Scientific Note

Comparison of Sampling Intensity to Estimate Infestations of Coffee Berry Borer on Hawaii

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Abstract. Sampling coffee fruit for coffee berry borer, *Hypothenemus hampei* (Ferrari), infestation can be a labor-intensive task. We compared three berry sampling intensities (count infestation on all berries per branch, the currently recommended procedure; on five berry clusters; and on three berry clusters), to determine whether reduced sample sizes resulted in a loss of accuracy in estimating proportion of berries damaged. Results show that sampling three or five clusters of berries per tree would significantly reduce sampling effort, with no significant change in accuracy of the estimated proportion damaged by coffee berry borer.

Key words: *Hypothenemus hampei,* sampling, IPM

The coffee berry borer, *Hypothenemus hampei* (Ferrari) (Coleoptera: Curculionidae), is widely considered as the most serious pest of coffee worldwide (Chaves and Riley 2001, Jaramillo et al. 2006). Females of this tiny beetle (adult size 1.2–1.8 mm) bore into developing or mature coffee fruit and construct irregular galleries to lay eggs (Baker et al. 1992). Drilling by egg-laying females and feeding by newly hatched larvae damages coffee beans inside the fruit. The pest was first detected in Hawaii in 2010 from coffee farms in the Kona district of Hawaii island (Burbano et al. 2011) and was subsequently detected from Oahu in 2014 (HDOA 2014). Genetic analysis of the beetles collected from Hawaii has suggested that the pest was accidently introduced from Latin America (Chapman et al. 2015). Being an introduced pest in Hawaii, no specialist natural enemies of this pest are currently known; however, predation by some flat bark beetles (Sylvanidae) has been reported (Follett et al. 2016). Integrated pest management practices for this pest are currently under development in Hawaii.

Several attempts have been made by various researchers to develop an effective sampling plan for coffee berry borer such as “a random sample of 80 clusters from a randomly selected coffee tree” (Puntener 1973), random sample of 2000 fruits (Decazy et al. 1989), and sampling based on attractive baited traps (Dufour and Frérot 2008, Fernandes et al. 2011). It is apparent from the published literature that there is no specific sampling method that is widely adopted because of various practical reasons (Wegbe et al. 2003, Fernandes et al. 2011). Current recommendations for managing CBB in Hawaii include sampling based on the CENICAFE method which primarily comprises a “modified 30 tree sampling” to detect pest incidence and damage and to identify optimum time to apply commercial formulations of *Beauveria bassiana,* an entomopathogenic fungus (Kawabata et al. 2015), coupled with field sanitation and agronomic practices. The CENICAFE process of sampling involves identifying a branch from the middle of canopy that has at least 45, or more, green coffee berries and counting all infested and non-infested berries. The process is then repeated for 30 trees selected randomly to estimate percent infestation rate in the field. It is not unusual to have hundreds of berries on a single branch and counting all of these berries is often a daunting task, and too labor intensive for many farmers. The objective of this study was to examine if the CBB infestation rate could be reliably estimated by counting a reduced number of berries in subsamples, compared to the whole-branch based method.

Materials and Methods

Study area. This study was conducted at two locations; Kona and Kau districts of Hawaii island in 2014. Two commercial coffee farms were selected at relatively high and low elevations from each of these locations. At Kona, the first farm (Farm-1) was about 580 m above MSL near Kaloko area and the second farm (Farm-2) was located at about 242 m above MSL near Napoopoo. At Kau, the first farm (Farm-3) was located at about 500 m above MSL and second farm (Farm-4) at about 300 m above MSL, both near Pahala. The elevations selected for the study represent major coffee growing areas in each of these two locations. Coffee plants grown at these farms were *Coffea arabica* L., variety 'Kona typica', planted at about 1–1.5 m (4–6 ft) apart in rows spaced at 3–3.5 m (10–12 ft) and had an age of about 4 to 18 years. Because shade has been reported as a factor in increasing CBB infestations (Mariño et al. 2016), most of the areas selected for the study from each of these farms were devoid of any significant shaded areas, except in farm-3, which was shaded to some extend from windbreaks on the boarder. Both farms at Kona were irrigated using drip irrigation, but the farms at Kau were rain-fed. Pruning at the time of the study was done using the Kona style, in which one or two vertical branches are pruned in successive years. All the farms selected had low to moderate levels slope, and the rows were planted across the direction of the slope. In general, all the farms had comparable landscape characteristics.

Plot layout. The experimental design was a randomized complete block design and the treatments were three sampling methods with 20 replications. Number of replications was based size of the selected plots and comparable to CENICAFE method, based on the area of the plot. Individual coffee trees were selected as experimental units. Experimental units (trees) were arranged in every third row in a block (9–10.5 m apart) and separated by two coffee trees (3–4.5 m apart) within a row. The blocks were arranged across the rows and along the slope of the study area. The three sampling methods included estimation of percent infestation by counting of all infested and non-infested green berries on 1) one branch per tree or 2) five clusters of berries on five different branches on a tree or 3) three clusters on three different branches on a tree. A cluster was defined as all the berries from a single node on a branch. A berry was considered infested when a clear entry hole was present at the blossom end. The first set of observations was taken on 6/13/2014 from Kona and on 7/2/2014 from Kau. A repeat observation was taken on 6/27/2014 from Kona and 7/17/2014 from Kau. We sampled each experimental unit (coffee tree) by randomly selecting a branch or nodes on a branch and counting all berries on a branch or berries from fixed number of nodes based on the experimental design. When berries from three or five nodes were sampled from a tree, we selected three or five different branches by walking around the tree and randomly selecting branches from the middle-height level of the canopy. Red cherries and small berries less than the size of a pea were not included in sampling.

Statistical analysis. Two variables were compared statistically: 1) proportion of infested berries detected by each method and 2) total number of berries counted by each method. Proportional data were arcsine transformed to normalize the data. No transformation was done on count data. Data from two sampling dates were averaged for the analysis. One-way ANOVA was carried out in SAS 9.3 (SAS Institute 2016) using PROC GLM and the LSMEANS statement was used to compare the means.

Results and Discussion

The four farms studied had significantly different levels of coffee berry borer infestation ($F_{3;471}$ = 239.71; P < 0.05). Mean proportion of infested berries were 0.11 ± 0.00 (11.6%), 0.02 ± 0.00 (2.1%), 0.48 ± 0.02 (48.5%) and 0.37 ± 0.02 (37.6%) respectively in Farm-1, Farm-2, Farm-3, and Farm-4. Overall, there was no significant difference in estimated mean proportion of berries infested based on sampling method ($F_{2;453}$ = 0.58; P = 0.56) (Fig.1). The experimental blocks had no significant effect on the estimation of infestation overall ($F_{19;453}$ = 0.59; P = 0.91). The results were similar when analysis was done based on individual farms except for Farm-1 (Table 1). Farm-1 had relatively more berries per cluster and this may have contributed to a different mean (Table 2). There was a significant difference among these three methods of sampling in mean total number of coffee berries (denominator) counted to estimate the proportion of infested berries ($F_{2;451}$ = 188.68; P < 0.05) (Fig. 2). The block effect was not a significant factor for denominator, overall ($F_{19;451}$ = 0.71; P = 0.81). Individual farms showed similar results when analyzed separately (Table 2).

The plots selected for the study had varying levels of infestation that ranged from low (Farm-1) to very high (Farm-3) at the time of sampling. Despite having varying levels of infestation, all the three sampling methods were equally effective in estimating the infestation levels in these plots. Labor-saving was clearly demonstrated, with 26.4% fewer berries sampled using the five-cluster method, and 53.6% fewer berries using the three-cluster method. Up to 205 berries/branch were counted in Farm-2 using the ‘one branch’ method and the maximum number of berries counted per branch was always above 100 in other fields. In contrast, the maximum number of berries counted did not exceed 95 with the “five clusters” method and did not exceed 58 with the “three clusters” method.

This study indicates that CBB infestation could be estimated with comparable accuracy by sampling and counting significantly fewer berries than required with the current recommended sampling plan (Kawabata et al. 2015). The current recommendation is to “randomly select a lateral branch in the middle of the tree with at least 45 berries (more is better).” Putting an upper limit for the number of berries to be counted/per branch would save considerable time and effort for the sampler. Coffee trees that are bearing for the first time after a pruning period usually have hundreds of berries per branch and very low damage early in the season (unpublished data). Counting all the berries on the branch to calculate percent infestation in these circumstances might be totally unnecessary and an overinvestment in effort. Based on this study, an upper limit of 50–60 berries per branch would be sufficient to reasonably estimate percent CBB infestation. The study was

Table 1. Mean proportion of berries infested (± SEM) estimated by three sampling intensity methods at four different coffee farms on Hawaii island.

Farm	Method of sampling	Mean ± SEM
Farm 1	One branch	0.11 ± 0.01a
	Five clusters	0.14 ± 0.02a
	Three clusters	0.09 ± 0.01b
Farm 2	One branch	0.01 ± 0.00a
	Five clusters	0.02 ± 0.00a
	Three clusters	0.03 ± 0.01a
Farm 3	One branch	0.49 ± 0.04a
	Five clusters	0.48 ± 0.03a
	Three clusters	0.48 ± 0.04a
Farm 4	One branch	0.41 ± 0.04a
	Five clusters	0.38 ± 0.04a
	Three clusters	0.34 ± 0.04a

Means followed by the same letter were not significantly different, P = 0.05.

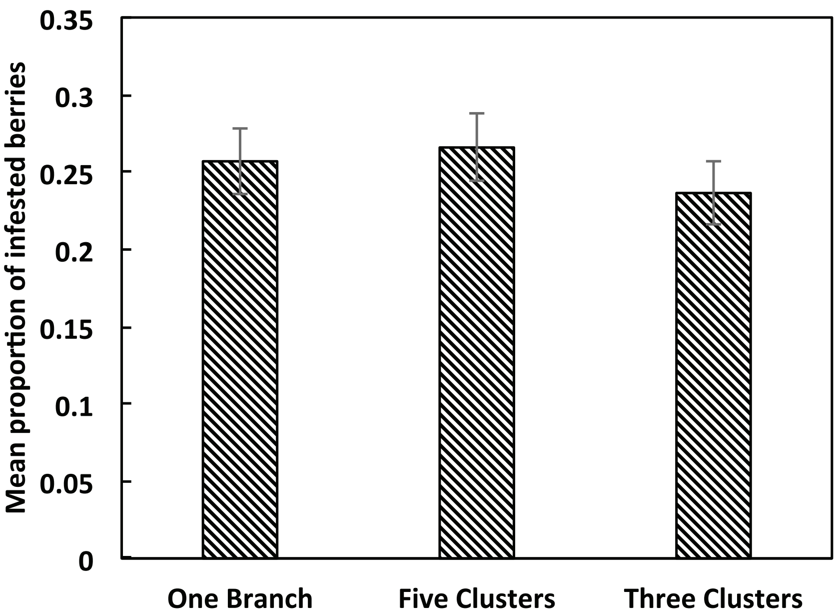

Figure 1. Mean proportion of infested berries ± SEM estimated by counting berries using three sampling intensities. The data are averages from four farms on Hawaii island.

Table 2. Mean total number of berries counted ± SEM to estimate proportion of berries infested by three sampling methods in four different coffee farms on Hawaii island.

Farm	Method of sampling	Mean ± SEM
Farm 1	One branch	82.63 ± 2.94a
	Five clusters	63.29 ± 1.92b
	Three clusters	41.50 ± 1.15c
Farm 2	One branch	92.88 ± 5.93a
	Five clusters	57.95 ± 2.12b
	Three clusters	36.70 ± 1.11c
Farm 3	One branch	56.85 ± 3.30a
	Five clusters	51.77 ± 1.32b
	Three cluster	31.33 ± 1.02c
Farm 4	One branch	72.83 ± 3.74a
	Five clusters	51.42 ± 1.11b
	Three clusters	32.03 ± 0.63c

Means followed by the same letter were not significantly different, P = 0.05.

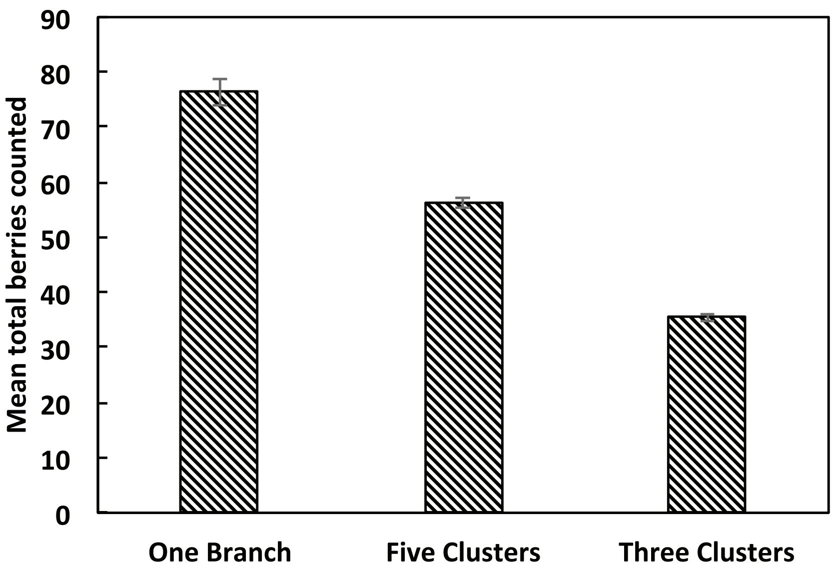

Figure 2. Mean total number of coffee berries counted ± SEM to estimate proportion of berries infested. The data are averages from four farms on Hawaii island.

conducted on a representative area (about 0.4 ha) of the selected farms and the total area of the farms ranged from 2 ha to 4 ha. Further studies are being conducted to compare these methods when field-wide samplings are done.

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