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E. MAZZONI, P. CRAVEDI

Observations on the overwintering and winter mortality of the San Jose scale in fruit orchards in Emilia Romagna (Northern Italy)

Abstract - To establish the developmental stage of overwintering San Jose Scale, *Quadraspidiotus perniciosus* (Comstock) (Homoptera: Diaspididae) specimens in Northern Italy, a monitoring programme was carried out in Emilia-Romagna during winter 1998-99, mainly on stone fruit. According to the data collected, most of the specimens spend the greatest part of the winter in the first stage. Only in march specimens begin to develop quite quickly towards the second and then the adult stage. The natural winter mortality recorded is significant but so is the fact that important differences exist between years. The importance of these data for timing insecticide treatment also with IGRs and for choosing the most satisfactory and suitable winter control strategy of the pest, is discussed.

Key words: *Quadraspidiotus perniciosus*, control strategies, development

INTRODUCTION

The economic importance of the San Jose Scale, *Quadraspidiotus perniciosus* (Comstock), has increased significantly in Italian orchards during the last few years and this species is now the most worrying scale insect in stone fruit orchards as the classic stone fruit orchard scale insect, the white peach scale *Pseudaulacaspis pentagona* (Targioni – Tozzetti), is now considered less important. In fact in the last few years the spread, incidence and economic importance of the latter species has reduced significantly even though its world-wide presence is spreading presumably due to global warming (Kozar *et al.*, 1997).

There have been several factors during the last few years which are still contributing to this phenomenon. There are environmental factors like the climatic changes that are taking place mainly producing milder winters, which can reduce natural mortality in many species. There are also substantial changes in control strategies applied against moths (*Cydia molesta* (Busk) and *Anarsia lineatella* (Zeller)) in many orchards: the use of IGR insecticides has increased in the last few years and there is a more extensive use of pheromone applications like mating disruption method.

This reduces the number of sprays with organophosphates against moth larvae, but some of these treatments were at least partially active against scale crawlers. Farmers are also less inclined to spray in winter with polysulphides and many farmers and technicians also find it difficult to recognise and evaluate the severity of early infestations of *Q. perniciosus*, which are certainly less evident than those of *P. pentagona*. But neglected infestations can kill plants and in many cases heavy crawler migration can produce significant damage on fruits.

In Emilia-Romagna, the number of farmers that use IGR (like buprofezin) to control SJS infestations is increased. However the efficacy of these insecticides can be substantially affected by the correct timing of the application. In fact, these insecticides need to be used against the young developing instars of *Q. perniciosus* to be effective. Moreover, among technicians and farmers, there are still many doubts about the overwintering stage of *Q. perniciosus* in Northern Italy in fruit orchards even though the development of this pest has been well investigated in the past (Gambaro, 1947). But the only recent available data about the overwintering of *Q. perniciosus* in Italy were collected in Southern Italy, where all the instars were found during winter (Russo, 1986; Longo *et al.*, 1989).

So the objective of the present paper is to collect and quantify information about the overwintering stages of *Q. perniciosus* in Northern Italy and also to gather data on the incidence of winter mortality in its populations.

MATERIAL AND METHODS

Overwintering

The analysis of the overwintering behaviour of *Q. perniciosus* was carried out in untreated orchards with significant San Jose Scale infestation during winter 1998-1999. Selected orchards were located near Faenza, a very important fruit growing district of Emilia-Romagna (Northern Italy) (Fig. 1).

On the whole 5 orchards were selected (Tab. 1): 3 peach orchards, 1 plum and 1 pear. Each orchard was sampled 4 times between leaf falling in autumn and the end of winter; so the samples were collected in November-December; mid January; mid February and March (Tab. 1).

Each time and in each orchard about 20-30 small young twigs were cut at random.

Twigs, soon after cutting, were inspected under a binocular microscope. Scale covers were gently removed with a pin and live specimens collected. Between 100 and 130 live specimens were collected per sample and directly slide mounted with Hoyer's medium (McClain *et al.*, 1990). Slides were observed with a Leica DMRB light microscope equipped with "interferential differential contrast".

Specimens were recorded according to the following developmental stage: first instar; second instar and adult female. No attempt was made to differentiate in young instars between males and females. First and second instar specimens were recorded

Fig. 1 - The dark spot shows the area of Emilia – Romagna involved in the present study

Table 1 - Orchards selected to monitor the overwintering stage of Q. perniciosus *in Northern Italy and the corresponding sampling date.*

Orchard	**Cultivar**	**1st sampling (November '98)**	**2nd sampling (January '99)**	**3rd sampling (February '99)**	**4th sampling (March '99)**
Peach 1	Sweet Red	25.11.1998	19.01.1999	13.02.1999	4.03.1999
Peach 2	Sweet Lady	1.12.1998	15.01.1999	11.02.1999	17.03.1999
Peach 3	Red Haven	2.12.1998	15.01.1999	13.02.1999	4.03.1999
Plum	==	25.11.1998	14.01.1999	11.02.1999	4.03.1999
Pear	Abate Fetel	15.12.1998	15.01.1999	13.02.1999	3.03.1999

also according to stylet formation and the detachment between the old and the new cuticle in pygidium area (Fig. 2).

Specimens were subdivided into 4 classes according to stylet development and into 2 classes of cuticle detachment. In the case of the cuticle only two classes were considered: where the separation between the old and the new cuticle was visible in the pygidium area a short time before moulting. In the white peach scale (*Pseudaulacaspis pentagona*) the place were the cuticle detachment was always seen first is the pygidium (Mazzoni, unpublished). The stylet renewal is a more complex

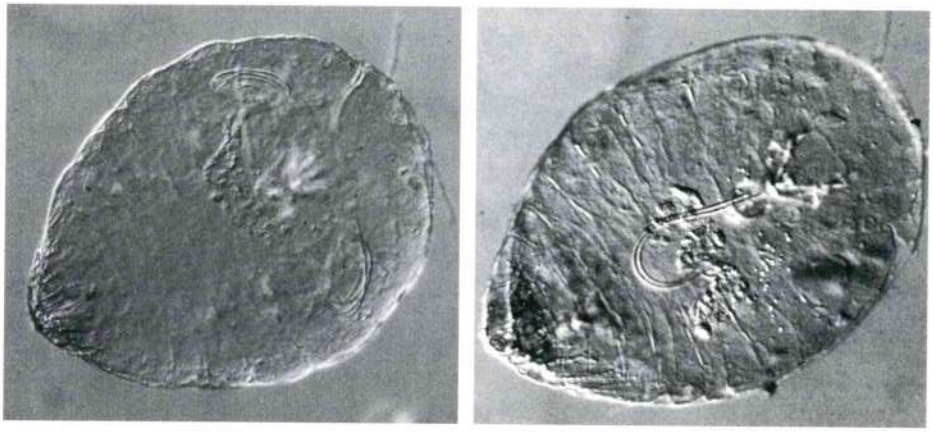

Fig. 2 - Slide mounted 1st instar specimens of *Q. perniciosus* with different development. On the left coils of the stylets are almost completed (class SC) and first sign of pygidium cuticle detachment can be observed (class PS). On the right the specimen is ready to complete its moult as new stylets have been transferred into the crumena (class SU) and the new pygidium is clearly visible (class PS).

phenomenon that is clearly linked to the moult and it is an important prerequisite because new stylets need to be ready before ecdysis (Tremblay, 1958). Different stages of development have been observed in this character and already used with *P. pentagona* (Mazzoni, unpublished data). Even though the formation of new stylets is a continuous process at least 4 distinct phases can be recognized: from the total absence of new stylet production till the despiralization and the transfer of the new formed stylets to the crumena (Tab. 2). This type of classification was also used to point out the beginning and the evolution of the moulting activity which is otherwise unidentifiable by external analysis of the body of the scale insect.

Analysis of the scale cover development was not carried out due to the difficulty of accurately separating the different instars using this characteristic.

Table 2 - List of the classes used to describe the evolution of development of Q. perniciosus.

Character	**class**	**Description**
Pygidium	PA	no detachment
	PS	clear separation in pygidium area
Stylets	NS	new stylets not yet developed
	ST	new stylets starting to develop and coils at least partly visible
	SC	new stylets fully developed and coils clearly evident
	SU	new stylets have been transferred into the crumena

Winter mortality

Mortality of *Q. perniciosus* was assessed during winters 1999-2000 and 2000-2001. A procedure very similar to that used in development analysis was adopted: small twigs or branches were cut and observed under a binocular microscope. Scale covers were removed and SJS specimens recorded as "alive" or "dead" according to their colour and turgidity / dryness or in doubtful cases the specimens were pricked with a pin to observe the presence of fluids inside of the body. No other methods were adopted mainly in order to determine as fast as possible (Ishaaya & Swirsky, 1990). As mortality was assessed during two open field evaluations of the efficacy of some insecticides, carried out during two different years, the number of samples collected was different. In the first year samples were collected 4 times in a peach orchard near Bagnacavallo (Ravenna), the first time at the end of autumn (1/12/1999), the second time in the middle of winter (13/01/2000), the third time at the end of winter before blossoming (14/03/2000) and the last time in spring (19/04/2000). In the second year samples were collected in a different peach orchard but in the same place (Bagnacavallo – Ravenna). Two sampling periods were chosen: at the end of winter, again before blossoming (13/03/2001), and in spring (11/04/2001). Each time 4 replicates were collected according to a randomized block scheme.

RESULTS

Overwintering

In the present study the average number of *Q. perniciosus* specimens collected and checked in each sample was 119 ± 6. The minimum was 104 and the maximum was 128. These statistics do not include the plum orchard in whose samples a limited number of live insects was found in January (only 20 specimens) and no live insects could be detected in the following samples (February and March), without any apparent reason.

The microscopic analysis of slide mounted specimens showed changes in two of the characteristics most involved in growth and development: stylet renewal and cuticle detachment and as a consequence also the evolution of the stage-structure of the *Q. perniciosus* populations observed.

In fig. 3 the average stage-structures of the observed populations are reported. As expected, in November adult females were still present in low numbers, but they were depositing crawlers and most of them were at the end of their life cycle. Many of the 1st instars collected were at the crawler stage or had just started to produce a scale. In January only a few 2nd-instar specimens were found but more than 99% of live scale insects collected were 1st-instar specimens. The same situation was observed in February. Only in March was there a substantial change, when about 25% of collected specimens were 2nd instars.

The single orchard data show that no important differences exist between plots

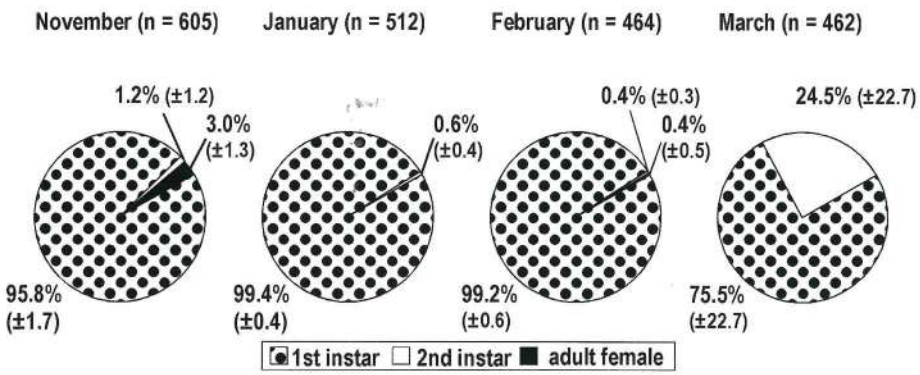

Fig. 3 - Live *Quadraspidiotus perniciosus* specimens collected in winter 1998-1999 in Northern Italy. Pies show the average percentage (±s.e.) of developmental stages present in each sampling period.

till the early days of March. But quite a sudden increase in insect activity was recorded in the following days. This fact is clearly shown by peach orchard no. 2 whose twigs were collected about two weeks later, in mid-March. Here a sudden change in the population structure was recorded as more than 92% of alive scale insects were 2nd instars (Fig. 4), which greatly influenced the instar ratio observed in March.

Live insects were also recorded according to classes of stylet renewal and cuticle detachment (Tab. 2) to show the changes in moulting activity of *Q. perniciosus* which are not detectable by external observation. Most of the specimens were always 1st-instar larvae (Fig. 3) and the frequency distribution of these specimens grouped according to the evolution of "stylets" and "cuticle detachment" is shown in fig. 5.

A progressive increase in the percentage of specimens included in the developed stylet classes is evident. In November only a few specimens showed any evidence of the presence of the characteristic coils of the newly developing stylets. In January, stylet development had started significantly and about 10% of the observed specimens were producing new stylets. In February, the percentage of 1st-instar specimens with clear development of new stylets was more than 25%. Between February and March there was the most significant increase in activity, as in the last sampling only less than 14% of specimens had no sign of metamorphosis. On the other hand, more than 75% of specimens showed fully developed stylets and were ready to moult. No signs of this kind of activity were detected in the few 2nd instar larvae collected.

The cuticle detachment also showed an increase towards the end of winter. While in November none of the 1st-instar specimens showed the detachment of the cuticle, in January, the phenomenon was observed for the first time in quite a low number of specimens (1.8%). But a significant change in this characteristic took place later and in March in more than half of *Q. perniciosus* observed the separation between the old and the new cuticle was observed in the pygidium. In general, the percentage of

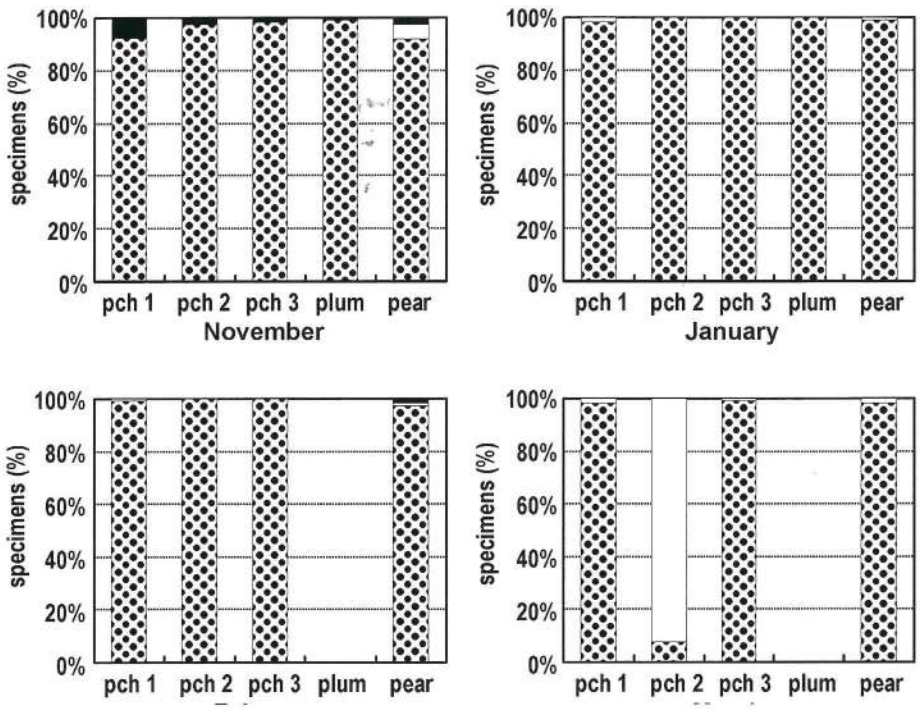

Fig. 4 - Percentage of developmental stages present in each orchard in the four sampling periods during winter 1998-1999, in Northern Italy. No live specimens were found in the plum orchard in February and March samplings (Legend: pch 1: peach orchard no. 1;. pch 2: peach orchard no. 2;. pch 3: peach orchard no. 3).

specimens, registered in the class "PS", was slightly lower than the percentage of specimens showing stylet renewal in progress.

There were little differences of development between *Q. perniciosus* populations collected in the 5 orchards but these differences seem to be linked with the particular orchard situation and not with crop influence (Fig. 4). The greatest differences in stylet development were recorded only in February when, in peach orchard no. 1 and no. 3, the percentage of specimens without any evidence of new stylet formation or new pygidium visible was about 30% lower than in the other orchards. Before and after no important differences were detected.

In general, the number of second instar specimens was quite low, with the single exception of the sample collected in March in peach orchard no. 2. In this case, more than 92% of specimens were at 2nd instar but all of them had moulted recently and no sign of new stylet formation could be detected.

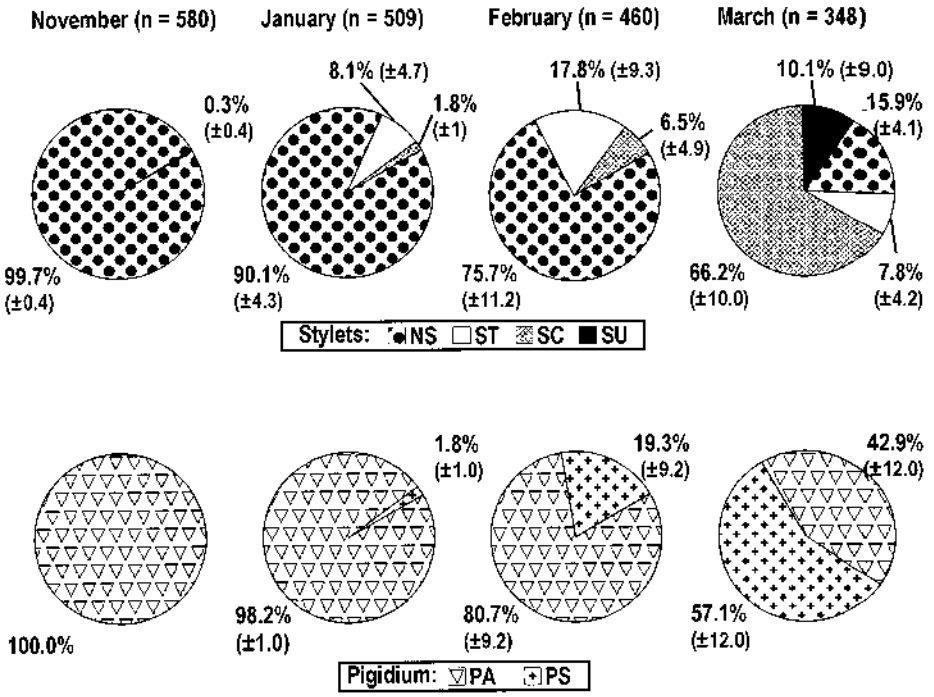

Fig. 5 - Distribution (average - percentage) of stylet development classes and cuticular detachment classes in 1st instar alive *Q. perniciosus* specimens collected in winter 1998-1999 in Northern Italy. (Legends [as in Table 2]: PA=no cuticular detachment; PS=clear separation between the old and the new cuticle in pygidium area; NS=new stylets not yet developed; ST=new stylets starting to develop; SC=new stylets fully developed; SU: new stylets in the crumena).

Winter mortality

To assess winter mortality 536 ± 38 (in winter 1999-2000) and 418 ± 25 (in winter 2000-2001) specimens were counted in each replicate. So respectively about 2000 and 1600 *Q. perniciosus* specimens were observed in each sampling.

In general, as reported in Fig. 6, it can be clearly observed that there is an increase in mortality during winter but this is significant only between January and March and between March and April. Analysis of variance (ANOVA) is extremely significant in 1999-2000 (F=17.8; df=3) and significant in 2000-2001 (F=7.4; df=1). Nevertheless important differences can be observed between years, because in 2000-2001 mortality was much lower than the preceding year. However, the great increase in mortality at the end of the winter is confirmed. In 1999-2000 the minimum temperature was generally lower than in 2000-2001. In fact the average minimum temperature in the

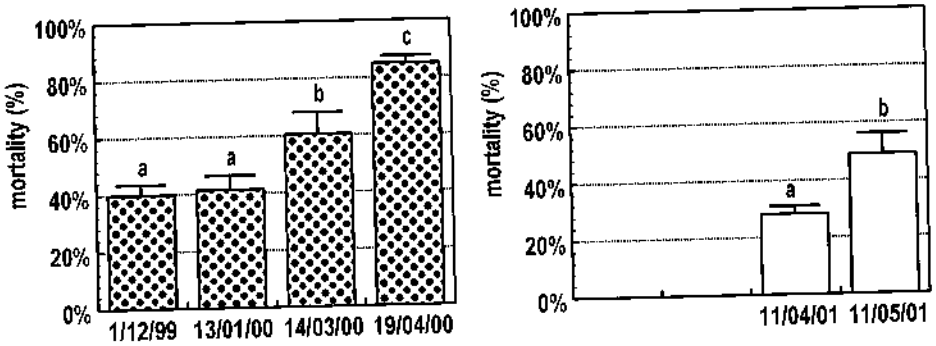

Fig. 6 - Average winter mortality in *Q. perniciosus* in 1999-2000 (left) and 2000-2001 (right). Bars showing the same letters do not differ significantly (ANOVA and Duncan's test for 1999-2000 data). Data are the average of 4 replicates; error bars show standard error.

period November 1st – mid April (last sampling) was 0.0 ± 5.0 °C as opposed to 4.9 ± 4.3 °C. Moreover, in 1999-2000 the absolute minimum temperature was –11.1 °C as opposed to –4.7 °C, but there was a rather long period with many more days with a minimum temperature below 0 °C. (Fig. 7).

CONCLUSION

In Emilia-Romagna the development of *Q. perniciosus* appears to stop during the middle of the winter, as reported earlier in Veneto (Gambaro, 1947). In this period the 1st-instar larvae are completely predominant, but they are not completely inactive as more detailed analysis of the renewal of their stylets shows that about 10% of specimens are producing new mouthparts. The pest increases its activity before the end of winter and between February and March most of the specimens start to moult. No important differences in development have been detected between specimens collected on peach and those collected on pear. Unfortunately data from plum are not comparable because of the sudden and total mortality observed in the considered plot. No significant reasons were found to explain this phenomenon. The presence of 2nd-instar larvae and of adult females seems to confirm that if any diapause is present, it is very short. As for 2nd instars, at present it is not possible to state if they were in a quiescent stage or simply a feeding stage but they did not show any sign of moulting activity. The presence of young adult females confirms that *Q. perniciosus* are able to have a very reduced quiescent period in winter in Emilia-Romagna, and to develop very quickly if environmental conditions are suitable. However, this phenomenon is certainly of very little importance.

The analysis of winter mortality shows that *Q. perniciosus* populations can be decimated by unfavourable climatic conditions like sudden temperature falls and in general the greatest mortality is linked with the moulting periods.

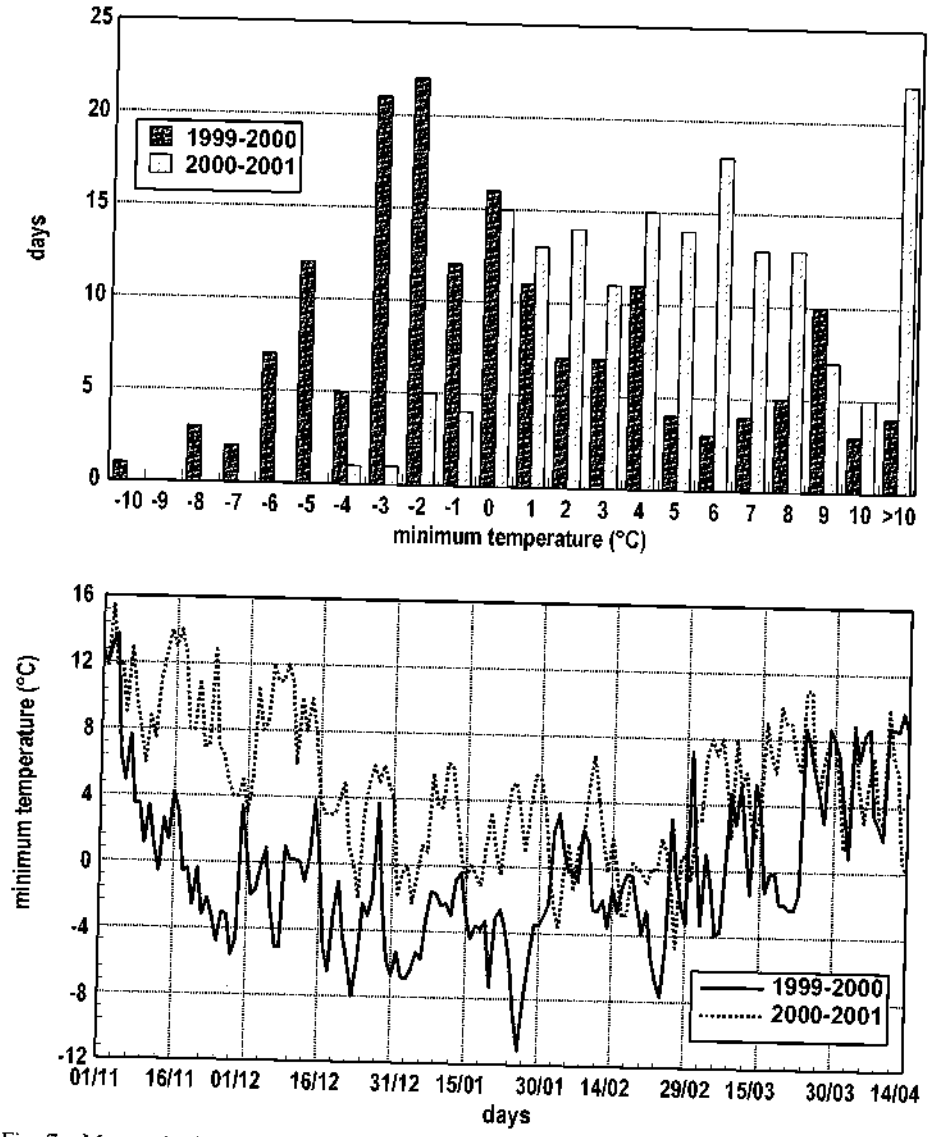

Fig. 7 - Meteorological data collected in Bagnacavallo. Up: number of days with minimum temperature below the indicated value. Low: daily minimum temperatures between November 1st and the last sampling in spring.

The collected data can have significant practical consequences on the SJS control strategies. In fact, even if IPM greatly improved in the recent years in Italian fruit orchards, winter sprays against scale insects are still based greatly on empiricism and

tradition but little support is given by scientific works and by monitoring. The knowledge of the moulting period is important to choose the best time to use IGR insecticides.

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DR. EMANUELE MAZZONI - Istituto di Entomologia e Patologia vegetale - Università Cattolica del Sacro Cuore, Via Emilia parmense 84, 29100 Piacenza, Italy. E-mail: mazzoni@pc.unicatt.it

PROF. PIERO CRAVEDI - Istituto di Entomologia e Patologia vegetale - Università Cattolica del Sacro Cuore, Via Emilia parmense 84, 29100 Piacenza, Italy.