

# Development and validation of sequential sampling plans for *Sitophilus* species associated with pet specialty stores

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## Abstract

Eight pet stores in Kansas were sampled between February and August 2002, using traps baited with food and pheromone lures for capturing multiple species of beetle adults. Thirty traps were arranged in a grid pattern in each store and were checked every 2–3 weeks. The most common and abundant beetles captured in traps in all eight stores were the *Sitophilus* spp. (rice, granary, and maize weevils). The rice weevil, *Sitophilus oryzae*, was the most common and predominant of the three weevils. Trap capture data from each store were used to calculate mean numbers of *Sitophilus* spp./trap/week and associated variance and weekly presence/absence of adults. About 60% of the weekly trap capture data from the 8 stores were used for developing fixed precision and binomial sequential sampling plans and the other 40% of the data were used for testing the performance of these plans through computer simulations using the "Resampling for Validation of Sampling Plans" software. Green's fixed precision sampling plan was used for estimating *Sitophilus* spp. density of 0.62 insects/trap/week that corresponded with 50% of infested traps. The actual precision and sample sizes needed for estimating density at the fixed precision levels of 0.25, 0.35, and 0.50 were determined. Wald's sequential probability ratio test plan and a fixed sample size binomial plan were developed to classify infestation level with respect to an infestation threshold (50% of infested traps). Operating characteristic and average sample number curves generated using the validation data sets were used to gauge performance of the binomial plan. In addition, the actual errors in classifying infestation levels were also determined. The development, performance, and utility of these sampling plans in retail stores are discussed.

**Keywords:** Retail stores; *Sitophilus* spp.; Trapping; Sampling plans

## Introduction

Estimation of pest density (number of insects/sample) or classification of infestation levels (proportion of infested sample units) with reference to an action threshold is central to any integrated pest management (IPM) program. In theory, when the pest population density reaches the economic threshold an intervention should occur to prevent the pest population from reaching the economic injury level

and causing economic loss (Pedigo, 1996). Methods for estimating pest density or classification of infestation levels can be time consuming due to aggregated pest dispersion patterns and sampling costs. Use of traps, a relative sampling method, is one way to reduce the time and labour needed for absolute sampling. A second time and labour-saving technique is the implementation of cost-effective sampling plans. Sequential sampling plans use variable numbers of sample units, making them less expensive than methods based on a fixed number of sample units (Waters, 1955). Sterling (1975) estimates that sequential plans generally need 40–60% fewer sample units than plans requiring a fixed number of sampling units.

Sampling plans that rely on a fixed number of samples have been developed for stored-product insects associated with bulk-stored grain (Hagstrum et al., 1985; Hodges et al., 1985; Subramanyam and Harein, 1990; Subramanyam et al., 1993; 1997). Subramanyam et al. (1997) developed a sequential sampling plan for estimating insect density of the rusty grain beetle, *Cryptolestes ferrugineus* (Stephens), infesting farm-stored wheat. Sampling plans for classifying infestation levels relative to a threshold have been developed based on presence/absence or binomial methods (Binns, 1994).

*Sitophilus oryzae* (L.), *Sitophilus granarius* (L.), and *Sitophilus zeamais* (Motschulsky) are internally infesting stored-product insects that were found to be abundant in 8 retail pet stores in Kansas during 2001 (Rennie Roesli, unpublished data). Generally, they are whole grain feeders (i.e. bird food), but we also observed them feeding on extruded products such as dog chow during the study. Adults and larvae consume large amounts of food and are adults are particularly adept at chewing holes through packaging. The complete life cycle takes less than a month at typical retail store temperatures ( $\geq 22^{\circ}\text{C}$ ) and the adults live for 5–8 months. Females typically lay 300–400 eggs, resulting in large pest populations in relatively little time.

To our knowledge, sampling plans for estimating insect density or classifying infestation levels in retail pet stores have not been developed. Our objectives were to develop sampling plans for estimating density or classifying infestation level of *Sitophilus* spp. in retail pet stores, and test the performance of these plans through computer simulations.

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## Materials and methods

Eight retail pet stores (mean floor surface area = 1,772 m<sup>2</sup>) located in eastern Kansas, USA, were sampled for insects using food-baited traps (Flit-Trak or Dome traps, Trécé, Inc., Salinas, CA, USA). Pet specialty stores exclusively sell raw and processed food products for dogs, cats, small animals, and birds. These stores also sell animal-care products such as shampoos, toys, cat litter, and animal byproducts. Thirty traps were placed underneath shelves or kick plates in each store in a grid pattern. Each trap was baited with 15 drops of food oil in addition to pheromone lures (Mullen, 1994). Each trap had three different pheromone lures for *Lasioderma serricornis* (F.), *Stegobium paniceum* (L.), *Trogoderma* spp., and *Tribolium* spp. The food oil was replaced every 2–3 weeks and the pheromone lures were replaced every 4–6 weeks. Traps were serviced every 2–3 weeks from February through August 2001. Adult insects captured in traps were identified to species and counted. *Sitophilus* spp. were the most abundant group of insects captured and comprised over one-third of the total insects captured in pitfall traps (Rennie Roesli, unpublished data).

Trap captures of *Sitophilus* spp. from each store during each sampling occasion were used to calculate the mean number of insects/trap/week and associated variance. Weekly trap captures were also used to calculate the proportion of the 30 traps which captured one or more *Sitophilus* spp. Across all 8 stores, sampling between February and August 2001 yielded 74 data sets. Weekly data sets were entered into spreadsheets and 60% of the data were selected at random for developing sampling plans, leaving 40% for validating the sampling plans.

### Establishing the relationship between *Sitophilus* spp. density and proportion of infested traps

The relationship between mean trap captures (relative density) and proportion of infested traps was established through a nonlinear regression using TableCurve 2D software (Anon., 1994). We are unaware of established action thresholds pertaining to pest density or proportion of infestation samples for use in retail stores. Therefore, we arbitrarily chose an action threshold (AT) of 50% of the traps containing at least one or more *Sitophilus* spp. for developing sampling plans. The corresponding insect density at 50% infested traps was estimated from this relationship.

### Green's fixed precision sampling plan for estimating density

A fixed-precision, sequential sampling plan (Green, 1970) was used to estimate density of *Sitophilus* spp. at three precision levels. Green's fixed-precision sampling algorithm requires that the data first be fitted to Taylor's Power Law [ $s^2 = Am^b$  (Taylor, 1961)] to model the relationship between the mean ( $m$ ) and variance ( $s^2$ ). Both  $A$  and  $b$  were estimated by regressing sample variances against sample means after transformation of both variable to log<sub>10</sub> scale ( $\log s^2 = \log a - b \log m$ ). The antilogarithm of  $a$  is  $A$ .  $A$  is a factor dependent on the sample unit and  $b$  is an index of aggregation with  $b < 1$ ,  $b = 1$ , and  $b > 1$  suggesting the uniform, random, and aggregated spatial distribution of insects, respectively (Southwood, 1978).

Green's fixed-precision stop lines for sequential estimation of density were calculated as:

$$\log_{10} T_n = (\log_{10}(D^2/A)/b-2) - (b-1/b-2) * \log_{10} n \quad (1)$$

where  $T_n$  is the cumulative number of insects in the sample units,  $D$  is the precision expressed as the ratio of the standard error of the mean to the mean, and  $n$  is the sample size. We chose fixed precision levels of 0.25, 0.35, and 0.50. Performance of the sampling plans was evaluated using the validation data sets through a resampling approach (Naranjo and Hutchinson, 1997). This approach uses a PC software program called "Resampling for Validation of Sampling Plans" or RVSP. This software is available at the following world-wide web site: <www.wcri.ars.usda.gov/software/software.htm>.

### Wald's sequential probability ratio test (SPRT)

Wald's (1947) binomial sequential plan allows pest density to be classified relative to an infestation threshold. The sampling plan is based on information on four parameters (Binns, 1994): the upper threshold,  $p_j$ ; the lower threshold,  $p_0$ ; the probability of exceeding the upper threshold when in fact the true infestation level is at or below the lower threshold,  $\alpha$  (action is taken); and the probability of falling below the lower threshold when in fact the true infestation level is at or above the upper threshold,  $\beta$  (action is not taken). For developing the sampling plan,  $p_j$  was set at 0.50 (or 50% of infested sample units),  $p_0$  was set at 0.40, and the action threshold (AT) was set at 0.45. The rationale for setting the AT below  $p_j$  was discussed by Subramanyam et al. (1997). The  $\alpha$  and  $\beta$  errors rates were set at 0.20.

For binomial count data, upper ( $T_U$ ) and lower ( $T_L$ ) sampling stop lines were calculated as:

$$T_U = (\text{intercept}) \ln[(1 - \beta)/\alpha] + n (\text{slope}) \quad (2)$$

$$T_L = (\text{intercept}) \ln[\beta/(1 - \alpha)] + n (\text{slope}) \quad (3)$$

The intercept was calculated as

$$1/\{\ln[(p_j q_0)/(p_0 q_j)]\} \quad (4)$$

where  $p_0 = 0.4$ ,  $p_j = 0.5$ ,  $q_0 = 1 - p_0$ , and  $q_j = 1 - p_j$ . The slope was calculated as

$$\ln(q_0/q_j)/\{\ln[(p_j q_0)/(p_0 q_j)]\} \quad (5)$$

Stop lines were generated by plotting  $T_U$  or  $T_L$  against  $n$ .

To use the sampling plan, data from the validation data set were compared with the stop lines generated above. The lower stop line crossed the  $x$ -axis at 9. This suggested that at least 9 samples should be taken to classify infestation level with respect to the AT. Samples are examined and the number of infested samples out of the total was plotted with reference to the stop lines. Sampling stops when one of the stop lines is crossed. Between stop lines sampling continues until the stop lines are crossed. If the upper stop line is crossed, indicating that the infestation level is above the upper threshold, then pest intervention is needed. If the lower stop line is crossed, no pest intervention is needed because infestation level is below the lower threshold.

Evaluation of the sampling plan was performed using the methods of Fowler and Lynch (1987). The operating characteristic (OC) and the average sample number (ASN) functions were generated for each infestation level of the validation data after 500 simulations with the RVSP software. Resampling of the data sets was performed with replacement and the minimum sample size was set to 5. The OC function indicates the probability of not taking an action,  $\beta$ , relative to the selected AT. The ASN function shows the average number of samples necessary to classify the population at a particular infestation level. OC or ASN values were plotted against the corresponding proportion of infested samples and then a smooth curve was fit to the plotted data using SigmaPlot® software. The observed  $\alpha$  error rate from the OC curve is calculated as  $1 - \beta$  at  $p_0$ .

### Fixed binomial sampling plan

This plan is based on a user-specified number of samples to determine the proportion of sample units infested with at least  $t$  insects. Using the OC function described above, the user can then decide how many samples (traps in our case) would be needed to make management decisions for a given AT. The  $\beta$  error rate can also be obtained from the OC curve at different sample sizes. We chose sample sizes of 20, 25, and 30 to examine the OC curves and resulting  $\beta$  error rates.

## Results and discussion

### Relationship between *Sitophilus* spp. density and proportion of infested traps

The proportion of infested traps increased rapidly with a small increase in mean trap capture or density (Fig. 1). About 90% of the traps were infested at a mean density of 2 insects/trap/week. The mean density was 0.62 insects/trap/week when 50% of the traps were infested. For the purposes of using the sampling plan that predicts insect density (Green's plan), we considered 0.62 insects/trap/week our (economic) threshold.

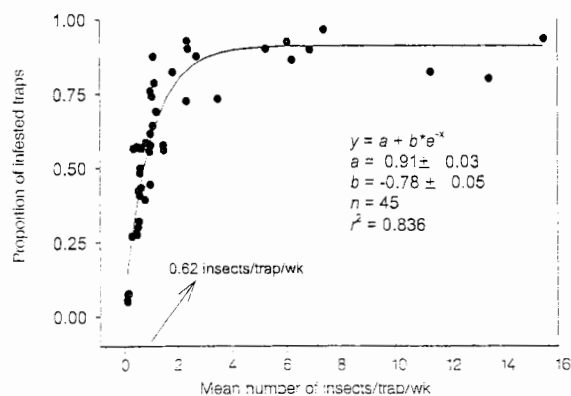


Fig. 1. Nonlinear relationship between the proportion of traps with one or more *Sitophilus* spp. and the mean density

### Green's sequential sampling plan

The log variance against log mean regression showed that the variance increased significantly with mean density ( $P < 0.01$ ;  $r^2 = 0.957$ ; Fig. 2). The antilogarithm of the intercept ( $A$ ), a factor dependent on the sample unit (Southwood, 1978) on arithmetic scale was 2.831. The slope ( $b$ ) value of 1.7 indicated that the distribution of insects in the traps was aggregated.

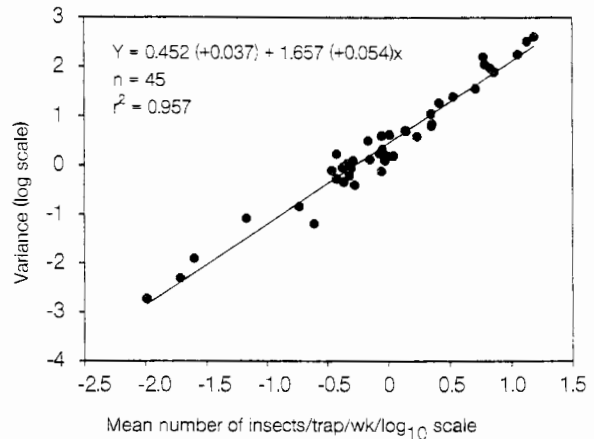


Fig. 2. Linear regression showing relationship between sample variance and mean density

Green's (1970) fixed-precision stop lines for estimating mean density at  $D = 0.25, 0.35,$  and  $0.50$  are shown in Fig. 3. These stop lines show that fewer samples are needed for estimating high densities and more samples are needed for estimating low mean densities. RVSP simulation with independent data showed that densities of  $>3$  insects/trap/week at a fixed precision of 0.25 can be estimated with 30 traps. However, more than 30 traps were necessary for estimating densities  $<3$  insects/trap/week (Table 1). A similar trend was observed at a fixed precision of 0.35; however, the density of 0.62 insects/trap/week could be estimated with 30 traps, while more than 30 traps were needed for estimating densities  $<0.62$  insects/trap/week. At a fixed precision of 0.50, the range of *Sitophilus* spp. densities of 0.03–17 insects/trap/week could be estimated with 20 traps. The observed precision in estimating *Sitophilus* spp. densities of 0.03–17 insects/trap/week was  $\leq 0.25$  at the fixed precision of 0.25 for 13 of the 26 data sets. At the fixed precision of 0.35, the observed precision was  $\leq 0.35$  for 12 of the 26 data sets. Finally, at a fixed precision of 0.50, the observed precision was  $< 0.50$  for 8 of the 26 data sets. Observed precision was at or below the fixed precision in cases where the sample variance for the independent data sets was equal to or less than the variance predicted by the Taylor's Power Law (Table 1). Generally, Taylor's Power Law did not accurately predict sample variance of independent data at low mean densities. Hutchinson et al. (1988) showed that both the precision and the number of samples needed for estimating mean densities are stochastic. Based on Green's plan, 30 traps can be used to estimate the threshold density of 0.62 insects/trap/week and densities  $>0.62$  insects/trap/week at a precision of 0.35 or 0.50.

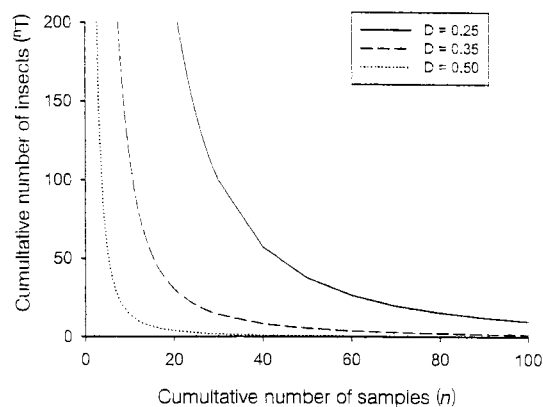


Fig. 3. Fixed precision stop lines for sequential estimation of *Sitophilus* spp. density. The precision (D) was expressed as the ratio of the standard error of the mean to the mean

Table 1. Performance of Green's (1970) fixed precision sequential sampling plan for estimating mean trap captures (density) of *Sitophilus* spp.

Data set	Mean <sup>a</sup>	Variance <sup>b</sup>	Predicted variance <sup>c</sup>	Fixed precision, D					
				D = 0.25		D = 0.35		D = 0.50	
				$n^d$	Obs D <sup>e</sup>	$n^d$	Obs D <sup>e</sup>	$n^d$	Obs D <sup>e</sup>
1	0.032	0.020	0.009	194	0.46	102	0.64	56	0.88
2	0.033	0.150	0.010	146	0.30	76	0.42	39	0.61
3	0.215	0.135	0.221	78	0.19	41	0.27	20	0.38
4	0.300	0.217	0.385	68	0.18	35	0.25	18	0.36
5	0.424	0.491	0.684	61	0.21	32	0.29	16	0.40
6	0.431	0.370	0.702	61	0.18	31	0.25	16	0.35
7	0.447	0.386	0.747	61	0.18	32	0.25	16	0.34
8	0.503	0.619	0.905	58	0.20	30	0.27	15	0.38
9	0.598	2.859	1.209	56	0.36	29	0.46	15	0.56
10	0.665	1.853	1.440	53	0.27	28	0.37	14	0.49
11	0.747	0.505	1.745	50	0.13	26	0.18	13	0.26
12	0.752	1.467	1.765	53	0.21	27	0.28	14	0.39
13	0.766	0.929	1.820	50	0.17	26	0.22	13	0.27
14	0.875	2.277	2.269	49	0.24	26	0.32	13	0.38
15	1.035	5.017	2.995	48	0.31	25	0.41	13	0.53
16	1.207	12.535	3.866	44	0.39	23	0.48	12	0.52
17	1.321	9.662	4.490	42	0.34	22	0.44	11	0.54
18	1.379	3.012	4.823	41	0.19	21	0.25	11	0.35
19	3.400	24.900	21.500	30	0.25	16	0.33	8	0.45
20	3.909	14.615	27.103	29	0.18	15	0.25	8	0.34
21	5.576	596.382	48.818	28	0.67	17	0.63	10	0.61
22	5.910	200.790	53.760	26	0.44	14	0.53	8	0.58
23	6.500	52.148	62.942	24	0.22	13	0.29	7	0.39
24	6.958	78.448	70.452	24	0.26	13	0.35	7	0.47
25	8.083	187.733	90.317	23	0.33	12	0.41	7	0.50
26	8.893	135.303	105.804	22	0.27	12	0.36	6	0.46
27	12.448	140.756	184.729	20	0.20	10	0.28	5	0.37
28	17.000	764.276	309.595	18	0.33	10	0.42	5	0.48

<sup>a</sup> Mean number of insects/trap/wk of independent data.

<sup>b</sup> Sample variance of independent data.

<sup>c</sup> Predicted variance,  $\sigma^2 =$ , was calculated using  $Am^b$ , where  $A = 2.851$ ,  $b = 1.657$ , and  $m =$  independent data means.

<sup>d</sup> Mean number of sample units required to estimate the mean as determined by the RVSP software.

<sup>e</sup> Observed mean precision level for estimating the mean as determined by the RVSP software.

### Wald's sequential probability ratio test binomial sampling plan

Wald's binomial sampling plan stop lines are shown in Fig. 4. A minimum of 9 samples is required for classifying infestation level with respect to the AT of 0.45. The OC curve showed that the actual error rate in classifying infestation as being below  $p_0$  when in fact it is above  $p_1$  (i.e.  $\beta$  error) was 0.25 at the preset error of 0.2 (Fig. 5). This suggested that one would fail to take action more than 20% of the time when in fact an action is warranted. The observed  $\alpha$  error rate was 0.275 at the preset error of 0.2. Thus, one would take action more often than needed. Therefore, the binomial sampling plan would either increase control costs or insect damage if used at the established thresholds and error rates of  $\alpha = \beta = 0.2$ .

The ASN showed that for classifying 0.001 – 1 infested traps about 10–52 samples were needed, with more traps being required for classifying infestation level at the AT (Fig. 6). Subramanyam et al. (1997) also reported a similar pattern in ASN values when classifying infestation levels of the rusty grain beetle, *Cryptolestes ferrugineus* (Stephens), associated with stored wheat. Uncertainty in classifying infestation levels at the AT generally results in a large number of samples being required to arrive at a decision. However, in our retail store example, we plan to use no more than 30 traps. With 30 traps, infestation levels  $\leq 0.30$  and  $\geq 0.62$  could be classified with respect to the AT. Therefore, Wald's binomial sampling plan is not useful for classifying an infestation level of 0.45 unless more traps are used. The proportion of infested sample units should be 0.62 or higher to use Wald's binomial plan to classify infestation levels of *Sitophilus* spp.

### Fixed binomial sampling plan

The fixed binomial plan with 20, 25, or 30 traps showed that the probability of not taking action (b) with respect to the threshold of 50% infested sample units was  $>0.40$  (Fig. 7). Therefore, losses could occur due to pest damage. Consequently, when the data indicated an intervention was necessary, pest damage beyond the threshold may have already occurred.

It is important to realise that the Wald's and fixed binomial sampling plans are based on the action thresholds that were chosen arbitrarily. More research is needed to establish realistic action thresholds, by relating infested trap or mean densities to mean densities or infestation levels in retail store products or to product recalls. However, the sampling plans presented here are a first step in developing more refined sampling plans as additional data become available.

### Acknowledgements

We appreciate Sharath Menon for entering much of the data into spreadsheets. We also thank the managers and staff of the 8 retail pet stores in Kansas for their cooperation on this project. This project was supported by the Nestlé Purina PetCare Company, St. Louis, Missouri, USA, and partially by CSREES-USDA under Agreement No. 00-51101-9674.

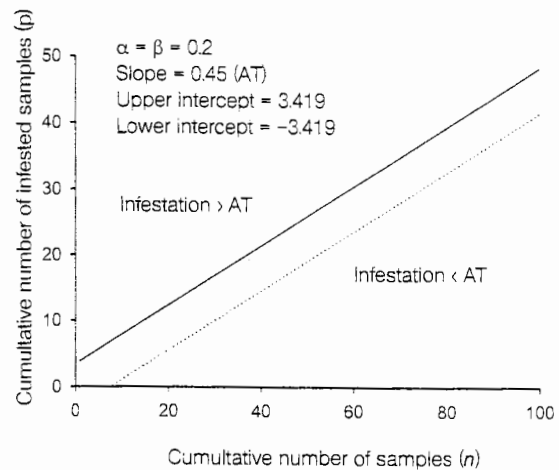


Fig. 4. Binomial sequential probability ratio test stop lines for classifying the infestation level of *Sitophilus* spp. relative to an action threshold (AT)

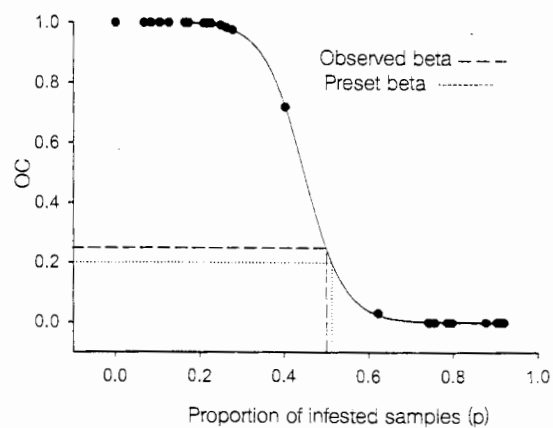


Fig. 5. Operating characteristic (OC) curve for the binomial sequential probability ratio test sampling plan ( $\alpha = \beta = 0.2$ ,  $p_0 = 0.4$ ,  $p_1 = 0.5$ )

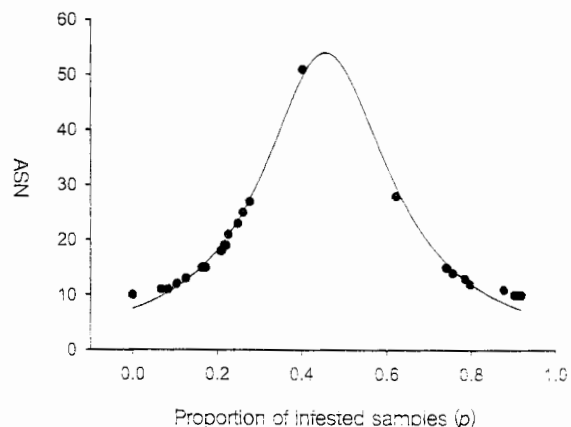


Fig. 6. Average sample number (ASN) curve for the binomial sequential probability ratio test sampling plan ( $\alpha = \beta = 0.2$ ,  $p_0 = 0.4$ ,  $p_1 = 0.5$ )

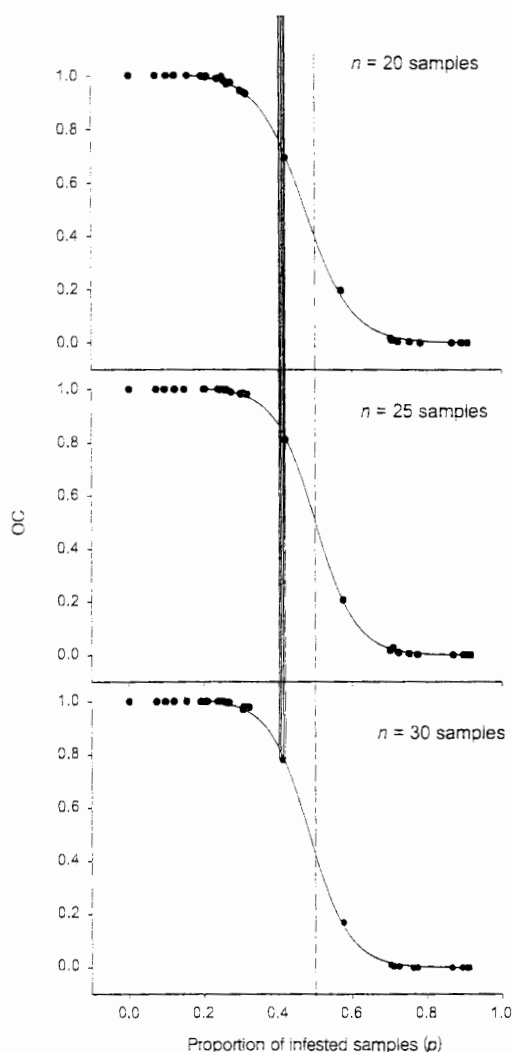


Fig. 7. Operating characteristic (OC) curve for fixed binomial sampling plans with 20, 25, or 30 traps

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