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Environmental factors and agronomic practices associated with Savi's pine vole abundance in Italian apple orchards

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Abstract The development of an integrated rodent pest management strategy requires the adoption of protocols that should be based on a good knowledge of species-habitat relationships. Vole damage to orchards has increased in Italy in recent decades, as new cultivation practices have been introduced, despite the use of rodenticides. To improve our understanding of factors influencing Savi's pine vole abundance in Italian apple orchards, we monitored the relationship between environmental and agronomic variables and population densities. Vole population sizes were influenced by the abundance of voles in the neighbouring fields and the presence of kiwifruit orchards, usually not treated with rodenticides, bordering on apple orchards. The type of irrigation system had the strongest influence on vole abundance and flood-irrigated fields had fewer voles than drip-irrigated fields. Apple tree age and tilling practices also had an influence on vole abundance. Our research provides evidence that vole populations are influenced mostly by agronomic practices and type and extension of fruit orchards. An integrated management strategy in the apple orchard district should include coordination in chemical treatments between farmers and the experimentation of flood irrigation and regular soil tillage management as a possible cost-effective way to reduce vole populations.

Keywords Abundance index • agro-ecosystems • Italy • *Microtus savii* • rodents • species-habitat relationship

Introduction

The development of an integrated rodent pest management strategy as an alternative to the heavy use of rodenticides, requires the adoption of protocols for species under specific production systems (Byers 1984; White et al. 1997; Singleton et al. 1999). These protocols should be based on a good knowledge of species-habitat relationships and the effects of environmental variables, as well as agronomic activities on species presence and abundance (Singleton et al. 1999; Palis et al. 2007, 2010). This information is generally derived either from intensive field work (Maisonneuve and Rioux 2001; Hansson 2002) or from habitat models (Bertolino et al. 2011). Model-based analyses of species-habitat relationships can help to clarify which factors influence the presence and abundance of species, thus helping to develop appropriate mitigation strategies (Bertolino and Ingegno 2009).

Voles are common rodent species in agro-ecosystems, where they can cause extensive damage to crops, orchards and forestry (e.g. Sullivan and Hogue 1987; White et al. 1997; Hansson 2002;

Wiman et al. 2009; Miñarro et al. 2012). Although the diet of voles consists primarily of grasses, sedges and forbes, they also feed on bark, vascular tissues and tree roots. High-density populations can develop very rapidly, due to the high reproductive output of the species. In these cases, damage to orchards might result in bark removal at the soil surface and in the root system (Santini 1986). This damage causes a reduction in yield, fruit size and quality that lasts several years, and often the tree must be replaced.

In Italy, the common vole (*Microtus arvalis*) and the Savi's pine vole (*Microtus savii*) have been reported to affect apple and citrus orchards and crop fields, especially in no-till fields (Santini 1986, 1988). The major vole species present in fruit orchards is the Savi's pine vole. This species is nearly endemic to Italy and is widely distributed throughout the Italian peninsula and the island of Sicily, from sea level up to 2,000 m a.s.l. Its presence is characterised by a well-defined trail system and burrows (Amori et al. 2008). The small litter size and the prolonged gestation time suggest that the Savi's pine vole could be considered a k strategist within the Microtinae (Caroli et al. 2000).

Damage to orchards has increased in Italy in recent decades, as new cultivation practices have been introduced (Santini 1988; Galliano et al. 1995). To date, a rational control programme has not been implemented, and in the presence of heavy attacks, rodenticide baits are usually distributed inside vole burrows. However, the results of field trials are conflicting and a great number of growers have reported continued rodent problems, in spite of the use of rodenticides (Santini 1986; Galliano et al. 1995; Capizzi and Santini 2008). Previous studies on orchards indicated that only short-term substantial control has been achieved with toxicants (Sullivan 1986; Merwin et al. 1999). A more effective way to control vole damage in fruit orchards could be the development of an ecologically based rodent pest management strategy that includes agronomic practices and habitat manipulation (Sullivan and Hogue 1987; Merwin et al. 1999; Singleton et al. 1999). This, however, requires a better understanding of the factors that influence settlement and development of vole populations in orchards and surrounding areas and the variables that are correlated with damage. While this information is available for many vole species (e.g. Sullivan and Hogue 1987; Sullivan and Sullivan 2006; Wiman et al. 2009), there is still a lack of knowledge for the Savi's pine vole.

To fill this gap of knowledge, we initiated research to gather information on the presence of the Savi's pine vole in different production contexts and to evaluate the environmental factors associated with vole abundance. The aim of the present study was to analyze the spatial distribution and abundance of voles in apple orchards according to habitat attributes of agro-ecosystems. The following questions were addressed: (1) are voles distributed equally across orchards? Is vole abundance related to (2) landscape features or (3) to agronomic practices?

Materials and methods

The study was conducted in the province of Cuneo in the Saluzzo district (Northwestern Italy, Fig. 1), where we monitored the Savi's pine vole in apple orchards. The only other vole species present in these orchards, although highly localised, is *Arvicola* sp. However, activity traces of this species might be easily distinguished from those of the Savi's pine vole.

In 2011, we evaluated the effect of environmental and agronomical variables on the abundance of voles in apple orchards. As a first step, we compiled a list of 195 orchards which were > 0.5 ha and younger than seven years old, since damage by voles in older orchards is limited. From this list, we randomly extracted 36 orchards (Fig. 1).

The orchards were located in a part of the Po plain mainly used for agriculture (orchards and crop fields) bordering the first part of the Alps in the southeastern part of the area where woodlands were concentrated (see Fig. 1 in Online Resource 1). The landscape − evaluated in a circular plot with a radius of 12 km from the centroid of the minimum convex polygon including all the sampled orchards − was composed by 60.4% of crop fields, 18.8% of woodlands and poplar plantations, 11.1% of orchards, 5.4% of meadows, pastures and other herbaceous habitats, while the remaining surface was mainly covered by urban areas, rivers and channels (Fig. 2 in Online Resource 1).

The abundance of voles was determined using the open-hole index (OEPP/EPPO, 1992; Tkadlec and Stenseth 2001; Lisická et al. 2007). This vole abundance index (V_{ai}) measures the presence or absence of a vole within an underground burrow, by relying on the vole's propensity to reopen tunnel entrances in its burrow system that were previously closed with soil, and it is considered a good index of relative population abundance. During August and September 2011, the vole abundance index was evaluated in each orchard as the mean of reopened tunnel entrances counted along six transects. A 50 by 2 m transect was established along a single row of apple trees counting the active tunnel entrances on the two sides of the plants. The six parallel transects were distributed every two rows starting from the second one. To graphically represent the index recorded in different apple orchards, values were categorized into 5 ranks: 1: < 4.0, 2: 4.1–8.0, 3: 8.1–16.0, 4: 16.1–32, 5: 32.1–64.0 (Fig. 1).

We used multiple-regression analyses to investigate whether environmental and agronomical variables influenced the abundance of voles in each apple orchard. Model selection should be based on a set of variables that plausibly have a direct link with the dependent variables. The following information was collected in each orchard: municipality, UTM coordinates, apple-tree age, the presence of kiwifruit orchards, other orchards, crop fields or fallows bordering the monitored apple orchard, and the presence and distance of neighboring riparian strips along rivers or canals. We also measured the number of active tunnel entrances in the neighboring areas (i.e. areas bordering the monitored orchard) to the north, east, south and west, as an index of vole abundance in the surrounding areas. The farmers gave us information on the type of irrigation system used (i.e. flooding or dripping), the use of chemical weed control, natural fertilization, and earthing up. The habitat composition in a circular area with a radius of 500 m centered in the sampled orchards was evaluated with 1:10,000 digitized maps of land cover imported to QGIS software (see Table 1 in Online Resource 1). The buffer areas were mainly composed by orchards (mean ± SD: 66.5 ± 37.7%) and crops (27.0 ± 34.2%), while grasslands (meadows and pastures) covered 0.31 ± 0.58%; these three variables were also considered in the analysis. A possible spatial autocorrelation was considered in the modelling procedure inserting the Universal Transverse Mercator (UTM) coordinates, first each alone (North or East) and then together and with the other variables. These variables were not selected in the best models, indicating a lack of trend correlated with latitude and longitude. The variables used in the analysis are described in Table 1, with references to studies that justify their possible influence on the presence and abundance of voles.

We considered models that included each variable alone, as well as models with multiple variables, until all combinations of possible variables had been tested. Explanatory variables were not correlated ($r < 0.5$); assumptions were checked with the Variance Inflation Factor VIF, the Tolerance Values and the analysis of residuals. An information-theoretic approach was used to select models that were most informative (Burnham and Anderson 2002). A model with a low Akaike Information Criterion (AIC) score compared with other models fitted to the same data represents a preferred model in terms of goodness-of-fit. All candidate models were ranked based on the AICc score for small sample sizes and we used delta AICc (Δ_i) and the Akaike weights (w_i) to assess the strength of evidence that a particular model was the best within the candidate set. The Δ_i is the difference between the AICc of a given model and the AICc of the highest ranked model (i.e. with the lowest AICc); a $\Delta_i < 2$ suggests substantial evidence for the model. The Akaike weights (w_i) indicate the probability that the model is the best among the whole set of candidate models. Summing sequentially the weights of the models, starting from the best (most parsimonious), until the total weight sums to 0.95 (w_i95%), the models obtained can be used as the equivalent of a 95% certainty interval (Burnham and Anderson 1998; Greaves et al. 2006). This represents the smallest subset of models for which there is a 95% confidence that the set contains the best approximating model to the true model.

A weighted average was calculated for the coefficients of parameters in the top ranked models ($\Delta_i < 2$) and the w_i95% certainty model average (Burnham and Anderson 1998; Greaves et al.

2006). For a group of models, the coefficient is averaged over all the models in which that parameter appears, multiplying its value by the model's weight recalculated based on the models included in the group, so that the sum of the weights is equal to one. The sum of these values is then divided by the sum of the weights of all models in which that parameter occurs.

Mean values of population indexes were compared using the Student's t-test. Variables were square-root transformed to meet normality when necessary. Means are reported ± SE unless otherwise stated. All statistical analyses were performed with SPSS version 17.0.1.

Results

The vole abundance index in apple orchards ranged from 2.36 ± 1.53 to 22.96 ± 5.83; values are presented in Fig. 1 with a 1–5 rank system. Orchards clustered in the central part of the study area (n = 17, Vai = 18.7 ± 3.9, included in the dashed circle in Fig. 1) hosted more abundant populations with respect to other fields (n = 19, Vai = 3.7 ±1.2; $t = 3.86$, $P < 0.01$).

Table 2 shows the top ten models with the lowest AICc values. The model with only the variable related to the abundance of voles in neighbouring fields (Vole NF) had the lowest AICc but a $w_i = 0.278$. This suggests that this model was not convincingly the best model. The second and third models had Flow or Kiwi as further variables. These three models had a $\Delta_i < 2$ and were therefore considered the most parsimonious; together, they had an Akaike weight of 0.50. Five models had a $\Delta_i < 4$, so they can be considered as useful to explain the variance in vole abundance in apple orchards. The sum of the weights of these models was 0.92, thus they could be considered as the equivalent of a 95% certainty interval.

Model averaging was performed on the three top ranked models (Δ_i <2) and the eight w_i95% certainty model averages ($\Delta_i < 4$). The coefficients for the averaging models are shown in Table 3; they were very similar between the two groups of models, except for the three additional variables in the second group. Flow and Kiwi were respectively the variables with the strongest effect, followed by Earthing up and Vole NF, while the effect of Age and Orchards 500 was limited. Orchards 500, in particular, was selected in only one model with a weighted average of the coefficients that was nearly zero, therefore it did not really affect vole abundance.

Flood-irrigated apple orchards had fewer voles ($V_{ai} = 2.60 \pm 1.93$) than drip-irrigated fields ($V_{ai} = 12.13 \pm 2.58$; $t = 2.96$, $P < 0.01$, Fig. 2 see also Table 1 in Online Resource 1). There were more voles in apple orchards bordering on kiwifruit orchards ($V_{ai} = 15.34 \pm 3.93$) than in orchards further away from kiwifruit orchards ($V_{ai} = 6.75 \pm 2.27$, $t = 1.89$, $P= 0.07$, Fig. 2). Considering only apple orchards bordering kiwifruit orchards, the vole abundance increased with the number of active tunnel entrances found in kiwifruit plots (R^2= 0.59, $F_{1,15}$ = 21.14, $P < 0.001$, Fig. 3). In general, vole abundance in monitored apple orchards increased with an increase in the number of active tunnel entrances counted in neighbouring fields (R^2= 0.48, $F_{1,34}$ = 31.04, $P < 0.001$, Fig. 3).

Discussion

We monitored the relationship between environmental and agronomic variables and Savi’s pine vole relative abundance in apple orchards. Our analysis provided evidence that vole populations in apple orchards were mainly influenced by agronomic practices and the extension of fruit orchards nearby. Rodent populations were more abundant in apple orchards clustered in the centre of the study area, a sector that was characterized by more intensive fruit farming. Three main variables influenced the Savi's pine vole population size in apple orchards: the abundance of voles in the neighboring fields, the presence of kiwifruit orchards bordering apple orchards and the type of irrigation system. The models included in the 95% certainty interval also considered tree age and earthing up practices.

The presence of voles in the neighboring fields influenced the abundance of voles in the apple orchards. Although damage is evident in orchards under seven years old, vole populations are present over larger areas, including older orchards. These fields are usually not treated with rodenticides because there is no damage and they might act as source areas for a rapid recolonisation of treated fields following the control of rodents. The high reproductive output and the mobility of voles make it easy for them to recolonise orchards after populations have been reduced (Miller and Richmond 1982). These authors showed that pine vole (*Microtus pinetorum*) reinvasion of an intensively trapped orchard led to a density higher than previously within a single year. The presence of kiwifruit orchards bordering on apple orchards represents a possible source of field colonization by voles after chemical treatments. Kiwifruit orchards are subject to less agricultural practices; the floor vegetation in these orchards might thus easily grow, to increase ground cover and form habitats where voles can establish populations that could colonise neighbouring apple orchards. Our results confirm the importance of a control treatment covering an area larger than a single field, to reduce reinvasion by nearby populations (Miller and Richmond 1982; White et al. 1997). Kiwifruit orchards bordering apple orchards should be included in the treated area if voles are present.

The irrigation system had the strongest influence on vole abundance. Flood-irrigated fields had fewer voles than drip-irrigated fields. Voles were more abundant in the central area of intensive farming involving drip irrigation to reduce the use of water. Damage caused by voles in Italian apple orchards has grown where over-canopy or dripping irrigation have become widespread and the traditional irrigation by flooding has been abandoned by many farmers (Santini 1986, 1988). Flood irrigation appears to be effective in reducing the presence of voles. Therefore, it could be useful in fields with drip irrigation, to test whether water submersion might contain vole populations, and to evaluate the periods of the year when the treatment is most effective; a possible effect on crop productivity should also be evaluated.

Tree age and earthing up practices appear to have some influence on vole abundance. Although we only selected fields younger than seven years old, the presence of voles increased with tree age. This is probably related to the deep ploughing necessary to prepare the soil for a new plantation, with the consequent heavy disturbance and the requirement of some years for voles to colonise new fields. Earthing up along the row probably increases the disturbance to rodents and delays the recolonisation and prevents high densities.

The presence of the Savi's pine vole is favoured by a good grass cover (Capizzi and Santini 2008) and its limitation within the orchards could negatively affect populations. In our study the number of chemical treatments repeated during the year to control weed development was not selected as an important variable. This is because the weed management is very similar among farmers: in general two or three applications of herbicide in the intra-tree row and the maintenance of the grass cover in the space between rows. Therefore, it should be useful to compare in a future study these orchards with others where weed control is more intensive and covers all the surface.

Environmental variables related to natural habitats are usually important determinants of vole abundance (e.g. Maisonneuve and Rioux 2001; Sullivan and Sullivan 2006; Witmer et al. 2009). We tested the influence of fallow areas bordering the orchards and the presence nearby of rivers and canals on the abundance of the Savi's pine vole. These variables are known to influence the presence and abundance of other vole species in agro-ecosystems (White et al. 1997; Maisonneuve and Rioux 2001; Heroldová et al. 2007; Yletyinen and Norrdahl 2008), however these variables were not selected in our best models. In our study areas, apple orchards are highly simplified agricultural habitats, where natural vegetation is sparse and limited. We suppose that the size of riparian strips along riparian banks is too limited to see a positive influence of vegetation cover on vole population abundance.

Conclusion

The reduction in regular soil tillage management, the decreased chemical weed control of the grass cover and the replacement of traditional irrigation by ground submersion with drip irrigation, has favoured the increase of Savi's pine vole populations in fruit orchards (Santini 1986, 1988). Our results provide evidence that vole management activities should be implemented over large areas. Rodents are able to move from non-crop to crop habitats and vice versa (Horskins et al. 1998); therefore, the management unit for rodent control should be expanded from the single field to a larger area, to include a buffer zone inhabited by rodents. An integrated rodent pest management strategy in the apple orchard district should include coordination of chemical treatments by farmers and the experimentation of flood irrigation, regular soil tillage and grass cover management as a possible cost-effective way to reduce vole populations.

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Table 1 Summary of the variables evaluated for monitored apple orchards and references that support their possible influence on vole populations

Variables	**Description**	**References**
Municipality	Municipality of the orchards	
UTM	UTM coordinates of the orchards	
Age	Age (years) of the plantation	
V_{ai}	Vole abundance index	
Vole NF	Number of active tunnel entrances counted along 30 m in areas neighbouring the orchards in the four cardinal directions (mean of four values)	Miller and Richmond, 1982; Sullivan and Sullivan, 2006; Yletyinen and Norrdahl, 2008
Other Orchards	Presence of orchards bordering the orchard monitored (presence/absence)	Sullivan and Sullivan, 2006
Crop	Presence of crop fields bordering the orchard monitored (presence/absence)	Yletyinen and Norrdahl, 2008; Heroldová et al. 2007; Wiman et al. 2009
Fallow	Presence of fallows bordering the orchard monitored (presence/absence)	White et al. 1997; Butet et al. 2006; Heroldová et al. 2007; Yletyinen and Norrdahl, 2008; Sullivan and Sullivan, 2009
Kiwi	Presence of kiwifruit orchards bordering the orchard monitored (presence/absence)	Jacob, 2003
Riparian strip	Presence of river or canals bordering the orchard monitored (presence/absence)	Canova, 1992; Maisonneuve and Rioux, 2001;
Orchards cover 500	Surface covered by orchards in a circular area with a radius of 500 m centered in the sampled orchards (ha)	Sullivan and Sullivan, 2006
Crops cover 500	Surface covered by crop fields in a circular area with a radius of 500 m centered in the sampled orchards (ha)	Yletyinen and Norrdahl, 2008; Heroldová et al. 2007; Wiman et al. 2009
Grasslands 500	Surface covered by meadows and pastures in a circular area with a radius of 500 m centered in the sampled orchards (ha)	White et al. 1997; Butet et al. 2006; Heroldová et al. 2007; Sullivan and Sullivan, 2009
Flow	Use of flood or drip irrigation	Santini, 1988
Chemical weed control	Number of chemical treatments to control weed development	Sullivan et al. 1998; Jacob 2003
Fertilize	Treatment with natural fertilizer	Morilhat et al. 2007
Earthing up	Earth up in the intra-tree row	

Table 2 The first ten selected models with lower AICc values and their Akaike weights (w_i); variable names and explanation are reported in Table 1

ID	**Model**	**K**	**AICc**	**Δ_i**	**w_i**
1	Constant + Vole NF	3	429.3	-	0.278
2	Constant + Vole NF + Flow	4	431.0	1.7	0.117
3	Constant + Vole NF + Kiwi	4	431.3	2.0	0.100
4	Constant + Vole NF + Age	4	431.4	2.1	0.097
5	Constant + Vole NF + Orchards500	4	431.4	2.1	0.097
6	Constant + Vole NF + Earthing up	4	431.6	2.3	0.087
7	Constant + Vole NF + Earthing up + Kiwi	5	431.7	2.4	0.083
8	Constant + Vole NF + Flow + Age	5	432.8	3.5	0.061
9	Constant + Vole NF + Flow + Kiwi	5	433.4	4.1	0.045
10	Constant + Vole NF + Flow + Earthing up	5	433.5	4.2	0.035

Table 3 Coefficients for AICc top ranked ($\Delta_i < 2$) models and AICc > 95% certainty model average ($\Delta_i < 4$)

	Models with $\Delta_i < 2$	Models with $\Delta_i < 4$
	Coefficient	
Constant	2.912	2.290
VoleNF	1.510	1.512
Flow	-4.337	-4.359
Kiwi	2.453	2.331
Earthing up		-1.532
Age		0.670
Orchards 500		0.053

Figure captions

Fig. 1 Study area in the Piedmontese region (Italy) and Savi's pine vole abundance reported with the rank system (see methods for rank values). The dotted circle indicates the orchards in the central part of the area with high vole abundance

Fig. 2 Comparison of Savi's pine vole abundance in apple orchards that were drip irrigated or flood irrigated (A) and of vole abundance in apple orchards with or without bordering kiwifruit orchards (B)

Fig. 3 Relation of Savi's pine vole abundance in apple orchards and the number of active tunnel entrances in neighboring kiwifruit orchards; only apple orchards bordering on kiwifruit were considered (A) and relation to the numbers of active tunnel entrances counted in neighboring fields (B)

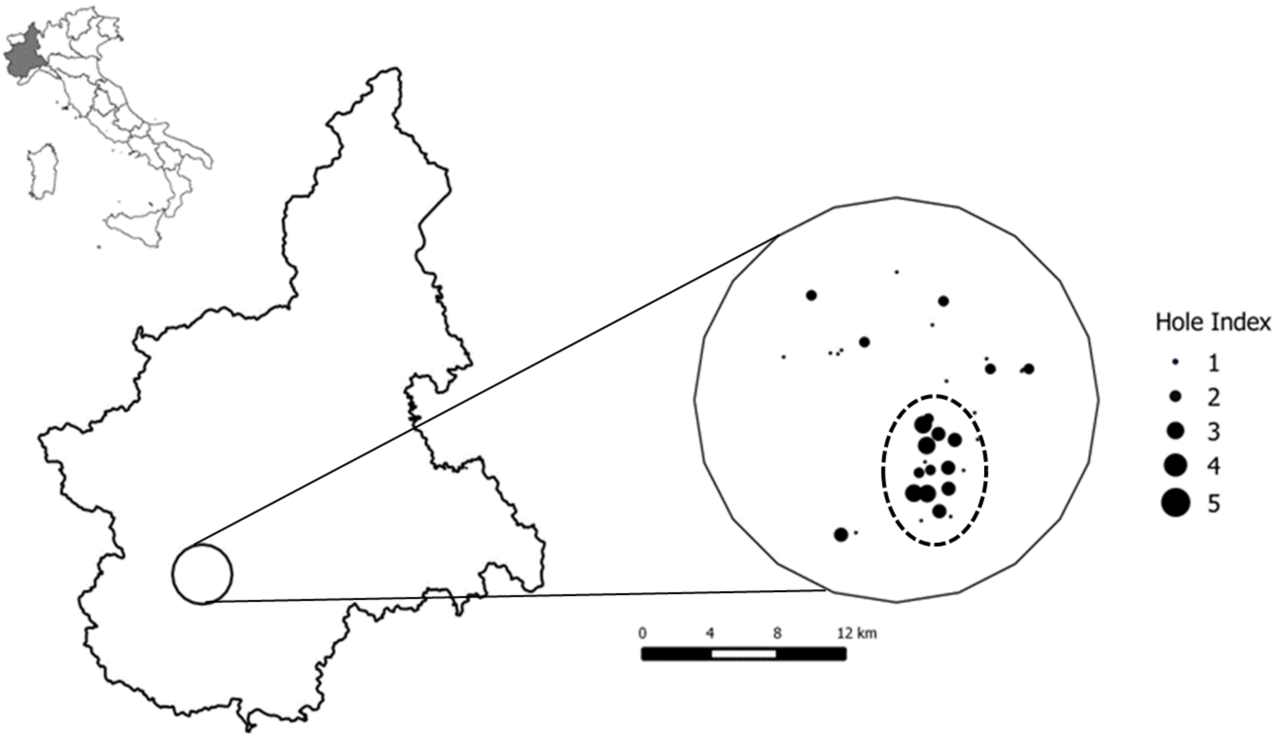

Figure 1

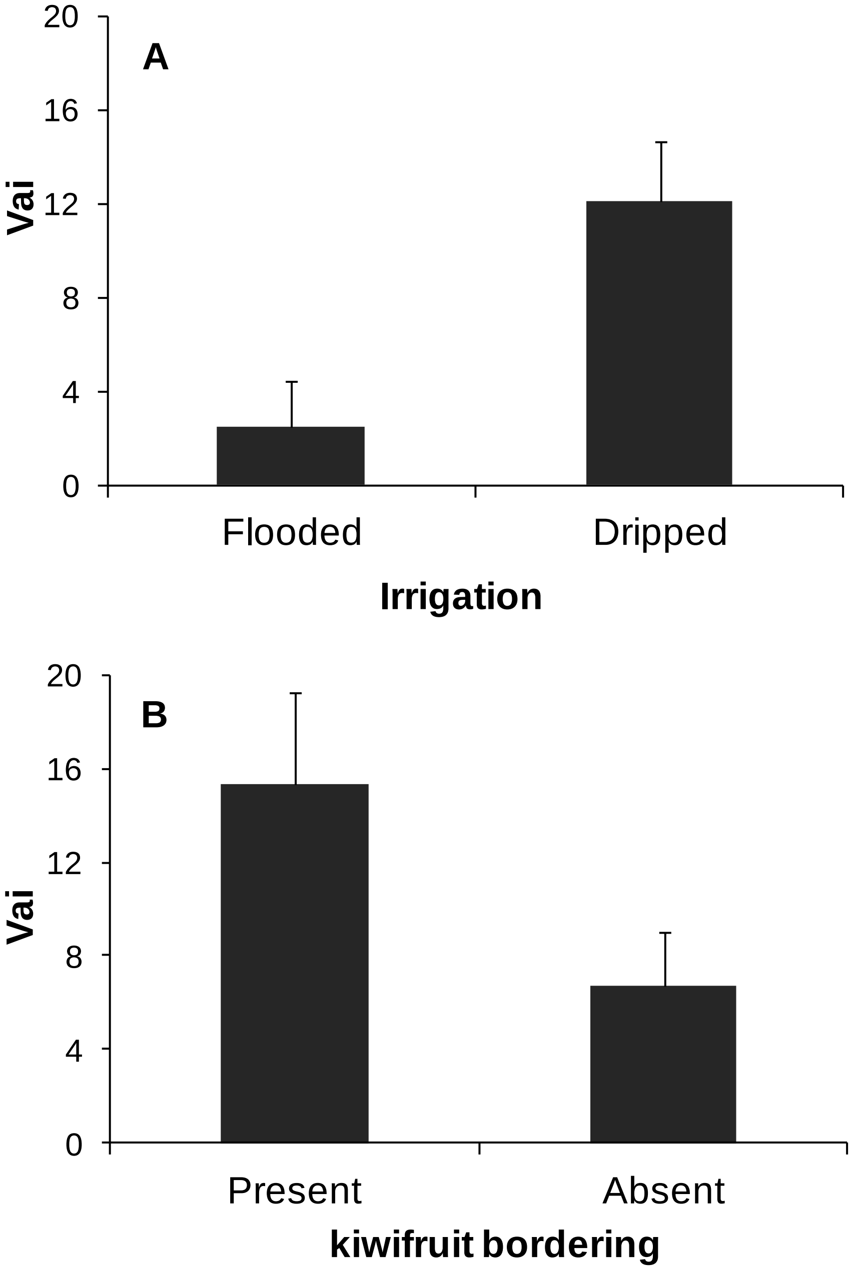
A
20
16
12
8
4
0
Vai
Flooded
Dripped
Irrigation
B
20
16
12
8
4
0
Vai
Present
Absent
kiwifruit bordering

Figure 2

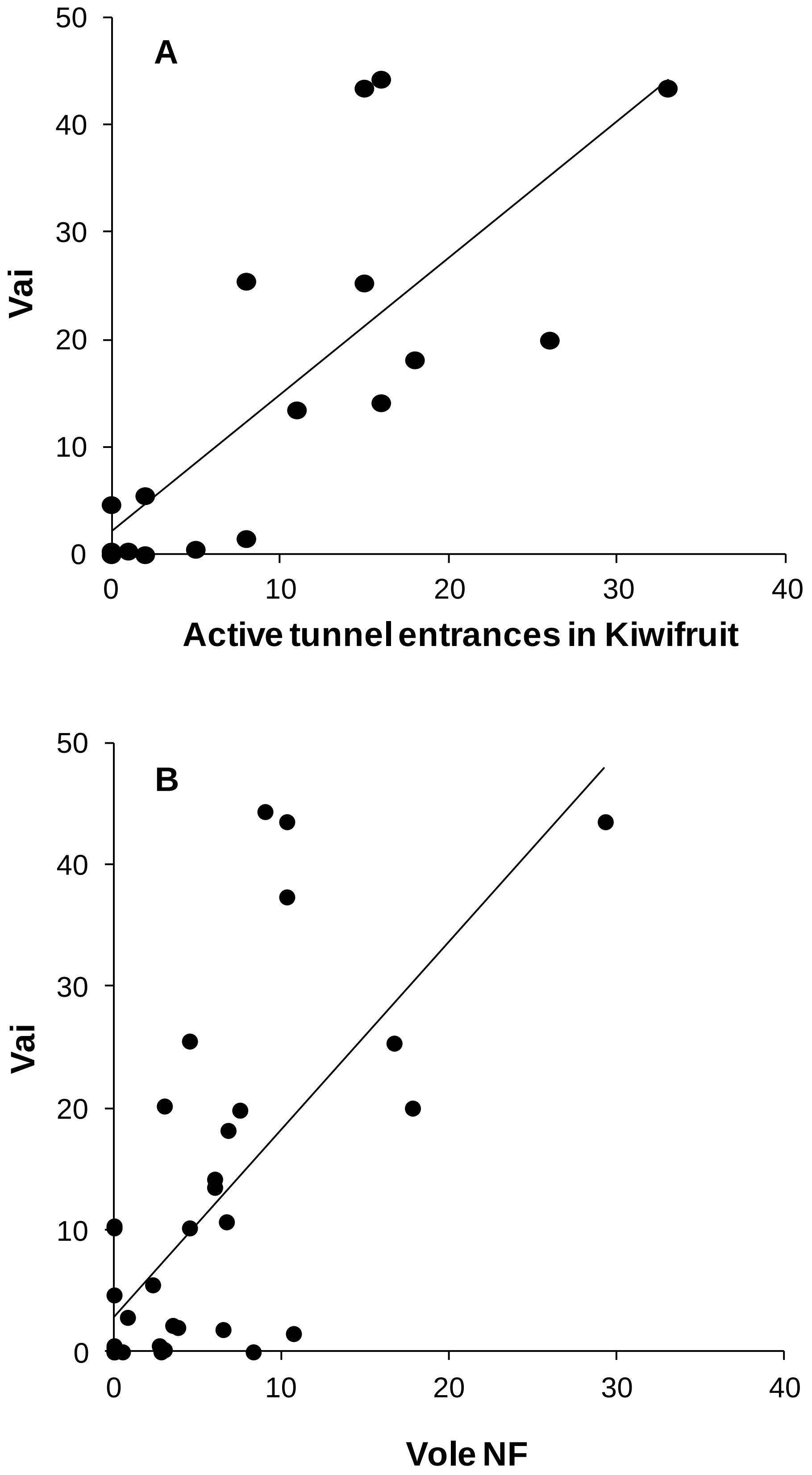
A
Vai
50
40
30
20
10
0
0
10
20
30
40
Active tunnel entrances in Kiwifruit
B
Vai
50
40
30
20
10
0
0
10
20
30
40
Vole NF

Figure 3