Chapter 6. CURRENT SITUATION IN RESISTANCE STRATEGY

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RESISTANCE OF *VENTURIA INAEQUALIS* TO STROBILURIN AND DODINE FUNGICIDES IN POLISH APPLE ORCHARDS

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Abstract

The monitoring of *V. inaequalis* resistance to strobilurin and dodine fungicides was conducted during 2004–2007. The high level (above 50%) of forms resistant to strobilurin was found in 8 of 53 monitored orchards. In 7 orchards the percentage of these forms amounted from 26 to 42.6% and in the remaining it was low (0–21.8%). The high resistance to dodine was observed in 7 of 64 monitored orchards (above 50% of resistant forms) but in 9 it varied between 30.1 and 49.7%. The low level (below 18.9%) of dodine resistant forms was found in 48 orchards. The results showed that the problem of *V. inaequalis* resistance to strobilurin and dodine fungicides occurs in considerable number of apple orchards in Poland, therefore the application of these fungicides should be reduced according to FRAC recommendations.

Key words: *Venturia inaequalis,* resistance, strobilurin, dodine.

Introduction

Apple scab (*Venturia inaequalis* (Cooke) Wint.) is the most important disease of apples. Application of fungicides remains the primary tool for managing this disease. However, their frequent and improper use can result in the development of the *V. inaequalis* resistance. The problem related to anilinopirimidin, ergosterol biosynthesis inhibitor (EBI), dodine and strobilurin was revived by /Köller, Wilcox, 2001/.

The first resistant forms of *V. inaequalis* to dodine in Poland were detected in 1990 /Nowacka, 1991/, while to strobilurin fungicides (QoI) in 2002 /Broniarek-Niemiec, Bielenin, 2003; 2004/. Because of their common application and specific mode of action the development of *V. inaequalis* resistant forms to QoI was noted in Western Europe for the first time in 2000, after 4 years of commercial use /Bartlett et al., 2002/.

The aim of our study was to determine the occurence of *V. inaequalis* forms resistant to strobilurin and dodine fungicides in apple orchards located in different regions of Poland.

Materials and Methods

The monitoring of *V. inaequalis* resistance to strobilurin and dodine fungicides was conducted during 2004–2007 in the selected orchards, in which, in spite of chemical control, severe apple scab symptoms on leaves and fruits have been observed.

For determination of resistance to strobilurins the germination test on detached apple leaves was used /Broniarek-Niemiec, Bielenin, 2005/. The samples of 50 leaves with visible scab lesions were collected from 53 orchards. The conidia were washed off and their water suspensions were used for inoculation of air dried leaves, originating from apple trees growing in greenhouse, previously treated with kresoxim-methyl at 100 µg a.i/l or trifloxystrobin at 75 µg a.i/l at rates recommended for the commercial control of apple scab. The control constituted leaves treated with water only. Next, leaves were placed on wet filter paper in Petri dishes and incubated for 24 hours at 200 C and 95–100% relative humidity and afterwards the samples of spores were taken from leaves using a celluloid adhesive tape and the determination of spores germination was performed under optical microscope. Ten replications for each sample with 100 spores in each replication were evaluated. Index of resistance level (IRL%) was calculated according to the formula:

IRL% = (A / B) x 100%

A – percentage of germinated spores on leaves treated with fungicide (kresoxim-methyl or trifloxystrobin)

B – percentage of germinated spores on control leaves treated with water.

The monitoring of resistance to dodine in population of *V. inaequalis* was conducted in 64 orchards. From each orchard 10 scabbed leaves were collected. Conidia from a single scab lesion from each leaf were placed directly onto potato dextrose agar (PDA) with addition of 1 ppm (mg/l) dodine and on fungicide-free PDA as control. After 24 h of incubation, the germination of conidia was checked. From each orchard 1000 conidia (100 from 10 lesions) were screened. Then index of resistance level (IRL%) was calculated according to the formula described above.

A – percentage of germinated spores on PDA with dodine

B – percentage of germinated spores on fungicide-free medium

Results and Discussion

The results of monitoring showed that the problem of *V. inaequalis* resistance to strobilurin fungicides occurs in considerable number of apple orchards in Poland. In 2004, in 2 of 10 monitored orchards above 60% of resistant forms in fungus population were found (IRL > 60%), while in the remaining 8 orchards it varied from 1.4 to 21.8% (Figure 1). In 2005, in 4 of 20 monitored orchards the percentage of resistant forms was above 60% and in 2 orchards it amounted to 32.2 and 42.6% respectively. In the remaining orchards the level of resistant forms ranged from 0 to 14.8% (Figure 3). In the season 2006, resistant forms to strobilurin were determined in 11 orchards. In 2 of them the percentage of resistant forms ranged above 50%, in 2 orchards – 26% and 34.1%, respectively. In the remaining 5 orchards it varied from 0.53 to 21%. It should be pointed out that only in 2 of the monitored orchards resistance to strobilurins was not

found (Figure 5). In 2007, in 3 of 12 orchards the level of resistant forms ranged from 27.2 to 36.5% and in the other 9 orchards it varied from 0.2 to 20.9% (Figure 7).

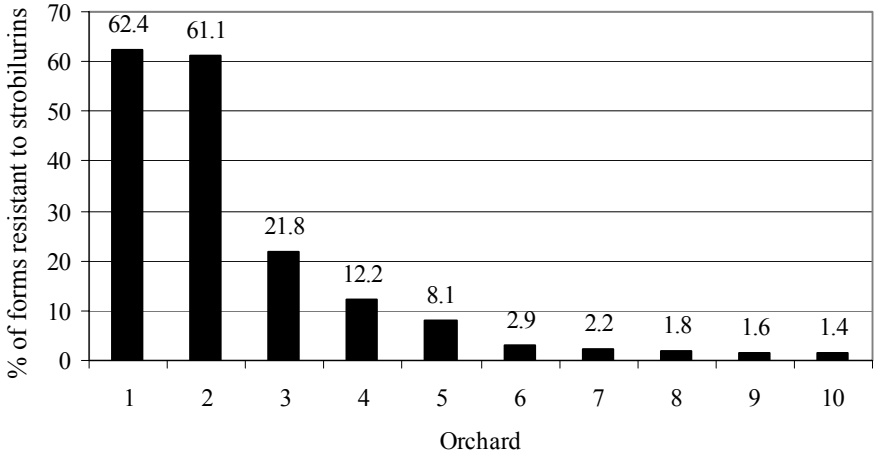

Figure 1. Percentage of strobilurin-resistant forms in *V. inaequalis* populations in 2004

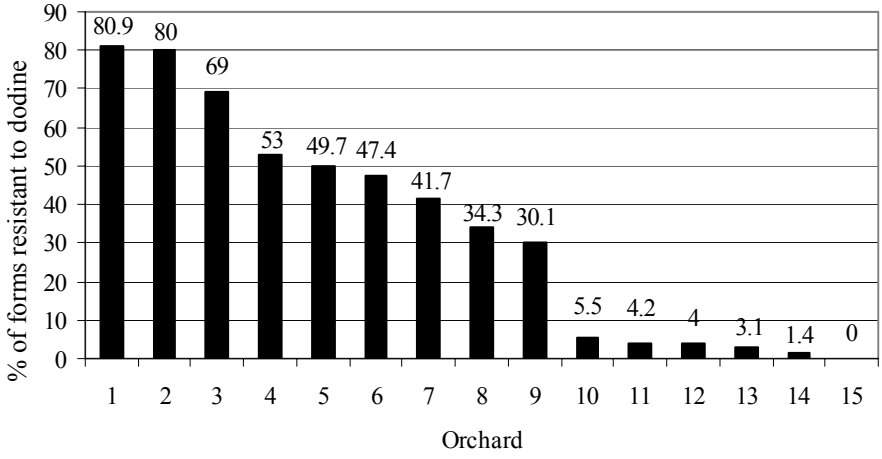

Figure 2. Percentage of dodine-resistant forms in *V. inaequalis* populations in 2004

This situation indicates that the use of strobilurin fungicides in apple orchards should be reduced. In orchards with *V. inaequalis* resistant forms, the use of strobilurins is not recommended at all. In other orchards these fungicides should be used only in a mixture with a fungicide of different mode of action and in strictly limited number of treatments (2–3 per season), according to Fungicide Action Committee (FRAC) recommendations /Dux et al., 2004/. In literature there is a lack of detailed information on the level of *V. inaequalis* resistance to various groups of fungicides. International FRAC QoI Working Group report from 2007 indicates that the levels of *V. inaequalis* resistance

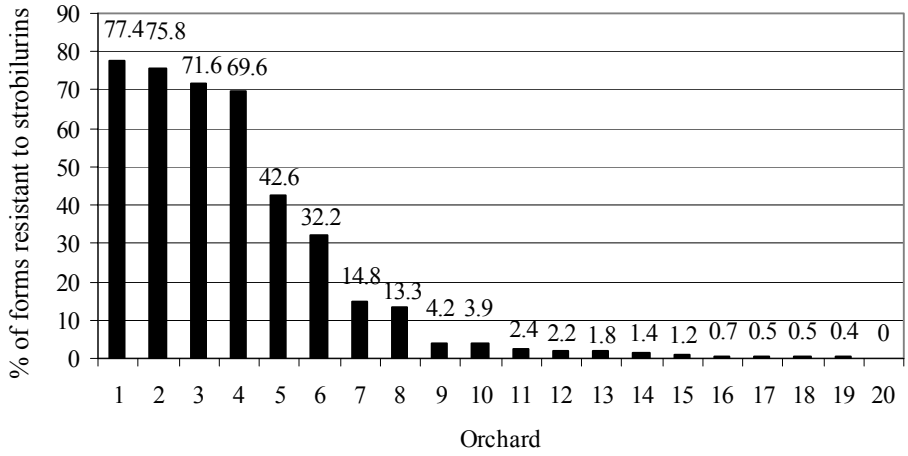

Figure 3. Percentage of strobilurin-resistant forms in *V. inaequalis* populations in 2005

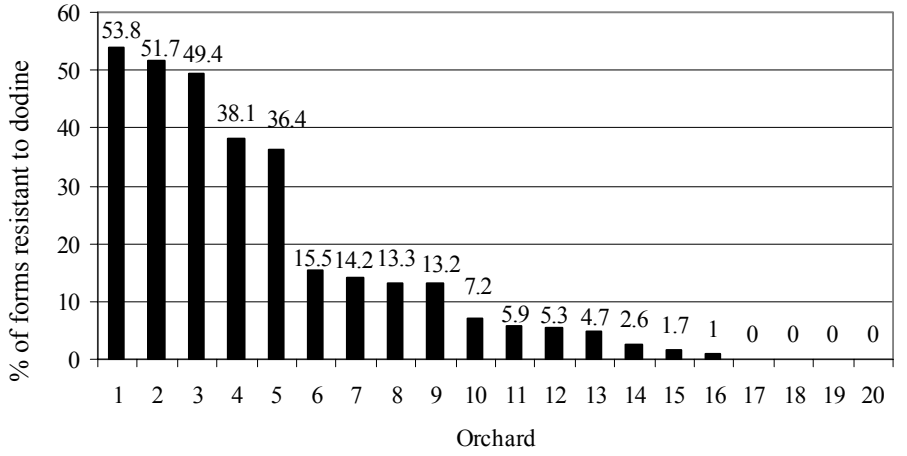

Figure 4. Percentage of dodine-resistant forms in *V. inaequalis* populations in 2005

to QoI are very heterogeneous. Moderate to high resistance was observed in Southern France, Northwest Italy (Piedmont and Po Valley), Poland and Eastern Germany. Low to moderate resistance was noted in Spain, Belgium and Western Germany. Low resistance was found in Northeast Italy (Terentino, Alto Adige) and Northern France (Loire Valley). No resistance was detected in the UK, Portugal, Lake Constance area of Germany, Canada, New Zealand. Resistance was also confirmed in the USA, South Africa and Greece. This information indicates that *V. inaequalis* resistance to QoI is a serious problem in many countries. The mechanism of *V. inaequalis* resistance to QoI is very complex. In the case of its mutation in cytohrom *b* gene and induction of alternative pathway respiration is observed. But mutation does not always explain the QoI resistant phenotype /Fernández-Ortuño et al., 2008/.

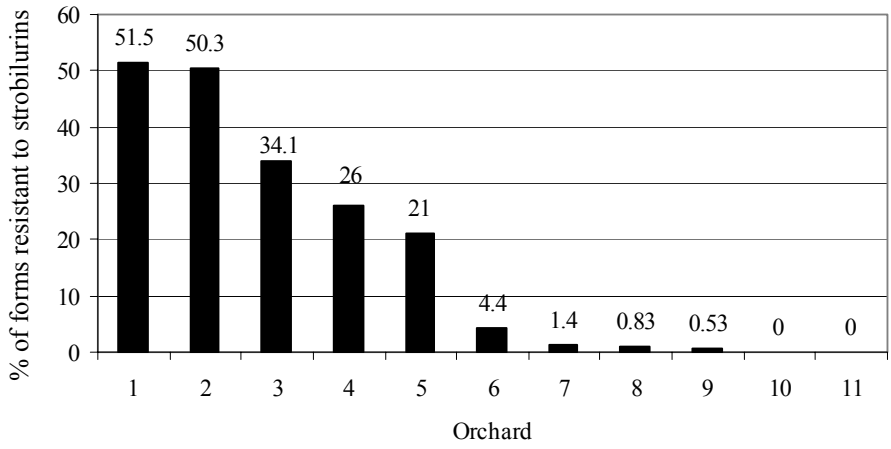

Figure 5. Percentage of strobilurin-resistant forms in *V. inaequalis* populations in 2006

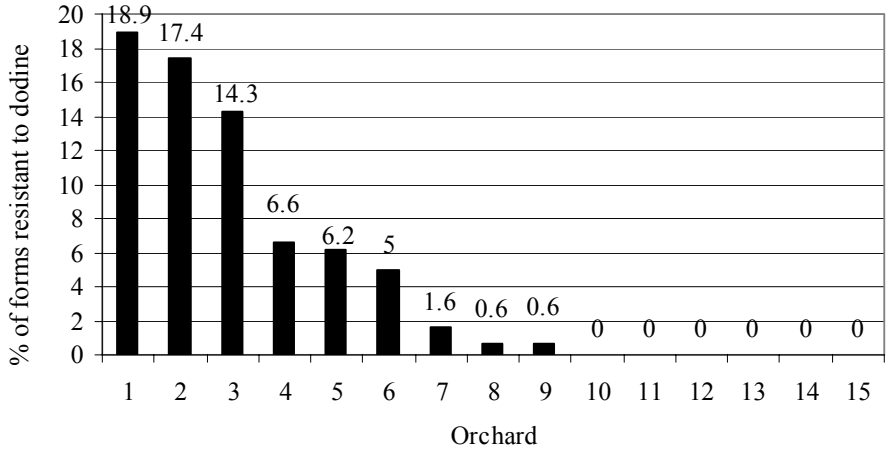

Figure 6. Percentage of dodine-resistant forms in *V. inaequalis* populations in 2006

Monitoring conducted in 1998–1999 showed that in some Polish orchards more than 50% of fungus strains in the population were resistant to dodine /Meszka, Bielenin, 2001/. In 2004, the conidial germination tests showed that a level of resistance in 4 of 15 monitored orchards was very high (above 50%). In 5 orchards it ranged from 30.1 to 49.7% and in the remaining 6 orchards it was low (below 6%) (Figure 2). In 2005, in 2 of 20 monitored orchards the percentage of resistant forms was above 50%, in 3 orchards it amounted from 36.4 to 49.4%, in the remaining 11 orchards it varied from 1 to 15.5%. Only in 4 orchards the resistance to dodine was not observed (Figure 4). In 2006, in 9 of 15 monitored orchards the resistant forms amounted from 0.6 to 18.9% and in the other 6 orchards, they were not detected (Figure 6). In 2007, only in 2 of 14 orchards resistant forms amounted to 57 and 32.9% and in the other 12 orchards the level of resistance was

low (from 1.1 to 18.8%) (Figure 8). The results of monitoring showed that the resistance of *V. inaequalis* to dodine is still a serious problem in Polish orchards. Probably dodine resistance gene may persist in the fungus population for a very long period. Practically *V. inaequalis* dodine resistance could recur rapidly in response to resumed dodine usage.

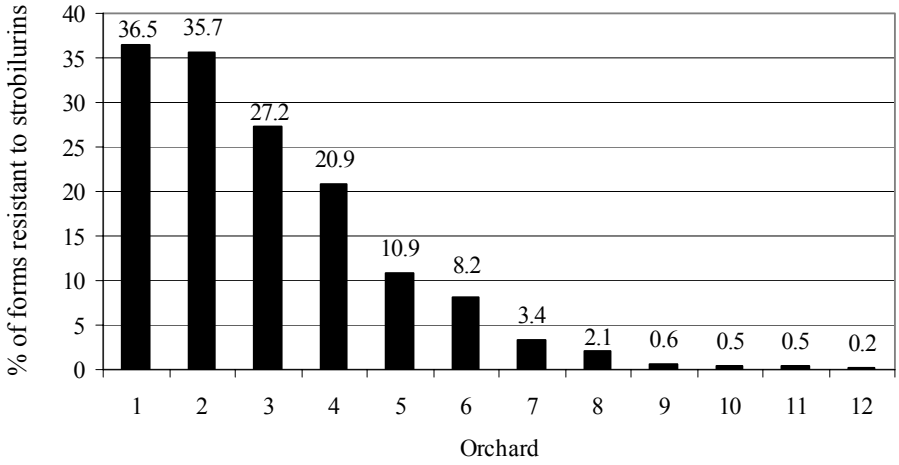

Figure 7. Percentage of strobilurin-resistant forms in *V. inaequalis* populations in 2007

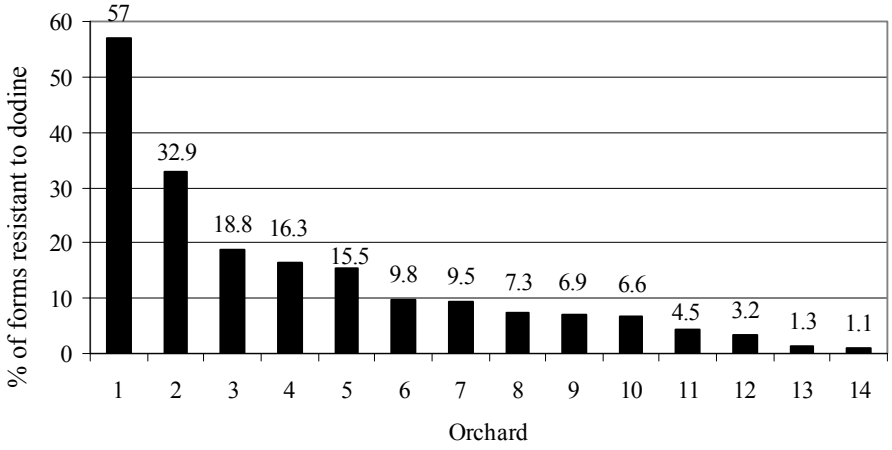

Figure 8. Percentage of dodine-resistant forms in *V. inaequalis* populations in 2007

Monitoring of commercial orchards in New York and Michigan revealed that dodine sensitivities were not uniform throughout regions where dodine resistance was widespread in the late 1970s /Szkolnik, Gilpatrick, 1973/. Sensitivities ranged from baseline to dodine-resistant and appeared to reflect the dodine use history at particular sites. Mechanism details of dodine resistance have not been elucidated, resistance development progressed through a pattern typical for polygenic resistance, with resistant phenotypes comprising the least sensitive part of continuous distribution of isolate sensitivities in baseline populations /Köller, Wilcox, 1999/. Scholberg et al. (1989) reported that

some conidia from half of 173 orchards in Canada were resistant to dodine. There are reports from another countries of *V. inaequalis* strains less sensitive to dodine /Cesari et al., 1985/, therefore applications of dodine fungicides should be still strictly limited. These fungicides may be used only in orchards, in which resistance to dodine is not observed or if it is present at very low level (up to 5%) and only 1–2 times per season.

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REFERENCES

1. Bartlett D. W., Clough J. M., Godzin J. R. et al. Review the strobilurin fungicides // Pest Management Science. – 2002, vol. 58, p. 649–662

2. Broniarek-Niemiec A., Bielenin A. Problem odporności *Venturia inaequalis* na fungicydy stosowane w sadach jabłoniowych w Polsce // Folia Horticulturae Suplement. – 2003, vol. 1, p. 590–592

3. Broniarek-Niemiec A., Bielenin A. A simple method for monitoring of *Venturia inaequalis* resistance to strobilurins // Abstracts 14th International Reinhardsbrunn Symposium "Modern Fungicides and Antifungal Compounds", 25–29April, 2004, Reinhardsbrunn, Germany. – 2004, p. 38–39

4. Broniarek-Niemiec A., Bielenin A. Monitoring odporności *Venturia inaequalis* na fungicydy strobilurynowe i dodynowe // Zeszyty Naukowe Instytutu Sadownictwa i Kwiaciarstwa. – 2005, vol. 13, p. 143–150

5. Cesari A., Fiaccadori R., Brandolini V. Résistance à la dodine chez *Venturia inaequalis* dans les vergers de pommiers de l'Italie septentrinale // EPPO Bulletinn. – 1985, 15, p. 473–476

6. Dux H., Sierotzki H., Gisi U. Sensitivity of *Venturia inaequalis* populations to anilinopyrimidyne, DMI and QoI fungicides // Abstracts 14th International Reinhardsbrunn Symposium "Modern Fungicides and Antifungal Compounds", 25–29 April, 2004, Reinhardsbrunn, Germany. – 2004, p. 40–41

7. Fernández-Ortuño D., Torès J., Pèrez-Gracía A. Mechanisms of resistance to QoI fungicides in phytopathogenic fungi // International Microbiology. – 2008, vol. 11, p. 1–9

8. Köller W., Wilcox W. F. Evidence for Predisposition of Fungicide-Resistant isolates of *Venturia inaequalis* to a Preferential Selection for Resistance to Other Fungicides // Phytopathology. – 2001, vol. 91, p. 776–781

9. Köller W., Wilcox W. F. Quantification, persistence, and status of dodine resistance in New York and Michigan orchard opulations of *Venturia inaequalis* // Plant Disease. – 1999, vol. 83, p. 66–70

10. Meszka B., Bielenin A. Decrease of *Venturia inaequalis* (Cook) Aderh. Sensitivity to Ergosterol Biosynthesis Inhibitor and dodine fungicides // Journal of Fruit Ornam. Plant Res. – 2001, vol. 9, p. 85–92

11. Nowacka H. A decrease of *Venturia inaequalis* (Cke.) Aderch. Sensitivity to fenarimol // Fruit Sci. Rep. – 1991, vol. 18, p. 139–142

12. Scholberg P. L., Yorston J. M., Warnock D. Resistance of *Venturia inaequalis* to benomyl and dodine in British Columbia, Canada // Plant Dis. – 1989, vol. 73, p. 667–669

13. Szkolnik M., Gilpatrick J. D. Tolerance of *Venturia inaequalis* to dodine in relation to the history of dodine usage in the orchard // Plant Dis. Rep. – 1973, vol. 57, p. 817–821