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Assessment of yield loss in Vanilla, *Vanilla Planifolia* Andrews (Orchidaceae) caused by the Giant African Snail, *Lissachatina fulica* (Bowdich, 1822)

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ABSTRACT: The impact of Giant African Snail, *Lissachatina* (*Achatina*) *fulica* (Bowdich, 1822) damage on quantitative and qualitative loss of Vanilla (*Vanilla planifolia* Andrews) beans produced in different cropping systems was assessed. Snails were active during night, crawled on the vanilla vines and fed on the column (united stamen and pistil) of pollinated flowers. During day time, snails hid under leaf mulch and in the irrigation channels. Feeding damage to the floral parts resulted in production of inferior quality beans with lower market marketable properties. An economic loss of Rs. 765 to Rs.1020 was recorded per inflorescence. However, the density of snails on plant surface did not indicate the corresponding level of pest damage.

Keywords: Vanilla, beans, column, *Lissachatina fulica,* damage

INTRODUCTION

Vanilla, the most widely used food flavourant is derived from beans (fruits) of the orchid, *Vanilla planifolia* Andrews (Orchidaceae), a native of South-eastern Mexico and the world's second most valuable spice crop next to saffron (Anonymous, 2020). It is used extensively to flavour ice-cream, chocolates, beverages, cakes, custards, pudding and other confectionery, also used in perfumery and to a small extent in pharmaceuticals. Traditionally, vanilla flavourings are extracted from the matured beans of *V. planifolia.* The beans are harvested before they fully ripe. Subsequently, they are fermented and cured (Purse glove, *et al.*, 1988). 'Vanillin' accounts for about 2% of the dry weight of cured vanilla beans and is the chief among about 200 other flavour compounds found in beans responsible for unique vanilla flavour. The flavour of vanilla beans is far superior to that of synthetic vanillin due to the presence of other flavour compounds in the natural product (Sudharshan *et al.*, 2006). The yield of the vanilla flavour from the processed beans is highly influenced by the length of the beans. Longer the beans, higher the price. Primarily the length of beans is an attribute of successful pollination and fertilization of vanilla beans. The main reason for shortness of beans is detachment of 'column' (united stamen and pistil) from the developing beans. *V. planifolia* has been successfully introduced as an inter-crop in arecanut (*Areca catechu* L.), coffee (*Coffea* spp.), black pepper (*Piper nigrum* L.) and cardamom (*Elettaria cardamomum* Maton) plantations in Western hilly areas of Southern India (Sudharshan and John, 2003).

Vanilla is known to plague by many annoying insect pests in India (Prakash *et al.,* 2002; Varadarasan *et al.,* 2002b). Infestation of molluscans from different parts of the world also has been reported on vanilla. Chiders and Cibes (1948) have reported the snail, *Thelidomus lima* Fer., and the slug *Veronicella kraussii* Ferussac damaging vanilla by removing outer portion or entire sections of buds, leaves, shoots and entire beans in Puerto Rico. In Southern India, incidence of *A. fulica* on *V. planifolia* was reported by Varadarasan *et al.,* (2002a).

The Giant African Snail, has been considered the most widely introduced and invasive land snail species in the world and a major agricultural and garden pest (Mead, 1979; Karnatak *et al.,* 1998; Raut and Barker, 2002; Sridhar *et al.,* 2013). In our pest surveillance studies in Karnataka, we observed that the Giant African Snail, *Lissachatina fulica* (Bowdich) was one of the factors behind shorter sized vanilla beans. Earlier reports have considered the direct damage caused by *L. fulica* on vanilla by feeding on the vegetative parts *viz.*, young leaves, tender shoots and younger plants (Varadarasan *et al.*, 2002a). Hitherto, no attempt has been made to estimate the crop loss caused by the snails obstructing the successful fertilization of the beans.

Hence, a study was undertaken to quantify the economic loss caused by the snail, *L. fulica.* In this paper the abundance of snails in different cropping systems associated with vanilla, nature of damage by the snails and the extent of economic loss incurred followed by snails has been documented.

MATERIALS AND METHODS

Study sites

Five fields were selected in two major vanilla growing districts of Karnataka in the hilly zone. Climate of Hilly zone is humid and tropical and receives an annual rainfall ranging from 719 to 5225 mm. Study was conducted during the flowering period from January-April, 2019. The locations and site characteristics are given in Table 1. Of five locations, vanilla was cultivated as an intercrop in three locations. Vines were grown under the natural shade of support trees. In areca based cropping system, vanilla vines were permitted to grow up to 1.2-1.5m on the support trees *viz*., *Erythrina indica* and *Gliricidia sepium,* even on areca itself allowed to hang down from the same. Coffee, Gliricidia and Erythrina trees were allowed to form branches to different directions to have an umbrella shaped appearance about 1.5-2.0m above the ground. At initial stages vanilla vines get attached to these support trees and are allowed to grow later. Support trees were pruned to regulate appropriate shade. In Kesuve, vanilla was cultivated as sole crop under the shade and support of Gliricidia, spacing of 2m between rows and 1.5m within a row was maintained. In Harekoppa, instead of live support trees hardwood posts and nylon ropes were used. Posts were put at 1.2 to 1.5m above the ground, ropes tied to posts and the vines being brought over them and looped up as they grow long. This artificial structure was housed under the shade of perforated agro-shade net. At all the locations mean age of the vines was 3.16±0.76 years and approximately 3000 vines occupied one hectare. Well decomposed organic matter and mulch in the form of dry leaves were placed to a thickness of 10-15cm at the base of the vines. Irrigation was provided using sprinklers. At all the sites, vanilla blooming commenced during January and February, reached its peak during March and ended during April. A trained worker carried out the hand pollination of vanilla flowers throughout the season at the respective locations. Observations were recorded twice in March at second and fourth week after flowering.

Assessment of abundance of snails

The mechanical wooden frames measuring 0.5 m^2 area were prepared and placed between plants/ structures supporting vanilla vines at 17:00 h. One such frame was placed at the centre between four palms/ vines approximately 4.5 m away from each plant base. Abundance of snails was randomly assessed from 25 marked spots in the reported plantations. At 22:00 h the number of snails in such wooden frames (0.5 m^2 areas) was counted and recorded as abundance of snail/0.5 m^2 area. Snail population was not recorded from the plant base, on the roads, near water source, near fence and near compost pits/heaps.

Assessment of abundance of snails on plant surface

At 23:00 h the number of snails that were present on 25 randomly selected vanilla vines was counted and recorded as *abundance of snail/vine.* Among 25, ten vines harboured snails were marked using coloured ribbons for assessing crop loss study. Snails of all sizes were considered for assessing abundance both on ground and plant surface.

Estimation of quantitative loss caused by snails

Assessment of crop loss due to snail damage

Ten racemes (each from a single vine) were randomly selected from the previously marked vines at 23.00h. The total number of beans (TB) and number of beans devoid of column (L1) were counted and recorded at 23:00 h. On the next day before commencement of pollination operation at 08:00 h, all the 10 previously marked racemes were re-examined and total number of beans devoid of column (L2) was counted and recorded. The number of beans devoid of column (L3) following snail infestation was estimated by calculating L3=L2-L1. The economic loss incurred was worked out for each location as Loss of column of one bean = loss of one good grade bean.

Table 1. Locations and site characteristics

District	Location	Latitude	Longitude	Altitude (m)	Main crop	Support tree/structure
Chikkamagalur	Kesuve	13.576438°	75.388038°	734	Vanilla	*Gliricidia sepium*
	Golgar	13.587221°	75.374865°	764	Area & coffee	Areca & coffee
Shivamogga	Araga	13.689496°	75.244989°	631	Areca	Areca
	Devangi	13.629998°	75.283904°	647	Areca & Banana	*Erythrina indica*, *G. sepium*
	Harekoppa	14.156828°	75.092699°	508	Vanilla	Hardwood posts

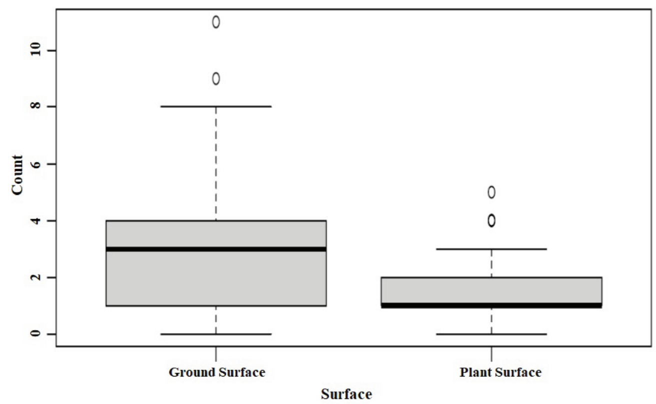

Fig 1. Distribution of snails on ground and plant surfaces

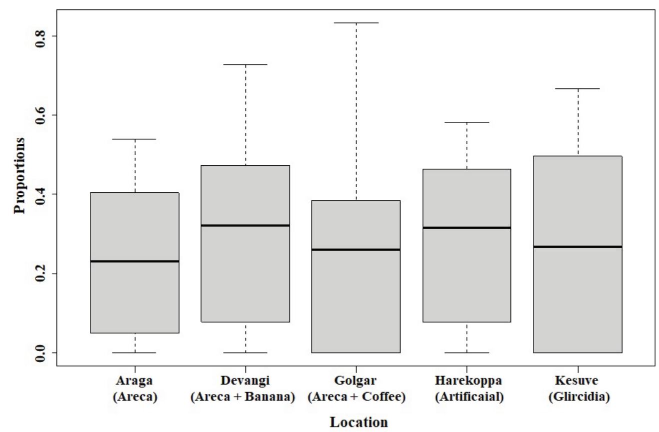

Fig 2. Proportion of yield loss in different cropping systems

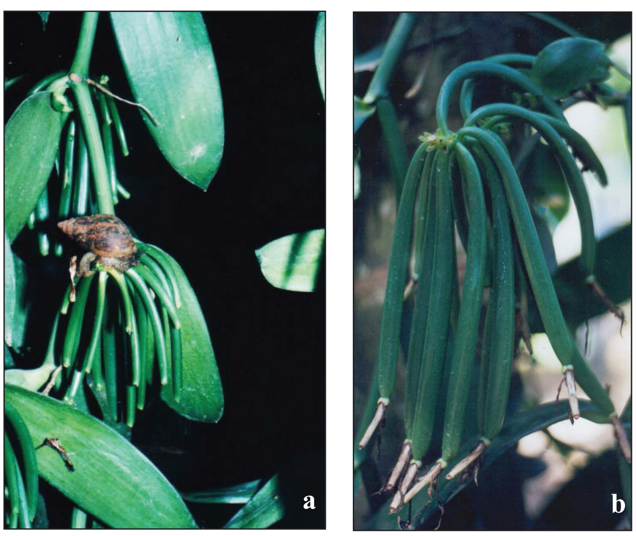

Plate 1. a.Snail on vanilla inflorescence b.Vanilla beans

Estimation of monetary loss

To estimate the monetary loss caused by the snails to vanilla, the price offered by the auctioneer for different grade of beans at auction centre was recorded from 2015 to 2018 (Anonymous, 2018). Further, the number beans that constitute one kg in different grade categories were also estimated.

Estimation of qualitative loss caused by snails

The normal (grade-I), medium quality (grade-II) and dwarf (grade-III) beans were subjected to curing and analysed for chief physical and chemical quality parameters.

Statistical analysis

The data analyses were performed using R software (Version: i386 4.0.3). The differences among different locations, weeks and surfaces were analyzed using three-way ANOVA. Mathematical pitfalls of using independent P-values of t-test to determine the utility of a multiple-variables hypothesis as a whole, *i.e* t-tests applied to three or more groups at the same time, inflates the Type I error rate, thus is considered as dishonest. Hence, Tukey's "Honestly Significant Difference" test was employed to have in-depth comparison (Hu *et al.,* 2021) between different combinations of location (cropping system), surface, and over weeks.

Table 2. Abundance of *Lissachatina fulica* in different cropping systems at study locations

Location	Abundance of snails					
	Snails on ground surface (No. of snails/0.5 m^2 area)			Snails on vine (No. of snails /vanilla vine)		
	II week	IV week	Mean	II week	IV week	Mean
Kesuve (Gliricidia)	2.36±1.46	1.80±1.22	2.08±1.34	1.32±1.08	1.28±0.97	1.30±1.02
Golgar (Areca+coffee)	2.48±1.58	2.30±1.65	2.40±1.61	1.08±0.95	0.80±0.91	0.94±0.93
Araga (Areca)	4.64±1.35	4.44±2.23	4.54±1.79	2.16±1.10	1.48±1.22	1.82±1.16
Devangi (Areca+Banana)	3.64±1.72	2.76±1.78	3.20±1.75	1.48±0.96	1.28±1.08	1.38±1.01
Harekoppa (Artificial)	1.36±1.41	0.92±1.03	1.14±1.22	1.32±0.98	1.64±1.11	1.48±1.04
3 Factor ANOVA (R^2: 0.75)						
Week	Surface	Location	Week x Location	Location x Surface	Week x Surface	Location x Week x Surface
5.264*	117.5***	21.8178***	11.1**	38.622***	3.16*	43.7***

Table 3. Results of post-hoc multiple comparison test: Tukeys HSD on abundance of snail over combinations of different weeks/ surface/ cropping systems

Location (Cropping System)	Tukey (HSD)	p-Adjusted
Comparison of over different locations (Cropping System)		
Devangi (Areca+Banana) & Araga (Areca)	-0.89 **	0.00
Golgar (Areca+coffee) & Araga (Areca)	-1.49 **	0.00
Harekoppa (Artificial) & Araga (Areca)	-1.87 **	0.00
Kesuve (Gliricidia) & Araga (Areca)	-1.49 **	0.00
Golgar (Areca+coffee) & Devangi (Areca+Banana)	-0.60 *	0.05
Harekoppa (Artificial) & Devangi (Areca+Banana)	-0.98 **	0.00
Kesuve (Gliricidia) & Devangi (Areca+Banana)	-0.60 *	0.05
Comparison for given week		
Plant surface:W2 & Ground surface:W2	-1.44 **	0.00
Plant surface:W4 & Ground surface:W4	-1.15 **	0.00
Comparison over week & surface		
Plant surface:W4 & Ground surface:W2	-1.62 **	0.00
Ground surface:W4 & Plant surface:W2	0.98 **	0.00
Comparison over week & cropping systems		
W2:Golgar (Areca+coffee) & W2:Araga (Areca)	-1.58 **	0.00
W2:Golgar (Areca+coffee) & W4:Araga (Areca)	-1.14 **	0.01
W2:Harekoppa (Artificial) & W2:Araga (Areca)	-2.06 **	0.00
W2:Harekoppa (Artificial) & W2:Devangi (Areca+Banana)	-1.22 **	0.00

W2:Harekoppa (Artificial) & W4:Araga (Areca)	-1.62 **	0.00
W2:Kesuve (Gliricidia) & W2:Araga (Areca)	-1.56 **	0.00
W2:Kesuve (Gliricidia) & W4:Araga (Areca)	-1.12 **	0.01
W4:Devangi (Areca+Banana) & W2:Araga (Areca)	-1.38 **	0.00
W4:Golgar (Areca+coffee) & W2:Araga (Areca)	-1.84 **	0.00
W4:Golgar (Areca+coffee) & W2:Devangi (Areca+Banana)	-1.00 *	0.05
W4:Golgar (Areca+coffee) & W4:Araga (Areca)	-1.40 **	0.00
W4:Harekoppa (Artificial) & W2:Araga (Areca)	-2.12 **	0.00
W4:Harekoppa (Artificial) & W2:Devangi (Areca+Banana)	-1.28 **	0.00
W4:Harekoppa (Artificial) & W4:Araga (Areca)	-1.68 **	0.00
W4:Kesuve (Gliricidia) & W2:Araga (Areca)	-1.86 **	0.00
W4:Kesuve (Gliricidia) & W2:Devangi (Areca+Banana)	-1.02 **	0.04
W4:Kesuve (Gliricidia) & W4:Araga (Areca)	-1.42 **	0.00
Comparison over different surface and cropping systems		
GS: Devangi (Areca+Banana) & GS: Araga (Areca)	-1.34 **	0.00
GS: Devangi (Areca+Banana) & PS: Araga (Areca)	1.38 **	0.00
GS: Golgar (Areca+coffee) & GS: Araga (Areca)	-2.10 **	0.00
GS: Golgar (Areca+coffee) & PS: Devangi (Areca+Banana)	1.06 **	0.00
GS: Harekoppa (Artificial) & GS: Araga (Areca)	-3.40 **	0.00
GS: Harekoppa (Artificial) & GS: Devangi (Areca+Banana)	-2.06 **	0.00
GS: Harekoppa (Artificial) & GS: Golgar (Areca+coffee)	-1.30 **	0.00
GS: Kesuve (Gliricidia) & GS: Araga (Areca)	-2.46 **	0.00
GS: Kesuve (Gliricidia) & GS: Devangi (Areca+Banana)	-1.12 **	0.00
GS: Kesuve (Gliricidia) & GS: Harekoppa (Artificial)	0.94 *	0.02
GS: Kesuve (Gliricidia) & PS: Golgar (Areca+coffee)	1.14 **	0.00
PS: Araga (Areca) & GS: Araga (Areca)	-2.72 **	0.00
PS: Devangi (Areca+Banana) & GS: Araga (Areca)	-3.16 **	0.00
PS: Devangi (Areca+Banana) & GS: Devangi (Areca+Banana)	-1.82 **	0.00
PS: Golgar (Areca+coffee) & GS: Araga (Areca)	-3.60 **	0.00
PS: Golgar (Areca+coffee) & GS: Devangi (Areca+Banana)	-2.26 **	0.00
PS: Golgar (Areca+coffee) & GS: Golgar (Areca+coffee)	-1.50 **	0.00
PS: Golgar (Areca+coffee) & PS: Araga (Areca)	-0.88 *	0.04
PS: Harekoppa (Artificial) & GS: Araga (Areca)	-3.06 **	0.00
PS: Harekoppa (Artificial) & GS: Devangi (Areca+Banana)	-1.72 **	0.00
PS: Harekoppa (Artificial) & GS: Golgar (Areca+coffee)	-0.96 *	0.01
PS: Kesuve (Gliricidia) & GS: Araga (Areca)	-3.24 **	0.00
PS: Kesuve (Gliricidia) & GS: Devangi (Areca+Banana)	-1.90 **	0.00
PS: Kesuve (Gliricidia) & GS: Golgar (Areca+coffee)	-1.14 **	0.00

RESULTS AND DISCUSSION

Snail's activity commenced at dusk around 08:00 h at all the locations. During day time, snails hid below the mulch and cooler parts in the plantations. Usually, snails took shelter in cooler irrigation channels in the vanilla garden. Snails crawled on the ground surface. Later they marched towards vanilla/areca crops and started climbing the trees. The snails took the vines in 2 to 3 hours. Snail abundance varied across the cropping pattern. In the intercropping system snail abundance was as high as 4.54±1.79 where as sole crop garden recorded 2.08±1.34 snails. Snail abundance was the highest in the areca and area+banana cropping system (Table 2) compared to artificial system.

Although, average number of snails observed was 1 to 2 on plant surface and 2 to 4 on ground surface, the variability was very high between weeks, surface and over different locations. Post-hoc test *viz*., Tukeys HSD test employed in the study captured higher variability present and enhanced the ability to detect the statistical significance. Comparisons between different combinations of locations (cropping system), surface, and over weeks were further tested using Tukeys HSD. The combinations which have exhibited significant difference are enlisted in Table 3. Other combinations not mentioned in the paper exhibit no significance difference with respect to snail infestation.

Snail abundance on vanilla grown as an intercrop in areca (vines trailed on areca at Araga) was found highly significant followed by vines trailed on Areca+Banana (Devangi) cropping system. Other cropping systems *viz.,* vanilla grown as sole crop (vines trailed on Gliricidia), as an intercrop (vines trailed on Areca+coffee) and a sole crop (vines trailed on hardwood posts/ artificial) cropping systems have similar snail density. Banana plants are heavy water feeder which makes a suitable microclimate for the development of moist loving snails (Umashankar *et al.*, 2016). This substantiates that infestation on Areca+Banana was more compared to Areca+coffee cropping system. However, Vanilla trailed on artificial support system had the least snail infestation (Table 3).

Abundance of snails on ground was significantly high compared to plant surface for a given week (Table 3). The week *per se* had no significant influence on number of snails either on ground or plant. However, within 2nd and 4th week, presence of snails on ground contributed to overall significance.

Irrespective of surface, abundance of snail reduced over the second fortnight and the reduction is significant. Vanilla vines trailed on areca at Araga harboured higher number of snails compared to all other cropping systems. This is followed by vines trailed on Areca+ Banana (Devangi) cropping system over both weeks. Abundance on Areca+Coffee cropping system was significantly less compared to Areca+Banana. Intrestingly, artificial support system had the least number of snails over both weeks and the reduction is non-significant (Table 3).

Irrespective of week, pattern of snail abundance was entirely different on plant and ground surfaces. Abundance was significantly least on ground in artificial support system, while on plant surface in Areca+Coffee cropping system (Table 3). While the abundance remained significantly high on both surfaces in Areca and Areca+Banana cropping systems. Snail abundance on vines in artificial support system was on par with vines trailed on Gliricidia and Areca+Banana cropping system.

It was clear that both the abundance of snails and variability in number of snails over ground surface was higher compared to its abundance and variability on plant surface (Fig 1). It is obvious that pest variability should be less for successful control of pests. In actual sense, when abundance of snails was considered, the control measures should be targeted on snails prevailed on ground surface, as the variability in this count is high that forces us to not just rely on one control measure with respect to ground surface. As, the abundance of snails was sparse on plant surface hand-picking and destroying would be more economical while, integrated approach would be highly effective with respect to ground surface.

Assessment of quantitative crop loss due to snail damage

Snails caused damage to column in following ways: (1) They crawled on the distal end of the beans at which point flower is borne; the column drops off by the virtue of snail's body weight (2) Snails landed on the shoot/beans, dragged the column portion of the surrounding beans and later devoured the column along with dried sepals and petals. Damage was limited to newly pollinated beans. An economic loss of Rs. 765 to Rs.1020 was recorded per inflorescence at all the location. The population density on plant surface did not indicate the corresponding level of pest damage and the economic loss caused by snails at the study sites (Table 4).

Proportion of yield loss was calculated by taking snail damage (loss of column) to the total bean observed (L3/Total). 2 Factor ANOVA showed that no significance difference (R^2 =0.053) between weeks, locations and over both week and locations (Table 5). Figure 2 indicates that proportion of loss over different locations have similar

Table 4. Impact of snail damage on vanilla beans

Location	II week after pollination (n=10 inflorescence)	IV week after pollination (n=10 inflorescence)	Total Number of beans damaged by snails	
	Number of beans damaged by snails (L3)	Number of beans damaged by snails (L3)	Mean Number of beans damaged by snails (L3)	Economic loss (Rs.)
Kesuve (Gliricidia)	3.3±2.71	3.0±2.62	6.3±2.21	945±332.03
Golgar (Areca+coffee)	2.3±2.31	2.8±2.52	5.1±3.90	765±585.02
Araga (Areca)	3.4±2.41	2.9±2.64	6.3±3.40	945±515.02
Devangi (Areca+Banana)	3.1±2.37	3.1±2.68	6.2±2.09	930±314.64
Harekoppa (Artificial)	2.7±2.16	4.1±2.76	6.8±2.52	1020±379.47

* The Price of one glade-I bean was Rs.150.

mean and variability though seems to be high, it is not enough to exhibit significance in its contribution towards loss. Albeit, the abundance of snails was the least on ground in artificial trailing system, loss incurred was high in artificial trailing system.

Assessment of qualitative crop loss due to snail damage

At the time of harvest, the beans needed to make one kilogram was assessed for all three grades. On an average 75.51±5.5 fresh beans constituted one kilogram in grade-I. However, more than 150 beans were needed to make one kilogram in grade II (146.9±3.80) & grade-III beans (169.83±6.20). The chief physical and chemical quality parameters of the cured beans varied among three grades. Grade-I beans underwent normal curing process and become fleshy, supple, very dark brown to black in colour, somewhat oily in appearance, strongly aromatic and yielded good quality beans (vanillin: 2.13%; moisture: 26.70%). The grade-II beans became hard, dry, thin, brown or reddish brown and possess a poor aroma and yielded less vanillin (1.58%) and low moisture content (21.30%). The grade-III beans failed completing the curing process and become dry and shrivelled after slow drying. These beans were disposed to prevent the spoilage of other good grade beans in the storage. Although, the intensity of snail's damage was not conspicuous at field level, it caused appreciable loss in the marketable parameters of the vanilla beans due to snail feeding. Vanilla is sold in different forms: beans, extract and essence, powdered and vanilla sugar. The processed vanilla beans are sold as whole pods packed in small plastic/glass containers in specialty grocery stores and health centres, called 'Gourmet quality or for extraction'. Gourmet quality beans ought to be top quality beans that are long, fleshy, supple, very dark brown to black in colour, somewhat oily in appearance. Though there can be about 18-20 racemes in a plant and as much number of flowers, it was suggested to pollinate first 10 or 12 flowers and removing terminal flowers. Similarly, it is advised to maintain only 10-12 racemes per plant in order to get beans with maximum size and of high-quality standards. In this context any kind of loss (quantitative and qualitative loss) incurred to vanilla beans adversely affects the economic value of the end product.

The major reason for shortness of beans is improper fertilization. Impact of failure of pollination is highly conspicuous in the field as it is evident by fall off of the column (united stamen and pistil) from the flower within 24 hours after the pollination. Maximum growth rate (about 80 per cent length) of beans was observed in the first 45 days after pollination. Our study showed that the snails preferred columns of developing vanilla beans of less than 15 days old (Plate 2). This clearly indicated the role of snail's feeding damage and further yield loss. In recent years, rapid spread of *L. fulica* in the vanilla gardens is posing severe threat to the production of quality vanilla beans. Hence, prevention of snail's damage during vanilla flowering period about 150 days (January to May) helps in production of good grade beans. Maintenance of high humidity in the vanilla garden during flowering period is an essential agronomic practice which extends the viability of the flower opening and receptivity time of the stigma. Humidity has been found to be a more reliable predictor of naturalization and activity of *L. fulica* than temperature, and continuous activity is restricted largely to areas with 80% relative humidity (Raut and Barker, 2002). Hence, suppression of snail's population during flowering period in vanilla gardens offers a new challenge.

Table 5. Proportion of yield loss in different cropping systems

Location	W2	W4	Average
Devangi (Areca+Banana)	0.331	0.280	0.306
Golgar (Areca+coffee)	0.281	0.206	0.244
Kesuve (Gliricidia)	0.342	0.214	0.278
Araga (Areca)	0.296	0.194	0.245
Harekoppa (Artificial)	0.273	0.315	0.294
Average	0.305	0.242	0.273
Week	Location	Week * Location	R^2
1.97^{NS}	0.32^{NS}	0.43^{NS}	0.053

In this paper we have documented for the first time the crop loss caused by the *L. fulica* on vanilla by feeding on floral parts under different cropping systems. Though abundance of snail varied across different cropping systems, weeks and locations, the loss incurred is almost same in all. This shows that the density of snail is just a number with less economic significance. It implies that the density of snail (threshold levels) should not be considered while deciding on the plant protection measures. Even lesser numbers can lead to serious concerns. This study suggests a need for further investigation on the management of the snails in the vanilla gardens.

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