# Influence of Pesticides on Yeasts Colonizing Leaves

Renata Vadkertiová* and Elena Sláviková

Culture Collection of Yeasts, Institute of Chemistry, Slovak Academy of Sciences, Dúbravská cesta 9, 845 38 Bratislava, Slovakia. Fax: +421-2-59410222.
E-mail: renata.vadkertiova@savba.sk

* Author for correspondence and reprint requests

Z. Naturforsch. **66 c**, 588–594 (2011); received April 11/September 6, 2011

The effect of nine different pesticides on the growth of yeasts isolated from the leaves of fruit and forest trees was investigated. Four insecticides (with the active ingredients: thiacloprid, deltamethrin, lambdacyhalothrin, and thiamethoxam) and five fungicides (with the effective substances: bitertanol, kresoxim-methyl, mancozeb, trifloxystrobin, and cupric oxychloride) were tested. The concentrations of chemicals were those recommended by the manufacturers for the spraying of trees. The yeast strains isolated from the leaves of fruit trees were not sensitive to any of the insecticides. The majority of yeast strains isolated from the leaves of forest trees were either not sensitive or only to a small extent. While *Rhodotorula mucilaginosa* and *Pichia anomala* were not affected by any insecticide, the strains of *Cryptococcus laurentii* and *Rhodotorula glutinis* showed the highest sensitivity. The effects of fungicides on the growth of isolated yeasts were more substantial. The fungicide Dithane® DG (mancozeb) completely inhibited the growth of all yeasts. All strains isolated from fruit tree leaves were more resistant to the tested fungicides than those isolated from the leaves of forest trees. The most resistant strains from the leaves of fruit trees belonged to the species *Metschnikowia pulcherrima*, *Pichia anomala*, and *Saccharomyces cerevisiae*, whereas *Cryptococcus albidus* and *C. laurentii*, originating from the leaves of forest trees, showed the highest sensitivity to fungicides.

*Key words:* Yeasts, Pesticides, Inhibition

## Introduction

All aerial plant surfaces are inhabited by diverse assemblages of microorganisms, including bacteria, filamentous fungi, yeasts, and algae. These organisms have profound effects on plant health and thus impact on the ecosystem (Lindow and Brandl, 2003). The yeasts form a major component of the population on leaves (Inácio *et al.*, 2002). Leaf surfaces are colonized by members of several genera of saprophytic yeasts that provide a natural buffer against plant pathogens (Fokkema, 1988). Furthermore, the yeasts, as eukaryotic cells, are considered to be good models for the assessment of the toxicity of many chemical compounds, among them pesticides (Ribeiro *et al.*, 2000).

Various insecticides and fungicides are applied to protect crops, vegetables, fruits, and nuts from insects and fungal diseases. These agrochemicals differ from each other by their chemical composition, physico-chemical properties, and mode of action. Insecticides generally target the nervous system, the growth, and development, or energy production of the pest (Brown, 2005), while fungicides act mainly on essential fungal functions such as respiration, sterol biosynthesis or cell division (Leroux, 2003). However, when these compounds are used in plant protection, they can also affect non-target microorganisms associated with the plant phyllosphere.

The effect of a particular pesticide on phyllosphere microorganisms depends on the specific chemical properties of the pesticide and the concentration at which the pesticide is used, the type of indigenous phyllosphere microorganisms present, and the environment in which the organisms grow (Walter *et al.*, 2007).

Many pesticides are non-specific in their mode of action, and little is known about their impact on the microbial community in the plant phyllosphere. Some agrochemicals can inhibit the growth of individual bacteria, yeasts, and fungi and/or whole populations of non-target microorganisms (Walter *et al.*, 2007; Zhang *et al.*, 2009).

The application of the insecticide cypermethrin on the pepper plant phyllosphere caused a decrease in the abundance of fungi and a shift in the

community composition towards bacteria (Zhang *et al.*, 2009). The study of the influence of pesticides on the yeasts isolated from agricultural soil showed that the insecticides cypermethrin and triazamate inhibited the growth of *Pseudozyma aphidis* very strongly; this yeast grew very well in the presence of the fungicides fluquinconazole and prochloraz (Sláviková and Vadkertiová, 2003).

Gildemacher *et al.* (2004) investigated the variability in the effects of apple scab fungicides. Dithianon and dodine were active against all tested species, while bupirimate and pyrimethanil were only slightly active. The red yeasts *Sporobolomyces roseus* and *Rhodotorula glutinis* were completely resistant to captan and only slightly affected by the higher doses of tolylfluanid. It was also reported that representatives of the genus *Sporobolomyces* and, to some extent, *Rhodotorula* were isolated more frequently from the leaves of fungicide-treated apple trees as compared to untreated ones (Andrews and Kenerley, 1978). The application of organic fungicides on wine grapes during ripening caused a drastic reduction in the yeast flora and a shift in the yeast population towards *Aureobasidium pullulans*, whereas *Hanseniaspora uvarum* and *Metschnikowia pulcherrima* were dominant on untreated grapes (Comitini and Ciani, 2008). The growth of *Hanseniaspora uvarum* was also inhibited by the fungicide pyrimethanil during the spontaneous wine fermentation which was confirmed by assays of toxicity *in vitro* (Čuš and Raspor, 2008). It was established that the effect of pesticides on the phylloplane fungi was closely related to their effect on the growth rates of these fungi *in vitro* (Southwell *et al.*, 1999).

The objective of the present research was to compare the sensitivity of eight yeast species most frequently isolated from the leaves of fruit trees (fungicide-treated trees) and forest trees (non-treated trees) to nine different pesticides *in vitro*, and to find out whether the pesticides used could reduce or restrict the growth of these yeast strains.

## Material and Methods

Fourty-eight yeast strains belonging to eight different yeast species were chosen in order to study their tolerance to nine pesticides. The set of chosen strains originated from the leaves of fruit trees and forest trees. The species listed below belong to the species most frequently isolated from both types of trees (Sláviková *et al.*, 2007, 2009):

*Aureobasidium pullulans* CCY 27-1-117 (pine), L10 (maple), L26 (spruce), CCY 27-1-118 (peach tree), CCY 27-1-119 (cherry tree), $L49_1$ (plum tree);

*Cryptococcus albidus* L19 (hornbeam), $L29_2$, L33 (pine), CCY 17-4-39, L255 (apple tree), CCY 17-4-40 (cherry tree);

*Cryptococcus laurentii* CCY 17-3-30 (maple), L33 (pine), L38 (linden), CCY 17-3-32 (cherry tree), 17-3-33 (apricot tree), L175 (peach tree);

*Metschnikowia pulcherrima* CCY 29-2-127 (maple), L9 (beech), L36 (oak), CCY 29-2-128 (peach tree), CCY 29-2-129 (plum tree), L152 (apple tree);

*Pichia anomala* CCY 38-1-33 (maple), CCY 38-1-34 (spruce), L39 (linden), CCY 38-1-35 (plum tree), $L98_2$ (peach tree), L236 (cherry tree);

*Rhodotorula glutinis* CCY 20-2-36 (ash), CCY 20-2-37 (pine), L3 (hornbeam), CCY 20-2-39 (cherry tree), CCY 20-2-41 (apricot tree), L 227 (apple tree);

*Rhodotorula mucilaginosa* CCY 20-1-33, L6 (maple), L36 (oak), CCY 20-1-34, CCY 20-1-35 (apple tree), L157 (plum tree);

*Saccharomyces cerevisiae* CCY 21-4-110 (oak), CCY 21-4-108 (pine), L8 (hornbeam), CCY 21-4-112 (plum tree), CCY 21-4-114 (peach tree), $L95_2$ (apricot tree).

Forest trees: ash (*Fraxinus excelsior* L.); beech (*Fagus silvatica* L.); hornbeam (*Carpinus betulus* L.); linden (*Tilia cordata* Mill.); maple (*Acer campestre* L.); oak (*Quercus robur* L. ex Simk.); pine (*Pinus silvestris* L.); spruce (*Picea abies* Karst.).

Fruit trees: apple tree (*Malus domestica* Borkh., cultivar Jonathan); apricot tree (*Prunus armeniaca* L., cultivar Maďarska); cherry tree (*Prunus avium* L., cultivar Karešova); peach tree (*Prunus persica* L., cultivar Redhaven); plum tree (*Prunus domestica* L., cultivar Stanley).

One hundred and thirty seven yeast strains belonging to 17 species were isolated from the leaves and needles of the forest trees (Sláviková *et al.*, 2007) and 155 strains belonging to 17 species were isolated from the leaves of the fruit trees (Sláviková *et al.*, 2009). The strains were maintainted on malt agar in a refrigerator and used in various studies. As the number of isolated strains was large, only some strains (with the acronym

CCY) were chosen to be deposited in the Culture Collection of Yeasts (Institute of Chemistry, Slovak Academy of Sciences, Bratislava, Slovakia).

Four insecticides and five fungicides were tested for their activities.

Insecticides: Calypso® 480 SC with the active ingredient thiacloprid; Decis® EW 50 with the active ingredient deltamethrin; Karate® Zeon with the active ingredient lambdacyhalothrin; and Actara® 25 WG with the active ingredient thiamethoxam. Fungicides: Baycor® 25 WP with the active ingredients bitertanol (25%) and alkylarylsulfonate (1%); Discus® with the active ingredient kresoxim-methyl; Dithane® DG with the active ingredient mancozeb; Zato® 50 WG with the active ingredient trifloxystrobin; and Kuprikol® with the active ingredient cupric oxychloride.

All pesticides (trademark compounds) were obtained from a local market. The fruit trees from which the yeast strains were isolated are routinely sprayed with the above mentioned pesticides. Trees in all tree localities were sprayed at the same time, either to rid the trees of pest or fungi, or as part of a regular spraying schedule, in accordance with the manufacturer's instructions.

The impact of the pesticides on the growth of yeasts was studied in the laboratory. The strains were cultured in a medium consisting of 6.7 g yeast nitrogen base (Difco, Houston, TX, USA) and 20 g of glucose per litre of distilled water. pH was adjusted to 6.5. The medium was sterilized by autoclaving at 121 °C for 15 min. The pesticides were sterilized by filtration and aseptically added to the medium to reach the final contents recommended by the manufacturers for spraying of fruit trees (Figs. 1 and 2).

The strains were cultivated in L-shaped tubes containing 9.5 ml of sterile medium and 0.5 ml of inoculum ($10^6$ cell/ml). The yeasts were grown aerobically at 22 °C on a shaker (100 rev/min). The biomass yields were measured after 7 d of cultivation and compared with the biomass production in control samples. The biomass yields in control samples were considered as the maximum, *i.e.* 100% of growth. Growth yield of the yeasts was determined as dry biomass (drying at 105 °C to constant weight). All experiments were repeated three times, and the mean value of three experiments for each strain was calculated. These data were used to compare each group of three strains of the same species and the same origin. Mean values and standard deviations for each group were calculated using Microsoft Excel 2007 (see Figs. 1, 2a, b).

## Results and Discussion

Six strains from every isolated yeast species (three strains isolated from the leaves of fruit trees and three strains isolated from the leaves of forest trees) were used for the study of pesticide effects on yeasts. Two classes of pesticides, four insecticides and five fungicides, were tested. The concentration of pesticides in the media corresponded to those recommended by the manufacturers for the particular spraying.

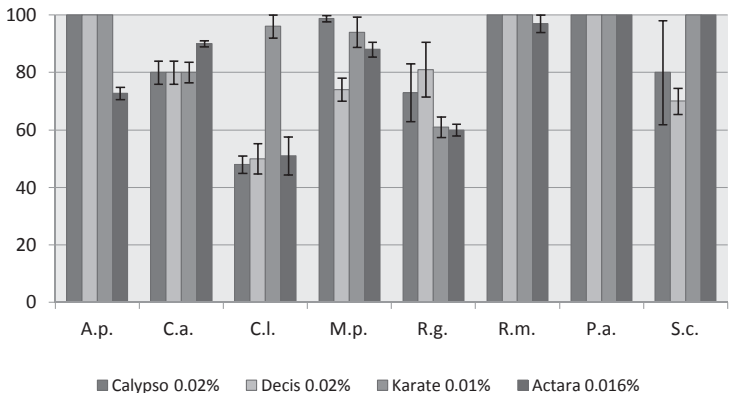


Fig. 1. The influence of insecticides on the growth of yeasts isolated from forest tree leaves. Values shown indicate growth in % of controls without pesticide. A.p., *Aureobasidium pullulans*; C.a., *Cryptococcus albidus*; C.l., *Cryptococcus laurentii*; M.p., *Metschnikowia pulcherrima*; R.g., *Rhodotorula glutinis*; R.m., *Rhodotorula mucilaginosa*; P.a., *Pichia anomala*; S.c., *Saccharomyces cerevisiae*. Each value is the mean of tree strains. Bars represent the standard deviation of three strains.

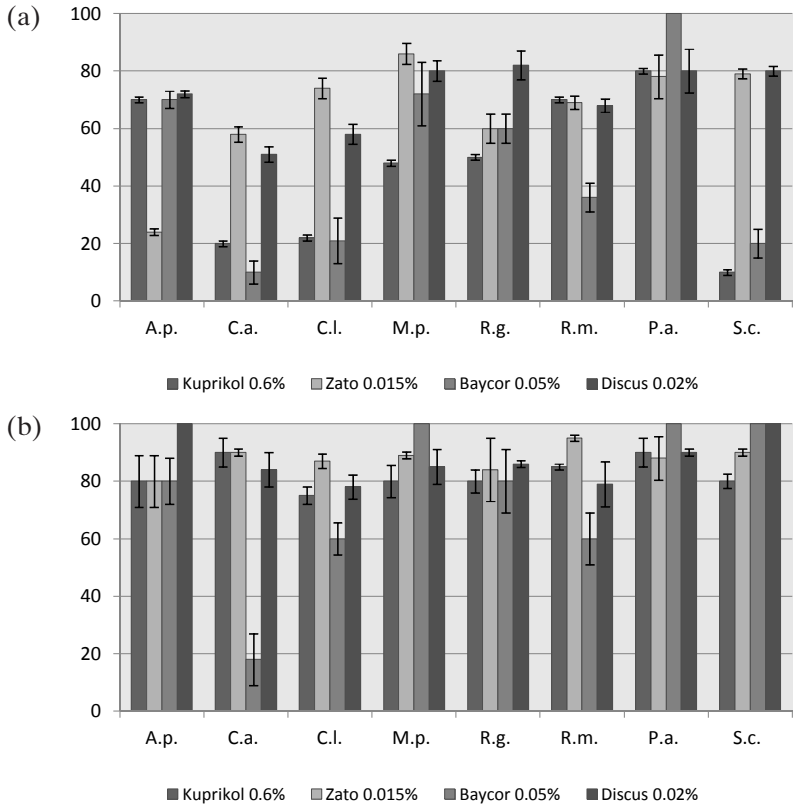


Fig. 2. The influence of fungicides on the growth of yeasts (no growth was observed in the presence of Dithane® DG): (a) Yeasts isolated from forest tree leaves. (b) Yeasts isolated from fruit tree leaves. Values shown indicate growth in % of controls without pesticide. A.p., *Aureobasidium pullulans*; C.a., *Cryptococcus albidus*; C.l., *Cryptococcus laurentii*; M.p., *Metschnikowia pulcherrima*; R.g., *Rhodotorula glutinis*; R.m., *Rhodotorula mucilaginosa*; P.a., *Pichia anomala*; S.c., *Saccharomyces cerevisiae*. Each value is the mean of tree strains. Bars represent the standard deviation of three strains.

The yeast strains isolated from the leaves of the fruit trees were not sensitive to the insecticides. The sensitivity of *Rhodotorula glutinis* to the insecticide Decis® EW 50 was insignificant. On the other hand, some, but not all, of the yeast strains isolated from the leaves of forest trees were sensitive to the insecticides (Fig. 1). The strains of two species showed a higher sensitivity: *Cryptococcus laurentii* was the most sensitive to Decis® EW 50, Calypso® 480 SC, and Actara® 25 WG, and *Rhodotorula glutinis* to Karate® Zeon. The strains of *Pichia anomala* and *Rhodotorula mucilaginosa* were not or hardly affected (Fig. 1). Krzepiłko and Święciło (2007) examined the effect of pyrethroids on the growth of *Saccharomyces cerevisiae* cells. They observed that deltamethrin (the active ingredient of Decis® EW 50) was the most toxic among pyrethroid insecticides. As our results show, the five species tested were sensitive to Decis® EW 50 (Fig. 1).

The ability to degrade some insecticides has been reported by Cabras *et al.* (1988) who noted absorption and degradation activity of *Saccharomyces cerevisiae* towards insecticides (among them deltamethrin), and Dai *et al.* (2010) reported the ability of *Rhodorula mucilaginosa* to degrade thiacloprid (the active ingredient of Calypso® 480 SC).

Conner (1983) pointed out that the yeast *Saccharomyces cerevisiae* is not as susceptible to insecticides and herbicides as it is to fungicides. Figs. 2a and 2b show that the influence of fungicides on the yeast growth was more pronounced. The effect of the fungicides on the biomass production was dependent not only on the respective fungicide but also on the yeast species.

The fungicide Dithane® DG (0.2%), with the active ingredient mancozeb, totally inhibited the growth of all strains. Mancozeb has very often been used as a protective fungicide as well as in treatments to control a wide spectrum of acute fungal diseases (Gandhi and Snedeker, 2000). It significantly reduced the fungal population on wheat and barley leaves (Southwell *et al.*, 1999) and was also very toxic to *Saccharomyces cerevisiae* strains (Conner, 1983). Our results also confirmed the high toxic effect of mancozeb. In its presence, none of the strains tested was able to grow.

Baycor® 25 WP (0.05%) and Kuprikol® (0.6%) had a higher inhibitory effect on the growth of yeasts isolated from leaves of the forest trees (Fig. 2a) than on that of yeasts isolated from leaves of the fruit trees (Fig. 2b). Both fungicides inhibited the growth of *Cryptococcus albidus*, *Cryptococcus laurentii*, and *Saccharomyces cerevisiae*, originating from leaves of the forest trees. On the other hand, the strains of *Pichia anomala* were not affected by Baycor® 25 WP, independent of their origin. None of the strains tested was completely resistant to Kuprikol®. Whereas, doses of Kuprikol® lower than recommended (0.05 and 0.3%) did not inhibit the growth of the yeast strains at all.

The fungicides Discus® (0.02%) and Zato® 50 WG (0.015%) belong to the strobilurins – fungicidal compounds produced by basidiomycetes and subsequently converted to commercial preparations by a synthetic process. Strobilurins inhibit the mitochondrial respiration of fungi and are active against a wide range of fungal plant pathogens (Dayan *et al*., 2009).

Similar to the previous fungicides, strobilurins also produced greater inhibition of the strains isolated from forest as compared to those from fruit trees, but the differences were not so pronounced (Figs. 2a, b). The species *Aureobasidium pullulans* and *Cryptococcus albidus* exhibited the highest variation in their sensitivity to strobilurins. The strains of two species – *Saccharomyces cerevisiae* and *Aureobasidium pullulans*, originating from the leaves of fruit trees – were not affected by Discus®, whereas none of the strains tested was completely resistant to Zato® 50 WG (Figs. 2a, b).

To summarize these results, it is evident that the yeast strains isolated from leaves of the fruit trees were insensitive to the insecticides tested and less affected by the fungicides than those isolated from leaves of the forest trees. The species *Rhodotorula mucilaginosa* and *Pichia anomala* were not affected by any of the insecticides. The strains of the species *Saccharomyces cerevisiae* isolated from fruit tree leaves showed the highest resistance among all the strains tested. They were not inhibited by two insecticides – Karate® Zeon and Actara® 25 WG, and two fungicides – Baycor® 25 WP and Discus®. Calhelha *et al.* (2006) also reported that *Saccharomyces cerevisiae* belonged to the yeasts most resistant to some fungicides. On the other hand, the strains originating from leaves of the forest trees were strongly sensitive to Baycor® 25 WP and Kuprikol®. Ribeiro *et al.* (2000) reported that the fungicide cymoxanil significantly inhibited *Saccharomyces cerevisiae*, whereas penconazol did not. Čuš and Raspor (2008) noted that a lower concentration of pyrimethanil (1 mg/l) did not affect the anaerobic growth of *Saccharomyces cerevisiae,* but a higher concentration (10 mg/l) diminished the initial growth of the yeast.

Similar to *Saccharomyces cerevisiae*, the strains of *Cryptococcus* species, isolated from leaves of the forest trees, were strongly affected by Baycor® 25 WP and Kuprikol®. It was previously found that the strains of the genus *Cryptococcus*, isolated from agricultural soil, were the most sensitive to pesticides among all the strains tested (Sláviková and Vadkertiová, 2003), and their populations were diminished by the fungicides captan and dithianon (Gildemacher *et al.*, 2004). On the contrary, Comitini and Ciani (2008) found *Cryptococcus* species, together with *Aureobasidium pullulans*, to be the prevalent species on grapes treated with organic fungicides.

On the basis of the above findings, it can be concluded that certain fungicides totally or significantly reduce the yeast population, while others have only a moderate or no effect on yeasts. The fungicide Discus® exhibited the most toxic effect.

Buck and Burpee (2002) suggested that the selection pressure caused by fungicide applications results in the formation of yeast populations highly resistant to a variety of fungicides. Mmbaga and Sauvé (2009) found that fungicide treatments did not kill all epiphytic microorganisms. When they did, the organisms killed were replaced by new ones and rapid re-colonization occurred.

The results of the present study confirmed the adaptability of yeast strains to pesticides. The yeasts isolated from leaves of the fruit trees, which

are regularly treated with pesticides, were much more resistant to these agrochemicals than the strains of the same species isolated from leaves of the untreated forest trees.

The saprophytic yeasts were found to have antagonistic activities against plant pathogens such as *Botrytis cinerea*, *Penicillium expansum*, and *Monilia fructicola*. Furthermore, the combination of antagonistic yeasts with fungicide can more significantly enhance the biocontrol capacity of the yeasts against pathogenic fungi (Dimakopoulou *et al.,* 2008; Chand-Goyal and Spotts, 1997; Buck, 2004).

It is possible that the yeasts originating from the leaves of fruit trees may have some biocontrol activities that can be used in combination with a fungicide. The pesticide resistance, therefore, could be an important factor in the development of biological control agents against plant pathogens.

Based on the strength of the inhibitory effects of pesticides on the yeast growth, the following orders can be arranged:
insecticides: Karate® Zeon < Calypso® 480 SC < Decis® EW 50 < Actara® 25 WG;
fungicides: Discus® < Zato® 50 WG < Kuprikol® < Baycor® 25 WP < Dithane® DG.

*Acknowledgements*

This work was supported by grant VEGA No. 2/0005/10 from the Grant Agency of the Slovak Academy of Sciences.

Andrews J. H. and Kenerley C. M. (1978), The effects of pesticide program on non-target epiphytic microbial population of apple leaves. Can. J. Microbiol. **24**, 1058–1072.

Brown A. E. (2005), Mode of action of structural pest control chemicals. In: Pesticide Information Leaflet Series, Vol. 41. Maryland cooperative extension. University of Maryland, pp. 1–8. http://www.entomology.umd.edu/directory/amyebrown

Buck J. W. (2004), Combination of fungicides with phylloplane yeasts for improved control of *Botrytis cinerea* on geranium seedlings. Phytopathology **94**, 196–202.

Buck J. W. and Burpee L. L. (2002), The effects of fungicides on the phylloplane yeast populations of creeping bentgrass. Can. J. Microbiol. **48**, 522–529.

Cabras P., Meloni M., Pirisi F. M., Farris G. A., and Fatichenti F. (1988), Yeast and pesticide interaction during aerobic fermentation. Appl. Microbiol. Biotechnol. **29**, 298–301.

Calhelha R. C., Andrade J. V., Ferreira I. C., and Estevinho L. M. (2006), Toxicity effects of fungicide residues on wine-producing process. Food Microbiol. **23**, 393–398.

Chand-Goyal T. and Spotts R. A. (1997), Biological control of postharvest diseases of apple and pear under semi-commercial and commercial conditions using three saprophytic yeasts. Biol. Control **10**, 199–206.

Comitini F. and Ciani M. (2008), Influence of fungicide treatments on the occurrence of yeast flora associated with wine grapes. Ann. Microbiol. **58**, 489–493.

Conner A. J. (1983), The comparative toxicity of vineyard pesticides to wine yeasts. Am. J. Enol. Vitic. **34**, 278–279.

Čuš F. and Raspor P. (2008), The effect of pyrimethanil on the growth of wine yeasts. Lett. Appl. Microbiol. **47**, 54–59.

Dai Y. J., Ji W. W., Chen T., Zhang W. J., Liu Z. H., Ge F., and Yuan S. (2010), Metabolism of the neonicotinoid insecticides acetamiprid and thiacloprid by the yeast *Rhodotorula mucilaginosa* strain IM-2. J. Agric. Food Chem. **58**, 2419–2425.

Dayan F. E., Cantrell C. L., and Duke S. O. (2009), Natural products in crop protection. Bioorg. Med. Chem. **17**, 4022–4034.

Dimakopoulou M., Tjamos S. E., Antoniou P. P., Pietri A., Battilini P., Avramidis N., Markakis E. A., and Tjamos E. C. (2008), Phyllosphere grapevine yeast *Aureobasidium pullulans* reduces *Aspergillus carbonarius* (sour rot) incidence in wine-producing vineyards in Greece. Biol. Control **46**, 158–165.

Fokkema N. J. (1988), Agrochemicals and the beneficial role of phyllosphere yeasts in disease control. Ecol. Bull. **39**, 91–93.

Gandhi R. and Snedeker S. M. (2000), Critical evaluation of mankozeb's breast cancer risk. Cornell University. Program on breast cancer and environmental risk factors in New York State. Crit. Evaluation **13**, 1–16. http://www.cfe.cornell.edu/bcerf/

Gildemacher P. R., Heijne B., Houbraken J., Vromans T., Hoekstra E. S., and Boekhout T. (2004), Can phyllosphere yeasts explain the effect of scab fungicides on russeting of Elstar apples? Eur. J. Plant Pathol. **110**, 929–937.

Inácio J., Pereira P., De Carvalho M., Fonseca Á., Amaral-Collaco M. T., and Spencer-Martins I. (2002), Estimation and diversity of phylloplane mycobiota on selected plants in a Mediterraneantype ecosystem in Portugal. Microb. Ecol. **44**, 344–353.

Krzepiłko A. and Święciło A. (2007), The influence of selected pyrethroid on the growth and number of *ρ*-mutants in *Saccharomyces cerevisiae* yeast. Pol. J. Environ. Stud. **16**, 403–406.

Leroux P. (2003), Modes d´action des produits phytosanitaires sur les organismes pathogenes des plantes – Mode of action of agrochemicals towards plant pathogens. C. R. Biol. **326**, 9–21.

Lindow S. E. and Brandl M. T. (2003), Microbiology of the phyllosphere. Appl. Environ. Microbiol. **69**, 1875–1883.

Mmbaga M. T. and Sauvé R. J. (2009), Epiphytic microbial communities on foliage of fungicide treated and non-treated flowering dogwoods. Biol. Control **49**, 97–104.

Ribeiro I. C., Veríssimo I., Moniz L., Cardoso H., Sousa M. J., Soares A. M. V. M., and Leão C. (2000), Yeasts as a model for assessing the toxicity of the fungicides penconazol, cymoxanil and dichlofluanid. Chemosphere **41**, 1637–1642.

Sláviková E. and Vadkertiová R. (2003), Effects of pesticides on yeasts isolated from agricultural soil. Z. Naturforsch. **58c**, 855–859.

Sláviková E., Vadkertiová R., and Vránová D. (2007), Yeasts colonizing the leaf surfaces. J. Basic Microbiol. **47**, 344–350.

Sláviková E., Vadkertiová R., and Vránová D. (2009), Yeasts colonizing the leaves of fruit trees. Ann. Microbiol. **59**, 419–424.

Southwell R. J., Brown J. F., and Welsby S. M. (1999), Microbial interactions on the phylloplane of wheat and barley after applications of mancozeb and triadimefon. Australas. Plant Pathol. **28**, 139–148.

Walter M., Frampton C. M., Boyd-Wilson K. S. H., Harris-Virgin P., and Waipara W. (2007), Agrichemical impact on growth and survival of non-target apple phyllosphere microorganisms. Can. J. Microbiol. **53**, 45–55.

Zhang B., Bai Z., Hoefel D., Tang L., Wang X., Li B., Li Z., and Zhuang G. (2009), The impact of cypermethrin pesticide application on the non-target microbial community of the pepper plant phyllosphere. Sci. Total Environ. **407**, 1915–1922.