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Working Group “Integrated Protection in Viticulture”

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Groupe de Travail “Lutte Intégrée en Viticulture”

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Giuseppe Carlo Lozzia

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The role of the I.O.B.C. in modern viticulture

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Abstract: Here is a brief summary on some of the activities of the O.I.L.B., with particular regard to those concerning the "Integrated Production and Protection in Viticulture" Working Group. The contribution given by the Organisation to the development of the "Integrated Farming" concept and its role in developing and promoting ecologically safe management measures are underlined

The use of know-how on the interaction between grapevine and pests or diseases to improve integrated protection strategies

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Abstract: The current pest (including diseases) control strategies employed in viticulture consider the plant as the growing substrate necessary to the fulfilment of their life cycle. This has led to a linear conception of the relation between quantitative presence of the pest, time and final damage. However, it is plausible and evident that this relation-ship is considerable altered by various host related factors, which in their turn are growth stage and environment influenced. In the current production system, a maximal quality-quantity crop production over a long period is requested. To achieve this object, the plant system of assimilate allocation, based on a ranking of sink priority, that change as the season progresses, is used and manipulated according to the production goals. The grapevine will react to any intervention such as pruning or any other stress modifying its sink priority and consequently the priority of the assimilate allocation. On this basis, the plant must be, therefore, placed at the centre of the vineyard crop system, where cultural practices, diseases and pests can be considered as stress factors. Consequently, the development of control strategies should be based on analysis of the complete crop system and its interactions. Therefore, it must consider: the damage analysis, the analysis of the compensation mechanisms applied from the plant, the analysis of the existing interactions between damage and cultural practices and the analysis of the ecological interactions. These analyses permit to elaborate decision rules to optimise the pest/disease control on ecological, cultural or chemical basis. We present here three cases, *Plasmopara viticola*, *Empoasca vitis* and *Guignardia bidwellii*, of the application of this approach.

Efficacy evaluation of biocontrol agents against downy mildew for copper replacement in organic grapevine production in Europe

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Abstract: Copper is traditionally used in agriculture to control several plant diseases. Copper has a fungicide activity against a broad range of pathogen due to its enzyme denaturant ability. On grapevine it is extensively used in traditional and organic viticulture especially against downy mildew. However the use of copper may have long-term consequences due to its accumulation in the soil, which appears incompatible with organic farming's objective of being sustainable and environmental friendly. Therefore the Annex II of Regulation (EEC) No 2092/91 was modified in 2002 limiting the dosage of copper to 6 kg/ha/year from 2006. The current alternatives to copper allowed in EU organic agriculture, are less effective and inconsistent in controlling downy mildew in particular with high disease pressure. The aim of this research, which is part of a European funded project (REPCO), is to find new alternatives to be used in organic vineyard. New products (two experimental plant derivatives, called EXP1 and EXP2, plant based alcohol extract, lignin derivatives, fungicide based on the lactoperoxidase system, biosurfactant, copper gluconate, *Bacillus subtilis*, potassium salt of fatty acid, chitosane, acylbenzolar-s methyl, Clinoptilolite, Lumbrico Humus, electrolyzed acid water, surfactant) were tested for efficacy against artificially inoculated *Plasmopara viticola* on grapes in greenhouse. The bets products were tested under natural condition in a vineyard (copper gluconate, potassium salt of fatty acid, chitosane, EXP1, clay, plant based alcohol extract, salix extract, biosurfactant, *B. subtilis*, acylbenzolar-s methyl) and compared to traditional copper formulations. EXP1, copper gluconate, salt of fatty acid, plant based alcohol extract show high control capacity of the disease in controlled greenhouse condition. In the vineyard trial, copper gluconate (containing 8% of Cu^{2+}) shows an efficacy comparable to copper hydroxide (containing 35% of Cu^{2+}) however none of the other tested products showed a comparable control. The greenhouse trials reveal the resistance inducer activity of acylbenzolar-s methyl (Bion 50 WG) and vineyard trial confirmed its efficacy.

Copper replacement and copper reduction in organic viticulture by the use of biopesticides and new copper formulations

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Abstract: In organic viticulture copper applications are recently under consideration due to ecotoxicological aspects. Therefore, a cluster project is seeking for sustainable and economically feasible alternatives for copper applications to control *Plasmopara viticola*, one of the most important pathogens in viticulture worldwide. Investigations on disease reduction of grey mould (*Botrytis cinerea*) by selected plant resistance improvers are included in the project as well.

Efficacy of salicylic acid against *Plasmopara viticola*

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Abstract: Copper doses allowed in Organic Farming to control downy mildew are limited. This leads to an increasing interest for new methods of control, especially enhancers of natural defences of the plants. That is why GRAB (Research Institute for Organic Farming) has tested effect of an aqueous extract of *Salix alba* on downy mildew, compared to a solution of synthetic salicylic acid and a non treated control, at several doses and applied 2 or 4 days before inoculation with *Plasmopara*. The disease is then monitored to evaluate effect of the treatments. It appears that *Salix* extract (pure at 0,14 µmol/l or diluted 10 times) is as efficient as the highest dose of synthetic salicylic acid (7 mmol/l). 4 days between elicitation and inoculation give the best result in term of limitation of the disease. *Salix* extract is a promising alternative to copper, with no risk for resistant races to *Plasmopara* to appear. Nevertheless, its action is strictly preventive and *Salix* extract should be applied before contaminating rains, which are impossible to forecast. All the more, this trial has been made under controlled conditions and be repeated on larger plots, under natural conditions.

Disease dynamics and genetic variability of *Plasmopara viticola* in three untreated vineyards

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Abstract: Downy mildew, caused by the diploid oomycete *Plasmopara viticola*, is one of the most significant grapevine diseases. Downy mildew epidemics were observed during three growing periods in untreated vineyards in Germany. Lesions from the vineyard were genetically identified. Genetic determination was based on genome identification by microsatellite PCR amplification.

Effects of plant extracts on downy mildew of vine

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(No abstract)

Development of decision rules for pest vineyard management

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Abstract: Vine crop protection is nowadays based upon systematic sprays. A new approach is thus necessary for the generalisation of Integrated Pest Management (IPM). This concept must clearly describe decisions to control diseases and insects pests. INRA develops such type of tool called Decision Rules (DR). The IPM strategy takes into account indicators that are activated through an “if-then” decision making process. Linking the value of the successive indicators along a DR-chain leads to decisions: spray, do not spray, or wait. Based on this scheme, we have developed DR for the management of diseases and for insect pests management. Available knowledge on arthropods pests was condensed into a single DR, our aim being:

- (1) to maintain a good functional quality of foliage throughout the season (leafhoppers and mites)
- (2) to produce grapes that are suitable to the high quality standards of AOCs (grape moths).

Outbreaks of Flavescence Dorée, against which regulations impose compulsory pesticide applications leads to two scenarios: with or without this disease. Monitoring of insect population is then used as a basis for management, based on injury thresholds. Management actions are firstly directed against the grape berry moths and secondarily against leafhoppers. The strategy is preventive, at the insect populations levels, and makes use of diverse insect traps and insect counts. Side-effects on beneficial organisms (such as predatory mites) are included in the criteria for selection of chemical compounds. Mating disruption technology is also used, and applied if the level of grape berry moths population exceeds a value which would require sprays at each generation.

The DR has been tested since 2003 on four plots in AOCs production, 0.25 to 0.5 hectare in size, planted with merlot or cabernet-sauvignon. Over three years of experiment, situation changed with various levels of populations of insect pests and with or without compulsory FD control. *Lobesia botrana* Den. & Schiff. was well controlled by use of *Bacillus thuringiensis* or Insect Growth Regulators, without mating disruption justification. No control of *Empoasca vitis* Goethe larvae was necessary. The *Typhlodromus pyri* Scheuten population level was high and no Tetranychidae was observed during the experiment.

This type of pest management based on the hierarchical importance of species, injury thresholds and side effects on beneficial organisms leads to use biological or no chemical control options. This point favours beneficials, including Phytoseiidae. In the future, transfer to growers needs to explore new decision models to simplify the system. A new approach using fuzzy logic is in progress.

(Abstract only)

Potential use of biocontrol agents to prevent *Plasmopara viticola* oospore germination

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Abstract: The Oomycete *Plasmopara viticola* (Berk. et Curt.) Berl. et de Toni is the causal agent of grapevine downy mildew and represents one of the most destructive pathogens in viticulture. A recent study of European downy mildew populations allowed the estimation of quantitative contribution of the primary infections to an epidemic. European populations were shown to be highly genetically variable and constituted by a large number of genotypes. Genetic heterogeneity indicated a high occurrence of oospores as a consequence of massive sexual reproduction. Subsequent periods of primary infection outbreaks, with different extents, were shown to occur from May to August, depending on the epidemic. For this reason biocontrol agents targeted to the reduction of the oosporic pool represent a possible control measure to reduce disease severity.

Selected microorganisms (110) isolated from different substrate in untreated or abandoned vineyards were tested during four years for their ability to reduce the primary inoculum of *P. viticola*. Infected leaves with high presence of oospores were treated with the microorganisms and left overwintering in natural conditions in the vineyard. The inhibition of oospore germination was tested in a controlled condition leaf disk test during spring. In the first year, 33 microorganisms (24 fungi, six bacteria and three yeasts) were evaluated. In the second year, 40 new microorganisms (21 fungi, 14 bacteria and five yeasts), plus 14 microorganisms (12 fungi, a bacterium and a yeast), which resulted effective in inhibiting oospore germination in the previous year, were tested. In the third year, the efficacy of 20 potential BCAs, which gave positive results in the previous two years and 20 new potential ones, was analysed. During the last year, nine already tested and 16 new microorganisms were evaluated.

In the first year, 14 (42.4 %) of the 33 tested organisms were effective in preventing germination of overwintered oospores in natural conditions, but only two of them were effective also in the following year. In the second year three (7.5 %) out of 40 new tested organisms were effective. During the third, year six out of 39 organisms were effective in the inhibition of oospore germination. In the fourth year 12 fungi and a bacterium out of 15 new tested organisms inhibit oospore germination. During the same year five BCAs assayed in previous years confirm their efficacy. Totally 27 fungi, three bacteria and three yeasts resulted effective in inhibiting oospore germination. Two fungi were effective in three years; five bacteria and a yeast were effective in two years. This raises again the issue of the influence of environmental conditions in the activity of BCAs in natural conditions. Before a practical application in vineyards the best environmental conditions and application methods of these potential BCAs must be identified.

Genetic diversity of *Armillaria mellea* in the Rotaliana Plane

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Abstract: In Trentino Region, in the North East of Italy, *Armillaria mellea* root rot is a severe and increasing problem on grapevine. In the vineyards the disease appears in patches of different dimensions. Infected areas and the incidence of the disease in a valley, called Rotaliana Plane, were evaluated. Genetic identification of individual mycelium (genet), and Geographic Information System (GIS) approach, were used to create a genets map of the area with the 56 dikaryotic isolates of *A. mellea* collected in 40 representative different locations (patches) of the valley. The isolates were classified in somatic incompatibility groups and RAPD polymorphisms determined with nine primer pairs. Somatic incompatibility analysis revealed the presence in the region of at least 23 different genets of *A. mellea*. Each genotype includes either one or many isolates. Three main groups (which include respectively 13, 8 and 6 isolates) were found. The RAPD analysis suggests that the population in the Rotaliana Plane was highly genetically polymorphic: 55 genotypes were identified among the 56 collected. The phylogenetic relationship showed a distinction among three major groups (clades 1-3), where the clade 2 included the majority of the individuals and was subdivided in three clusters (2.1, 2.2 and 2.3). The vegetative incompatibility group 5 was exclusively present in clade 1, the group 3 was exclusivity of the subclade 2.1 while group 8 was mainly correlated with group 2.2. Since *A. mellea* spores infection of grapevine has to be considered irrelevant we hypothesise that the area was infected long time ago by *A. mellea* starting from several locations (forest and fruit trees, bushes, etc.).

Efficacy evaluation of integrated strategies for powdery and downy mildew control in organic viticulture

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Abstract: In organic viticulture the protection against powdery and downy mildew is mostly based on toxic compounds like sulphur and copper. Control strategies that can help to reduce the use of these fungicides consider the use of biocontrol agents. Therefore we tested the integration of the commercial preparation of the mycoparasitic fungus *Ampelomyces quisqualis* against powdery mildew and sulphuric acidic clay minerals plus plant extract (Mycosin) against downy mildew in the common copper and sulphur based strategies. The study was carried out in two different locations (Southern and Northern Italy). During the growing season 2005, weather conditions were more favourable to *Uncinula necator* than to *Plasmopara viticola* in both the locations. In consequence, downy mildew infections were few and severity low and the tested strategies did not differ from each other. Regarding powdery mildew, similar results were obtained in the two different locations: the strategy using AQ10 treatments early in the season showed lower efficacy against powdery mildew especially on grapes, compared to the reference (sulphur). The reference strategy based on a new formulation of sulphur (Heliosoufre) gave the best results.

Efficacy evaluation and phytotoxicity assessment of copper peptidate on seven grapevine varieties and identification of the potential factors that induced copper damages on leaves

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Abstract: In organic viticulture downy mildew control is based mainly on copper. The European Commission has fixed a ceiling in copper used as fungicide. Copper peptidate can allow a four times reduction of the copper dosage, maintaining a good control of the diseases, but several studies have shown that the risk of phytotoxicity in the field can be high. The factors that increase the risk of phytotoxicity in the field are still unclear. The aim of this work was to evaluate the efficacy and the phytotoxicity of copper peptidate on seven different grape varieties (Victoria, Italia, Red Globe, Primitivo, Negroamaro, Uva di Troia and Chardonnay) and to identify the potential factors that are involved in increasing the risk of copper damages on the leaves. Copper peptidate (Naturam 5) was compared to a traditional formulation of copper hydroxide (Kocide 2000) in a Mediterranean environment (Bari, Italy). In greenhouse controlled conditions the influence of temperature (5, 15, 25, 35° C), leaf wetness and number of treatments (1, 3, 5, 7) on copper peptidate phytotoxicity was evaluated. Copper peptidate is more phytotoxic than copper hydroxide, even if the sensibility varies among the tested varieties. In particular the grapevine varieties, Primitivo and Negroamaro, were the most tolerant and the table grape variety Italia was the most susceptible. In controlled conditions, low temperatures, but also high ones, if associated with leaf wetness, induced phytotoxic effects. Phytotoxicity increased with the increasing of the number of treatments. Dry conditions induced only a slight phytotoxicity. When long leaf wetness periods combined with low or very high temperatures are frequent and on susceptible varieties it is necessary to be very careful in using copper compounds, as copper peptidate. Copper peptidate could be useful to reduce the annual copper amount per hectare, but it must be used only on grapevine varieties that are less susceptible to copper phytotoxicity, avoiding extremely cold and warm periods of the season and repeated applications. The use of copper peptidate along all the season as an alternative to traditional copper compounds in organic viticulture is not possible until a less phytotoxic formulation of copper peptidate will be available on the market.

Efficacy evaluation and phytotoxicity assessment of traditional and new copper compounds used in copper reduction strategies in organic viticulture in northern and southern Italy environments

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Abstract: Downy mildew is one of the most destructive diseases of grapevine, particularly in warm and wet climate. In organic viticulture *Plasmopara viticola* control is based almost exclusively on copper. Copper is considered highly toxic for the environment especially because it has long residual activity against a wide range of micro and macro organisms in the soil. This is why in Europe there were adopted rules for its reduction. The expected restrictions on copper use in organic agriculture in the European Union have stimulated the research of alternatives and the optimisation of allowed copper quantity use. Several researches have been done to compare efficacy against downy mildew of different copper compounds and low dosages based strategies have been developed, but only copper chelated to amino acids and peptides seems to be promising in reducing copper dosage, by enhancing copper penetration into the pathogen cell. On the other hand the fast and enhanced penetration through the cell membrane of copper chelated to peptides also increases the risk of copper toxicity to plant cells. The aims of this research were the evaluation of copper peptidate (Naturam 5) efficacy against *P. viticola* and the comparison of phytotoxicity on grapevine between copper peptidate and traditional copper compounds under vineyard conditions in two different climates (northern and southern Italy). Cabernet Sauvignon and Uva di Troia were the used varieties respectively in Rovereto (northern Italy) and Andria (southern Italy). Copper peptidate, applied at a concentration of 0.25 g Cu²⁺/l gave a good control of the disease, comparable to copper hydroxide used at 0.5 g Cu²⁺/l, but the phytotoxicity on plants was high in both the locations. In 2005 in both the experimental vineyards rains and wet periods were frequent and this could be one of the factors involved in the copper toxicity to grapevine.

Powdery mildew on grapevine: the date of primary contamination affects disease development on leaves and damage on grape

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Abstract: The temporal evolution of the disease on leaves and damage on grape was monitored at different scales in order to study the effect of early primary contamination on epidemic development and its relationship with damage on grape. At a vine scale, on vines artificially inoculated with 12 days delay, the disease evolution on leaves was delayed. At flowering, early contaminated vines were 60% more diseased than late contaminated vines. On bunches, the progression and final disease were significantly different with, at veraison, an average severity of 99% for early contaminated vines versus 62% for late contaminated ones and 29% for uncontaminated ones. A significant difference for the Incidence-Severity relationship at the leaf scale was quantified indicating the difference in symptoms according to the date of contamination. At the plot scale (330 vines), we compared the maps of frequency of diseased leaves per vine at different scoring dates with the maps of frequency of bunches with a given level of damage. Epidemics initiated earlier were characterised by higher severity for a given level of frequency of diseased leaves (at the vine scale) and higher final diseased leaves frequency (at the plot scale). Early difference in the number of diseased leaves led to significant differences in the number of highly damaged clusters (> 25%) and in the average clusters severity. The analysis of disease maps demonstrated the spatial relationship between the frequency of diseased leaves at flowering and the frequency of severe damaged bunches.

Controlling powdery mildew near harvest

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Abstract: Two applications of a highly refined paraffinic oil six weeks before harvest controlled established powdery mildew infections on bunches and almost completely eradicated the disease from leaves. Rates of at least 1L/100L of oil were most effective when applied in spray volumes of around 1000L per Ha. The efficacy of the oil spray depended on the level of spray coverage.

Effects of fungicides on the germination of *Ampelomyces quisqualis* AQ10, a biological antagonist of the powdery mildew of the grapevine

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Abstract: AQ10 is a biological control agent based on the mycoparasite *Ampelomyces quisqualis* used to control powdery mildews in grapes and other crops. In order to optimize the use of AQ10 in the IP strategy for powdery mildew control, the effect of 17 organic and inorganic fungicides on the growth of *A. quisqualis* was tested *in vitro*. Sulfur, strobilurines, mancozeb and SBI inhibit *Ampelomyces*, whereas copper and iprodione do not show an effect. Because of these inhibiting effects of widely used fungicides and the need for precipitation for the growth of *Ampelomyces*, the application of AQ10 is recommended only for the fall.

Evaluation of new control agents against grapevine powdery mildew under greenhouse conditions

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Abstract: Pathogen resistance to fungicides, concerns regarding pesticide residues and revocation of some widely used fungicides have increased research efforts for developing biocontrol agents of foliar fungal pathogens. Non chemical fungicides that are available for the control of grapevine powdery mildew are few and moderately effective. Moreover, no powdery mildew resistant varieties of grapevine are known. This is why there is an urgent need to develop new alternatives to chemical pesticides against this pathogen. The effect of some bacteria, yeasts, a plant extract, an enzyme based fungicide and electrolyzed acid water on grapevine powdery mildew was evaluated under controlled greenhouse conditions. The results showed that some of the tested yeasts and bacteria were effective in powdery mildew control, being able to significantly reduce the symptoms. Among natural products, only the enzyme based fungicide, when sprayed three or seven days after powdery mildew inoculation, showed a good efficacy. Better results were obtained using an experimental wetting agent (X22) with daily sprays during one week after inoculation.

Colonization of grapevine Powdery mildew cleistothecia by the mycoparasite *Ampelomyces quisqualis* in Trentino, Italy

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Abstract: the role of *Uncinula necator* cleistothecia in the epidemiology of grape powdery mildew in Trentino province (northern Italy) was studied. The presence of *Ampelomyces quisqualis* as mycoparasite in the vineyards was evaluated and wild strains were isolated to eventually develop a local strain for disease control. The results showed high presence of *U. necator* in 2004 (31 to 45 cleistothecia per square centimetre of leaf) and high percentage of potential mature cleistothecia (68 to 81%). The bark is an important overwintering site for the cleistothecia that can cause the new infections in the following season. Among the sampled vineyards there was a high variation in the presence of *U. necator* mature cleistothecia on the bark (2 to 41 cleistothecia per disk of filter paper of 12 cm diameter) and a low presence of *Ampelomyces* spp. conidia in them. *Ampelomyces* spp. conidia were only found in cleistothecia collected in an untreated vineyard. From this material an *Ampelomyces* spp. strain was isolated. The efficacy trials in greenhouse and vineyards of *A. quisqualis* based biofungicide (AQ10) showed no statistically significant differences between treated (AQ10) and untreated plants.

Observations of Black Dead Arm symptoms in Bordeaux vineyards: evolution of foliar symptoms, localisation of longitudinal necroses, questions, hypotheses

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(see addendum)

Blackrot on the hybrid *vitis* cultivar Isabella

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Abstract: Black rot caused by *Guignardia bidwelli* is present since almost 20 years in South Switzerland without causing damages in the vine producing Merlot vineyards. However since a few years in the old traditional vineyards of the hybrid cultivar Isabella damages can be high. Wrong diagnostic and missing know-how of the non professional winegrowers is the main cause. We describe here how the disease can be controlled efficiently under these conditions with as few as two correctly timed fungicide treatments.

First detection of *Eutypa lata* (Pers.:Fr.) Tul. with PCR directly out of grapevine trunks in Germany

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Abstract: Since 1999, symptoms of *Eutypa lata*, discovered in a Franconian vineyard, were observed year by year for every rootstock. Whereas in 1999 only 16% of all vines showed typical symptoms of eutypa dieback in 2003 the incidence of symptomatic vines increased to 58%. Almost one third of all vines (29%) were dead or already removed. Another 11% developed poorly and 18% showed typical signs of the eutypa dieback. When cutting the grapevine trunk vertically the degree of typical dark regions inside of the trunk was equivalent to the severity of external symptoms.

Small pieces of wood were taken from the dark region of infected grapevine trunks for molecular analysis. After isolating DNA by means of a purification kit and an extensive denaturing step, species-specific ITS-primers were added. DNA of *E. lata* that was possibly present in the eluate was amplified by means of polymerase-chain-reaction (PCR). Following PCR, fragments of DNA were separated in an agarose gel electrophoresis. A prominent band at 382 bp was typical for positive detection of *E. lata*. The DNA sequence analysis showed a match of 99 % between the tested DNA and the reference-sequence of *E. lata* described in literature. The same conformity was found between the tested grapevine DNA and that one of an *E. lata* control isolate.

Do chestnut stakes promote grapevine root rot caused by *Armillaria mellea*?

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Abstract: Chestnut (*Castanea sativa*) stakes were traditionally used in Southern Switzerland as a support for grapevines in vineyards. Despite their high durability and aesthetic role in the vineyard-based landscape, chestnut stakes are being replaced with iron or cement stakes because suspected to promote *Armillaria* root rot (*Armillaria mellea*). In this experimental study we investigated the ability of *A. mellea* to colonise chestnut stakes, false acacia (*Robinia pseudoacacia*) stakes, and Norway spruce (*Picea abies*) stakes, with and without copper treatment. After three years, 30% of the chestnut, 17.5% of the false acacia, and 2.5% of the treated Norway spruce stakes were colonised by *A. mellea*. Most of the colonised stakes were located near symptomatic or newly planted grapevines that replaced plants killed by *A. mellea*. Our results show that chestnut and false acacia stakes in vineyards affected by *Armillaria* root rot can effectively be colonised by *A. mellea*. However, their role in the spread of the disease is still unclear.

Control of esca of grapevine in Italy: what we can learn from past experience in view of an integrated management approach

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Abstract: The control of esca disease is at present still based only on cultural methods which attempt to reduce losses and limit the spread of the disease. Although several research groups, in Italy and elsewhere, are testing different approaches in controlling esca and related diseases, since sodium arsenite was banned, no efficient control method has been available. Ongoing progress in the aetiology and epidemiology of the disease has changed the outlook, with attention being directed towards prevention of infection in the nursery through the use of *Phaeoconiella* and *Phaeoacremonium*. To this end studies in the nursery are now focusing on the efficiency of the hot water treatment technique (HWT) and also on the beneficial effects of applications of *Trichoderma* thanks to the wide number of purposes for which it can be used. In the vineyard, the use of *Trichoderma* for the protection of pruning wounds, not least because of its persistence after application, and of Fosetyl Al as a plant defence response enhancer, are being evaluated on the basis of their success in preventing or limiting the spread of esca. On the whole it seems unlikely that esca can be countered effectively without an integrated, organic approach, starting from the selection of propagation material and taking due account of vineyard age and disease incidence.

Investigations on the control of ESCA disease by means of stem injection

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Abstract: Several plant protection agents were examined *in vitro* for their efficacy against *Fomitiporia* sp., *Phaeoacremonium* sp. and *Phaeomoniella* sp.. *In vitro* effective fungicides and fungicide mixtures were tested via StemJect[®] injection in the vineyard. The treatments were conducted in the dormancy period between January and March. 8,3 % of the vines of the treated vineyard plot showed typical ESCA symptoms. Disease incidence was recorded in the following growing periods (2003 to 2005). These field trials gave no definite effect of fungicide application via StemJect[®] to control ESCA disease. Both, untreated control and water control, showed effects, which were comparable to fungicide treatments. Substantially larger differences have been found between the different years of observation.

Fungi associated with young vine decline in Portugal: results of nine years surveys

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Abstract: In Portugal, surveys of young vine decline have been carried out in some of the most important wine producing regions. Obtained results showed that, at the base of the rootstock, *Cylindrocarpon* spp. and *Phaeomoniella chlamydospora* were dominant but other wood decay fungi like *Sphaeropsis* sp., *Pestalotiopsis* sp., *Phomopsis* sp., *Truncatella* sp. and *Botryosphaeria* sp. were also isolated in percentages which vary from 24% to 34%. At the graft union, both *Cylindrocarpon* spp. and *Pa. chlamydospora* percentages decreased and, conversely, the percentage of the other wood decay fungi increased. Finally, from the above tissues of the graft union, the wood decay fungi were prevailing but *Cylindrocarpon* spp. were also present and *Pa. chlamydospora* was not detected. *Cylindrocarpon* isolates were identified as *C. destructans* and *C. macrodidymum*. Pathogenicity tests carried out with selected *Cylindrocarpon* spp. (*C. destructans* and *C. macrodidymum*) and *Pa. chlamydospora* isolates on grapevine potted plants revealed their ability to induce disease and plant mortality. In conclusion, decline and dead of young vines should be considered as the final result of a complex interaction between pathogens and the host plant, since no relevant stress factors could be identified.

Esca disease: spore trapping, symptom evolution and incidence in Trentino Region, Italy

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Abstract: Esca is a complex disease of grapevine, present in almost all grape cultivation areas in Italy. In spite of recent and intensive researches in different parts of the world, several aspects of disease epidemiology are still unclear. In 2004 an Italian national project, coordinated by University of Firenze, has been funded with the aim of finding new tools for disease diagnosis and control. This project comprises 11 Regions and the Autonomous Province of Trento. We report here the first-year monitoring results obtained in Trentino Region, Northern Italy. The area was selected within the project because it could represent the first stage of the epidemic and help in understanding its development. In fact, even if the incidence of esca disease can be considered very low, symptomatic plants were increasingly observed in the last years, in old vineyards, but also in young plantations. In 2005 two vineyards near S. Michele all'Adige, were weekly monitored to assess esca incidence and the spatial distribution of infected vines. In the same two vineyards a spore trap and glass slides with sticky surface were used to sample airborne spores throughout the entire year. No spores of the three main fungi were found. Esca incidence in the whole Province of Trento was assessed in a total of 100 randomly chosen vineyards in five regions with different cultivar and age. Esca incidence is still relatively low and ranges from 0.5% to a high of 2.5% depending on the region.

Grape cultivar affects larval and female fitness of the European grapevine moth, *Lobesia botrana* (Lepidoptera: Tortricidae)

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Abstract: The reproductive performance of phytophagous capital breeder insects is strongly influenced by the food ingested by the juvenile stages and females may adapt their reproductive output in accordance with the diet they ingested as larvae. We studied this for the grapevine pest *Lobesia botrana* (Denis and Schiffermüller), which larvae feed on fruits or berries of different plant species. Larvae were offered one of eight grape varieties as food and their performance and adult reproductive output was measured. The individual performance of *L. botrana* was determined by measuring a suite of life history traits, from egg hatching to adult death. Larval development time, adult mating success, female fecundity, egg size and egg fertility were all significantly influenced by the variety of grape on which the larvae were reared. Interestingly, adult females adapted their reproductive output depending on which variety they had fed and this was correlated with their rate of development. Compared to slow-developing females, females that developed faster laid more eggs, but this was balanced by lower egg fertility. It further presents evidence for diet dependent plasticity in reproductive output in this grape pest. The average reproductive output was highest for females that were reared on cultivars on which larval development was intermediate. Therefore, variation in the performance of *L. botrana* is the result of differential suitability of larval food. This could influence the number of efficient reproductive adults on the next generation, and thus the cultivar on which larvae develops should be considered as a factor affecting the amount of European grapevine moth damages.

Parasitoids of *Lobesia botrana* (Den. & Schiff.) in Tuscany

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Abstract: An assessment of the parasitoid insects associated with *Lobesia botrana* (Den. & Schiff.) in Tuscany was undertaken in the nineties and pursued up to now, with the aim of defining the taxonomy of the involved species and the role played by each parasitoid in the host natural control. Parasitoids were obtained from eggs, larvae and pupae of *L. botrana*, collected in different localities of the region, throughout the three generations. *Trichogramma evanescens* Westwood (Trichogrammatidae) is the only egg-parasitoid recorded in our study. Its activity is particularly evident in the first generation where it can reach an egg parasitization rate of about 25%. As regards larval parasitoids, we collected *Campoplex capitator* Aubert (Ichneumonidae), *Ascogaster quadridentata* Wesmael (Braconidae), *Elachertus affinis* (Masi) (Eulophidae), *Hockeria* sp. (Chalcididae), *Goniozus gallicola* (Kieffer) (Bethyridae) and *Phytomyptera nigrina* (Meigen) (Tachinidae). *C. capitator* is widespread and effective, often reaching a parasitization rate of about 10%. This value is sometimes reached also by *P. nigrina* in some areas of the Arno Valley. In Chianti, young larvae of the second generation of *L. botrana* suffer the attack of *E. affinis* with parasitization rates up to 30%. The frequency of larval parasitoids, fairly high in the first and second generation, usually decreased noticeably in the third generation. Larvo-pupal or pupal parasitoids, which are mainly associated with the over-wintering generation, include the pteromalid *Dibrachys affinis* Masi and several ichneumonids, i.e. *Dicaelotus inflexus* Thomson, *Pimpla spuria* Gravenhorst, *Ischnus alternator* (Gravenhorst), *Theroscopus hemipterus* Fabricius, *Itoplectis maculator* (Fabricius), *Phytodiaetus* sp., *Triclistus* sp. and *Exochus* sp. The most efficient ones are *P. spuria* and *D. affinis* that show, every year, a parasitization rate of about 10%. Our study shows that in Central Italy the parasitoids and the role played by each species in the natural control of *L. botrana* are greatly variable in space and time.

Mating disruption for vine moths control in Tuscany: results of 2003 and 2004

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Abstract: Pheromone mating disruption (MD) with Isonet-L plus dispensers has been applied in Tuscany (Italy) in the last four years on about 150 hectares. Results of 2003 and 2004 are reported in this survey, with particular reference to the trials carried out at Castello di Ama Winery (Gaiole in Chianti, Siena). In both years, MD covered 40 hectares in three vineyards named Bellavista, Casuccia and S. Lorenzo. Some similar surrounding vineyards were considered as untreated controls. Pheromone traps (Traptest Isagro) were used to monitor the adult population, whereas larval density and fruit damage were evaluated by field and laboratory observations. In the pheromone treated vineyards, male captures were absent or highly limited. In 2003, the first and second generation population levels were similar to those observed in 2001 and 2002, whereas the high temperatures which occurred in July and August dramatically reduced third generation *L. botrana*, affecting the first generation of the following year. In 2004, the infestation rate recorded in the MD areas showed very low values in comparison with the control, particularly with regard to the second and third generation. Similar results were obtained in the same years in Fontodi Estate (Panzano in Chianti, Florence) where MD has been applied since 2002 on about 45 hectares. The 2004 results for the first time corroborate one method postulate, i.e., a real reduction in the target pest population can be obtained in areas where MD is applied for several consecutive years on wide surfaces.

Mating Disruption in viticulture – Dream or Reality

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Abstract: The use of pheromone Mating Disruption to control key grape pests *L. botrana* and *E. ambiguella* is a solid reality in areas where the technology transfer was supported with suitable extension service combined to reliable and affordable dispenser system. Fifteen years of direct experience and continuous field monitoring shows that the basics of this method are often disregarded, particularly in viticulture. Furthermore, the need to focus on field rather than on material, having its relevant quality and reliability confirmed, is still far to be achieved.

Mating disruption in viticulture will probably remain a dream for those who will not change their approach and pay more care to check the field and to understand the affecting local environmental factors.

Industry responsibilities are to supply guaranteed materials manufactured according to high quality standards and to provide detailed and clear information to support researchers and technicians in the long process required for putting this method into wide practice.

(Only abstract)

Mating disruption for the control of European grapevine moth *Lobesia botrana* (Den. Et Schiff.) in a plastic film greenhouse table grape vineyard

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Abstract: Results of *Lobesia botrana* (Den. & Schiff.) mating disruption trials on table grape under greenhouse condition by means of Isonet L dispensers (Shin-Etsu Chemical Co. Ltd.) are reported.

The trial was located in a table grape vineyard in Central Italy (Latium region) and the dispensers were applied before the first moth trap catch. The grapevine cultivar was Superior seedless trained with Geneva Double Curtain (GDC) system. The recommended application rate on head training system is 750 dispenser/ha while on espalier training system is 500 dispensers/ha.

The aim of the trial was to verify the effectiveness of a 500 dispensers/ha application rate in comparison with the farm standard control strategy.

Influence of grapevine presence in different ecosystems on the density of *Scaphoideus titanus* Ball (Homoptera: Cicadellidae)

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Abstract: The presence of the leafhopper vector *Scaphoideus titanus* Ball was studied during 2004 in northwestern Italy, in vineyards with different management: cultivated vineyards with pest management (type 1), cultivated vineyard without pest management (type 2), neglected vineyards (type 3), and brushwood with American grapevine rootstocks (type 4). Yellow sticky traps were used to capture and count leafhoppers in different types of sites. Captures were significantly higher in type 2 (131.6 adults per site) and 3 (72.4) than in type 1 (10.3) and 4 (20.9). Differences between sites of the same type were also found. It is likely that brushwood, where grapevine is quite sparse, is not a suitable environment for *S. titanus* to develop, whereas non-treated and neglected vineyards could become a serious source of infestation.

Prediction of the flight of *Hyalesthes obsoletus*, vector of stolbur phytoplasma, using temperature sums

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Abstract: A method to predict the flight of the planthopper *Hyalesthes obsoletus*, the vector of stolbur-phytoplasma that causes Bois noir disease in grapevine, is presented. The parameters required for the calculation of temperature sums were determined by multiple calculations of temperature sums till the observed dates of the start of flight activity of adult planthoppers in different years and the identification of combinations of starting dates and temperature thresholds for day-degree accumulation that led to a minimum relative standard error. The method can be currently used for populations of *H. obsoletus* that live on bindweed (*Convolvulus arvensis*), only. It needs to be adjusted for populations on nettle (*Urtica dioica*) that show a different flight activity. The procedure described here could be useful for other insects, too, where relevant data of the life cycle cannot be determined in laboratory rearing.

The role of the soil coverage on the occurrence of occasional ampelophagous planthoppers and leafhoppers in vineyards.

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Abstract: A preliminary study on the effect of vineyard soil coverage on the occurrence of not strictly ampelophagous planthoppers and leafhoppers on vine has been undertaken during the biennium 2003-04, in two vineyards of Western Tuscany, one covered by spontaneous grass (Matraia), the other (Riparbella) periodically tilled. In total, 152 species of "auchenorrhyncha" were identified, with 43,6% of taxa shared by both localities. The study of the population structure, from the ecological point of view, showed a substantial similarity between the two agroecosystems. The methods adopted to monitor the vineyards and the biodiversity indexes underlined a significantly higher rate of occurrence on vine-shoots of not strictly ampelophagous species and vectors or potential vectors of phytopathogenic agents at Matraia. It descends that a correct soil management, for instance by choosing appropriate cover crops, could regulate this phenomenon significantly.

Planting dog roses – an efficient method to promote mymarid populations in vineyards?

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Abstract: Dog roses planted along vineyards to substitute natural surrounding habitat structures established and promoted populations of the mymarid *Anagrus atomus*, but not any of the other egg parasitoids of the grape leafhopper *Empoasca vitis*. Only young shoots of the dog roses were used as egg laying sites by cicadellid host species and, consequently, by *A. atomus*. In the third study year, planted dog roses were almost continuously used for reproduction over the growing season and intensely as overwintering sites with a mean parasitisation rate of the cicadellid host eggs of 59%. Once the planted dog roses had reached a height > 2 m they housed as many mymarids as wild dog roses in the adjacent hedge. Furthermore, with an increasing biomass of the planted dog roses, the *A. atomus* population in the wild dog roses doubled each year but not in the other shrub species.

Surveys of the presence of *Hyalesthes obsoletus* Signoret (Rhynchota: Cixiidae) and other hoppers in Lombardy (Northern Italy)

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Abstract: A monitoring programme was carried out in Lombardy from 2000 to study the presence and biology of the leafhoppers involved in phytoplasma transmission to grape. From 2003 also biomolecular analyses were performed to assess the frequency of “positive” insects using PCR. Special attention was given to *Hyalesthes obsoletus* Signoret (Rhynchota: Cixiidae), a leafhopper whose presence in vineyards is worrying because it is well known from literature that this insect can transmit to the grape the “bois noir”, a phytoplasma disease whose symptoms are very similar to those produced by the “golden flavescence”. During the surveys this species was only occasionally collected directly from grape, but it was quite abundant on weeds, mainly *Urtica* sp. and in a lesser extent, *Convolvulus arvensis* and *Calystegia* sp.. The percentage of “positive” specimens was quite different in the various localities but on the average the percentage of positive ones was rather low. According to these results, *H. obsoletus* does not seem to play a major role in transmitting the “bois noir” in Lombardy, as the insect can be found only occasionally on grape, and moreover because the percentage of “positive” specimens, at least in some areas, is very low.

Preliminary observations on the role of botanical diversity on the presence of egg parasitoids of grape leafhoppers in Northern Italy

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Abstract: The botanical diversity is a key factor in the vineyard agro-ecosystem. In northern Italy research has been carried out to verify if different soil management techniques, by increasing the botanical diversity, could raise the presence of egg parasitoids of the grape leafhoppers. To this aim the presence of leafhoppers, both of the vine and of species that live in the grass, and their parasitoids was monitored in five lots. The density of the grape leafhoppers was very low and similar among all the vineyards. The cultural techniques have influenced the botanical diversity of weeds and the complexity of the leafhopper community which has settled there but it did not register a higher number of beneficial insects. Also the presence of the bramble in the surrounding of the vineyards has not always had beneficial effects. This preliminary data seems to indicate that an increase in the botanical diversity in itself is not enough to guarantee a suitable presence of egg parasitoids.

Potential leafhopper and planthopper vectors of phytoplasmas in wine vineyards of the Marche region (Central Italy)

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Abstract: A survey was carried out to determine the occurrence and distribution of leafhopper and planthopper vectors, or suspected (“potential”) vectors, of phytoplasmas in the most important viticultural areas of the Marche region of Italy. The investigations also determined the herbaceous plant hosts of the planthopper *Hyalesthes obsoletus*. Many species of Auchenorrhyncha were identified, some of which are known to be vectors, or potential vectors, of phytoplasmas. *Scaphoideus titanus* was not detected in this region. *H. obsoletus* larval instars were detected only on stinging nettle roots. From June to August, the cixiid adults were found on stinging nettle and field bindweed, rather than on grapevines.

Distribution and sampling methods of soft-scale-insects in vineyards

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Abstract: In southwest German vineyards scale insect studies were carried out between the year 1998 and 2005 on the species *Parthenolecanium corni* and *P. persicae*. Their phenology was studied and described. The distribution within a plant as well as the distribution within whole vineyards was found to be inhomogeneous and clumped. The negative binomial distribution describes *Parthenolecanium* species repartition within a vineyard. For *P. corni* a common k-Value (kc) is probable while for *P. persicae* it's not. Usually clumping is so extreme that random sampling doesn't make sense as the the number of grapevines to be sampled is often higher than the number of grapevines available within a vineyard. Density and population dynamic measurement in plant protection experiences can be achieved by the preevaluation of distinct plants. Parameter free ranking statistics may be usefull to apply, as the number of scale insects between two plants can change within three orders of magnitude.

Utilisation of the sexual pheromones of *Planococcus ficus* and *Planococcus citri* in vineyards

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Abstract: In 2005, observations were conducted in a vineyard of Northern Sardinia, heavily infested by populations of *Planococcus citri* and *P. ficus*. Males of these species were sampled using traps activated with the *P. ficus* pheromone at two concentrations (0.25 mg and 1 mg) and with that of *P. citri* (0.25 mg). Male of *P. ficus* were first caught during the last ten days of May and reached maximum density values in July and September. Captures were always higher in traps activated with 0.25 mg of pheromone (860.8 adults/trap from May to September) than in those activated with 1 mg of pheromone (429.1 adults/trap from May to September). Males of *P. citri* were captured in the same period of *P. ficus* captures, but at lower densities with a total of 232.1 adults/trap from May to September. Our results show that pheromone traps discriminate the mealybug species and *P. ficus* is more abundant than *P. citri* in the vineyard agroecosystem.

Effects of cover crop management on grape pests in a Mediterranean environment

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Abstract: A study was conducted in a vineyard of Southern Sardinia from 2002 to 2004 to determine the effect of soil management on grape pest populations. Observations were carried out in two plots of 0.5 ha each of the white berry cultivar “Malvasia”: one with soil cultivation and weed removal, and the other with natural ground cover since 1995. In that environment, during summer, resident vegetation dries up and does not compete with the grapevine for water and nutrients. The results showed that leafhoppers and thrips populations were not influenced by the ground cover. The attacks of European grapevine moth (*Lobesia botrana*) were always higher in the tilled plot, which showed a percentage of damaged bunches at harvest of 12%, 42% and 14% in 2002, 2003 and 2004, respectively. In contrast, these values were of 2%, 13% and 8%, respectively, in the cover crop plot. These differences could be due to the fact that grapevines of the cover crop plot had less vigorous sprouts that produced smaller and less clustered bunches, less preferred by the moth. On the contrary, the infestation of Vine mealybug (*Planococcus ficus*) on bunches of the cover crop was about twice as high as it was in the tilled plot (28% vs. 12% of bunches damaged by colonies in 2004, respectively). This was probably caused by a more abundant population of ant species, which protect the mealybugs from their natural enemies, in the ground cover plot than in the tilled one. In particular, the prevalent ant species found in the cover crop plot were *Tapinoma simrothi* and *Tetramorium* spp., while *Lasius niger* was the most abundant one in the tilled plot.

Augmentative releases of beneficials in vineyards: factors affecting predatory mite (Acari: Phytoseiidae) persistence in the long-term period

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Abstract: During the last years, phytoseiid mites were widely released on grapes to control phytophagous mites in North-eastern Italy. For a successful conservation biological control strategy the improvement of conditions that allow predatory mite persistence is required. For a better understanding of factors involved in beneficial mite settlement, field observations in commercial vineyards were carried out during 15 years after the release of a *Typhlodromus pyri* strain, suspected to be resistant to organophosphates. The predatory mite colonized the vineyard and competed successfully with other species. The role of alternative food (pollen or downy mildew) availability, leaf morphology and selective pesticides for the predatory mite persistence is discussed.

Faunistic and ecological aspects of spiders (Araneae) on vineyards ¹

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Abstract: In 2002 a research was carried out to test the population density of spiders (Araneae) on 4 different grapevine cultivars in 1 standard “tendone” and two screen-house vineyards sited in the same experimental farm near Bari. Samples were collected during the year once a week by beating 10 vine-shoots per cultivar. Furthermore 10 vine inflorescences or clusters were also examined from blooming until vintage once a week. 3065 spider specimens were collected from the three vineyards: 2018 from the standard “tendone” and 957 from both screen-houses. Thirteen spider families were altogether recognized: Clubionidae, Araneidae and Salticidae were the most widely represented with 914, 748 and 694 specimens, respectively. The identified genera were 35 with the dominance of *Cheiracanthium*, *Neoscona* and *Icius* with 834, 743 and 620 specimens respectively. The most frequently collected species were *Icius hamatus* (C.L. Koch), *Cheiracanthium mildei* L. Koch and *Meioneta rurestris* (C.L. Koch) with 202, 44 and 10 mature specimens, respectively.

The population trend of the whole spider community showed a peak of adults at the beginning of May, while spiderlings were plentiful since July until December. The population density of each dominant family and genus during the year was also described. Eleven taxa of spiders were found to overwinter in the vineyard. By comparing standard “tendone” and screen-house vineyards, the former showed a higher population density, particularly during the second half of the year, mainly due to the increment of Araneidae spiderlings. Only 10.5% of all collected spiders were found on vine clusters, with no more than a single spider per cluster: hunting spiders mainly represented this percentage.

Trapping *Lobesia botrana* females with apple juice: a valuable tool to predict oviposition ?

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Abstract: We evaluate through a cooperative work between INRA (Bordeaux), Plant protection service (Cognac), ‘Chambre d’Agriculture’ (Saint Jean d’Angely) and the vinegrowers cooperative of the Ré Island, the efficiency of trapping *Lobesia botrana* females. The objective is to gain accuracy in the prediction of the oviposition dates and the level of damages. Our results indicate that food traps containing apple juice are efficient in order to trap adults *L. botrana*. We could describe quite efficiently the dynamics of the 2nd flight and first captures of females which occurred in the 2 vineyards surveyed few days before the first eggs observed. Food traps may represent a valuable tool in order to predict the onset of oviposition occurrence. The validity of this method is studied and discussed.

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Observations of Black Dead Arm symptoms in Bordeaux vineyards: evolution of foliar symptoms, localisation of longitudinal necroses, questions, hypotheses

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Abstract: The development of Black Dead Arm (BDA) symptoms was studied in 5 vineyards (cultivars Cabernet Sauvignon and Cabernet Franc) at Médoc and Entre-Deux-Mers, France. Symptoms were recorded from June to mid-July in 2004, and from mid-June to September 2005. Foliar symptoms were very variable according to the vines and showed different levels of deterioration (interveinal drying and/or red wine pigmentation) in their early stage. They developed on one or more shoots, on one or both arms. The most damaged leaves (wilted) drop down rapidly. The less damaged leaves showed after approximately two weeks typical tiger-striped foliar symptoms of esca, indicating an evolution throughout summer from BDA symptoms towards esca symptoms. The bark of the trunk of vines showing recent leaf symptoms was peeled off and exhibited typical longitudinal stripes. At their early development, the altered wood was yellowish-orange and very similar to those of oxidized tissues. They extended to the base and along the affected canes and sometimes down to the graft union. There was no wound (entry) and no necrotic tissue occluding the vessels. Thus, the origin of this peculiar wood symptom seemed questionable and some hypotheses (fungal invasion?, sap disruption?) are discussed in relationship with the architecture of esca-affected vines.