

## **Different fungicide combinations against apple scab helping to avoid fungus resistance**

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IOBC guidelines for integrated fruit production prescribe the use of forecasting systems in direct plant protection. In Latvia, LPPRC, model RIMpro for apple scab (*Venturia inaequalis*) control was tested from 2003. Following to FRAC guidelines, in order to reduce the risk of fungus resistance developing, from 2007 efficacy of fungicide mixtures (Chorus, a. i. cyprodinil + Dithane NT, a. i. mancozeb; Effector, a. i. dithianon + Candit, a. i. krezoxy-methyl) and alternately curative or strobilurine – protective fungicide use was tested. In all cases the first protective application before scab ascospores discharge was carried out with Cu product Champion 50. In case of emergency Effector was used during secondary scab infection period.

Fungicides registered in Latvia for apple scab control were effectively used as mixture of protective/curative or strobilurine products, not alternately, except strobilurine Candit (Qo inhibitor), which was applied separately, because fungus resistance appeared in the 3rd season of Candit use. Efficacy of Candit/Effector mixture corresponded other treatments. Curative product Chorus did not lose its efficacy after 6 seasons of use, applied no more than 3 times per season. Nevertheless, further strategy of resistance preclusion has to be considered, what request minimal use of risky products separately. In all cases fungicide applications, even of Chorus/Dithane mixture, were more effective before infection. Weather forecast not always was precise; in such cases the number of necessary applications increased. Most frequently under Latvia conditions there are three or four severe scab infection periods during the total primary infection period, subsequently 3 or 4 fungicide applications were necessary in addition to the first Champion treatment.

**Key words:** apple scab, fungicides, mixtures, resistance, risk groups, use in alternation.

**Introduction.** Development of pathogenic fungi resistance to several fungicides has become a considerable problem in plant protection, including apple scab (*Venturia inaequalis* (Cooke) Wint.) control in apple orchards. Fungicide Resistance Action Committee (FRAC) has developed fungicide resistance management guidelines to prolong the effectiveness of risky fungicides. The problem relates mainly to site-specific products disrupting single metabolic processes of definite fungus. In control of apple scab the most harmful products are demethylation inhibitors in sterol biosynt-

hesis (SBI, DMI) (Scheinpflug, 1988; Szkolnik, 1981), strobilurins – quinone outside inhibitors (QoI) (Broniarek-Niemec, Bielenin, 2008; Turechek, Köller, 2004), and anilinopyrimidines (Dux et al., 2004; Köller, Wilcox et al., 2005). According to anti-resistance management strategies, risky products are used in mixtures or in alternation with protective low risk fungicides, e. g. mancozeb, also they are used in sufficiently high dosages, only when justified and mainly protectively (Köller, Parker et al., 2005; Köller, Wilcox, 2001, Staub, 1991). FRAC is developed fungicide Code List according to resistance risk level (FRAC Code List, 2007).

In Latvia traditionally minimal amount of pesticides was used on horticultural crops. During the last decades, apple plantations quickly enlarged and growers intensified pesticide use to save yield quality. The choice of fungicides is poor. Therefore, recommendations of suitable anti-resistance plant protection strategy are important.

From 2003 apple scab warning model RIMpro was tested in Latvian Plant Protection Research Centre for adoption under conditions of Latvia. In the first years only curative fungicide Chorus 75 WG (a. i. cyprodinil, anilino pyrimidine group) was used shortly after RIMpro warning about risky infection. During the last years, FRAC (Fungicide Resistance Action Committee) baselines and recommendations about strategies to prevent fungus resistance were considered: use of fungicide mixtures with different modes of action or their use in alternation, as much as possible not several times in turn, use of protective products before forecasted infection as well as all applications before infection. There was little experience of fungicide mixtures efficacy in apple scab control in Latvia up till now. In 2007 and 2008 field trial was arranged to compare efficacy of different fungicide combinations in scab control taking into account RIMpro indices of risky infection.

**Object, methods and conditions.** Field trial was carried out in peasant farm “Kalnanoras”, Ikšķile, Ogre district in 2007–2008. There was investigated apple cultivar ‘Spartan’, on dwarf rootstock B 396, planted in 2000. Trial plot: 6 trees, 4 replicates in randomized blocks. Untreated control plots located out of blocks. In the trial the mostly used fungicides registered for apple orchards in Latvia and recommended mixtures were employed (Table 1).

**Table 1.** Fungicides used in trial  
**1 lentelē.** Bandyne panaudoti fungicidai

Trade name Prekinis pavadinimas	Active ingredient Aktyvi sudedamoji dalis	g kg <sup>-1</sup>	Chemical group Cheminė grupė	Mode of action Poveikio būdas	FRAC code FRAC kodas	Resistance risk Atsparumo pavojus
1	2	3	4	5	6	7
Champion 50 SP	Copper hydroxide Vario hidroksidas	770	Copper Varis	Protecting Apsauginis	M1	Low Mažas
Dithane NT WG	Mancozeb	750	Dithio- carba- mate	Protecting Apsauginis	M3	Low Mažas
Effector WG	Dithianone	700	Quinone	Protecting Apsauginis	M9	Low Mažas

**Table 1** continued  
**1 lentelės tęsinys**

1	2	3	4	5	6	7
Chorus 75 WG	Cyprodinil	750	Anilino- pirimi- dine	Systemic, post-in- fection Sisteminis, poinfek- cinis	9	Medium Vidutinis
Candit WG	Kresoxim-metyl	500	Strobilu- rin, QoI	Locally systemic, mostly protecting Vietiškai sisteminis, daugiausia apsauginis	11	High Didelis

These are mostly all fungicides registered in Latvia for apple scab control with exception of Score 250 (a. i. difenoconazole, SBI, DMI), against which considerable fungus resistance was observed in trial orchard after 5 years of use.

In anti-resistance management strategy the prophylaxis in advance of primary apple scab infection period at green tip stage of apple trees with protecting spray is recommended (Köller, Parker et al., 2005). In the trial this application was carried out with copper fungicide Champion 50 in all treatments.

All products were used in maximal rates registered in Latvia: Champion 4.0 (a. i. 3.08) kg ha<sup>-1</sup>, Dithane 3.0 (a. i. 2.25), Effector 0.6 (a. i. 0.42), Chorus 0.3 (a. i. 0.225), Candit 0.25 (a. i. 0.125) kg ha<sup>-1</sup>. In mixture 75 % of each product was used. Water volume was 600 l ha<sup>-1</sup>. The biggest attention was paid to primary scab infection control. If effectiveness of applications was insufficient, fungicide use was continued during secondary scab infection period.

Real beginning, intensity and the end of ascospores release was determined on spore traps – glass slides placed on old apple leaf layer in trial orchard. Slides were collected after every rain and spores counted under microscope. Discharge of scab ascospores commonly is ending in the middle of June.

In 2007 Champion was sprayed on 18.04. (GS 53, green tip) in all the treatments, except untreated control. The first scab ascospores were released on 20.04.

In 2007 the trial was carried out using systemic fungicide Chorus (alone, no more than 3 times per primary infection period), mixture of Chorus and protective Dithane 1–2 days after RIMpro showed risky infection level (above 70 RIM), as well as Dithane and Chorus in alternation accordingly shortly before and after infection. Before or shortly after infection (24 hours) protective Efector, mixture of Efector and strobilurine Candit, as well as Effector and Candit (2 times) in alternation were used (Table 2).

RIMpro shown the amount of efficient residues (more than 25 %) after each application was taken in account too. Chorus in trial territory up till now was used for 6 years 2–3 times per season without symptoms of efficacy decrease, Candit – for 3 years with efficacy decrease symptoms.

**Table 2.** Fungicide combinations for control of primary apple scab infection, 2007

**2 lentelė.** Fungicidų deriniai pirminės obelių rauplių infekcijos kontrolei, 2007 m.

Dates of critical infection periods Kritinių infekcijos laikotarpių datos	09–10.05	15.05	26–28.05	01.06		
RIM risk values per day in average Vidutinės RIM rizikos vertės per dieną	832	337	652	117		
Precipitation sum Kritulių suma (mm)	17.0	13.2	27.4	1.4		
Dates of fungicide applications Fungicido panaudojimo datos	27.04	10.05	16.05	24.05	28.05	30.05
Apple growth stages (GS), BBCH Obelių augimo tarpsniai (AT)	55 (mouse-ear) (žiediniai pumpurai dar uždari)	57 (red buds) (raudoni pumpurai)	59 (ballon stage) (dauguma žiedų su vainiklapiais)	65 (full flowering) (visiškas žydėjimas)	67 (end of flowering) (žiedai vysta)	69 (end of flowering) (žydėjimo pabaiga)
Treatments Variantai						
Control – untreated Kontrolė – neapdorota	-	-	-	-	-	-
Chorus (Ch) after infection Chorus (Ch) po infekcijos	-	Ch	Ch	-	Ch	-
Dithane (D) before, Chorus after infection Dithane (D) prieš, Chorus po infekcijos	D	Ch	Ch	-	Ch	-
Dithane / Chorus mixture after infection Dithane / Chorus mišinys po infekcijos	-	D/Ch	D/Ch	-	D/Ch	-
Effector (E) before forecasted or shortly after infection Effector (E) prieš numatytą infekciją arba tuoj po jos	E	E	-	E	-	E
Effector / Candit mixture for 3 times Effector / Candit mišinys 3 kartus	E/Ca	E/Ca	-	E/Ca	-	E
Effector in turn with Candit (Ca) Effector pakaitomis su Candit (Ca)	E	Ca	-	Ca	-	E

Weather conditions during May were rainy, especially in the 3rd 10-day period, when precipitation sum was 49.8 mm, 293 % from normal, and mean temperature

19.4° C, 6.7 °C above normal, being the warmest period in Latvia during 84 years. Such conditions promoted primary scab infection. Rainy period started again from the middle of June and July and additional fungicide application during secondary infection period was necessary. Additional applications with Effector were carried out on 19.06 and 12.07 in the 7<sup>th</sup> treatment where Candit alone 2 times was used, and on 27.07 in all treatments. It is permitted to use the low resistance risk product Effector for 6 times per season in Latvia.

In 2008 all fungicide combination schedules were repeated, except 7<sup>th</sup> treatment, where clearly appeared efficacy loss; strobilurine Candit was used alone for 2 times. Following to FRAC recommendations, all fungicide treatments were carried out before forecasted precipitation and scab infection (Table 3).

**Table 3.** Fungicide combinations for control of primary apple scab infection, 2008

**3 lentelė.** Fungicidų deriniai pirminės obelų rauplių infekcijos kontrolei, 2008 m.

Dates of precipitation periods Kritulių laikotarpių datos	02–05.05	08, 12.05	15–18.05	11–20.06
Precipitation sum Kritulių suma (mm)	2.2	0.4	14.2	26.8
Dates of fungicide applications Fungicido panaudojimo datos	29.04	09.05	13.05	10.06
Apple growth stages (GS), BBCH Obelų augimo tarpsniai (AT)	56 (green buds / žali pumpurai)	58 (pink buds / rausvi pumpurai)	61 (beginning of flowering / žydėjimo pradžia)	71 (fruits of 10 mm in diameter / 10 mm skersmens vaisiai)
Treatments Variantai				
Control – untreated Kontrolė – neapdorota	-	-	-	-
Chorus (Ch)	Ch	-	Ch	Ch
Dithane (D) in turn with Chorus Dithane (D) kartu su Chorus	D	Ch	-	D
Dithane / Chorus mixture Dithane / Chorus mišinys	D/Ch	D/Ch	-	D/Ch
Effector (E)	E	E	-	E
Effector/ Candit mixture Effector/ Candit mišinys	E/Ca	E/Ca	-	E/Ca

In 2008 Champion was sprayed on 16.04. (GS 53) in all treatments including untreated control for partly decrease of high scab infection level in control plots. The first scab ascospores were released on 17.04.

Weather conditions in May were comparably dry and cool; some days it rained, noteworthy that on 18.05 it rained 12.2 mm and the mean temperature in the 2<sup>nd</sup> 10-day period was 10 °C. The 3<sup>rd</sup> 10-day period of May and the 1<sup>st</sup> of June were completely without precipitation. Rainy period lasted from June 11<sup>th</sup> until the middle of July. Apple scab widely spread in unprotected areas. In treated plots no fungicide applications were carried out during secondary infection period. For weather forecast data of Latvian Environment, Geology & Meteorology Agency (LEGMA) and web page www.gismeteo.ru was employed.

Apple growth stages are characterized according to BBCH scale.

Scab incidence and extent level was assessed on 100 leaves and fruits per plot from the beginning of disease appearance in control treatment, before each fungicide application and further after rainy periods, on leaves – till the end of July, on fruits – until harvesting. Disease extent was evaluated according to % scale of damaged surface.

The data were subjected to analysis of variance. Significant differences at the probability of 95 % are shown in the tables by letters. Data of control treatment are not included in processing.

**Results.** In 2007 there were four noteworthy critical primary apple scab infection periods. At the end of April – beginning of May precipitation appeared later than forecasted, subsequently the first test treatment was carried out too early in schedules with planned fungicide application shortly before infection (see Table 2). As a result, according to the schedule of planned applications, before infection four treatments were carried out and after infection – three fungicide treatments during primary apple scab infection period.

Five tested schedules significantly decreased scab infection level on the leaves without significant difference among them, but in the 7<sup>th</sup> treatment, where 2 times strobilurine Candit was used, there was clearly visible the loss of effectiveness, what indicated fungus resistance development. After two additional Effector sprays on 19.06 and 12.07, scab infection leveled with other treatments until the end of July (Table 4).

**Table 4.** Apple scab incidence on apple leaves using different fungicide combinations, Latvia, 2007

**4 lentelē.** Obelų raupliu paplitimas ant obelų lapu naudojant skirtingus fungicidų derinius, Latvija, 2007 m.

Treatments Apdorojimai	Dates of assessments Apskaitu datas			
	24.05	19.06	06.07	27.07
	scab incidence (amount of infected leaves) raupliu paplitimas (pažeistu lapu kiekis) (%)			
<i>I</i>	2	3	4	5
Untreated control Neapdorota kontrolē	0.2	25.3	38.7	55.2
All schedules, except treatment with Candit Visos shemas, išskyrus apdorojimā su Candit	0	0.60 a	0.55 a	3.35 a

**Table 4** continued  
**4 lentelės** tęsinys

<i>1</i>	<i>2</i>	<i>3</i>	<i>4</i>	<i>5</i>
Candit for 2 times separately in alternation with Effector	0	2.75 b	5.00 b	5.00 a
Candit 2 kartus atskirai, kaitaliojant su Effector				
LSD <sub>95</sub> / R <sub>95</sub>	-	1.52	2.12	4.22

According to fruit infection extension, the same tendency appeared as on the leaves: after the schedule of two sprays with Candit alone there was significantly higher scab infection level in the beginning of July in comparison with other schedules. There was also tendency of higher fruit infection in the treatment with Dithane/Chorus mixture for 3 times after primary infection periods, though on leaves, as infection source, such tendency did not appear. Possibly mixture with Dithane could be used before primary infection period. After additional fungicide applications during secondary scab infection period until the harvest time fruit infection leveled without significant difference between treatments. There remained the tendency of higher infection in Dithane/Chorus mixture treatment. The decrease of infection in Candit treatment was due to dropping of more damaged fruits (Table 5).

**Table 5.** Apple scab incidence on apple fruits using different fungicide combinations, Latvia, 2007

**5 lentelė.** Obelių rauplių paplitimas ant obuolių naudojant skirtingus fungicidų derinius, Latvija, 2007 m.

Treatments Variantai	Dates of assessments Apskaitų datos			
	19.06	06.07	27.07	24.09
	scab incidence (amount of infected fruits) rauplių paplitimas (pažeistų vaisių kiekis) (%)			
Untreated control Neapdorota kontrolė	29.0	35.0	59.9	82.3
All schedules, except treatment with Candit use and Dithane/Chorus mixture Visos schemos, išskyrus apdorojimą su Candit ir Dithane/Chorus mišini	0	1.62 a	3.75 a	6.0 a
Dithane/Chorus mixture for 3 times Dithane/Chorus mišinys 3 kartus	0	2.5 ab	6.25 b	8.0 a
Candit for 2 times separately in alternation with Effector Candit 2 kartus atskirai, kaitaliojant su Effector	0	5.0 b	5.25 ab	3.75 a
LSD <sub>95</sub> / R <sub>95</sub>	-	3.09	2.24	4.63

In the tables there are shown only more characteristic results of 10 assessments. The extent of damaged leaf surface showed the same tendency as the amount of damaged fruits.

In 2008 there were 4 noteworthy precipitation periods during primary scab infection period, like in 2007. Applying all fungicides 1–4 days before forecasted rain period effectiveness of all treatments was similar to scab infection control on the leaves in total. There was some significant difference between treatments in separate assessments, but the tendency did not maintain during assessment period. At the end of July there was no significant difference of leaf infection level between treatments (Table 6). Decrease of leaf infection level on 11.07 in comparison with previous assessment was due to forming young uninfected leaves.

**Table 6.** Apple scab incidence on apple leaves using different fungicide combinations for 3 times before forecasted primary infection, 2008, Latvia

**6 lentelē.** Obelų raupliņu paplitimas ant obelų lapu naudojant skirtingus fungicidų derinius po 3 kartus prieš numatytą pirminę infekciją, Latvija, 2008 m.

Treatments Apdorojimai	Dates of assessmentsApskaitu datos			
	10.06	04.07	11.07	31.07
	Scab incidence (amount of infected leaves) raupliņu paplitimas (pažeistų lapu kiekis) (%)			
Untreated control Neapdorota kontrolē	11.8	27.5	40.75	35.0
Chorus 75 separately Chorus 75 atskirai	0	3.50 a	3.50 a	4.75 a
Dithane NT for 2 times, Chorus 1 time Dithane NT 2 kartus, Chorus 1 kartā	0	3.25 a	2.75 ab	5.25 a
Mixture of Dithane/ Chorus Dithane/Chorus mišinys	0	4.25 ab	1.75 b	5.75 a
Effector separately Effector atskirai	0	5.50 b	2.50 ab	5.75 a
Mixture of Effector/ Candit Effector/Candit mišinys	0	4.00 ab	1.75 b	3.50 a
LSD <sub>95</sub> / R <sub>95</sub>	-	1.65	1.44	2.81

For control of fruit infection the use of Chorus alone during primary scab infection period was significantly less effective. Such results did not indicate the appearance of fungus resistance because this schedule effectively controlled leaf infection, but post-activity of systemic product was too short to protect fruits until harvest (Table 7).

**Table 7.** Apple scab incidence on apple fruits using different fungicide combinations, 2008, Latvia  
**7 lentelė.** Obelių rauplių paplitimas ant obuolių naudojant skirtingus fungicidų derinius, Latvija, 2008 m.

Treatments Variantai	Dates of assessments Įvertinimo datos			
	11.07	31.07	05.09	30.09
	scab incidence (amount of infected fruits) rauplių paplitimas (pažeistų vaisių kiekis) (%)			
Untreated control Neapdorota kontrolė	12.5	21.0	63.5	76.8
Chorus 75 separately Chorus 75 atskirai	0	0.75 a	5.25 a	7.50 a
Dithane/Chorus in alternation and Mixture of Effector/Candit Dithane/Chorus pakaitomis ir Effector/Candit mišinys	0	0.25 b	4.50 a	3.88 b
Mixture of Dithane/Chorus and Effector separately Dithane/Chorus mišinys ir Effector pakaitomis	0	0.12 b	2.75 b	4.38 b
LSD <sub>95</sub> / R <sub>95</sub>	-	0.49	1.65	1.80

**Discussion.** The use of protecting and risky fungicide mixtures during the primary scab infection period, according to the results of several authors, is the most effective component in apple scab protection schedules. Mixture of anilinopyrimidine with mancozeb has been more effective in comparison with mancozeb alone and especially anilinopyrimidine alone that provided unacceptable level of scab control (Köller, Wilcox et al., 2005; Köller, Parker et al., 2005). Such resolution agreed with our research results, by comparison of effectiveness of Dithane/ Chorus mixture and Chorus alone. According to strobilurins, several trial results showed that before resistance emergence strobilurin products alone in higher registered dosage were more effective in comparison with strobilurins/ mancozeb mixture and mancozeb alone (Turechek, Köller, 2004). However, if the symptoms of resistance are observed, strobilurins will have to be used only in mixture with another scab fungicide (Köller, Parker et al., 2005). Protecting product dithianon (Effector WG registered in Latvia) is recommended for the mixture with krezoxym-metyl products by manufacturer company BASF. Therefore, this mixture was tested in our trial and although the decreased effect of separately used krezoxym-metyl (Candit WG) appeared already in the trial orchard the use of Effector/Candit in protection schedule had similar effectiveness in comparison with other schedules.

**Conclusions.** 1. The use of protecting, low resistance risk fungicide Dithane NT (a. i. mancozeb) 1–4 days before primary apple scab infection in alternation with systemic, medium risky product Chorus 75 (a. i. cyprodinil) shortly before or after infection, as well as the use of Dithane/ Chorus mixture at dosage of 75 % from full rate of each product before infection in resistance preventive protection schedule similarly effectively controlled apple scab infection on leaves and fruits.

2. High resistance risk strobilurin fungicide Candit (a. i. krezoxym-metyl) showed clearly visible loss of effectiveness used the 4th season for 2 times separately in alternation with Effector, while the use of Effector/Candit mixture for 3 times shortly before or after infection at dosage of 75 % from full rate of each product was similarly effective in comparison with other resistance preventive protection schedules.

3. Medium risky product Chorus 75, used the 7<sup>th</sup> and 8<sup>th</sup> season for 3 times by turns during primary scab infection period was similarly effective in resistance preventive schedules controlling apple leaf infection, but lost the effect of fruit infection control during secondary scab infection period. Such schedule is not recommended in integrated plant protection.

4. Low risk protecting fungicide Effector (a. i. dithianone) used for 3 times by turns shortly before or after primary scab infection was similarly effective with other resistance preventive schedules, but such schedule is not recommended in integrated plant protection.

5. Planning fungicide treatments before infection can increase the number of sprays if weather forecast is not precise.

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### **Įvairūs fungicidų deriniai nuo obelų rauplių, padedantys išvengti rauplėgrybio atsparumo**

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#### *Santrauka*

Auginant vaisius integruotai, jų produkcijai taikomos IOBC nuorodos rekomenduoja augalų apsaugai naudoti prognozavimo sistemas. Latvijoje, Augalų apsaugos tyrimų centre, nuo 2003 metų buvo tirtas RIMpro modelis obelų rauplių (*Venturia inaequalis*) kontrolei. Pagal FRAC nuorodas, siekiant sumažinti rauplėgrybio atsparumo išsivystymo pavojų, nuo 2007 metų buvo tirtas fungicidų mišinių (Chorus, a. i. cyprodinil + Dithane NT, a. i. mancozeb; Effector, a. i. dithianon + Candit, a. i. krezoxy-methyl) ir pakaitomis naudojamų gydomojo arba strobilurino – apsauginio fungicido veiksmingumas. Visais atvejais pirmasis apsauginis apdorojimas prieš rauplių askosporų pasirodymą buvo atliktas su Cu produktu Champion 50. Esant reikalui, antrinės rauplių infekcijos laikotarpiu naudotas Effector.

Latvijoje registruoti fungicidai nuo obelų rauplių buvo veiksmingi naudojami ne pakaitomis, o kaip apsauginių / gydomųjų arba strobilurino produktų mišinys, išskyrus strobilurinę Candit (Qo inhibitorių), kuris buvo naudojamas atskirai, nes trečiuoju Candit naudojimo sezonu atsidaro atsparumas rauplėgrybiams. Candit / Effector mišinio veiksmingumas buvo toks pat kaip ir kitų fungicidų. Gydomas produktas Chorus po šešių naudojimo sezonų neprarado savo veiksmingumo, jei buvo panaudotas ne daugiau kaip 3 kartus per sezoną. Vis dėlto reikia apsvarstyti tolimesnę strategiją, kaip užkirsti kelią atsparumo išsivystymui, t. y. kaip minimaliai naudoti pavojingus produktus atskirai. Visais atvejais fungicidai, net Chorus/Dithane mišinys, buvo veiksmingesni prieš infekciją. Orų prognozės ne visada pasitvirtindavo – tada fungicidus tekdavo naudoti dažniau. Latvijos sąlygomis bendru pirminės infekcijos laikotarpiu būna trys ar keturios stiprios infekcijos, taigi, be pirmojo apdorojimo su Champion, reikėjo naudoti dar 3–4 fungicidus.

**Reikšminiai žodžiai:** atsparumas, fungicidai, mišiniai, naudojimas pakaitomis, obelų rauplės, rizikos grupės, .

