APPLIED BREEDING OF APPLE ROOTSTOCKS CONDUCTED AT THE NATIONAL INSTITUTE OF HORTICULTURAL RESEARCH, SKIERNIEWICE, POLAND

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The National Institute of Horticultural Research
Skierniewice, Poland

20 YEARS OF POLAND IN THE EUROPEAN UNION

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Baltic Sea
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Olsztyn
Kaunas
Vilnius
Berlin
Poznan
Warszawa
SKIERNIEWICE
Lodz
Leipzig
Dresden
Wroclaw
Lublin
Praha
Krakow
Brno
Regensburg
Ivano Frankovsk
Kosice
200 km
Linz
YEARS
OF POLAND
IN THE EUROPEAN UNION

SHORT INFORMATION ABOUT POLAND

- **occupies an area of 313 000 km^2**
- **38 million inhabitants**
- **mean yearly temperature ranges from 6,0^{o}C to 8,5^{o}C**
- **warmest month is July, the temp. exceeds + 30^{o}C**
- **coldest month is January, the temp. drops below - 30^{o}C**
- **late spring frosts in May are quite frequent**
- **average yearly precipitation is 500 - 600 mm**
- **about 60 % of the agric. land are rather poor podzolic soils consisting of clayey sand and boulder clay**

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FRUIT CROPS BRED AT THE NATIONAL INSTITUTE OF HORTICULTURAL RESEARCH

TREE FRUITS

- Apple
- Plum
- Sour cherry
- Sweet cherry
- Peach
- Apricot
- Rootstocks (for apple)

SMALL FRUITS

- Strawberry
- Raspberry
- Blackcurrant
- Gooseberry
- Blueberry
- Blackberry
- Saskatoon (*Amelanchier alnifolia*)

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MAIN BREEDING GOALS

BREEDING DIRECTIONS

- ADAPTABILITY TO THE LOCAL ENVIRONMENT AND MODERN CULTIVATION METHODS
- HIGH PRODUCTIVITY AND FRUIT QUALITY
- BREEDING FOR RESISTANCE

APPLE ROOTSTOCKS BREEDING PROGRAMM - HISTORY

- The breeding of vegetative apple rootstocks has a long tradition in Poland.
- The first breeding programm began already in 1954 at the former Institute of Pomolgy and, apart from a few short breaks, and they/it have/has been continued up to the present time at the Department of Horticultural Crop Breeding of the National Institute of Horticultural Research (InHort) in Skierniewice, central Poland.
- The aim of the apple rootstocks breeding programs is to obtain genotypes with the properties comparable to the well-known M.9 and M.26, better adapted to soil and climatic conditions changes in Poland, as well as less susceptible to dangerous pests and diseases.
- Since, the annual production of apples in Poland is already more than 4 million tons it is assumed that development of such rootstocks enables its intensification.
- The introduction of better rootstock genotypes may also stimulate the overall apple production, decrease production costs and increase competitiveness of Polish apples on world markets.

APPLE ROOTSTOCKS BREEDING PROGRAMM – CURRENT STATE

- The current stage of the applied breeding of vegetative apple rootstocks (started in 2000) aimed obtaining more divers genotypes, semi-dwarf and dwarf, which are frost tolerant and have better resistance to diseases, including fire blight.
- New parental forms have been introduced in the crossing programms, such as Bemali, B9, PB-4, Jork 9, Mark, P 14, P 22, P 66, P 67, Pajam 1, Pajam 2, Supporter 1vf, Supporter 2vf, Supporter 3vf, G.11, G.13, G.16, G.41, as well as wild species such as *Malus pumila*, *Malus robusta* and *Malus prunifolia*, being the donors of favorable apple agronomical traits.
- In the frame of conducted programms over 1500 hybrids are still under successive stages of assessment.

APPLE ROOTSTOCKS BREEDING PROGRAMM – CURRENT STATE

- Under final assessment are also the newly developed Polish genotypes: P 68 (A2 x B9), PJ-173/2012 (BW x Pajam 1) and PJ-191/2016 (Jork 9 x Bemali).
- These genotypes will be submitted as the new apple rootstocks to the Polish National List of Fruit Plant Varieties.

Szampion/PJ-191/2016

Szampion/PJ-173/2012

Szampion/M.9

VEGETATIVE APPLE ROOTSTOCKS USED IN POLISH NURSERY INDUSTRY (certified plants – about 28 million pieces)

Name of rootstock	%
M.9 and subclones (M.9 EMLA, M.9 T337, M.9 RN29)	50.0
M.26	22.0
Polish rootstocks (P series)	17.5
M.7	7.5
A 2	1.7
MM.106	0.6
Other	0.7
Total	100.0

- **In presented studies Polish apple rootstocks were evaluated in regard to fire blight resistance (experiment 1)and plant tolerance to cold hardiness (experiment 2).**

EXPERIMENT 1

- **The aim of this study was to assess the degree of susceptibility to fire blight of nine Polish-bred vegetative rootstocks for apple, four rootstocks originating from the UK and one from the USA, after artificial inoculation of the shoot tips of these rootstocks growing in containers in a tightly closed plastic tunnel.**

MATERIALS AND METHODS

➢POLAND – 9 rootstocks

ROOTSTOCK	PARENTAGE	GROWTH VIGOUR
P 2	M.9 x Antonovka	dwarf
P 14	M.9 x OP	semidwarf
P 16	Glagierowka x M.11	dwarf
P 22	M.9 x Antonovka	very dwarf
P 59	A 2 x B 9	very dwarf
P 60	A 2 x B 9	dwarf to semidwarf
P 66	P 22 x M.26	dwarf
P 67	A 2 x P 2	dwarf to semidwarf
P 68	A 2 x B 9	dwarf

MATERIALS AND METHODS

- England, UK – 4 rootstocks

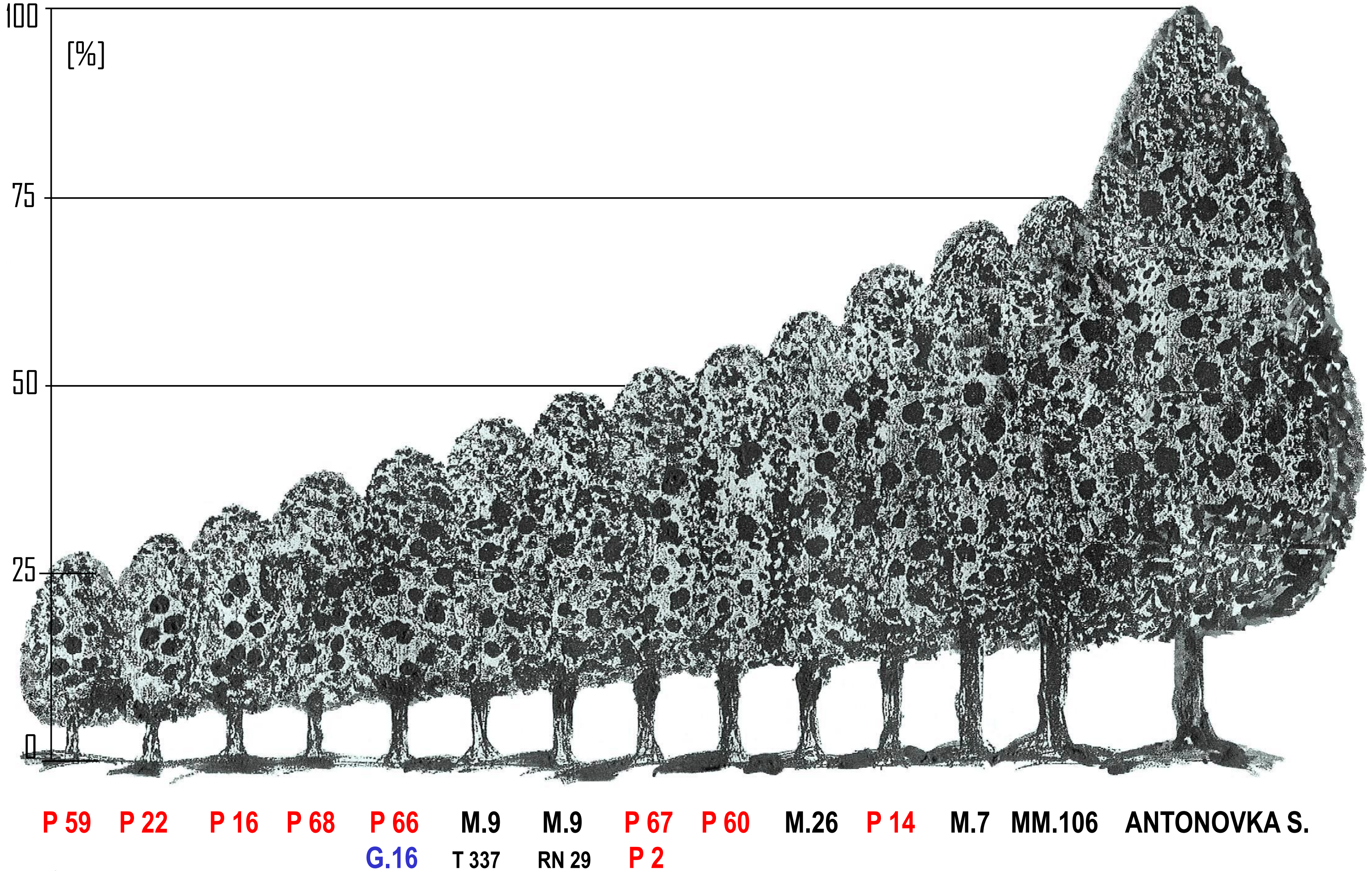
ROOTSTOCK	PARENTAGE	GROWTH VIGOUR
M.9	from a wild apple *Malus paradisica*	dwarf
M.26	M.16 x M.9	semidwarf
M.7	unknown	semidwarf
MM.106	Northern Spy x M.1	vigorous

MATERIALS AND METHODS

➢USA – 1 rootstock

ROOTSTOCK	PARENTAGE	GROWTH VIGOUR
G.16	Ottawa x *Malus floribunda*	dwarf

CLASSIFICATION OF APPLE ROOTSTOCKS ACCORDING TO GROWTH VIGOR



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MATERIALS AND METHODS

- **The rootstocks for the study came from stool-beds of the National Institute of Horticultural Research in Skierniewice (Central Poland), all with a virus-free health status.**

- **They were taken from the nursery in late autumn and placed in a cold store at 0°C.**

PRODUCTION OF PLANT MATERIALS

- ✓ **The rootstocks were planted into 5 litre plastic containers (in winter time of 2009 and 2010), filled with a mixture of peat substrate and compost soil at a volume ratio of 1:1, and the shoots of the rootstocks were cut back at a height of 5 cm above the soil level in the container. The containers with the pruned rootstocks were placed in a sealed high plastic tunnel.**

- ✓ **During the vegetation period, the plants were fertilized by applications to the soil (2 weeks after planting, there was a one-off application of slow acting fertilizer Osmocote at 20 g per container).**

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Growing rootstocks in high plastic tunnel

each genotype was represented by 20 rootstocks

Growing rootstocks in plastic tunnel – June 2009 and 2010

Inoculation of plants

Inoculation was done on 16th of June, 2009 and 14th of June, 2010. The terminal plant shoot tips were inoculated with a highly virulent strain (Ea 659) of *Erwinia amylovora* (10^7 cfu/ml) using infected scissors.

After inoculation, the tips of the inoculated shoots were covered with plastic bags for 24 hours to prevent the inoculum from drying up.

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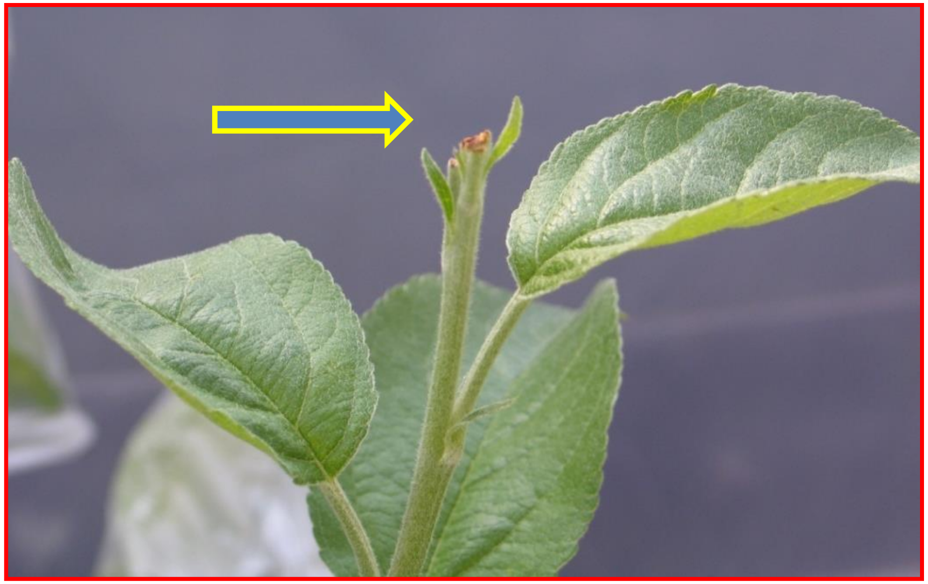
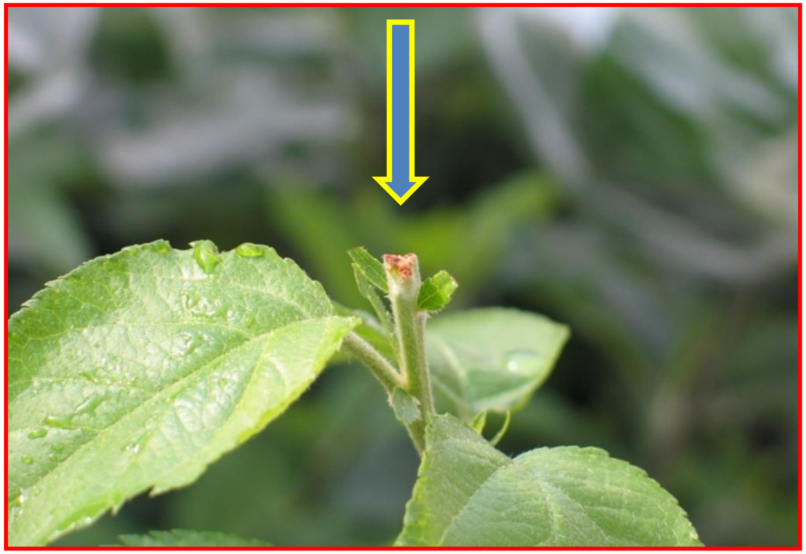
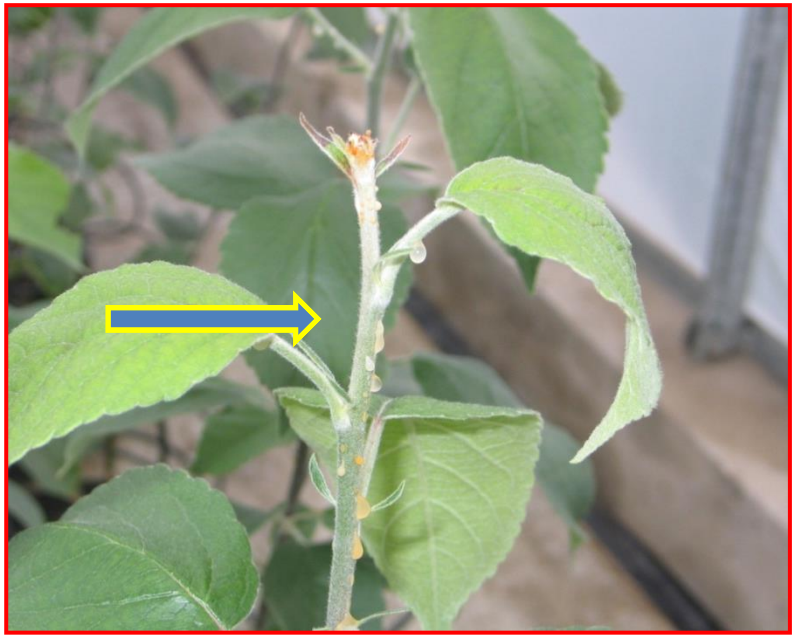
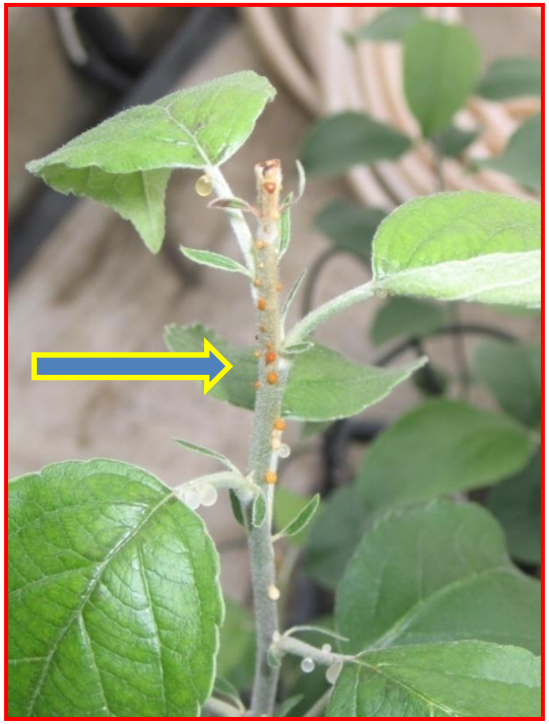
Fire blight symptoms









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Observations of the development of fire blight on the shoots and measurements of the length of the infected part of the shoots were performed 2, 4 and 6 weeks after the inoculation.

MATERIALS AND METHODS

The susceptibility of the rootstocks to fire blight was determined by comparing the length of the diseased part of the shoot with the total length of the shoot, which was expressed as a percentage. The calculated disease severity values (percentages of shoot necrosis) were presented as disease susceptibility scores according to the scale proposed by Le Lezec et al. (1997), where:

- ✓ 1 – very low susceptible (0-20% of shoot length necrotized) – class 1
- ✓ 2 – low susceptible (>20-40%) – class 2
- ✓ 3 – moderately susceptible (>40-60%) – class 3
- ✓ 4 – susceptible (>60-80%) – class 4
- ✓ 5 – very susceptible (>80-100%) – class 5

RESULTS

Rootstock	2009-2010		
	Percentage of shoot necrosis after:		
	2 weeks	4 weeks	6 weeks
very low susceptible plants – class 1			
G.16	9.9 a	10.6 a	10.9 a

In both years, six weeks after inoculation, the rootstock G.16 showed the lowest susceptibility with the very small (about 11%) necrosis observed in the entire length of the shoot. The rootstock G.16, developed in Geneva was assigned in the class 1 and represent very low susceptibility to fire blight.

6 weeks after inoculation

RESULTS

Rootstock	2009-2010		
	Percentage of shoot necrosis after:		
	2 weeks	4 weeks	6 weeks
low and moderately susceptible plants – class 2 and 3			
M.7	13.3 a	25.3 b	28.1 b
P 59	37.9 b	45.3 c	52.2 c

In both years, six weeks after inoculation, the rootstocks M.7 (class 2) and P 59 (class 3) were moderately susceptible to fire blight.

6 weeks after inoculation

RESULTS

Rootstock	2009-2010		
	Percentage of shoot necrosis after:		
	2 weeks	4 weeks	6 weeks
susceptible and very susceptible plants – class 4 and 5			
P 14	56.4 c	69.9 d	80.6 d
P 16	59.0 c	72.2 d	81.6 d
P 67	40.0 b	79.6 d	91.7 de
M.26	55.4 c	75.0 d	91.9 de
P 66	61.6 cd	82.2 d	94.9 de
M.9	59.9 c	80.4 d	95.0 de
P 68	63.4 cd	87.5 d	97.8 e
MM.106	72.2 cd	84.2 d	99.7 e
P 60	65.0 cd	83.1 d	99.9 e
P 22	63.7 cd	85.1 d	100.00 e
P 2	77.8 d	89.7 d	100.00 e

All the rootstocks were classified as very susceptible to fire blight (class 5).

6 weeks after inoculation

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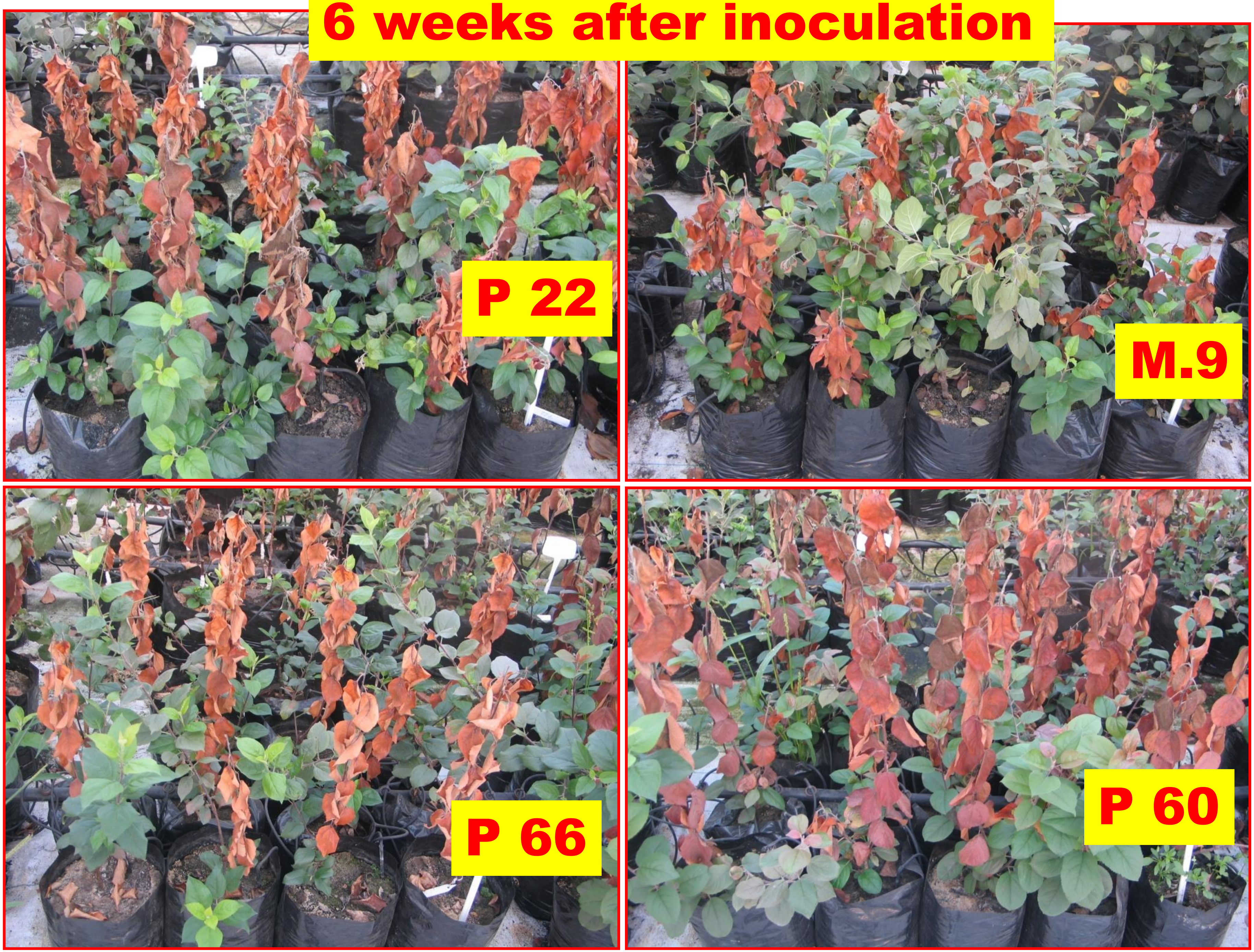
6 weeks after inoculation

6 weeks after inoculation



SUMMARY

Based on two years of evaluation the studied rootstocks can be classified as follows:

- **G.16 is very low susceptibility genotype.**
- **P 59 and M.7 are moderately susceptible genotypes.**
- **The rest of genotypes: P 2, P 14, P 16, P 22, P 60, P 66, P 67, P 68, M.9, M.26 and MM.106 are very susceptible to fire blight.**

EXPERIMENT 2

- In the second field experiment, nine Polish apple rootstocks (P 2, P 14, P 16, P 22, P 59, P 60, P 66, P 67 and P 68) as well as M.7, M.9, M.26, MM.106 (series from the UK) and Antonovka seedling (Russian origin) were tested in regard to cold hardiness.

MATERIALS AND METHODS

- **The rootstocks with a shoot diameter of 8-10 mm were packed in tightly closed plastic bags and placed in a cold store at 0°C for acclimatization.**
- **Before freezing, the rootstocks were sealed in plastic bags for protection against dehydration.**
- **Low temperature treatment of the pool of rootstocks (10 plants for each genotype and each temperature) was carried out at the temperatures of -8°C, -10°C and -12°C for 3 hours with the temperature decreasing 2°C/h, in the freezing chamber (BINDER GmbH, Germany) on 3rd - 5th March 2009 and 15th - 17th February 2010.**

MATERIALS AND METHODS

- Then again, the rootstocks were placed in the cold storage room at 0°C, before planting in the field, (mid-April of 2009 and 2010).
- Untreated rootstocks represented controls.
- Immediately after planting, all the rootstocks were pruned 5 cm above the soil surface.

-12ºC -10ºC -8ºC control

MATERIALS AND METHODS

In two consecutive seasons (2009-2010), the following field measurements and observations were conducted:

- Diameter (mm) of the rootstocks after planting and at the end of the growing season (late October) of the leading shoot of the rootstocks at a height of 5 cm from the ground, to calculate the increase in the diameter in the vegetation season,
- Growth vigour (cm) of the leading shoot of the rootstocks at the end of the growing season (late October) using a 1-5 ranking scale: 1 - no shoot (the plant has died), 2 - height of shoot up to 10 cm, 3 - height of shoot from 10.1 cm to 25 cm, 4 - height of shoot from 25.1 cm to 40 cm, 5 - height of shoot above 40.1 cm.

Increase in shoot diameter (mm) of rootstocks at a height of 5 cm above the ground after artificial freezing at different temperatures (Skierniewice, average for 2009-2010)

Rootstock	Control	Temperatures of freezing -8°C	-10°C	-12°C	Mean	% of the control
P 2	5.4 j[z]	3.1 h	1.9 i	0.7 c	1.9 a-c	35.2
P 14	3.8 f	1.9 b	1.1 d	0.8 d	1.3 ab	34.2
P 16	3.8 f	2.0 b	0.7 c	0.4 a	1.0 a	26.3
P 22	3.3 a	3.0 g	1.4 e	1.1g	1.8 a-c	54.6
P 59	3.8 f	3.3 i	1.6 g	1.5 h	2.1 cd	55.3
P 60	5.1 h	3.3 i	1.8 h	1.5 i	2.2 cd	43.1
P 66	4.1 g	2.6 f	2.1 k	2.1 k	2.4 cd	58.5
P 67	4.1 g	3.1 h	1.5 f	1.1 f	1.9 bc	46.3
P 68	5.9 k	5.1 k	2.9 m	3.1 l	3.7 e	62.7
M.7	3.5 e	1.5 a	1.9 j	1.0 e	1.5 a-c	42.9
M.9	2.7 c	2.1 c	0.3 a	1.1 fg	1.2 ab	44.4
M.26	2.5 b	2.4 e	0.8 c	0.6 b	1.2 ab	48.0
MM.106	3.4 d	2.3 d	0.6 b	0.6 c	1.3 ab	38.2
Antonovka seedling	5.3 i	4.5 j	2.4 l	1.6 j	2.8 d	52.8

[z] means marked with the same letter in columns are not significantly different at P = 0.05

Apple rootstocks treated with the temperature -8°C had the highest regeneration ability in comparison to the controls. Major differences in plant vigour, were found in pool of tested rootstocks frozen at -10°C and -12°C in relative to the control plants. The average for the two years of the study showed that among the tested rootstocks, after artificial freezing at -8°C, -10°C and -12°C, those that regenerated best the rootstocks P 59, P 60, P 66, P 67 and P 68. These rootstocks are thus relatively resistant to freezing in the applied temperatures, more resistant than the standard M.9 and M.26 rootstocks.

Growth vigour of the shoot of rootstocks after artificial freezing at different temperatures (Skierniewice, average for 2009-2010)

Rootstock	Control	Temperatures of freezing -8°C	-10°C	-12°C	Mean	% of the control
P 2	4.2[z] d-f[y]	2.6 ab	2.2 a	1.4 a	2.1 a	50.0
P 14	3.6 b	2.5 a	2.1 a	2.0 b	2.2 ab	61.1
P 16	3.6 b	3.1 c	2.8 b	2.4 b	2.8 c	77.8
P 22	3.7 bc	3.2 c	2.7 b	2.5 bc	2.8 c	75.7
P 59	4.2 d-f	3.5 d	3.2 c	3.1 de	3.3 d	78.6
P 60	4.6 f	4.5 g	4.4 e	4.3 g	4.4 f	95.7
P 66	4.1 c-e	4.0 e	3.3 c	3.0 d	3.4 de	82.9
P 67	4.5 ef	4.3 fg	4.2 e	3.9 fg	4.1 f	91.1
P 68	4.6 f	4.5 g	4.4 e	4.2 g	4.4 f	95.7
M.7	4.2 d-f	4.0 e	3.8 d	2.3 b	3.4 de	81.0
M.9	3.8 b-d	3.5 d	3.3 c	2.9 cd	3.2 d	84.2
M.26	3.1 a	2.8 b	2.7 b	2.2 b	2.6 bc	83.9
MM.106	4.2 d-f	4.1 ef	3.6 d	3.5 ef	3.7 e	88.1
Antonovka seedling	4.0 b-d	3.7 d	3.3 c	2.9 cd	3.3 d	82.5

[z] 1-5 ranking scale, where 1 - no stem (the plant has died), 2 - height of stem up to 10 cm, 3 – height of stem from 10 cm to 25 cm, 4 – height of stem from 25 cm to 40 cm, 5 - height of stem greater than 40 cm
[y] means marked with the same letter in columns are not significantly different at P = 0.05

Based on the average values of the test parameters (increase in shoot diameter during the growing season, growth vigour) for the three applied temperatures, the evaluated rootstocks were divided into two groups – frost tolerant and sensitive. The first group included rootstocks: P 59, P 60, P 66, P 67, P 68, M.7, MM.106 and the Antonovka seedlings, and the second - P 2, P 14, P 16, P 22, M.9 and M.26.

CONCLUSIONS

- The tested rootstocks were found to have different levels of resistance to low sub-zero temperatures.
- The applied temperatures at which the rootstocks were frozen, even the temperature of -12°C, did not kill them under the conditions of the experiment, but severely weakened the growth of the rootstocks, and thereby also the growth vigour and fruiting of the grafted cultivar.
- Both parameters used in the experiment to evaluate the sensitivity of rootstocks to low sub-zero temperatures, i.e. the increase in stem diameter during the growing season and growth vigour of the stem assessed at the end of the growing season, make it possible to assess reliably the tolerance of rootstocks to frost.

CHARACTERISTICS OF APPLE ROOTSTOCKS

Rootstock	Parentage	Relative Vigour [1]	Growth Vigour	Apple Scab	Powdery Mildew	Fire Blight	Winter Hardiness	Crown Rot	Soil Adaptability
P 14	M.9 x open pollination	50-55	semidwarf	low susceptible	moderately susceptible	very susceptible	medium to high	moderately susceptible	well drained soils
P 16	Glagierowka x M.11	25	dwarf	low susceptible	moderately susceptible	susceptible	medium to low	moderately susceptible	fertile soils
P 22	M.9 x Antonovka	23	dwarf	low susceptible	low susceptible	very susceptible	medium to high	resistant	well drained soils
P 59	A 2 x B 9	18-20	dwarf	low susceptible	low susceptible	moderately susceptible	medium to high	moderately susceptible	well drained soils
P 60	A 2 x B 9	35-40	dwarf to semidwarf	low susceptible	moderately susceptible	very susceptible	medium to high	moderately susceptible	well drained soils
P 66	P 22 x M.26	26	dwarf	low susceptible	low susceptible	susceptible	medium to high	moderately susceptible	fertile soils
P 67	A 2 x P 2	35	dwarf to semidwarf	low susceptible	moderately susceptible	susceptible	medium to high	moderately susceptible	fertile soils
P 68	A 2 x B 9	24	dwarf	low susceptible	low susceptible	susceptible	medium to high	moderately susceptible	fertile soils
M.7	unknown	66	dwarf to semidwarf	low susceptible	low susceptible	moderately susceptible	low	slightly susceptible	well adapted on most soils except heavy clay
M.9	from a wild apple *Malus paradisica*	29	dwarf	low susceptible	moderately susceptible	very susceptible	low	resistant	well drained soils
M.26	M.16 x M.9	50	semidwarf	low susceptible	low susceptible	very susceptible	medium to low	moderately susceptible	well drained soils
MM.106	Northern Spy x M.1	75	vigorous	low susceptible	moderately susceptible	very susceptible	low	very susceptible	best in loam and sandy loam soils, avoid poorly drained soils
G.11	M.26 x *Malus Robusta* 5	22	dwarf	low susceptible	low susceptible	resistant	medium to high	moderately resistant	well adapted on most soils
G.16	Ottawa 3 x *Malus floribunda*	26	dwarf	low susceptible	low susceptible	resistant	medium to high	tolerant	well adapted on most soils
Antonovka Seedlings	unknown	100	very vigorous	moderately susceptible	moderately susceptible	very susceptible	high	resistant	widely adapted

1–Classification of Polish P series rootstocks– Jakubowski T., Zagaja S.W., 2000. 45 years of apple rootstocks breeding in Poland. Acta Hort. 38, 723–727

P 67

- P 67 was selected in 1980 from a population of 144 seedlings obtained from a cross made in 1976 between A 2 x P 2.
- Since 2015 P 67 rootstock is protected in territory of the European Union (decision number 40350).
- P 67 is considered as a semi-dwarf rootstock, however trees on this rootstock are less vigorous than on M.26 and more vigorous than on M.9. P 67 has a good propagation ability in stoolbed (better than M.9 and M.26) and shows very good compatibility with the tested cultivars.
- The rootstock also shows high yield productivity (not lower than M.9 and M.26).
- Root system has a good anchorage ability, better than M9, however trees on P 67 in the orchard require additional support.
- P 67 has a good winter frost tolerance, better than M.9 and M.26. It is also low susceptible to apple scab, moderately susceptible to apple mildew and collar root of apple but is susceptible to fire blight.
- Our observations confirm that the P 67 rootstock stimulates more intense coloration of the skin of apple fruits.

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Thank you for your attention

The research conducted in the frame of subsidy of the Ministry of Agriculture and Rural Development special-purpose – Task 3.14: “*Production of breeding materials for apple rootstocks (Malus Mill.), resistant to ring rot of apple trunk, frost tolerant and thornless*”.

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