Bionomics and ecology of Bemisia tabaci (Sternorrhyncha: Aleyrodidae) in Italy

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Aleyrodidae, Bemisia tabaci, developmental time, cold resistance, overwintering, Italy, distribution

Abstract. The development of a B-biotype *Bemisia tabaci* Italian colony was studied on bean at 9 constant temperatures (15, 16, 17, 20, 23, 26, 29, 32, 35°C). The developmental time from egg-to-adult varied from 70 days at 16°C to 22 at 26°C and higher temperatures. A thermal requirement for egg-to-adult development of 307 day-degrees was calculated, based on a lower developmental threshold of 11.53°C. The survival of egg, nymph and adult whiteflies was investigated at 0, 2, 4, and 6°C on broad bean for periods of 1–8 days. The adult was the most cold-sensitive stage, while the egg and nymph showed a similar level of cold resistance. The effect of sub-lethal cold stress of 4–8 days at 4°C on eggs and nymphs was studied. After exposure to low temperatures, whiteflies needed longer developmental times, from 5 to 8 days more. The presence of *B. tabaci* under outdoor conditions in Italy was investigated with field surveys and correlated with climatic data; the whitefly species was found in open field conditions only south of the 41° parallel, in areas characterised by less than 5 frost days per winter and by annual mean temperatures >16°C.

INTRODUCTION

The cotton whitefly, *Bemisia tabaci* Gennadius, has been known, until early 1980's, as a tropical and subtropical pest (Cock, 1986); its presence was also reported in the Mediterranean basin and in Italy (Patti & Rapisarda, 1981), where it was not regarded as a pest, so that Iaccarino (1981) described the species in a publication on the "poorly known whiteflies of Italy". In a few years, the reports of *B. tabaci* infestations in open field and in greenhouse crops throughout the world have multiplied. In southern USA, a new aggressive biotype appeared (Cohen et al., 1992), with a wider host range (Brown, 1994) and a wider range of transmitted viruses (Bedford et al., 1994). Formerly called B-biotype, it was then described as *B. argentifolia* (Bellows et al., 1994). It spread very fast and progressively displaced the pre-existing biotypes of *B. tabaci*. In Italy, after the dangerous appearance of tomato yellow leaf curl geminivirus (TYLCV) epidemics in tomato crops in Sardinia and Sicilia (Luisoni et al., 1989; Credi et al., 1989), *B. tabaci* became known as an important agricultural pest, also because of its specific role in vectoring TYLCV (Rapisarda, 1990).

Soon after its appearance in the USA, studies on the biology of the new "biotype" (species) were undertaken in that country. In Europe, the basic biological characteristics of this pest have been investigated only in Switzerland (Arx et al., 1983), using whiteflies from Sudan, and in Denmark (Enkegaard, 1993), using a whitefly colony previously grown in Germany (origin of which was not specified). Detailed knowledge on the ecology and

temperature-relationships of local populations, and on geographical distribution of this species in Italy (so far only preliminary information are available in Bosco et al., 1993), are essential to develop management strategies for greenhouse infestations and to forecast the risk of the establishment of *B. tabaci* in outdoor conditions. The present field surveys and laboratory experiments on the temperature-dependent developmental rate were undertaken to put in evidence the Italian distribution of *B. tabaci* together with bionomic characteristics, in order to forecast the possibility of whitefly overwintering and establishment in open field conditions in Italian agro-ecosystems.

MATERIAL AND METHODS

Field surveys

Surveys for the presence of *B. tabaci* in crops and weeds were made during 1992–1995 in the following areas: Albenga, Sanremo and Sarzana (Liguria Region), coastal areas of Toscany, Marche, Abruzzi and Molise Regions, Foggia and Brindisi provinces (Apulia Region), Metaponto (Basilicata Region), Sibari, Lamezia S. Eufemia and Baroncello (Calabria Region), Messina Paradiso, Giardini Naxos, Sigonella and Lampedusa island (Sicily Region), La Nurra, Decimoputzu, Pula, S. Margherita di Pula (Sardinia Region). A presence of the whitefly adults was monitored by visual inspection of plants, and identification of pupae was made under a stereomicroscope in the laboratory. Adults were captured and isolated on cucumber plants for oviposition, in order to obtain pupae for species identification.

Developmental rates

A colony of *B. tabaci*, belonging to the B-biotype (according to esterase pattern) was established on cucumber from individuals collected on poinsettia plants in Sardinia. After some years on cucumber plants, whiteflies were reared on French bean (*Phaseolus vulgaris* cv Saxa) in climatic chambers at different temperatures. A number of adult whiteflies were allowed to oviposit for 24 h at 26°C on bean. Plants with eggs were maintained in rearing cages at constant temperatures of 15, 16, 17, 20, 23, 26, 29, 32 and 35°C (\pm 0.5°C) with a 16L: 8D photoperiod. The relative humidity varied from 60 to 90%. To determine the developmental time/rate, two plants per temperature were infested; newly emerged adults were counted daily and removed. The calculated lower temperature threshold (LDT) for egg-to-adult development (the temperature at which the development rate equals zero, T_0) was estimated by simple linear regression of the mean developmental rate (1/days) against temperature. Since simple linear regression does not take into account the heterogeneity of the variance (Hart et al., 1997), the weighted linear regression (Draper & Smith, 1981) was also used. Developmental time on a day-degree (°D) time scale or Sum of Effective Temperatures (SET) was computed as °D = Σ days (T-T₀) for T>T₀. The day-degree requirement was estimated from the reciprocal of the slope of the regression lines.

Chilling experiments

The survival of eggs, nymphs and adults of *B. tabaci* at temperatures of 0, 2, 4, and 6°C was studied on broad bean (*Vicia faba* cv Aguadulce), which was used because of its good resistance to low temperatures. Lower temperatures were not investigated because they were not suitable for the host plant. The developmental time of the whitefly has been determined by rearing a large number of individuals at the optimal temperature 26°C. To study the survival, plants were exposed to adult *B. tabaci* for 5 days at 26°C for oviposition, so that eggs and nymphs exposed to the chilling periods were not of the same age. In the egg experiments, after oviposition, plants were freed of adults, moved to cabinets at low temperature for chilling periods of 1, 2, 4 and 8 days and finally kept at 26°C. To study the survival of nymphs, after oviposition and removal of adults, plants were kept for 8 days at 26°C to allow the development of nymphs, and then treated as described for eggs. The emergence of adults within 40 days from the start of oviposition was registered. Survival of adult *B. tabaci* was studied by exposing groups of 20 male and female whiteflies to the chilling periods (as above) in glass tubes containing broad bean leaves. The presence of living adults, and the ability of females to lay eggs, were checked the next day following chilling. Mortality during chilling experiments was always very high, but mortality data are not shown, since at

temperatures close to 0° C, some plant leaves, with eggs or nymphs, were damaged, thus preventing a correct count of surviving whiteflies; only the presence/absence of surviving whiteflies are reported.

In order to investigate if the whitefly eggs or nymphs show some forms of dormancy, the developmental times of the whiteflies surviving chilling periods, in day-degrees, were compared to that of the same whitefly colony reared under the same conditions, but at the constant temperature of 26°C.

Climatic data

Monthly and annual mean temperatures, as well as the annual number of frost days for 20 localities throughout the country were obtained from "Atlante Tematico d'Italia" (Pinna, 1989). Minimum and maximum daily temperatures in wintertime were recorded from December 1st 1995 to February 29th 1996 at Villa San Pietro (Sardinia Region) and Albenga (Liguria Region). In this latter locality, temperatures were also recorded during the same period in cold greenhouse. Winter daily mean temperatures were used to obtain the sum of effective temperatures (SET), in day-degrees (°D), for the development of *B. tabaci*.

RESULTS

Host plants and distribution

B. tabaci was found in greenhouses on poinsettia, tomato, cucumber, sweet basil, gerbera, vervain, and on Amaranthus retroflexus L., Solanum nigrum L. and Sonchus asper (L.) Hill. In the open field, the whitefly was found on tomato, eggplant, French bean, pole bean, melon, zucchini and chrysanthemum crops and on weeds of the genera Aster, Conyza, Datura, Inula, Sonchus, Solanum, Chenopodium and Parietaria.

In open field, *B. tabaci* was found at Sanremo and, close to infested greenhouses, at Albenga in Liguria; at Metaponto, in Basilicata; at Sibari, Lamezia S. Eufemia and Baroncello in Calabria; at Messina Paradiso, Giardini Naxos, Sigonella and Lampedusa island in Sicilia; at Pula and Decimoputzu in Sardegna. A few specimens were also found in one occasion in a crop near Brindisi in Puglia (Fig. 1).

Developmental times and rates

Developmental times of *B. tabaci* on bean plants are showed in Table 1. The minimum time was attained at 26°C. Higher temperatures did not shorten the life cycle while resulting in a higher mortality; at this temperature, 323 day-degrees were needed for egg-to-adult development. The minimum temperature at which the whitefly completed the whole pre-imaginal cycle was 16°C. The developmental rate on the same host-plant is shown in Fig. 2; only the range 16–26°C, where the relationship between temperature and rate of development is linear, was considered for calculation of regressions of mean developmental rate against temperature. The weighted linear regression gave an estimate of an LDT of 11.53°C and of a SET requirement of 307°D. The simple linear regression gave an estimate of an LDT of 11.21°C and of a SET requirement of 324°D. Mean developmental times in days, as well as in day-degrees, together with minimum and maximum developmental times at each experimental temperatures, are shown in Table 1.

Chilling experiments

Survivals of different *B. tabaci* life stages at low temperatures are summarised in Table 2. Adults appeared the most sensitive stage, being killed by 1 day at 0°C, 4 days at 2°C, and 8 days at 4 and 6°C. Eggs and nymphs showed higher chill resistance, being able to survive 4 days at 0 and 2°C, and 8 days at 4 and 6°C. Eggs suffered a lower mortality than

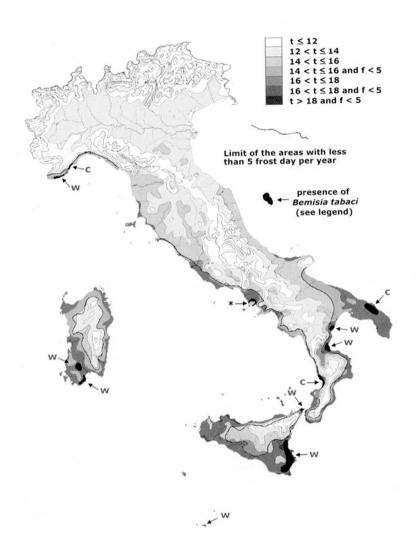


Fig. 1. Climatic map of Italy and distribution of *Bemisia tabaci* in open field conditions. The different climatic areas are characterised by different annual mean temperatures (t) and different numbers of annual freezing days (f). C – B. tabaci on crops only, W – on weeds and crops, * – reported by Iaccarino (1981).

the nymphs. The results of the comparisons between whitefly developmental time on broad bean at a constant temperature of 26 $^{\circ}$ C and at the same conditions with a chilling period of 4 or 8 days are shown in Table 3. Developmental times of eggs and nymphs subjected to a chilling period were always significantly longer than in the control (P < 0.05).

Table 1. Developmental time for *Bemisia tabaci* on French bean in days (\pm SE) (range indicates the day of the first and of the last emergence) as well as in day-degrees (°D) at the experimental temperatures.

Temperatures (°C)	Mean ± SE (days)	Range (days)	°D
16	70.3 ± 0.67	66–72	314.2 ± 2.99
17	54.0 ± 0.62	51–55	295.4 ± 3.37
20	38.0 ± 0.23	26-44	321.8 ± 1.98
23	26.9 ± 0.18	20-32	308.6 ± 2.08
26	22.3 ± 0.11	18–26	323.2 ± 1.59
29	22.0 ± 0.30	18–27	383.6 ± 5.32
32	23.2 ± 0.58	22–26	475.5 ± 11.87
35	21.2 ± 0.42	19–23	497.6 ± 9.78

Table 2. Survival of *Bemisia tabaci* at low temperatures. Filled circle – surviving, empty circle – not surviving.

Temperature		0,	°C		·	2	°C			4	°C			6	°C	
Days	1	2	4	8	1	2	4	8	1	2	4	8	1	2	4	8
Eggs	•	•	•	0	•	•	•	0	•	•	•	•	•	•	•	•
Nymphs	•	•	•	0	•	•	•	0	•	•	•	•	•	•	•	•
Adults	0	0	0	0	•	•	0	0	•	•	•	0	•	•	•	0

Whitefly distribution in relation with climatic data

Monthly and annual mean temperatures, annual number of frost days, together with data of *B. tabaci* distribution, showed that the whitefly spreads in open field conditions in areas characterised by annual mean temperature above 16°C, with less than 5 frost days per year (Table 4), indicating that winter temperatures, more than heat unit accumulation, are critical for whitefly spreading. To investigate the possibility of whitefly development during

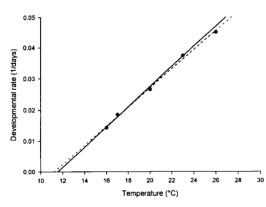


Fig. 2. Developmental rates of *Bemisia tabaci* on bean plants, fitted by simple linear regression (dotted line) and weighted linear regression (solid line).

the wintertime, the Sum of Effective Temperatures (SET) for B. tabaci has been calculated on the basis of daily temperatures from December-February in Southern Sardinian and Ligurian localities. In Sardinia, at Villa San Pietro, SET was 101.2°C, while in Liguria, Albenga, it was only 3.7°C. At Albenga SET was also calculated from daily temperatures in cold greenhouse, resulting in 229.9°C. In all cases, the number of day-degrees accumulated during the winter were less than the required for the completion of one generation. During the Italian winter,

B. tabaci should be able to survive, without completing a whole generation, only in the warmer areas, while a population increase in wintertime should be possible only in heated greenhouses.

Table 3. Developmental times of *Bemisa tabaci* on broad bean in days (\pm SE) as well as in day-degrees (°D), at constant 26°C (control) and at the same temperature following a sub-lethal cold stress. Values from all treatments are significantly different from the control (P < 0.01).

Treatment	Days	°D	Sample size (n)
Control	21.7 ± 0.06	314.4 ± 0.90	1,260
Eggs 4 days at 4°C	26.5 ± 0.35	383.1 ± 5.11	152
Eggs 8 days at 4°C	25.85 ± 0.18	374.1 ± 2.58	172
Nymphs 4 days at 4°C	28.3 ± 0.40	409.0 ± 5.78	61
Nymphs 8 days at 4°C	29.6 ± 0.40	428.5 ± 5.73	94

Table 4. No. of frost days per year, annual and monthly mean temperatures (°C) (from Pinna, 1989), and presence of *Bemisia tabaci* in open field conditions, in different Italian areas.

Locality	Frost days	Annual mean	January mean	July mean	Bemisia tabaci presence
Torino	75	12–14	0–2	24-26	_
Udine	50	12-14	2–4	22-24	_
Bologna	50	14–16	2-4	24–26	_
Foggia	25	14–16	6-8	26-28	_
Ancona	10	14-16	4–6	24–26	_
Livorno	10	14-16	6–8	24-26	_
Genova	10	14-16	6–8	24-26	_
Roma	10	16-18	6–8	24-26	_
Alghero	< 5	14–16	8-10	24-26	_
Lecce	< 5	16–18	6–8	24–26	+
Sanremo	< 5	16-18	8-10	24–26	+
Napoli	< 5	16-18	8-10	24-26	+
Taranto	< 5	16–18	8-10	26-28	+
Cagliari	< 5	16-18	> 10	24-26	+
Catania	< 5	16-18	> 10	26-28	+
Crotone	< 5	16-18	> 10	26-28	+
Palermo	< 5	> 18	> 10	24–26	+
Reggio C.	< 5	> 18	> 10	26-28	+
Messina	< 5	> 18	> 10	26-28	+

CONCLUSIONS AND DISCUSSION

The Sardinian biotype of *B. tabaci* showed a linear response to temperature for egg-to-adult development in a narrower range, compared to the biotypes described by Butler et al. (1983), Enkegaard (1993) and Wang & Tsai (1996). Assuming that development of all immature stages is a linear function of the temperature within the 16–26°C range, a developmental threshold of 11.53°C was estimated (but 16°C was the minimum experimental temperature allowing the completion of the whole development). The maximum

developmental rate was reached at 26°C; warmer temperatures did not increase the developmental rate while they caused a higher mortality.

In our study, the threshold temperature estimated by weighted regression was only 0.3°C higher than that calculated by simple linear regression. Nevertheless, as already observed by Hart et al. (1997), the weighted fitting passes closer to the experimental point for the lowest temperature tested, therefore estimating the developmental threshold more accurately. We conclude that the use of the weighted regression is advisable for LDT estimation and, therefore, for SET calculation.

The response to temperature increase was similar to those reported in most studies on developmental times/rates on different hosts. Shorter developmental times are reported on cotton at temperatures varying from 25 to 32°C (Butler et al., 1983; Powell & Bellows, 1992; Wagner, 1995), and longer times on poinsettia (Enkegaard, 1993). When comparing development on bean, our data were in good agreement with those of Coudriet et al. (1985) at 26°C. Differences in developmental times among populations can be partly explained by the different host plants. Compared to most investigated populations, the Sardinian population has one of the lowest temperature thresholds, it develops slowly at high temperatures, thus not taking advantage of high temperatures (at 26°C developmental time levels off). Many works concerning different insect orders have shown the existence of a negative relationship between LDT and SET (in Honěk, 1996a). The number of daydegrees needed to complete the egg-to-adult development on bean, 307 and 324°D using weighted and simple regression respectively, compares well with data from cotton, 316°D (Zalom et al., 1985) and 325°D (Butler et al., 1983, calculated from their data), and from poinsettia, 327°D (Enkegaard, 1993). Nevertheless, data from Powell & Bellows (1992) and Wang & Tsai (1996) allow for a calculation of much lower thermal requirements for their populations. Since cold-adapted populations show a faster development at low temperatures and a slower one at high temperatures (Honěk, 1996b), we conclude that the Sardinian population is more adapted to low temperatures than most B. tabaci-argentifolia strains.

The survival of different life stages to sub-lethal cold stress experiments showed that the adult was the most sensitive stage while eggs and nymphs showed a similar resistance to chilling. Nevertheless, a higher resistance of eggs to lower temperatures can be hypothesized, since at the experimental temperatures of 2 and 0°C, we found a lower mortality of eggs compared with nymphs. Wagner (1995) found a lower mortality of eggs at all temperatures, including those close to the lower threshold. Thus, the egg should represent the stage most adapted to cold. The experiments also pointed out that adults are not able to survive the frost condition.

We showed that whiteflies exposed to low sub-lethal temperatures, as egg or nymph, had subsequently higher thermal requirements compared to those reared at continuous optimal temperature. This could be explained as a repair of slight cold injury. Thus, only egg and nymph, the stages with the most extended range of resistance, survive to cold stresses, although suffering high mortality. The survival value of this quiescence for the population is probably very limited, but the phenomenon indicates that some kind of adaptation to low temperatures also occurs within this tropical and sub-tropical species. A selective pressure favouring more cold-adapted individuals needing more day-degrees for development (Honěk, 1996a) seems unable to explain such important differences in thermal

requirement (Table 3), although it cannot be completely ruled out. It is worth noting that a similar effect has been reported for *Frankliniella occidentalis* (Pergande) (McDonald et al., 1997).

In Italy, the presence of *B. tabaci* in outdoor conditions seems to be limited to the warmer regions, namely the areas south of 41°N, where the whitefly is probably able to overwinter outdoor, although suffering from a high mortality. On the other hand, the whitefly appears ubiquitous in greenhouse conditions, where it multiplies on a variety of protected crops, mainly on poinsettia, and where it can find suitable overwintering crops such as tomato (particularly in Sardinia and Sicily), gerbera and basil (particularly in Ligurian Riviera). The results of the present surveys suggest that in northern and central Italy, the whitefly can be occasionally found in open field only close to infested protected crops. The spreading of the whitefly from greenhouse to outdoor appeared very limited; if not so, epidemics of TYLCV, specifically transmitted by this species and introduced in our country since 1988, would probably occur also outside Sardinia, Sicily and Calabria. The absence of such epidemics in Campania and Apulia regions, where the whitefly has been found in outdoor conditions, probably indicates that these areas represent the northern boundary of *B. tabaci* distribution, where the whitefly can be found only occasionally in open field.

The climatic parameters that appear to limit the distribution of B. tabaci in Italy are the number of frost days per winter (<5) and the annual mean temperatures (>16°C). As it can be seen in the map, B. tabaci has the potential for damaging in some other crop areas in Italy in addition to Sardinia, Sicily and Calabria. Inside such areas, growers should consider the risk of B. tabaci infestations and subsequent damages (i.e. virus epidemics). Outside of these areas it can be reasonably expected that only greenhouse crops can be injured. However, even a relatively low increase of mean temperatures, due to general heating of the Earth, for example, or a further adaptation to lower temperatures, would dramatically expand the areas of establishment of B. tabaci in Italy.

ACKNOWLEDGEMENTS. We thank D. Marian for skillful technical assistance, G. Minuto and B. Fresu for providing climatic data from Albenga (Liguria) and Villa San Pietro (Sardinia), and S. Demichelis for determining the whitefly biotype. The work was funded by the University of Torino, research grant ex 60% "Insetti vettori di agenti fitopatogeni".

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