

## Availability of alternative foods can influence the impact of pesticides on predatory mites (Acari): a summary of the evidence\*

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### Abstract

Generalist predatory mites belonging to the Phytoseiidae play a major role in keeping phytophagous mites below economic threshold levels in European apple orchards and vineyards. Apart from their primary prey, these phytoseiids can exploit a range of other foods, among which pollen and plant pathogenic fungi are very important. The ability of generalist predatory mites to feed on alternative foods is of importance for their persistence in perennial crops. Pesticides can exert dramatic effects on mite communities, and these effects can be more severe on predators than on phytophagous mites, with practical consequences for pest management. Several factors of intrinsic (e.g. resistance to stress) and extrinsic nature (e.g. immigration) can influence the response of predatory mite populations to pesticide applications. Environmental conditions, in particular availability of alternative foods, could affect the resilience of predatory mite populations after pesticide applications. Here we evaluate the role of availability of alternative foods on the response of predatory mite populations to pesticide applications in two crop systems: apple and grape. In the former, increasing the abundance of pollen through appropriate grass management resulted in less pronounced negative effects of some insecticides on the predatory mite *Kampimodromus aberrans* (Oudemans). In a controlled laboratory experiment, we demonstrated that availability of fresh pollen reduced the effect of pesticides on the fecundity of that predator. In the grape system, we found a positive effect of the plant pathogen grape downy mildew (GDM) on *Amblyseius andersoni* (Chant) populations, while some pesticides had a negative impact. GDM availability on plants favored the colonization by beneficial mites of pesticide treated plants with positive implications for mite persistence in vineyards.

**Key words:** Phytoseiidae, biological control, alternative foods, pollen, grape downy mildew, pesticides.

### Introduction

Generalist predatory mites belonging to the Phytoseiidae play a major role in keeping phytophagous mites (Tetranychidae and Eriophyidae) below economic threshold levels in European orchards and vineyards. The biology of the phytoseiids *Typhlodromus pyri* Scheuten, *Amblyseius andersoni* (Chant) and *Euseius finlandicus* (Oudemans) has been widely investigated (e.g., Overmeer, 1981; Dicke *et al.*, 1990; Duso & Camporese, 1991; Wei & Walde, 1997; Schausberger, 1998; Broufas & Koveos, 2000) while other phytoseiids, such as *Kampimodromus aberrans* (Oudemans), have received less attention (e.g., Schausberger, 1998; Kasap, 2005; Broufas *et al.*, 2007). Some phytoseiid species can develop and reproduce on pollen as well as on tetranychids and eriophyids (Helle & Sabelis, 1985). Pollen is a fundamental food source for generalist predatory mites (McMurtry, 1992; McMurtry & Croft, 1997) and leaves are excellent pollen traps (Eichhorn & Hoos, 1990; Duso *et al.*, 1997). Populations of *T. pyri*, *K. aberrans* and *A. andersoni* increase when pollen is abundant on grape leaves (Engel & Ohnesorge, 1994a, b; Duso *et al.*, 1997, 2002, 2004) and a similar trend has been reported on apple leaves (Girolami *et al.*, 2000). Plant pathogenic fungi can also be food sources for certain predatory mites (Pozzebon & Duso, 2008; Pozzebon *et al.*, 2009). The presence of grape downy mildew (GDM), *Plasmopara viticola* Berk. et Curtis ex. de Bary, in vineyards increases the persistence of *A. andersoni* and *T. pyri* (Duso *et al.*, 2003; Pozzebon *et al.*, 2010).

Pesticides can have a remarkable impact on mites, involving lethal and sub-lethal as well as indirect effects. The capacity of predatory mite populations to react after pesticide application and reach populations comparable to those existing earlier depends on various factors. The availability of pollen or fungi could modify the response of predatory mite populations to detrimental pesticides.

In this paper, we review a series of experiments conducted to investigate: a) the effect of alternative food sources on predatory mite species; b) the effects of pesticides commonly used in apple orchards and vineyards on these species; c) the role of alternative foods for predatory mites in mitigating harmful pesticide side effects, with emphasis on *K. aberrans* in apple orchards and *A. andersoni* in vineyards.

## Mites of apple orchards

### Effect of grass management on predatory mites in apple orchards

Experiments were conducted in an orchard at the experiment station of E. Mach Foundation (FEM, S. Michele all'Adige, Trento, Italy), where *K. aberrans* was the completely dominant phytoseiid species (Baldessari, 2005). Three types of grass management (treatments) were compared, each replicated three times: a) low mowing frequency (twice in May); b) high mowing frequency (weekly); and c) high tillage frequency (weekly, with rotary tiller). Management activities occurred from mid June to the end of August 2004. Sampling was conducted every 7–14 days, from July to September. Leaf samples were transported to the laboratory to identify mites and determine their abundance. Data were analyzed with a repeated measures REML (Restricted Maximum Likelihood) model. We evaluated the effect of grass management practices using F test ( $p=0.05$ ). A pairwise t-test ( $p=0.05$ ) was also applied to the least-square means of treatments.

Pollen flow in the orchard was estimated using a Hirst volumetric sampler (VPPS 1000, Lanzaroni, Italy). Pollen grains were captured on a slide and then their numbers and taxa were assessed under a compound microscope at 40x magnification (Mandrioli, 1999; Mandrioli *et al.*, 1978). Data on pollen grains were analyzed with a Friedman test and compared between treatments as described by Ipe (1987).

Grass management significantly affected *K. aberrans* populations (Baldessari, 2005). Population densities declined from June to early August but increased from late August to September. The effects of grass management started in July but became noticeable in August and September. *Kampimodromus aberrans* population densities reached higher levels in plots mowed only twice than in plots mowed weekly or in tillage plots. The latter treatments did not differ between themselves. During the experimental period, pollen flow was 5–6 times higher in plots mowed only twice than in other plots. Pollen of Poaceae was dominant among pollen taxa.

### Effect of insecticides on *Kampimodromus aberrans* on apple trees

The effect of insecticides on *K. aberrans* was evaluated in an apple orchard located at FEM experiment station. The most common insecticides used in Italian apple orchards were used in this experiment: chlorpyrifos, etofenprox, thiacloprid, acetamiprid, lufenuron, indoxacarb and methoxyfenozide. All of them were applied twice, in June and July, except acetamiprid, which was applied only in June. A randomized block design was adopted with four replicates per treatment. Sampling was conducted before the first insecticide application and approximately every seven days afterwards. Apple leaf samples were taken from each plot and transported to the laboratory where mite stages were identified and counted. Data were analyzed with a repeated measures REML model and effect of pesticide application was evaluated with F test ( $p=0.05$ ). A pairwise t-test ( $p=0.05$ ) was also applied to the least-square means of treatments.

The first insecticide application affected *K. aberrans* populations. Predatory mite numbers were significantly reduced by etofenprox compared to other treatments. There were no differences between treatments immediately before the second insecticide application, but differences emerged in the subsequent sampling dates. Lower *K. aberrans* numbers were recorded in etofenprox treated plots compared to the control, chlorpyrifos and indoxacarb. *Panonychus ulmi* (Koch) populations reached low population levels. There were no significant differences among spider mite densities recorded in the various treatments (Duso *et al.*, unpubl. data).

### **Grass management, pesticides and predatory mites in apple orchards**

Experiments were carried out in two contiguous orchards located at FEM experiment station. In the first orchard (SMO, Single Mowing), mowing was conducted once in June while in the second (FMO, Frequent Mowing), mowing was conducted weekly from late May to late August. Three treatments, namely applications of chlorpyrifos or of spinosad, and untreated control, were compared in each orchard in a randomized block design with four replicates per treatment; insecticides were applied in late June. Leaf samples were taken from May to August and processed as mentioned previously. Pollen flow was estimated using the above mentioned procedure. Data were analyzed separately for each orchard using a general linear model with F test ( $p=0.05$ ). Treatment means were separated with Tukey's test ( $p=0.05$ ).

*Kampimodromus aberrans* population increased first in FMO orchard and then in SMO orchard, independently of pesticide use (Baldessari *et al.*, 2005). In July, mite densities fluctuated at similar levels in both orchards but the population decline in the summer was faster in FMO orchard. Pollen flow in SMO orchard appeared to be about four times higher than that in FMO orchard. Insecticide application significantly affected predatory mite populations in SMO orchard; considering all sampling dates together, the numbers of predators in this orchard were similar in the control and chlorpyrifos plots but more abundant in these plots compared to spinosad treated plots. In FMO orchard, predatory mites were more abundant in the control than in spinosad and chlorpyrifos plots while there were no differences between the latter 2 treatments. Hence, spinosad was detrimental to predatory mites in both orchards while chlorpyrifos exerted negative effects in FMO orchard only. Reduced mowing frequency resulted in higher pollen flow and this situation favored the recovery of predatory mite populations in the chlorpyrifos treated plots of SMO orchard.

### **Suitability of *Typha* pollen and *Panonychus ulmi* as food for *K. aberrans* in the laboratory**

*Kampimodromus aberrans* colonies were established using females collected from FEM apple orchards. The mites were reared on detached leaves and fed with *Typha* pollen. Sixty eggs were transferred singly to experimental units maintained at controlled conditions ( $23 \pm 3^\circ\text{C}$ ,  $70 \pm 10\%$  R.H., 16L: 8D photoperiod) and reared to adulthood on either the tetranychid *P. ulmi* or *Typha* sp. pollen. *Panonychus ulmi* juvenile stages and *Typha* sp. pollen were provided daily. Time period of all developmental stages and their survival rate on both diets were recorded every 12 hours. Mite oviposition was monitored every 24 hours at the same laboratory conditions. The effect of food source on phytoseiid development and oviposition was analyzed with t-test ( $p=0.05$ ).

Survival of *K. aberrans* was higher on pollen than on spider mites (91% vs. 72%). Development of females (from egg to adult) was longer on *P. ulmi* than on pollen (231.7 vs. 183.5 h). Total fecundity was higher on pollen than on *P. ulmi* (0.8 vs. 0.6 eggs/female/day) (S. Ahmad, M. Lorenzon, A. Pozzebon, C. Duso, unpubl. data).

### **Effect of pollen and pesticides on *K. aberrans* survival and fecundity in the laboratory**

This experiment was setup in a factorial design, in which the factors were insecticide application (chlorpyrifos, spinosad and control), amount of *Typha* pollen (low and high) and frequency of pollen application (low: once at the start of the experiment; high: every 48 h after the first 72 h). Prior to the

experiments, apple leaves were immersed in the pesticide solutions or in distilled water (control) for 30 seconds; after pesticide residues dried out, leaves were used to construct the experimental units. Pollen and then 2 coeval females of *K. aberrans* were transferred onto each unit. A total of 40 females was used per treatment. Female survival was recorded 72 and 168 h after the beginning of the experiment; fecundity was recorded only after 168 h. Data on survival were analyzed with a factorial logistic regression ( $p = 0.05$ ), while data on fecundity were analyzed with a REML model ( $p = 0.05$ ).

In the first assessment, survival was higher in the control than in chlorpyrifos or spinosad treatments and there was no significant difference between the latter treatments. Survival was not affected by different pollen amounts. In the second assessment, mortality was 100% in the spinosad treatment, and a significant mortality (about 13%) was induced by chlorpyrifos compared to the control, regardless the amount of pollen applied and the frequency of application. Fecundity was higher in the control than in chlorpyrifos (0.8 vs. 0.6 eggs/female/day). Predatory mite fecundity increased by augmenting pollen amount or increasing application frequency. The interaction “insecticide\*pollen application frequency” was significant: at low pollen application frequency, fecundity was higher in the control than on chlorpyrifos, while no effects of insecticides were observed at high pollen application frequency (C. Duso, A. Pozzebon, P. Tirello, unpubl. data).

## Mites of vineyards

### Effect of fungicides on predatory mites in vineyards

The effect of three fungicide strategies to control grape downy mildew, based on mancozeb, folpet or copper hydroxide, on *A. andersoni* populations was assessed in a randomized block experiment in a vineyard located at CRA – Viticulture Station, Conegliano (Northern Italy). An untreated control was also included. Fungicides were applied weekly, from May to June; in addition, wettable sulphur was applied in all plots to control grape powdery mildew, *Uncinula necator* (Schw.) Burr., on the same dates. Leaf samples were taken from May to July and processed in the laboratory. This evaluation period (here designated as “period a”) is consistent with standard procedures (Blümel *et al.*, 2000). Data on predatory mite densities were analyzed with a repeated measures REML model. Differences between treatments were evaluated with contrasts ( $p = 0.05$ ).

For an additional assessment, observations were also conducted between August and September (period b) to evaluate whether GDM spread can influence the recovery of predatory mites after the use of detrimental pesticides. Data on predatory mite densities and leaf area with GDM sporulations were analyzed with a repeated measures REML model. Differences between treatments were evaluated with contrasts ( $p = 0.05$ ).

In period (a), fungicide applications affected predatory mite populations (Pozzebon *et al.*, 2008). Lower (by about 30%) *A. andersoni* densities were observed in mancozeb than in control plots while no effects were determined for other fungicides compared to the control.

In period (b), predatory mites increased in number and their densities were higher in the control than in fungicide treated plots. Higher *A. andersoni* densities were found in copper hydroxide than in folpet or mancozeb plots, and the differences between folpet and mancozeb determined earlier in the season disappeared. GDM incidence was higher in the control than in treated plots. Among the treated plots, GDM incidence was similar between copper hydroxide and mancozeb plots and higher on these plots than on folpet plots. The high incidence of GDM in late season favored predatory mite populations in plots treated with mancozeb, a fungicide known for its detrimental effects on *A. andersoni*.

### Suitability of GDM, pollen and spider mites as foods for *A. andersoni*

We investigated the effect of *Typha* pollen, the tetranychids *P. ulmi* and *Eotetranychus carpini* (Oudemans), and GDM on *A. andersoni* biological parameters. The experimental procedures re-

ported for *K. aberrans* were applied to an *A. andersoni* strain collected from vineyards. Grapevine leaf sections were used for the experiments. Data were analyzed using one-way ANOVA with F test ( $p=0.05$ ). Treatment means were separated using Tukey-Kramer test ( $p=0.05$ ).

*Amblyseius andersoni* females developed and reproduced on GDM mycelium (Lorenzon *et al.*, 2011). Developmental periods (egg-adult) of predators fed on GDM or *P. ulmi* were similar, longer on these treatments compared to pollen, and intermediate on *E. carpini*. Oviposition rates were similar for mites fed on pollen and tetranychids, but lower (at least 58%) on GDM mycelium.

## Discussion

Laboratory studies have demonstrated the suitability of *Typha* pollen as food for *K. aberrans*. Grass pollen has been shown to be suitable (Engel & Onhesorge, 1994b) for predatory mites having the same life style of *K. aberrans*. Pollen availability on leaves may depend on grass management; pollen flow was much higher in plots mowed only twice than in plots mowed or tilled weekly. High pollen presence in airflow is likely to induce high pollen deposit on leaves and thus increase food availability to phytoseiids. Implications for predatory mite population dynamics were clear in mid-summer, when prey availability diminishes and windborne pollen becomes the most frequent food source for generalist predatory mites (Addison *et al.*, 2000). Frequent mowing in the summer is a common cultural practice in European apple orchards; this reduces grass flowering and pollen availability in orchards (Girolami *et al.*, 2000; Madinelli *et al.*, 2002). Reducing mowing frequency will enhance pollen deposition in the canopy, thereby increasing the provision of alternate food for phytoseiids. Pesticides caused dramatic effects on *K. aberrans* in apple orchards, but grass management in the summer influenced the outcome of insecticide application on predatory mite abundance. Detrimental effects of chlorpyrifos appeared to be less pronounced in plots where grass management allowed a high pollen flow (Baldessari *et al.*, 2005). The laboratory experiment with pollen and pesticide applications provided possible explanations for the phenomenon observed in experimental fields. Chlorpyrifos applications were detrimental to *K. aberrans* in terms of survival and fecundity. However, pollen augmentation resulted in higher fecundity of that predator, independently of the presence of that insecticide. The effect of pollen application frequency was observed on predatory mites exposed to leaf residues of chlorpyrifos, suggesting that fresh pollen availability can reduce the effect of that insecticide.

Laboratory experiments confirmed that GDM is an alternative food for *A. andersoni*, allowing its development and reproduction (Pozzebon & Duso, 2008). Some pesticides can be detrimental to predatory mites in vineyards, but the presence of GDM on plants can play a role in alleviating the effect of pesticides on predatory mites. The patterns observed in a field experiment supported the idea that the effects of a detrimental fungicide (mancozeb) could be mitigated by late season occurrence of GDM. While mancozeb did lead to lower predatory mite densities early in the season compared to other fungicides, the increased GDM incidence late in the season favored colonization of treated plots by *A. andersoni*. Managing GDM could therefore provide a way of promoting predatory mite persistence in vineyards. In late season, GDM infection can occur below economic threshold with no consequences on yield, while potentially promoting predatory mite population resurgence when detrimental pesticides are applied.

The reported studies improved our knowledge on some mechanisms influencing predatory mite population dynamics in perennial crop systems. Habitat management practices that enhance the availability of alternative food sources can increase predatory mite persistence and resilience after pesticide use.

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