāros marķierus, tika veikta 33 mājas plūmju genotipēšana. Lietotie marķieri uzrādīja labu starpsugu pārnesi un nodrošināja augstu ģenētisko daudzveidību (konstatēti 14–37 amplifikācijas fragmenti, vidējā heterozigotāte — 0,953). Izmantotie molekulārie marķieri uzrādīja augstu izšķirtspēju un nodrošināja visu pētījumā iekļauto mājas plūmju šķirņu identifikāciju. Pamatojoties uz iegūtajiem rezultātiem, praimeru pāri EM-PC2consFD/ EM-PC3consRD, PasPcons-F1/ PaC1cons-R1 un F-Box50A/ F-Box intronR tiek rekomendēti kā papildus marķieri plūmju augu materiāla raksturošanai un identifikācijai. Lai gan pētījumā lietotie molekulārie marķieri nespēja grupēt plūmju šķirnes pēc to piederības pašsaderības gupām (pašsaderīgas, daļēji pašsaderīgas un pašnesaderīgas), tika identificēti katrai no šīm grupām unikāli amplifikācijas fragmenti, kas izmantojami turpmākajā darbā, izstrādājot saderības grupām specifiskos marķierus.