Incidence, Size and Structure of Grapevine Infesting Groups of *Kalotermes flavicollis* (Isoptera: Kalotermitidae) in Sherry Vineyards (Spain)

by

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ABSTRACT

Grapevine trunks in Sherry vineyards (Andalusia, SW Spain), yielding important registered designation of origin wines, are seriously affected by the drywood termite *Kalotermes flavicollis*. In order to analyze the patterns and extent of occurrence of the infesting groups, a 16-month-long investigation was performed, during which termites collected from each colonized grapevine trunk were recognized and counted according to the developmental stage or caste and the variation of caste proportions according to group size and season of occurrence was investigated. Results showed that different grapevine trunks exhibited different degrees of infestation, ranging from minuscule (0-50 individuals) to very large (more than 1000). Evaluating the composition and the occurrence of the colonizing groups, it was possible to determine that: a) minuscule groups, composed especially by eggs, reproductives, younger developmental stages and no alate adults, were probably 1-3 year old incipient colonies and occupied 12.6% of the infested grapevines; b) all groups with more than 50 individuals exhibited higher percentages of nymphal stages and, during fall, a considerable fraction (from 1/12 to 1/3) of the population was represented by alates, suggesting that they were mature colonies at different degrees of development. The greater part (64.2%) of infested vines were attacked by small-medium size colonies with a few hundred termites (51-700) and about 8% alates. Groups with the highest potential of infestation such as large colonies (up to 1000 units and 20% alates) and very large ones (1000-1692 individuals and 34% alates) were found respectively

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in 14.7% and 8.4% of termite colonized grapevines. The importance of these findings for the management of Sherry vineyards is discussed.

Keywords: termites, vineyard pest, caste ratio, colony size, vineyard management.

INTRODUCTION

Kalotermes flavicollis (Isoptera: Kalotermitidae) is a polyphagous species living in regions across the Mediterranean basin (Harris 1970). As in most kalotermitids, this strictly xylophagous species can feed on the wood of living trees (Lee & Wood 1971) and it has been reported to occasionally infest several species of ornamental and fruit trees such as poplar, elm, oak, pine, olive, chestnut, pear and almond (Monastero 1947, Prota 1965, López *et al.* 1996, Rossi & Rossi 1977, Ferrero 1973, Lebrun 1976, Kervina 1972).

The importance of *K. flavicollis* as a pest of grapevine trunks has been recognized since the end of the 19th century in Italian and French vineyards (Grassi & Aloi 1885, Mayet 1907, Bournier 1956, Springhetti 1957, Ferrero 1959). More recently, this kalotermitid has been recognized as one of the major pests in some Spanish and Portuguese vineyards (Pérez 1982, Lara & Cordero 1992, Castillo 1998, López *et al.* 2000), and the problem is particularly heavy in Sherry vineyards, SW of Spain (Andalusia), which yield the important registered Designation of Origin wines named "Jerez-Xérex-Sherry" and "Manzanilla de Sanlúcar de Barrameda", and the vinegar "Vinagre de Jerez".

K. flavicollis is a drywood termite with a typical "single wood piece life type" (Abe 1987), nesting and feeding in the initially selected wooden substrate. Its slow-development colonies, usually made of relatively low numbers of individuals, start to produce swarming alates after 3-4 years from their foundation (Grassi & Sandias 1893, Silvestri 1945, Grassé & Noirot 1958). The information is very scarce on the proportions of the different developmental stages and castes in wild colonies, and it usually refers to termites occasionally found in dead trees (Mazzantini 1953, Grassé 1986).

In the vineyards, *K. flavicollis* penetrates as dealate imagoes into the grapevine trunks across the pruning wounds, crevices and holes. The gradual increase of the colony and relative expansion of the galleries in the deeper tissues of living parts, progressively debilitates the vine causing the sequential loss of the arms and finally the death of the whole stock (López & Ocete 1999). The present study was performed to investigate the occurrence, size and seasonal social composition of grapevine infesting groups of *K. flavicollis* in order to get the degree of potential danger to which Sherry vineyards are exposed, in the view to provide useful hints for its management.

MATERIALS & METHODS

Randomly selected grapevine trunks were collected twice a month during a 16 month period from differently aged vines (one 4, 6, 9, 15, 20 year old and two/four 30 year old) of the "Palomino fino" variety from eight different districts of the Sherry area. The total amount of collected vines (198) was reduced, in order to conserve plants in productive plots.

Termite infested trunks, which were all aged 15 years or older (López *et al.* 2006), were categorized into five different groups according to the degree of colonization, namely the total number of individuals (excluding eggs) detected in the whole vine. The groups (in brackets the number of individuals) were: M= minuscule (0-50), S= small (51-300), I= intermediate (301-700), L= large (701-1000), V= very large (> 1000). Dates of sampling were grouped into seasons (WINTER= December, January, February; SPRING= March, April, May; SUMMER= June, July, August; FALL= September, October, November).

All the termites and eggs collected from each trunk were recognized, separated and counted according to the following classification (Noirot 1985): eggs (EGGS), larvae (LA 1-2), older larvae (LA 3-4), pseudergates (PSE), nymphal stages (N1, N2 and N3, respectively young, older and mature nymphs), alate adults (ALA), reproductives (REP) (males and females primary or secondary reproductives), white soldiers (WS) and soldiers (SOL). For each trunk the proportion of different stages was calculated. The total number of termites (including eggs) detected on grapevine trunks in different seasons are reported in Table 1.

For each stage, a 2-way ANOVA, followed by Duncan post-hoc test, was performed to detect if and how caste proportions (transformed into arc-sine square root) in grapevine colonized trunks were affected by the group size and by the seasons. All analyses were performed by means of Statistica 5.5 (Statsoft Inc., Tulsa, OK). Untransformed means are shown in the figures.

Table 1. Details on grapevine trunks colonized by *K. flavicollis* in Sherry vineyards.

Season	Group size	No. colonized trunks	Total No. termites (incl. eggs)
	M	6	331
	S	10	2672
Winter	I	15	8653
	L	3	2701
	V	2	2344
	M	1	4
	S	8	1956
Spring	I	6	3255
	L	3	2615
	V	1	1197
	M	2	40
	S	5	764
Summer	I	5	2553
	L	5	4410
	V	3	3470
	M	3	113
	S	5	991
Fall	I	7	3392
	L	3	2583
	V	2	3225

RESULTS

K. flavicollis was detected in 47.9% of the collected grapevine trunks, in 7 out of the 8 districts examined in the Sherry area. As shown in Fig. 1, the majority (64.2%) of the infested vines were attacked by small-medium size groups with a few hundred termites (51-700), whereas bigger groups (701-1692 individuals) and minuscule ones were found respectively in 23% and 12.6% of the termite affected vines.

Features on the social composition of vine infesting groups and on their variation along the year were obtained by means of statistical analyses (Table 2) and are shown in Figs. 2-3. For almost all stages the proportions were significantly different in grapevine colonizing groups of different size, with the exception of younger larvae, pseudergates and soldiers, whose percentages (respectively 13 ± 2%,

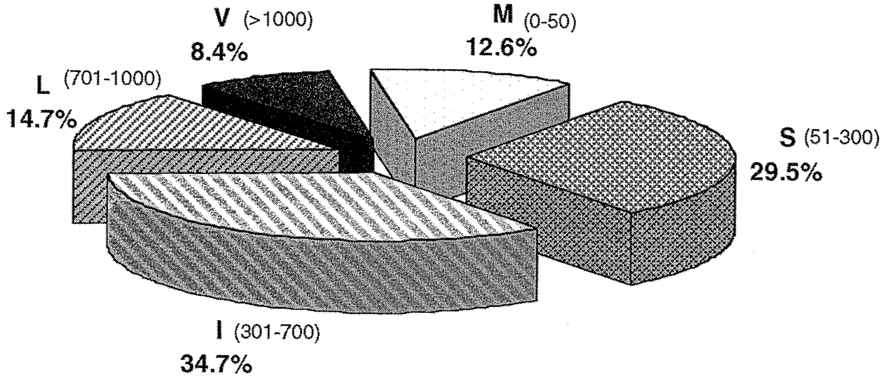


Fig. 1. Proportion of different colonization groups (M= Minuscule, S= Small, I= Intermediate, L= Large, V= Very large) of *K. flavicollis* detected in Sherry vineyards. Numbers in brackets indicate the group size in terms of total number of individuals (excluding eggs).

Table 2. Summary table for results obtained with 2-way ANOVA performed for each stage. (LA 1-2= larvae, LA 3-4= older larvae, PSE= pseudergates, N1-N2-N3= nymphal stages, WS= white soldiers, SOL= soldiers, ALA= alates, REP= reproductives).

	Season (df= 3)		Group size (df= 4)	
	F	P-value	F	P-value
EGGS	4.77	0.004	3.21	0.01
LA 1-2	1.77	0.15	1.64	0.17
LA 3-4	3.10	0.03	2.74	0.03
PSE	1.29	0.29	1.45	0.22
N1	0.98	0.41	12.22	< 0.001
N2	0.70	0.55	13.17	< 0.001
N3	14.18	< 0.001	15.68	< 0.001
ALA	64.05	< 0.001	4.88	0.001
REP	1.18	0.32	14.53	< 0.001
WS	2.29	0.08	5.46	< 0.001
SOL	2.29	0.03	2.28	0.07

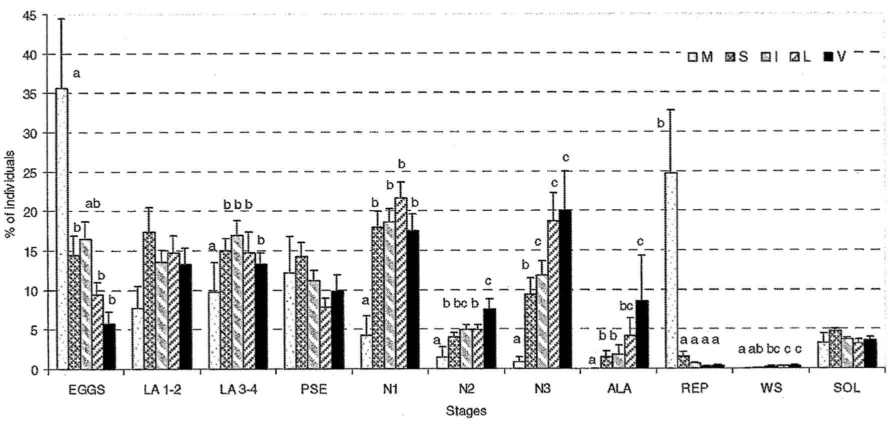


Fig. 2. Mean percentage of individuals (+ SE) of each developmental stage of *K. flavicollis* detected on grapevine infesting groups during different seasons, independently from the group size. Columns marked by the same letter within a group (or with no letters) are not significantly different at the 0.05 level determined by Duncan post-hoc test. (LA 1-2= larvae, LA 3-4= older larvae, PSE= pseudergates, N1-N2-N3= nymphal stages, WS= white soldiers, SOL= soldiers, ALA= alates, REP= reproductives).

11 ± 2% and 3.7 ± 0.6%) were roughly stable. The proportion of eggs was particularly high (36 ± 9%) in minuscule groups, where also detected was the highest percentage of reproductives (25 ± 8%), no alates were ever found and the percentages of all other castes were significantly lower than in other vine-infesting groups. The proportions of older larvae, N1 and N2 were

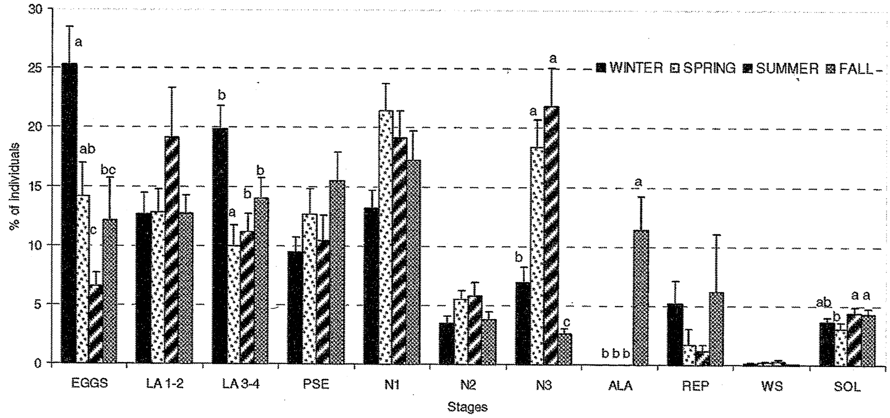


Fig. 3. Mean percentage of individuals (+ SE) of each developmental stage of *K. flavicollis* detected on grapevine infesting groups of different size (M= Minuscule, S= Small, I= Intermediate, L= Large, V= Very large), independently from the season of sampling. Columns marked by the same letter within a group (or with no letters) are not significantly different at the 0.05 level determined by Duncan post-hoc test. For acronyms see Fig. 2.

similar in all groups with more than 50 termites (respectively 15 ± 2%, 19 ± 2% and 5 ± 0.8%) and those of mature nymphs, alates and white soldiers were significantly higher in large and very large groups (respectively: N3= 20 ± 4%; ALA= 6.3 ± 4%; WS= 0.3 ± 0.1%).

Considering the effect of the seasons, for most of the stages no significant differences were detected, with the exception of: 1) eggs, whose percentage in winter was almost 4 times higher than in summer; 2) older larvae, whose proportion in winter was double than in spring; 3) mature nymphs, whose percentages peaked (20 ± 2.8%) in spring and summer and dropped in fall (2.5 ± 0.4%); 4) alates, that were detected exclusively during fall; 5) soldiers, whose proportion in spring was significantly lower than in other seasons.

Trends of the social composition of different grapevine colonizing groups for each season are reported in Fig. 4 a-d and most relevant results can be summarized as follows:

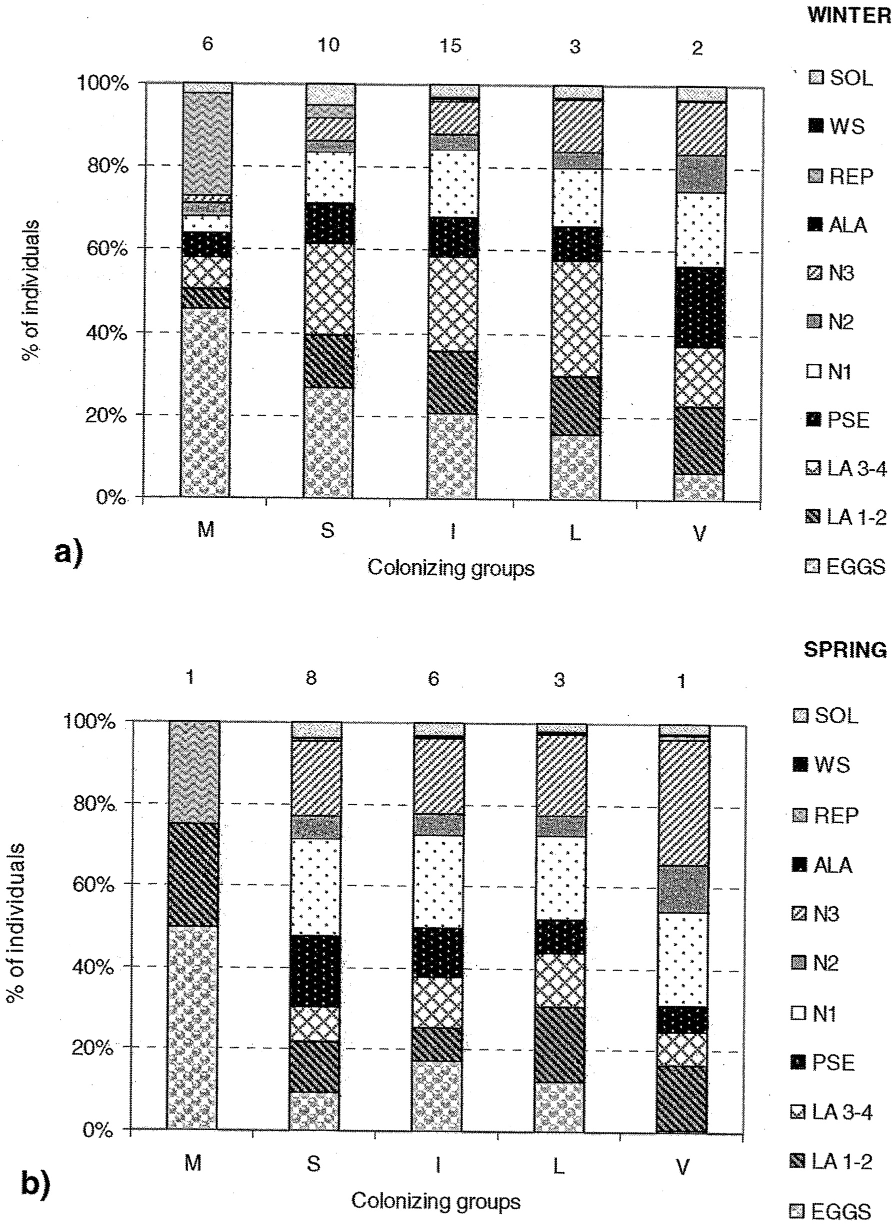
a) In minuscule groups the percentage of reproductives was always very high, ranging from 37% in fall to 7% in summer. Eggs represented half of the individuals detected during winter and spring; the percentage of larval stages was lower during the coldest seasons (12%) and tripled in summer; pseudergates proportion was high in summer and fall (22%) and low in winter (6%); the fraction of nymphal stages (with a bigger occurrence of N1) never exceeded 1/10 of the individuals; the highest percentage of soldiers was found during summer (7.5%).

b) Seasonal trends of all groups with more than 50 individuals showed that: during winter most of the population was represented by eggs and larval stages; during spring-summer there was a progressive increase of the percentages of young and mature nymphs, especially in bigger groups; during fall, together with the severe drop of N3, there was the appearance of alates, in proportions ranging from 8% in small and intermediate groups to 20 and 34% in large and very large groups.

DISCUSSION

The objective of this study was to obtain information on the patterns of infestation of *K. flavicollis* in Sherry vineyards. Our investigation showed that this drywood termite was detected in almost half of the collected grapevine trunks, all aged 15-30 years old (López *et al.* 2006), in colonizing groups of different size, ranging from less than 50 up to 1692 individuals. Colonies of *K. flavicollis* are reported to be made of relatively low numbers of individuals, usually a few hundreds and very rarely up to 1600-2500 (Grassi & Sandias 1893, Mazzantini 1953, Springhetti 1964). The development of an incipient colony of this species is relatively slow, since one year after its foundation it usually has no more than 50-60 individuals and it takes almost 2 years before the appearance of the first nymphs; however, after the first 2-3 years, the growth of a colony is exponential (Silvestri 1945, Grassé & Noirot 1958). In order to characterize vine-infesting groups in the Sherry area, the proportion of each developmental stage/caste for each colonized trunk was calculated and the statistical analyses allowed to detect variations in the social composition related to the group size and the season of sampling.

A characteristic of drywood termites, such as Kalotermitidae, is that a single piece of wood is usually occupied by a single colony, carrying out its whole life cycle inside of it, from the time of the initial colonization by a pair of dealate reproductives until the death of the colony several years later, when the wood is used up or becomes vulnerable to predation by birds and ants (Luykx 1986). Assuming that a higher degree of wood infestation might correspond to a mature-old colony, smaller degrees of colonization could represent either incipient colonies (1-3 years after their foundation)


WINTER
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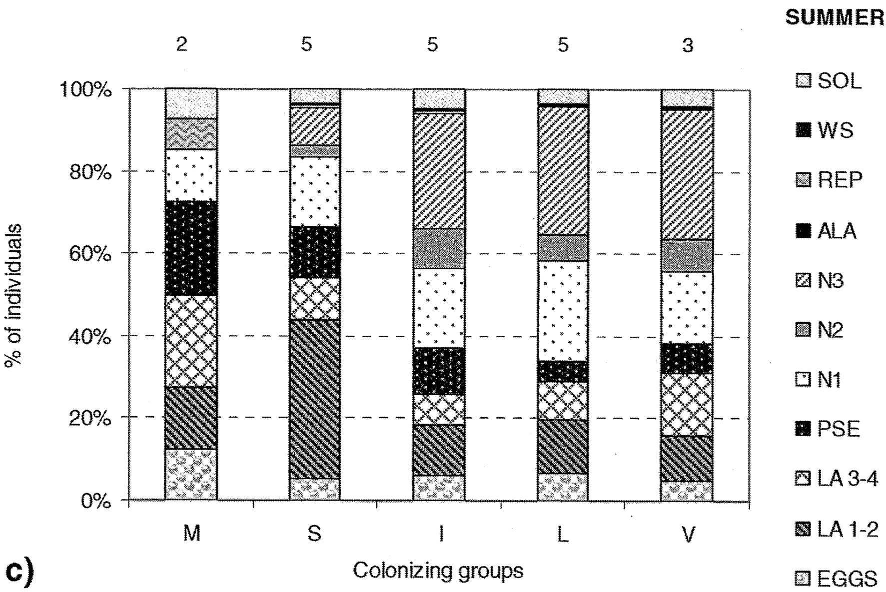

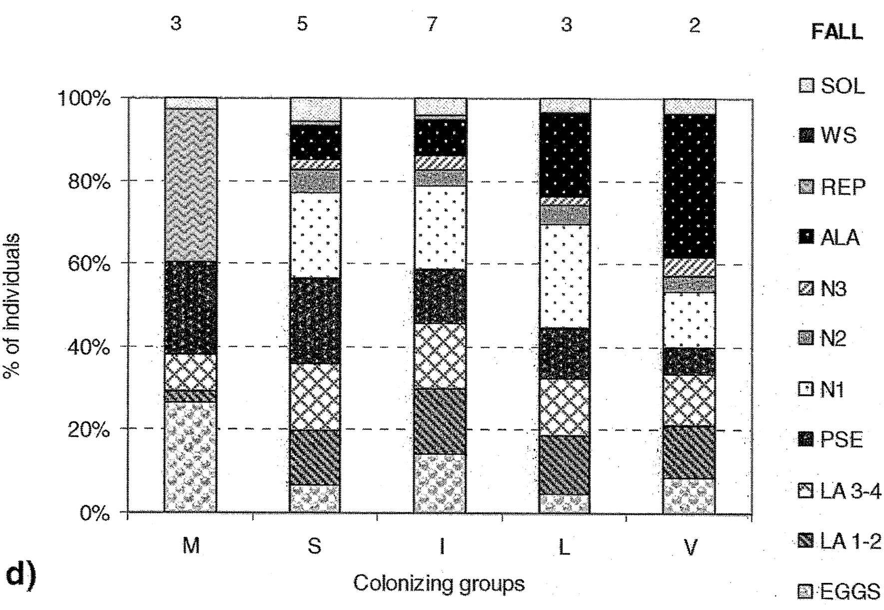

Fig. 4 a-d. Average seasonal composition (expressed as percentages of each developmental stage/caste) of different colonization groups of *K. flavicollis* in Sherry vineyards (N is indicated by the numbers on top of each column). For acronyms see Fig. 2.

or colonies close to extinction. From our investigation it emerged that the average social composition of minuscule groups (with less than 50 individuals) was characterized by a majority of eggs, a prevalence of younger developmental stages over nymphs, absence of alate imagoes, and by the highest proportion of reproductives, especially in the vines collected during winter. It is therefore most likely that less colonized vine trunks were occupied by incipient colonies. The higher percentage of reproductives in winter might be explained by multiple attempts of colony foundation by adult males and females swarmed during the previous fall, most of which were destined to become unsuccessful. In all vines colonized by more than 50 individuals, the percentage of eggs and reproductives was much lower, the proportion of nymphal stages (especially mature ones) prevailed over younger ones and alate imagoes always appeared during fall in percentages from 8 to 34% of the total vine trunk population, thus confirming that greater degrees of infestation might correspond to older, mature colonies of *K. flavicollis*, at different degrees of development. Our findings reveal that the proportions of younger larvae, pseudergates and soldiers did not vary in colonizing groups of different size, and this is possibly related to their biological constrains and functional role inside the colony. Younger larvae might represent a reserve of young "totipotent" undifferentiated individuals; pseudergates probably represent the major work force for colony labor and nestmates care (Noirot & Pasteels 1987); soldiers proportion, corresponding to the average value (2 and 5%) found in wild colonies for this species (Haverty 1977, Grassé 1986), is probably a compromise between the need of colony defense and the nutritional dependence of this caste on other nestmates. Seasonal trends detected for the proportions of eggs, younger larvae and soldiers show that in *K. flavicollis* winter seems the most important period for egg production and that during warmer seasons the differentiation to castes takes place, according to the colony degree of development and its specific needs, such as, for example, the increased proportion of soldiers during summer and fall, necessary to protect emerging alates (Howard & Haverty 1981, Luykx 1986). The appearance of alates only during fall, together with the peak of N3 detected during summer, indicate that in this species alate production occurs only once a year and confirms that in the Sherry area swarming takes place between September and October (López *et al.* 2003b, López *et al.* 2003a).

From this study it emerged that most of the grapevine trunks colonized by *K. flavicollis* were occupied by small (29.5%) and intermediate colonies (34.7%), probably aged 3-5 years, whose potential of infestation, in terms of alate production, was around 20-60 male and female imagoes a year. Mature older colonies (large and very large groups) with a much greater potential of infestation (about 100-600 alates/colony a year) were found in 23% of the infested vines. These data should be considered seriously, together with the estimates from recent observations performed in Marco del Jerez (a district of the Sherry area), indicating that *K. flavicollis* was found in 45% of the vines aged 5 years, in 60% of 10 year old ones and in 100% of vines older than 15 years (López *et al.* 2003b). Among the possible reasons for this increasing termite infestation, two hypothesis can be formulated and may well interact, and both refer to the last serious drought periods (1979-83 and 1988-95) (López & Ocete 1999) occurring in the Sherry area: a) the weakening and premature aging of the vines as a consequence of the water shortage sustained, rendered the plants more vulnerable to termites; b) it is possible that drier conditions could favor colony development and alate production in this species, thus increasing the infestation rates. The management of Sherry vineyards should therefore include strong recommendations for the control of *K. flavicollis*, such as the adoption of preventative measures, like the use of sealing paste on the pruning wounds (Pérez 1982, Kashyap *et al.* 1984, Castillo 1998) and the prohibition to use pinewood supporters (potential receptacles of active termites) or extreme measures such as the uprooting and burning of termite-infested grapevines (Castillo 1998).

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