Theses of Doctoral Dissertation

FLORAL BIOLOGY AND FRUCTIFICATION FEATURES OF DISEASE RESISTANT APPLE VARIETIES AND CANDIDATES

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1. INTRODUCTION

Apple is produced in the world in the fourth largest amount but is the most important fruit species in the moderate climate. It is also the most important fruit species in Hungary, according to the production volume.

Cultivation problems (e.g. extreme weather conditions, changing market trends) increasing year by year raise the attention to varieties with higher ecological tolerance and disease resistance.

Allogamy is a widespread phenomenon in the plant kingdom. Species ‘utilize’ this phenomenon in order to enhance variability (Halász , 2008). In the case off apple the majority of varieties provide maximum 1-2% self compatibility. This value is not enough for reaching a maintainable crop production. That is why apple is called practically self incompatible. When planting an orchard considering market aspects is not sufficient. Variety association plans are needed, in order to find proper pollen donor for each variety in proper distance. Suitability as pollen provider depends on many aspects. Biological and technological aspects must be taken into consideration as well.

The knowledge of floral and fructification features are essential for orchard planning. On the other hand these attributes have outstanding role in the productive phase of the orchard. For example the common blooming time of the associated varieties determine the timing of plant protection or fertigation elements.

We discuss floral phenology and fructification features of scab resistant apple varieties and multi-resistant candidates (resistance against scab, powdery mildew and fire-blight) in this dissertation. We would like to give solutions for proper variety associations in new orchards. We would also like to recommend variety associations for ecological orchards, that presents opportunity to exploit the real benefit of resistant varieties.

2. OBJECTIVES

Our main objectives were the followings:

1. evaluation and development of floral penology and fructification observations
   - data sheet and function system compilation in order to simplify data collection and working up; software development
2. in the case of scab resistant apple varieties and multi-resistant candidates bred in Hungary:
   - determination of blooming time and floral phenology
     - blooming overlap determination
     - blooming time group creation, variety classification into the groups
   - determination of self compatibility level
   - determination of cross compatibility level
     - cross compatibility evaluation with traditional methods after manual crossings
     - fructification values comparison with literature data (*S*-allele compatibility system)
   - *In vitro* determination of pollen germination
   - Determination of ploidy level
3. Metaxenic effect evaluation on fruits from crossings
4. Variety association plan elaboration according to results of the above listed objectives.

3. MATERIAL AND METHOD

3.1. Introduction of the test orchards

Field evaluations (floral phenology observation, self compatibility evaluation, and crossing) were carried out in the two test orchards of the Corvinus University of Budapest.

The orchard of Szigetcsép (N lat. 47°15'; E long. 18°58') was planted in 1997 on Rootstock M9 with wired supporting system in spacing 4 × 1,5 m. trees were trained as slender spindle.

The orchard of Soroksár (N lat. 47°23'; E long. 19°08') was planted in 2003 on rootstock M9 supported with stakes in spacing 3,5 × 1 m. Trees were also trained as slender spindle.

Both orchards are equipped with dripping irrigation system but regular water supply was provided only in Soroksár.

3.2. Varieties and candidates involved

In our experiment scab resistant apple varieties and multi-resistant candidates bred in Hungary were involved (table 1.).

Table 1. Varieties involved

Hungarian candidates	Susceptible control varieties	German resistant varieties	Resistant varieties from the USA	French resistant varieties	Resistant varieties from the Czech Republic
MR-03 MR-09 MR-10 MR-11 MR-12 MR-13	Golden Reinders Delicious Idared	Reanda Reglindis Reka Remo Renora Resi Retina Rewena	Freedom Liberty Prima Priscilla	Baujade Florina X6398	Produkta Rajka Resista Rubinola Topaz

3.3. Floral attributes examination

3.3.1. Floral phenology observations

200-500 flowers on 1-3 branches of 1-3 trees were labelled in red bud stage from each variety. Selected branches were labelled with nursery labels. One year old twigs were excluded according to the methods of Wertheim (1996). Number of buds opened and worn flowers were counted daily during the whole blooming period. Observations were carried out consistently in morning hours in the same variety order (Tóth et al., 1975). Flowers were considered as buds till petal open, as far as the reproductive floral parts were hidden. Flowers were considered as worn flowers when more than the half of the anthers was withered. Data were treated as percentage values in order to the easier comparison. Namely the ratio of buds, opened flowers and worn flowers were determined as the percentage of the total number of the observed flowers of a variety.

3.3.2. Creation of blooming time groups.

According to the ratio of opened flowers varieties were classified into blooming time groups. According to the literature we also created four groups (early, mid-early, mid-late, and late). The grouping was carried out with cluster analysis. SPSS software was used for the cluster analysis (Vargha, 2007).

As we wanted to create four groups SPSS was instructed to create 3-5 groups. In most cases the four groups were taken into consideration; however 5-cluster-classification can be useful in some years when one of the varieties bloom extremely early or late.

3.3.3. Evaluation of blooming overlap

Blooming overlap of the varieties were determined by the overlap of their blooming phenograms. The common area of opened flowers phenograms shows the real blooming overlap; namely the percentage of the overlap of one variety to and contrary.

In order to accelerate and simplify the calculation of the phenogram areas we elaborated software with MATLAB software package.

3.4. Fructification features examinations

3.4.1. Self compatibility and parthenocarpy level determination

100-150 flowers on 1-3 branches of 1-3 trees were labelled in red bud stage from each variety. Selected branches were isolated with paper bags, when the flowers were in red bud stage or maximum in balloon stage. Paper bags were removed after petal fall. This is how we provided flowers from alien pollen. Although we also excluded the opportunity of bee pollination; the effect of wind and the manual shaking of paper bags helped the own pollen to get on the stigmas.

As we had no opportunity to examine the seed content, fruits derived from self pollination and parthenocarpy were treated together.

Fruit set values were treated as percentage values in order to the easier comparison. Fruit set was evaluated three times according to the natural fruit drop periods (first drop, June drop, and pre-harvest drop).

3.4.2. Crossing – evaluation of cross compatibility

For crossing only non-opened flowers can be involved, in order to exclude alien pollen. That is why we carried out the crossing in balloon stage. According to the method elaborated in the Department of Pomology two lateral flowers of a cluster were emasculated, namely the petals and anthers were removed. Pollination was carried out once with pollen stored for one year in refrigerator. Isolator bags were not used. Fruit set was evaluated in the above mentioned three times.

3.4.3. Ploidy level examination

Ploidy level plays a significant role in the suitability of a variety as pollen provider. Ploidy level was examined on samples prepared from the youngest leave tips. Flow citometer was used for the measurements.

A wide spread puffer was used for cell isolation (Galbraith et al., 1983). After this the suspension was filtered on a 30 μm CellTricks filter (Partec GmbH). Samples were stored on 4 ºC. Propidium-iodid dye was added immediately before the flow citometer measurement (20 μl/sample).

The principle of flow citometer measurement is the following: diploid plants cells before DNA replication (in G1 phase) give trace at zone 200, while cells after DNA replication give trace at zone 400; in the case of triploid plant cells give trace at zone 300 and 600 respectively.

3.4.4. Pollen germination tests

Pollen germination tests were carried out *in vitro*. Pollen was reached out from flowers in balloon stage. Anthers were stored (dried) at room temperature for 2-3 days. After anther dehiscence a few pollen was placed onto microscope slide. 10 % sucrose solution was added in order to enhance pollen germination. After six hours germination time 1-2 drops of acetocarmine was added.

400-500 pollen grains of each variety were examined. Pollen germination was expressed in percentage values.

3.5. Fruit quality evaluation / metaxenic pollen effect examination

Metaxenia examinations were carried out on fruits derived from crossings. As self compatible control was not available (due to the practically self incompatibility of apple!!!) 10 pieces control samples from open pollination of mother and father varieties were used.

Physical (fruit diameter, height, weight, colour, stalk length, consistency) and chemical (sugar and acidic content) parameters of the fruits samples were measured.

Analysis of variance was used in the case of normal sample distribution and large sample number. In the case of lower sample numbers and Kruskal-Wallis and Mann-Whitney non-parametric probes were used.

4. RESULTS AND DISCUSSION

4.1. Floral phenology and blooming order of investigated varieties, creation of blooming time groups

Between 2006 and 2009 a larger variety group was observed in Szigetcsép. A less group of varieties was observed in Soroksár between 2007and 2009. Floral phenology is introduced in table 2. in the case of our 2006 Szigetcsép observations. Floral phenology is illustrated with the ratio of opened flowers of each day. Days with over than 25 % opened flower ratio were considered as full bloom period (indicated with grey colour). Blooming order was determined according to the full bloom, as this is the most important feature considering blooming overlap and cross pollination.

Table 2. Floral phenology of investigated varieties (Szigetcsép 2006)

fajta	4. 4.	4. 5.	4. 6.	4. 7.	4. 8.	4. 9.	4. 10.	4. 11.	4. 12.	4. 13.	4. 14.	4. 15.	4. 16.	4. 17.	4. 18.	4. 19.	4. 20.	4. 21.	4. 22.	4. 23.	4. 24.	4. 25.	4. 26.	4. 27.	4. 28.	4. 29.	4. 30.
Reglindis	2	4	8	17	21	24	35	33	27	23	20	17	14	9	5	3	2	1									
MR-13	2	3	6	11	17	23	28	34	35	24	20	17	12	8	6	4	2										
Prima	1	4	9	13	17	20	24	45	42	23	21	18	15	10	6	4	2	1									
MR-12		2	5	9	15	21	24	30	33	33	22	17	15	11	8	5	1	1									
Liberty			3	8	11	17	21	25	30	33	31	28	22	20	16	12	8	4	3	1							
Retina		1	4	7	16	20	22	23	28	31	34	22	20	16	11	9	7	5	2								
Topaz			2	5	8	14	20	24	26	30	35	32	24	19	20	17	12	8	6	3	1						
MR-09			1	6	9	15	20	22	23	29	40	37	24	20	17	15	10	6	2								
Idared					2	5	8	13	20	24	28	25	25	20	18	16	14	10	8	5	3	1					
Produkta					1	6	10	15	21	23	38	40	24	20	17	15	10	8	4	2	1						
Priscilla						2	5	7	13	23	35	31	22	21	18	14	11	9	6	5	3	3	1				
Reka							1	6	10	18	26	33	27	22	20	19	17	14	10	6	2						
X6398								3	8	16	25	29	33	31	22	20	16	14	12	8	4	2	1				
Rubinola								3	6	12	19	26	30	33	38	31	21	16	12	8	5	3	2				
Resi							2	4	9	15	20	23	26	35	39	22	20	16	13	7	4	1					
MR-11							3	5	7	10	17	21	30	33	34	29	24	20	17	16	11	8	5	3			
Resista								1	7	10	16	20	24	33	39	34	24	21	20	18	13	10	6	4	1		
Renora								3	8	13	19	21	23	30	30	34	31	22	19	16	12	10	8	3	2		
MR-10									2	4	7	11	19	23	30	35	31	20	16	12	10	8	5	4	2		
Freedom									1	6	10	16	20	22	24	30	38	30	24	20	17	15	9	6	4	1	
Baujade											2	5	8	17	23	27	35	34	30	28	24	20	16	11	8	3	1

*grey marking: full bloom period (opened flower ratio over 25%)

Blooming time groups were created according to the cluster analyses in each year. Statistic probes supported our grouping in most cases. However in some years cluster analysis revised our grouping especially in the mid-late and late blooming time groups. This can be explained with the fact that meteorological features have larger and larger impact on blooming time with the progress of blooming. A multiple variable statistical test can follow this more exactly (figure 1.).

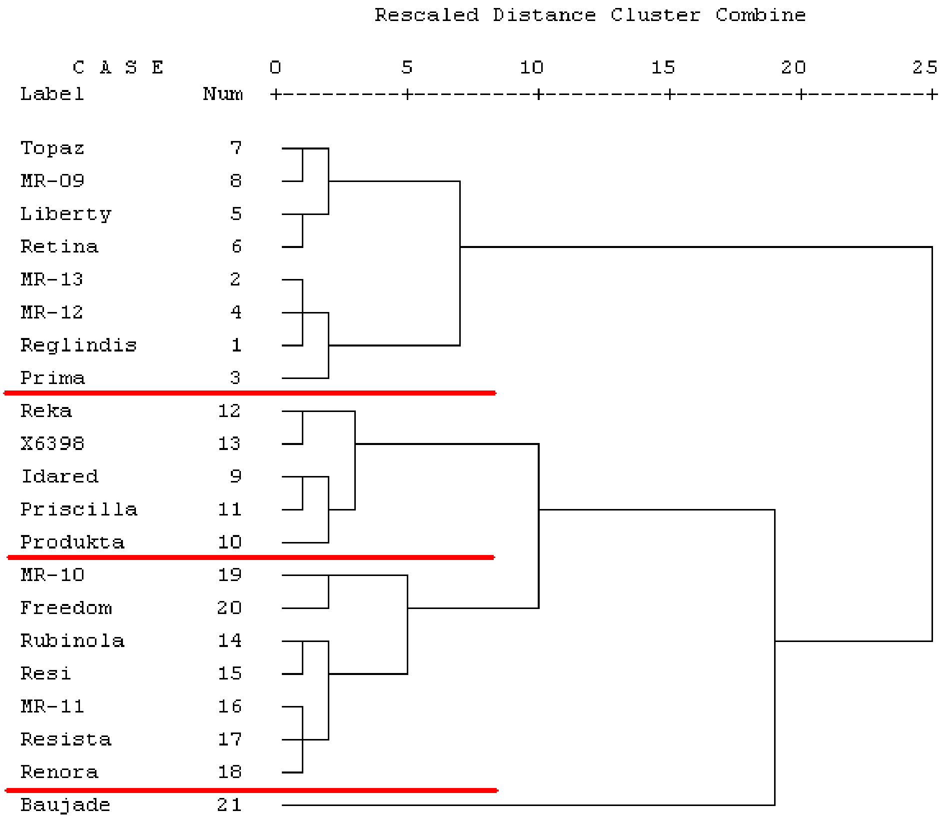

Figure 1. Blooming time groups according to the cluster analysis (Szigetcsép 2006)

We did not observed totally identical blooming order in the four investigation years in Szigetcsép and three years in Soroksár. However most of the varieties took relatively stabile place in the blooming order and in the blooming time group.

Varieties with stabile blooming time are suitable as reference varieties in further floral phenology investigations (table 3.). As there is a lack of information in blooming time of disease resistant apple varieties, determination of reference varieties is a very important gap filling research.

According to our results we would like to enhance varieties ‘Reglindis’ and ‘Baujade’. ‘Reglindis’ proved to be the earliest blooming variety in each observation years in Szigetcsép, and was overtaken only by ‘Prima’ MR-12 and MR-13 in Soroksár in one year.

‘Baujede’ proved to be the latest blooming variety in all observation years. In one year it bloomed so late that it was classified in a separate group by the cluster analysis. On one hand it makes the variety suitable as reference variety on the other hand it forecasts the occasional fructification problems of the variety.

Table 3. Blooming time group and blooming stability of the investigated varieties and candidates.

variety	blooming time group	number of observations	stability
Reglindis	**Early**-midearly	7	6-1
Prima	**Early**-midearly	7	6-1
MR-12	**early-midearly**	6	3-3
Retina	**early**-midearly-midlate	4	2-1-1
Topaz	**early**-midearly-midlate	4	2-1-1
MR-13	**early**-midearly-midlate	6	4-1-1
Liberty	**early**-midlate	4	3-1
MR-09	early-**midearly**-midlate-late	7	1-3-1-1
Produkta	**midearly**-midlate	4	3-1
Reka	**midearly**-midlate-late	7	5-1-1
Rubinola	**midearly-midlate**	4	2-2
X6398	**midearly-midlate**	4	2-2
Idared	early-midearly-**midlate**	6	1-2-3
MR-03	midearly-**midlate**	5	2-3
Freedom	midearly-**midlate**-late	7	1-4-2
Renora	**midlate**-late	4	3-1
MR-11	**midlate**-late	5	3-2
MR-10	midearly-**midlate-late**	7	1-3-3
Resi	**midlate-late**	4	2-2
Resista	**midlate-late**	4	2-2
Priscilla	**midearly-late**	4	2-2
Baujade	**late**	4	4

We recommend these above mentioned two varieties as reference varieties in further floral phenology investigations especially in the case of disease resistant ones. These two varieties represent a 'frame' for blooming period; this is how inaccurate grouping of other varieties can be avoided.

More difficult task is to find proper reference variety from the mid-blooming period. 'Produkta' might be on with its relatively stabile blooming time.

Varieties with relatively stable booming time like 'Freedom' or 'Prima' can be classified correctly with the help of proper reference varieties; other varieties with unstable blooming time like 'Priscilla' need more years of observation.

4.2. Booming overlap

In order to determine blooming overlap of the varieties we have upgraded the so called EV-1 method of Soltész (1992). We also fit the opened flowers phenograms of two varieties, and calculated the common area of the two curves. Blooming overlap was determined by the ratio of the common area and the total area of both varieties. We developed function system in order to simplify and accelerate the calculation of the phenogram areas. The ratio of opened flowers in the case of two following days determines trapezoids. The sum of the area of these trapezoids equals the total area of the phenogram (figure 2.).

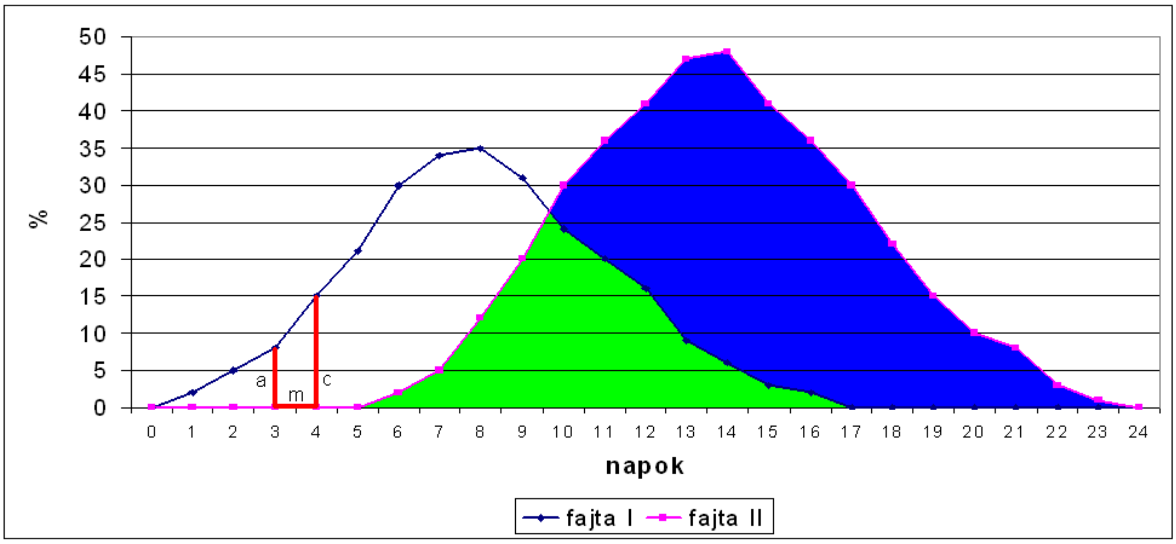

Figure 2. Determination of the common area of blooming phenograms

Average blooming overlap of the investigated varieties is shown in table 4. columns contain varieties as pollen providers while rows contain pollen receivers. The values of the table show the average overlap of the pollen provider varieties with the pollen receiver ones. For example MR-13 provides 83% overlap for 'Reglindis' while this value is 96% contrary. According to the blooming overlap four categories were created:

- Blooming overlap under 25% - variety is not suitable as pollinator for the other variety (red marking)
- Blooming overlap between 26-50% - suitability as pollen provider is risky; only in favourable years (yellow marking)
- Blooming overlap between 51-75% - the variety is suitable as pollen provider (blue marking)
- Blooming overlap 76% - the variety is excellent pollen provider (green marking)

We pointed out that correct blooming order helps grouping varieties, and four blooming overlap category can be separated easily from each other. Live up to the expectations the greater the distance of two varieties in the blooming order, the lower the blooming overlap is.

On the other hand blooming time groups create sharp dividing lines in the blooming order. In many cases it occurs that two following varieties in the blooming order are categorized in different blooming time groups, however they provide higher overlap than the two edge varieties of the same blooming time group.

Despite this blooming time groups have great significance as they represent frequency, namely it shows the group of a variety in the case of the majority of years. It is essential for the correct grouping. Furthermore blooming time groups are easier and more practical for growers.

Table 4. Average bloming overlap of the investigated varieties (Szigetcsép 2006-2009)

		Pollen provider																							
		Reglindis	Prima	Liberty	Retina	Topaz	MR-09	Idared	Produkta	Priscilla	Reka	X-63-97	Rubinola	Resi	Resista	Renora	MR-10	Freedom	Baujade	MR-13	MR-12	MR-11	Rajka	MR-03	Golden R.
Pollen receiver	Reglindis	A	79	66	76	67	57	58	49	32	55	52	54	37	31	38	34	32	13	83	77	43	83	72	23
	Prima	90	A	69	79	69	66	69	52	36	65	58	60	42	37	43	40	38	13	86	88	57	89	79	32
	Liberty	79	73	A	83	81	82	77	69	49	68	67	61	54	46	57	51	47	23	72	82	54	81	84	41
	Retina	77	70	74	A	83	66	64	59	47	65	61	70	44	38	52	37	41	19	68	76	56	97	80	28
	Topaz	67	59	69	81	A	64	64	62	51	70	66	79	57	48	63	49	50	22	58	64	58	85	80	30
	MR-09	71	72	85	82	80	A	82	78	53	76	77	66	64	56	69	62	58	27	68	79	77	81	94	57
	Idared	72	73	82	81	83	82	A	76	52	81	79	75	61	54	67	57	56	23	69	79	76	91	97	43
	Produkta	74	68	86	87	90	93	89	A	68	89	89	78	73	64	79	68	66	33	68	71	82	88	98	60
	Priscilla	51	50	64	66	70	65	61	65	A	66	68	67	76	74	72	63	81	52	33	37	86	64	70	64
	Reka	62	64	66	77	81	69	74	69	55	A	87	86	70	63	75	63	64	32	60	69	87	83	91	50
	X-63-97	63	62	69	75	80	74	77	73	58	91	A	85	77	69	81	66	70	37	57	63	91	87	100	56
	Rubinola	55	53	54	73	82	53	60	54	51	78	73	A	68	61	72	54	60	32	46	54	82	84	85	37
	Resi	41	42	51	49	61	55	52	52	65	66	70	70	A	89	88	74	87	50	33	35	86	57	77	77
	Resista	30	32	40	37	47	43	41	41	59	53	56	57	80	A	80	70	89	58	24	28	80	44	74	78
	Renora	40	39	50	57	63	55	53	54	57	67	70	70	83	82	A	79	80	47	36	39	83	53	80	78
	MR-10	40	37	45	47	54	52	51	52	56	61	62	58	72	72	83	A	76	55	41	40	71	32	57	71
	Freedom	31	33	40	39	48	43	42	41	63	55	57	56	77	87	76	71	A	64	23	25	78	47	72	74
	Baujade	14	12	22	21	23	23	19	24	51	27	32	30	50	62	49	59	70	A	12	14	42	16	35	57
	MR-13	96	88	70	91	69	72	68	64	36	64	58	52	39	29	42	39	29	14	A	88	36	88	64	12
	MR-12	85	86	77	94	71	78	72	62	38	66	57	54	38	31	43	38	28	14	84	A	40	84	67	20
	MR-11	51	62	58	50	68	68	69	55	51	82	85	89	90	90	91	76	87	45	31	34	A	81	100	73
	Rajka	81	76	72	82	89	55	64	45	30	71	61	84	45	43	50	37	47	14	73	92	63	A	80	32
	MR-03	61	58	68	62	76	59	62	51	34	70	64	74	53	59	64	54	59	27	65	91	48	72	A	44
	Golden R.	26	31	44	28	37	45	35	36	58	48	46	42	74	90	87	96	88	59	14	30	57	18	62	A

4.3. Self compatibility and parthenocarpy evaluation

Live up to the expectations very low fruit set was observed in our self compatibility experiments. In the case of self compatibility observations a later evaluation is more favourable as the high values immediately after blooming can be misleading. Apple is considered practically self incompatible, therefore economic production can be achieved only with proper cross pollination.

High values just after blooming can be resulted by the specific micro climate inside the isolator bags. Stress may cause high ratio of fruit development from self compatibility and parthenocarpy, however the majority of these fruits fall down during the first fruit drop period.

The second evaluation in June may be more informative, by that time the majority of insufficient fruits have fallen down.

According to our results ‘Resista’ can be emphasized with significant fruit set in all three observation years (2.5; 4.5 and 6.4%). Besides ‘Idared’ and ‘Resi’ provided at least 1% fruit set.

Some varieties like ‘Freedom’ and ‘Topaz’ produced outstanding values (5-8%) in one year, but very low fruit set in the other years. Fruit set values in the case of self compatibility and parthenocarpy proved to reasonably low, and not sufficient for economic growing. That is why we did not used statistical probes for this experiment.

The final fruit set was evaluated in August before ripening, however we had no opportunity to investigate the seed content of the fruits that is why we could not divide fruits from self compatibility and parthenocarpy from each other. As to us inclination to parthenocarpy should be clarified with further experiments.

4.4. Fruit set from crossings – cross compatibility of the investigated varieties

We prepared crossings among resistant varieties and candidates in three years (2005b in Szeigetcsép; 2008 and 2009 in Soroksár). On one hand crossing is very time and labour consuming task; on the other hand the plant material was limited and the proper time for do the crossings are only a few days. This is why we have chosen only some scientifically important combinations, and made reciprocal crossings only in 2009.

According to Fischer (2002) a combination with fruit set under 8% was considered incompatible. Although some author recon that 0% fruit set means cross incompatibility (Nyéki 1996). No combination resulted fruit set under 8% in 2005. Six combinations (MR-03

× MR-10, MR-03 × MR-13, MR-09 × MR-03, MR-10 × MR-13, MR-10 × ’Rubinola’ and MR-11 × MR-13) in 2008 and nine (MR-10 × MR-13, MR-10 × Florina, MR-10 × ’Prima’, MR-11 × MR-03, MR-11 × Liberty’, MR-12 × MR-03, MR-12 × MR-10, MR-13 × MR-09 and MR-13 × ’Prima’) in 2009 resulted fruit set under 8%.

Cross compatibility or incompatibility of apple varieties is determined by *S*-allele system; a multi allelic locus present in several plant families (De Nettancourt, 1997). According to Hegedűs (2008) and Halász (2009) MR-10 and MR-13 should be cross incompatible as they have identical *S*-alleles (S_7S_{10}). Our experiments support these findings only partially. Fruit set in combination MR-10 × MR-13 was only 5% in 2008 and was also low in 2009. However reciprocal crossing in 2009 resulted significantly higher fruit set. This result emphasizes the importance the further clarification of the relationship between compatibility and the *S*-genotype of the varieties.

Except the above mentioned combinations no incompatibility was found in our experiment.

4.5 Ploidy level

Ploidy level of the varieties was investigated by flow citometer. According to the literature ‘Idared’ was used as diploid control while ‘Jonagold’ as triploid. Live up to the expectations controls gave trace at zone 200-400 and 300-600 respectively.

Although ‘Liberty’ is mentioned as triploid in the literature and some of our candidates were thought to be triploid as well; all investigated variety proved to be diploid.

This is a favourable result considering the variety association; pollen sterility of triploids can be left out of consideration.

4.6. Pollen germination tests

We would like to answer two questions with the pollen germination tests. On one hand we wanted to prove that pollen germination does not decrease significantly during the one year storage in fridge. On the other hand we wanted to examine pollen germination of fresh pollen of the disease resistant apple varieties *in vitro*.

No significant pollen germination capacity loss was found after one year storage. Furthermore; such a huge amount of pollen is used for crossings that insures proper fructification even with 5% pollen germination.

In the case of fresh pollen only ‘Jonagored’ resulted in pollen germination under 1%. As it is triploid variety, it met our expectations. An interesting result is the 30% and 15% pollen germination of triploid presumed ‘Liberty’ in 2009 and 2007 respectively (figure 3.).

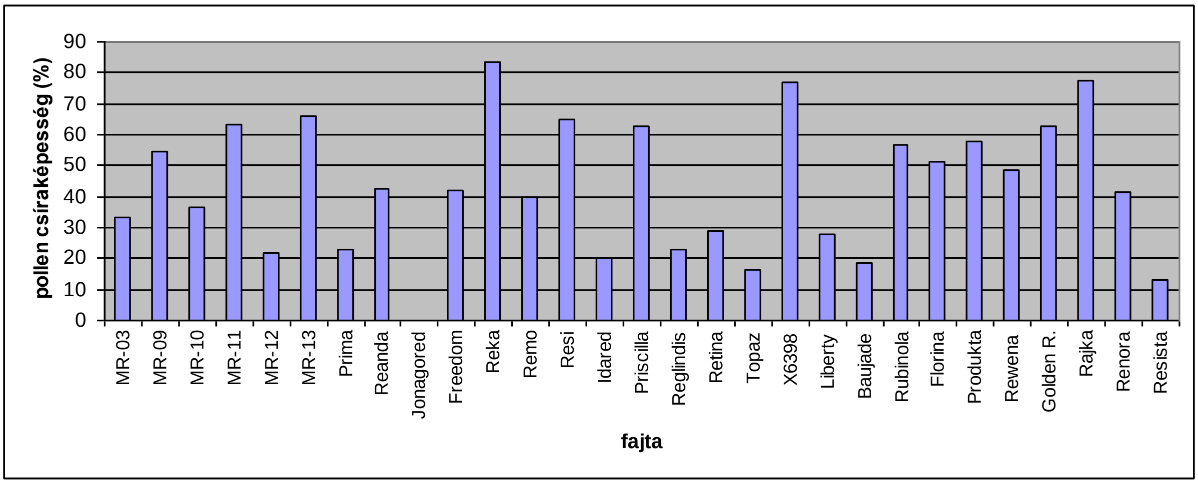

Figure 3. Pollen germination in 2009-ben

According to the literature a variety is suitable as pollen provider when pollen germination reaches at least 15-30%. Considering the 15% threshold all varieties proved to be good pollinizer in 2009 except the triploid ‘Jonagored’ and ‘Resista’. In 2007 some other varieties have fallen under the 15% threshold.

Further investigations are needed as significant differences were found between the two years.

4.7. Fruit quality / metaxenia evaluation

We have proved the existence of metaxenia - on physical and chemical parameters of the fruits - with our experiments. For example figure 4. shows the metaxenic effect on fruit size, colour and stem length.

On the other hand we pointed out that aspects of variety association are numerous; so the occasional metaxenic effects can be taken into consideration only as additive aspects. Some main principle can be authoritative in the case of different production aims:

- ❖ Production aim: fresh consumption → appearance e.g.
  - fruit colour: blush coloured varieties and varieties with only ground colour should be planted in separate plots
  - fruit shape: association of a flattened and an elongated fruited variety may result globular fruit size

- fruit size: less sized main variety can be affected by greater sized pollinizer variety

- ❖ Production aim: Fresh consumption after cold storage → appearance + consistency
- ❖ Production aim: industrial process → inner content values e.g.
  - Sugar and acidic content, juiciness and absorbance: a pollinizer variety with lower productivity but with outstanding inner content values may affect the fruit quality of the main variety favourable.

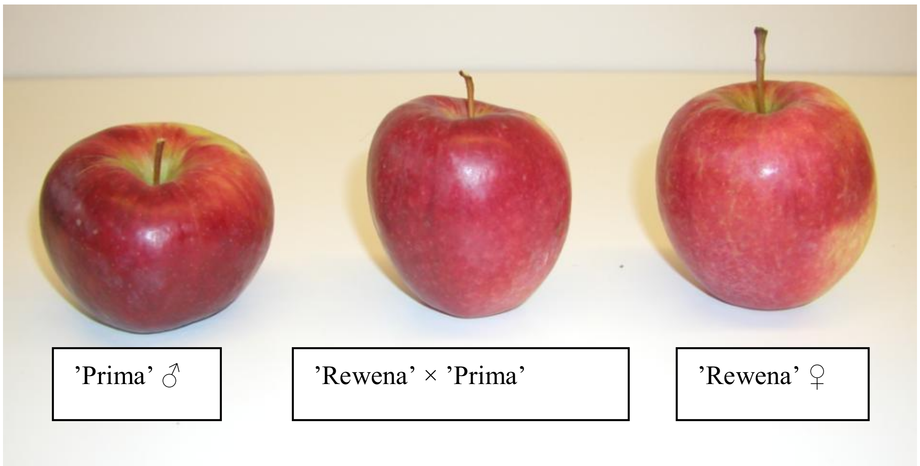

Figure 4. ’Prima’ as pollinizer resulted deeper fruit colour and shorter stem on crossed fruits.

4.8. recommendations for variety association

We elaborated variety association plans as a summary of our experiments. The core of our research program was the floral phenology observation and the evaluation of the blooming overlap. We recommend to associate varieties in an orchard or in a plot from the same blooming time group. Involving more varieties is highly recommended in the case of instable varieties.

We agree with the classical variety association principles, namely the three variety association version according to the ploidy level of the varieties (Tóth, 2001). As all the investigated varieties are diploid, any two varieties can be associated according to the first version; theoretically. This is why we do not indicate ploidy level in table 5. in the case of involving further occasionally triploid disease resistant varieties we recommend to indicate ploidy level in the variety descriptions, and variety tables.

Varieties with identical *S*-genotypes are cross incompatible in principle; however this was not unambiguously proved in our experiments. We recommend further evaluation of the relation of *S*-genotype and cross compatibility.

Next aspect is the pollen germination capacity. Although we have only partial results, very low pollen germination capacity was not found in the case of the investigated varieties except the triploid ‘Jonagored’. As to us involving further varieties is not needed considering this aspect.

The most important technological aspect is the fit of plant protection and the harvest. We recommend to associate varieties from the same blooming time group which results a maturity order. For example in the case of four varieties; planting of two-two varieties in separate plots are recommended. The variety pair in one plot should have approximately similar ripening time.

We stated that metaxenia is an existing phenomenon and may affect the fruit (crop) quality; however the aspects of variety association are numerous; so the occasional metaxenic effects can be taken into consideration only as additive aspects. In table 5. blush colour of the fruits is indicated. In the case of apple for fresh consumption colour is a very important quality feature. Green apple should be 100 green with no metaxenic blush. For example ‘Baujade’ as a green apple is a disease resistant alternative for ‘Granny Smith’. As ‘Baujade’ is a late blooming variety; ‘Golden Delicious’ from the mid-late blooming time group might be a proper pollinizer for it. On the other hand disease resistant varieties require also disease resistant pollinizers to maintain the real benefit of the resistance. ‘Produkta’ and ‘Resista’ meet these above mentioned requirements, although ‘Produkta’ belongs to the midearly–midlate group. So thus ‘Produkta’ is not the best pollinizer for ‘Baujade’.

To summarize our results further disease resistant varieties should be involved in complex evaluations; in order to elaborate further variety association plans. Our previously evaluated variety group should be investigated in further years in order to classify instable varieties into blooming time groups as well. Table 5. introduces floral phenlogy and fructification features of several disease resistant varieties. We recommend publishing this table in new variety descriptive books in order to ease variety selection for growers. The table can be completed with additional columns considering the production aim.

Table 5. Complex table for floral phenology and fructification features of the investigated disease resistant varieties

Variety	Blooming timegroup	Blooming stability	*S*-genotype	Pollen germination capacity*	Ripening time	Blush colour
Reglindis	**early**-midearly	6-1	?	o	end VIII.	+
Prima	**early**-midearly	6-1	S_2S_{10}	o	end VIII.	+
MR-12	**early**-**midearly**	3-3	S_2S_{23}	o	beg. X.	+
Rajka	**early**-midearly	1-1	?	++	mid-end IX.	+
Retina	**early**-midearly-midlate	2-1-1	?	o	mid. VIII.	+
Topaz	**early**-midearly-midlate	2-1-1	S_3S_5	o	mid-end IX.	+
MR-13	**early**-midearly-midlate	4-1-1	S_7S_{10}	++	end. VIII.	+
Liberty	**early**-midlate	3-1	$S_3S_5S_{10}$	o	beg. X.	+
MR-09	early-**midearly**-midlate-late	1-3-1-1	S_9S_{10}	+	mid. IX.	+
Produkta	**midearly**-midlate	3-1	?	+	end. IX. - beg. X.	-
Reka	**midearly**-midlate-late	5-1-1	?	+	end. IX.	+
Rubinola	**midearly**-**midlate**	2-2	S_2S_3	+	mid-end IX.	+
MR-03	midearly-**midlate**	2-3	S_2S_7	o	beg-mid. IX.	+
Freedom	midearly-**midlate**-late	1-4-2	$S_5S_?$	+	mid. IX.	+
Renora	**midlate**-late	3-1	?	+	beg. X.	+
MR-11	**midlate**-late	3-2	$S_{10}S_?$	++	mid-end IX.	+
MR-10	midearly-**midlate**-**late**	1-3-3	S_7S_{10}	o	end VIII. - beg IX.	+
Resi	**midlate**-**late**	2-2	?	++	end IX.	+
Resista	**midlate**-**late**	2-2	?	o	mid. IX.	slight blush
Priscilla	**midearly**-**late**	2-2	S_3S_9	++	mid. IX.	+
Baujade	**late**	4	?	o	mid. X.	-

*over 60% (very good): ++; between 40-60% (good): +; between 10-40% (moderate): o; below10% (poor): -

5. NEW SCIENTIFIC RESULTS

1. Floral phenology investigations; software elaboration for blooming overlap calculation; which is faster and more accurate than the previous methods.
2. Determination of blooming time, blooming overlap and blooming time group of 21 scab resistant apple varieties and candidates.
3. Determination of pollen germination capacity of 21 scab resistant apple varieties and candidates. We found no variety with low pollen germination, that makes all variety suitable as pollinizer; theoretically.
4. Determination of ploidy level of 21 scab resistant apple varieties and candidates. All variety proved to be diploid that makes all variety suitable for the classical 'two diploid variety association' model; theoretically.
5. Determination of ploidy level of 'Liberty' apple. The variety proved to be diploid according to our ploidy and pollen germination tests.
6. Elaboration of variety association plans. Compilation of a complex table for growers in order to ease variety selection.
7. Recommendation of reference varieties for further floral phenology observations: 'Reglindis' as stabile early blooming variety; 'Baujade as stabile late blooming variety.

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