UTILZATION OF SPOROPHYTIC COMPATIBILITY FOR ESTABLISHING THE POLLENIZERS FOR 'URIAŞE DE VÂLCEA' HAZELNUT CULTIVAR

UTILIZAREA COMPATIBILITATII SPOROFITICE PENTRU STABILIREA POLENIZATORILOR LA SOIUL DE ALUN 'URIASE DE VALCEA'

VICOL ADINA, BOTU M., BOTU I., NEAGOE A.
Fruit Growing Research and Development Station Vâlcea

Abstract. *The hazelnut has self-incompatibility and inter-incompatibility pollen. The research work carried out till now allowed to establish that pollen incompatibility in hazelnut cultivars is controlled by a series of alleles of a single sterility gene named "S". The pollen compatibility can be determined with the help of female inflorescences, pollen tester and florescence microscope. The establishing of hazelnut pollenizers on basis of sporophytic compatibility study is important for assuring of high fruit yield, achievement of a correct planting scheme. In this paper it is presented the pollen compatibility determination carried out in the laboratory for 'Uriaşe de Vâlcea' cultivar, this being pollinated with tester pollen from the following cultivars: 'Tonda Gentile delle Langhe' ($S_2\underline{S_7}$), 'Tonda Romana' ($\underline{S_{10}S_{20}}$), 'Vâlcea 22' ($S_2\underline{S_{10}}$), 'Butler' ($S_2\underline{S_3}$), 'Ennis' ($\underline{S_1}S_{11}$), 'Segorbe' ($\underline{S_9}S_{23}$), 'Uriaş de Halle' ($S_2\ \underline{S_5}$). As result of this work it was established that cultivars like: 'Vâlcea 22', 'Butler', 'Ennis' and 'TGDL' can be used as pollenizers for 'Uriaşe de Vâlcea'. Depending on the pollinating compatibility, the hazelnut cultivar 'Uriaşe de Vâlcea' can have $S_{11}\underline{S_{20}}$ allelic formula. The research on this topic will continue in order to confirm the allelic formulas.*

Rezumat. *Alunul prezintă fenomenul de auto-incompatibilitate si inter-incompatibilitate polinică. Cercetările efectuate până în prezent au permis să se stabilească faptul că incompatibilitatea polinică la alun este controlată de o serie de alele ale unei singure gene de sterilitate numită "S". Compatibilitatea poate fi determinată cu ajutorul inflorescentelor femele, a polenului tester si a microscopului cu fluorescentă. Stabilirea polenizatorilor, pe baza studiului compatibilităţii sporofitice, este importantă pentru asigurarea unei productii de fructe ridicate, realizarea unor scheme de plantare corecte si realizarea unor programe de ameliorare.Determinarea compatibilităţii a fost efectuată în laborator, la soiul 'Uriaşe de Vâlcea', acesta fiind polenizat, pe rând, cu polen tester cunoscut din soiurile: 'Tonda Gentile delle Langhe' ($\underline{S_7}S_2$), 'Tonda Romana' ($\underline{S_{10}S_{20}}$), 'Vâlcea 22' ($\underline{S_{10}}S_2$), 'Butler' ($\underline{S_3}S_2$), 'Ennis' ($\underline{S_1}S_{11}$), 'Segorbe' ($\underline{S_9}S_{23}$), 'Uriaş de Halle' ($\underline{S_5}S_2$). În urma determinărilor efectuate s-a stabilit că polenizatori buni pentru acest soi pot fi: 'Vâlcea 22', 'Butler', 'Ennis' şi 'TGDL'.În funcţie de compatibilitatea la polenizare a rezultat că soiul românesc 'Uriaşe de Vâlcea' poate avea următoarea formulă alelică $\underline{S_{20}}\ S_{11}$. Cercetările vor fi continuate pentru a se putea confirma formula alelică..*

Corylus genus is included into the *Betulaceae* Family, *Fagales* Order. All the *Corylus* species are monoecious and dichogamous. *Corylus avellana* and *Corylus maxima* species contributed substantially to the forming of hazelnut cultivars.

Also, it is known that hazelnut has self-incompatibility and inter-incompatibility pollinating systems.

Studies carried out by Thompson (1979a) and Germain (1981) showed that pollen incompatibility in hazelnut it is controlled by an allelic series of a single sterility gene named "*S*". The system is of sporophytic type because the pollen reaction is determined by the 2 alleles that are present into the anther tissue.

The *S* alleles of a number of hazelnut cultivars have been determined and published by Thompson (1979b), Mehlenbacher and Thompson (1988), Erdogan et al. (2005), etc.

Until now, 26 *S* alleles have been identified for *C. avellana* and over 40 allelic formulas have been emphasized at various cultivars. Compatibility can be determined with the help of female inflorescence, tester pollen and florescence microscope. Hazelnut pollinators' establishing for cultivars on basis of sporophytic compatibility is important for assuring a high fruit yield, achievement of a correct planting scheme and for breeding.

The study of the pollen compatibility has a major importance for the Romanian hazelnut cultivars that were obtained in the last years, for this reason we had in view to establish the allelic formula and the suitable pollinators for 'Uriaşe de Vâlcea', a cultivar with perspectives for spreading into culture due to its large fruits and high productivity.

MATERIAL AND METHODS

For the present study plants of Romanian cultivar 'Uriaşe de Vâlcea' from the national hazelnut collection from SCDP Vâlcea have been used. In February, the catkins (before opening) have been removed from tree branches and the emasculated branches were isolated with paper bags.

In the same time, tester pollen from the pollinators cultivars was harvested: 'Tonda Gentile delle Langhe' (S_2S_7), 'Tonda Romana' ($S_{10}S_{20}$), 'Vâlcea 22' (S_2S_{10}), 'Butler' (S_2S_3), 'Ennis' (S_1S_{11}), 'Segorbe' (S_9S_{23}), 'Uriaş de Halle' (S_2S_5). The pollen was introduced into the refrigerator (at 3°C or 4°C) for storing till pollination time.

When the styles of the female flowers from the isolated and emasculated branches have riched 2 - 6 mm, the glomerules were detached and introduced into the tester pollen and then were leaved on a wet filter paper into a Petri dish. After 16 hours the styles were detached and squashed into a drop of blue aniline (0.1g blue aniline, 0,71g K_3PO_4, 100 ml distilled water), and then observed at the fluorescence microscope (after Mehlenbacher and Thompson, 1988).

In the case of incompatibility, the pollen grain that was germinated is producing short pollen tubes, which cannot penetrate the stigma surface, most of them forming a bulb at the end of the tubes. If pollen compatibility occurs, then the pollen tubes penetrate the stigma surface and a mass of long, parallel and light colored tubes can be observed.

RESULTS AND DISCUSSIONS

Pollinating tests carried out at 'Uriaşe de Vâlcea' with various pollinators and results observed at fluorescence microscope have showed different reactions. The results obtained allowed us to classify the pollination combinations into compatible or incompatible (not compatible). The 'Uriaşe de Vâlcea' *x* 'Segorbe' and 'Uriaşe de Vâlcea' *x* 'Uriaş de Halle' combinations did not show clear results under the microscope and couldn't be included into the compatible or not compatible groups (Table 1).

Table 1

Pollinating scheme of 'Uriaşe de Vâlcea' cultivar and the results obtained

No. crt.	Pollinated cultivar	Pollinator	Results obtained at fluorescence microscope
1	'Uriaşe de Vâlcea'	'Tonda Gentile delle Langhe' (S_2S_7)	+
2	'Uriaşe de Vâlcea'	'Tonda Romana' ($S_{10}S_{20}$)	-
3	'Uriaşe de Vâlcea'	'Vâlcea 22' (S_2S_{10})	+
4	'Uriaşe de Vâlcea'	'Butler' (S_2S_3)	+
5	'Uriaşe de Vâlcea'	'Ennis' (S_1S_{11})	+
6	'Uriaşe de Vâlcea'	'Segorbe' (S_9S_{23})	Not observed
7	'Uriaşe de Vâlcea'	'Uriaş de Halle' (S_2S_5)	Not observed

„+" - compatible
„-" - incompatible
The underlined alleles are dominant or codominant.

'Uriaşe de Vâlcea' hazelnut cultivar has as genitors the cultivars 'Ennis' (S_1S_{11}), where the allelic formula is known, and 'Red Lambert', with unknown allelic formula.

Testing the pollen compatibility of 'Uriaşe de Vâlcea' with 'Ennis' the results obtained confirmed that these are compatible. It seems that 'Uriaşe de Vâlcea' has inherited from 'Ennis' the S_{11} non dominant allele. In the case when S_1 dominant allele is inherited from 'Ennis', no compatibility between these 2 cultivars exist. After testing the crossing 'Uriaşe de Vâlcea' *x* 'Tonda Romana' ($S_{10}S_{20}$) it was established that they are not compatible, so 'Uriaşe de Vâlcea' cultivar can have one of the two allelic formulas: $S_{11}S_{20}$ or $S_{10}S_{11}$. When testing 'Uriaşe de Vâlcea' *x* 'Vâlcea 22' (S_2S_{10}) it was obvious that compatibility exists when 'Uriaşe de Vâlcea' has $S_{11}S_{20}$ allelic formula. If 'Uriaşe de Vâlcea' has $S_{10}S_{11}$ formula, then no compatibility with 'Vâlcea 22' (S_2S_{10}) exists. The last pollen test crosses between 'Uriaşe de Vâlcea' ($S_{20}S_{11}$) and 'Butler' (S_3S_2) and 'Uriaşe de Vâlcea' ($S_{20}S_{11}$) and 'TGDL' (S_7S_2) proved to be compatible (Fig. 1)
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'Ennis'	*x*	'Red Lambert'	→	'Uriaşe de Vâlcea'
(S_1S_{11})		*(S? S?)*		*(S?S?)*
'Uriaşe de Vâlcea'	*x*	'Ennis'	Compatible →	Allelic proposal for 'Uriaşe de Vâlcea'
(S?S?)		*(S_1S_{11})*		*($S?S_{11}$)*
'Uriaşe de Vâlcea'	*x*	'Tonda Romana'	Incompatible →	Allelic proposal for 'Uriaşe de Vâlcea'
($S?S_{11}$)		*($S_{10}S_{20}$)*		*($S_{11}S_{20}$)* or *($S_{10}S_{11}$)*
'Uriaşe de Vâlcea'	*x*	'Vâlcea 22'	Compatible →	Allelic proposal for 'Uriaşe de Vâlcea'
($S_{11}S_{20}$) or *($S_{10}S_{11}$)*		*(S_2S_{10})*		*($S_{11}S_{20}$)*
			Incompatible →	Allelic proposal for 'Uriaşe de Vâlcea'
				($S_{10}S_{11}$) x (S_2S_{10})
'Uriaşe de Vâlcea'	*x*	'Butler'	Compatible →	Allelic proposal for 'Uriaşe de Vâlcea'
($S_{11}S_{20}$)		*(S_2S_3)*		*($S_{11}S_{20}$)*
'Uriaşe de Vâlcea'	*x*	'TGDL'	Compatible →	Allelic proposal for 'Uriaşe de Vâlcea'
($S_{11}S_{20}$)		*(S_2S_7)*		*($S_{11}S_{20}$)*

Fig.1. Mode of determining of the allelic formula for the 'Uriaşe de Vâlcea' hazelnut cultivar

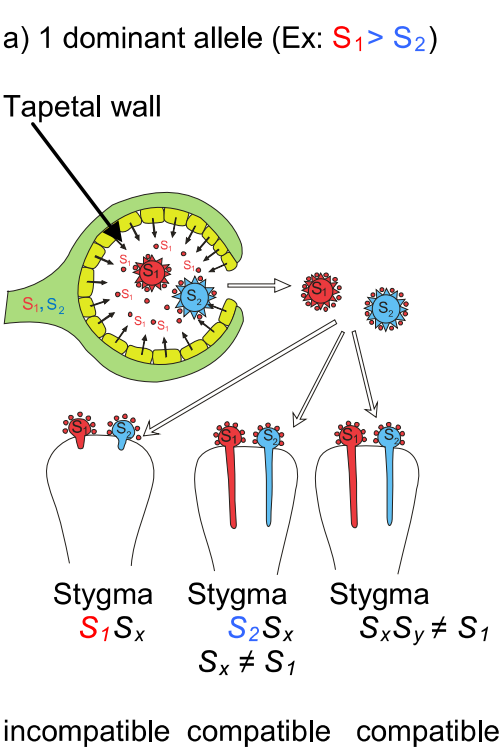

Fig. 2. Anther of a S_1S_2 cultivar with S_1 dominant allele at the level of tapetal wall for producing an incompatibility protein that is fixed on the external wall of the pollen grain.

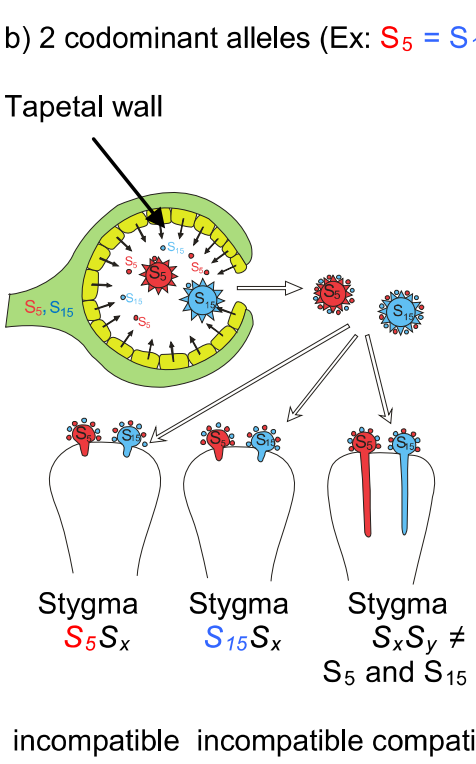

Fig. 3. Anther of a S_5S_{15} cultivar with S_5 and S_{15} codominant alleles at the level of tapetal wall for producing an incompatibility protein that is fixed on the external wall of the pollen grain.

Determinations carried out have respected the cases regarding dominance and codominance mechanisms for sporophytic pollen compatibility (after Germain, 2004).

The emphasize of the sporophytic compatibility of 'Uriaşe de Vâlcea' Romanian hazelnut cultivar and of other cultivars will be continued, in this manner the allelic formulas can be confirmed.

CONCLUSIONS

The establishing of the sporophytic compatibility with the help of fluorescence microscopy is a quick and efficient method for the Romanian hazelnut cultivars.

The cultivar 'Uriaşe de Vâlcea' proved to be compatible at pollination with 'Ennis', Valcea 22, 'Butler' and 'TGDL' and is incompatible with 'Tonda Romana'.

The allelic formula proposed for the hazelnut cultivar 'Uriaşe de Vâlcea' is $S_{11}S_{20}$.

The allelic formula determined based on sporophytic compatibility study can be used for establishing planting schemes (hazelnut cultivars with the right pollinators), that will assure high fruit yields, and also in the breeding work.

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