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CARCASS DISAPPEARANCE AND ESTIMATION OF MORTALITY IN A SIMULATED DIE-OFF OF SMALL BIRDS

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ABSTRACT: Carcasses of day-old chicks were placed randomly in grazed, mixed-grass pasture at a density of 50/ha each day for 5 days. Randomly chosen circular plots with a combined area equal to 10% of the total were searched each day for 10 days. The search method detected 95.2% of carcasses placed the day of the search but remains were found of only 20.8% of chicks placed 24 hr prior to searches. Seventy-six percent of carcasses that were observed daily disappeared completely within 24 hr. Although 250 carcasses were placed over a 5-day period, only two intact carcasses that had been in place for \geq 24 hr were found during searches. Estimates derived by extrapolation from the sampled area underestimated cumulative "mortality" on the entire area and did not reflect the course of the simulated outbreak well. There was a low degree of precision among repeated samples from the same area.

Key words: Epizootic, investigation, mortality, estimation, sampling, scavenging.

INTRODUCTION

Determining the extent of a disease occurrence in a population of wild animals is difficult. Often a count of carcasses is the only information available. Such counts are influenced by the rate at which carcasses decompose or are removed by scavengers, the time interval between occurrence of mortality and the search, and the accuracy and precision of the search method. The rate at which carcasses disappear is highly variable (Rosene and Lay, 1963; Payne, 1965; Woronecki et al., 1979; Wobeser et al., 1982; Bartmann, 1984; Balcomb, 1986; Humburg et al., 1986; Stutzenbaker et al., 1986; Tobin and Dolbeer, 1990; Linz et al., 1991; Pain, 1991) and is influenced by factors such as the density and visibility of carcasses, type and number of scavengers in the area, weather, and vegetation in the area. Carcass searches may be inaccurate and/or imprecise (Robinette et al., 1954; Anderson, 1978; Humburg et al., 1986; Stutzenbaker et al., 1986) and their success is influenced by many of the same factors that affect the disappearance of carcasses, as well as by the timing, method and intensity of the search procedure. Our objective was to determine the effectiveness of daily searches of 10% of an area as a method for assessing a simulated die-off of small birds in which the extent and timing of carcass placement were known. A secondary objective was to measure the rate of carcass disappearance under the experimental conditions.

MATERIALS AND METHODS

Surplus male domestic chicks that had been killed after hatching were obtained from a commercial hatchery and immediately frozen. The chicks had pale yellow down and weighed approximately 38 g (mean weight of 20 = 37.9 g; SD = 3.3). Before use, chicks were marked by removal of a toe specific for each day of the trial and allowed to thaw in a cooler overnight.

The study was conducted in a mixed-grass pasture used for beef cattle research at Goodale Experimental Farm, University of Saskatchewan, near Saskatoon, Canada (52°4'N, 106°34'W). The area has gently rolling topography with numerous small ephemeral ponds. The surrounding countryside is used principally for small grain production. Groves of aspen (Populus tremuloides) were present in the pasture and surrounding area but not on the study site. The study site had been grazed until a few days before the trial. Grass ranged from 7 to 15 cm in length and was relatively uniform in density over the site. Three adjoining study units were established and marked by perimeter stakes (Fig. 1). Units A and B were 50×100 m (0.5 ha), unit C was 20×50 m (0.1 ha).

The study, conducted from 12 to 21 June 1991, consisted of three trials. Trial 1 was intended to measure the accuracy of the search technique in detecting carcasses, the disappearance rate of carcasses from the site where they were placed, and the accuracy of the technique

for estimating daily and cumulative mortality on the area. (The term mortality is used in reference to the number of carcasses placed during the simulated epizootic.) In this trial 25 chicks were placed in each of units A and B between 0630 and 0800 on each of 12 through 16 June (days 1 to 5). Thus, the daily mortality on the combined area of units A and B was 50 on each of 12 to 16 June and the cumulative mortality increased from 50 on 12 June to 250 on 16 June. The location for placement of each chick was determined in advance using a method described by Fredrickson et al. (1977): a base line was established along one edge of the unit, coordinates for each site were then chosen by selecting a random number between 1 and 100 to identify a point along the baseline and a second random number between 1 and 50 to identify a distance perpendicular to the baseline at this point. The location for each chick was determined in the field by measuring with tape measures. The location of chicks was not marked.

Immediately after chicks were placed on each unit, or at the same time of day on days 6 to 10 when no chicks were placed, an area equal to 10% of each unit was searched for carcasses. Ten randomly chosen 50 m² circular plots on each unit were searched per day. The central point of each plot was determined by choosing coordinates in the same manner used for placement of chicks except that a random number between 4 and 96 was chosen for locations along the baseline and a second random number between 4 and 46 was chosen for the distance perpendicular to the baseline. This was done so that all plots were entirely within the study area. A stake was placed at the point identified by the coordinates and the area circumscribed by a 3.99 m cord attached to the stake was searched by the same observer in all cases. Presence and condition of remains within plots were recorded. Date of placement of carcasses found was determined by presence of a clipped toe. Carcasses were left in position and the stake was removed after the plot was searched.

Following completion of the trial, the location where each chick was placed and of circular plots searched were plotted on maps. Using these maps, the number of carcasses expected to be found during each search was calculated, assuming that all carcasses remained where placed until the search was conducted. This assumption was valid for birds placed immediately prior to the search but the validity for birds placed ≥ 24 hr earlier is unknown. Accuracy of the search method was assessed by comparing the number of carcasses found to the number expected to be found; this also provided information on the persistence of carcasses at the site where they were placed. Daily and cumulative mortality



FIGURE 1. Units used for studies of carcass disappearance.

were estimated by extrapolation from the number of carcasses detected on the sampled area.

Trial 2 was conducted on unit C simultaneously with trial 1. The purpose was to determine the rate at which individual carcasses disappeared. Five chicks were placed on the unit in the morning of each of 12 through 16 June. Methods for choosing sites for chicks and for marking chicks and the density of chicks/ha were the same as in trial 1. The location of each chick in unit C was marked with a 15 cm stake placed 1 m from the carcass. The marked sites were examined daily and the condition of each carcass was noted. If a carcass was not found where it had been placed, the area within a 10 m radius was searched; if no remnant was found in this area the carcass was considered to have disappeared completely and the corresponding stake was removed.

Trial 3 was conducted on 21 June on units A and B following completion of trial 1 on 21 June. The intent was to determine the precision (repeatability) of estimates obtained using the search technique. For this trial, 25 chicks were distributed randomly on each unit in the same manner used in trial 1. Immediately after placement of the chicks, ten surveys, each consisting of searches of ten randomly chosen circular plots with an area of 50 m², were conducted on each unit. This design was equivalent to sampling with replacement and since both the location of the chicks and the sites searched were chosen randomly the samples should be independent. However, bias might exist if there was an increased likelihood of detecting carcasses that had been found in prior searches. The location of carcasses and of the plots searched in trial 3 were plotted on maps and the accuracy of the searches was determined as in trial 1. The accuracy in trials 1 and 3 was compared to detect such bias.

RESULTS

Evidence of dead birds was found in all but one of the searches in trial 1 (Table 1). The most commonly encountered remains were tiny pieces of skin with attached feathers. Intact carcasses were found beyond the day of placement on

Date	Cumulative number of carcasses placed	Number and type of remains found	Estimated number of dead birds		
			Present <24 hr	Present >24 hr	Cumulative
June 12	50	4C*	40		40
June 13	100	5C ⁶	40	10	50
June 14	150	5C, 3F°	50	30	80
June 15	200	7C, 1P, ⁴ 4F	70	50	120
June 16	250	4C, 1P ^d	40	10	50
June 17	250	4F		40	40
June 18	250	1C, ^e 1P, ^d 5F		70	70
June 19	250	1P,' 2F		30	30
June 20	250	4F		40	40
June 21	250	5F		50	50

TABLE 1. Results of carcass searches of an area equal to 10% of the total area of units A and B combined in trial 1. The estimated number of carcasses present on the total area was obtained by extrapolation from the number found on the sampled area.

· C-intact carcass, placed same day unless specified.

" One intact carcass was from previous day.

¹ F, feathers.

^d P, partial carcass from June 14.

' One intact carcass from June 16.

'Partial carcass, date of placement unknown.

only two occasions in trial 1. One was found the day after placement, the other persisted intact for at least 2 days. Four partial carcasses were found; of these, one had persisted for 1 day, two had persisted for 2 days, and one had persisted for at least 2 days. Two partial carcasses consisted only of a single foot. The search technique was 95% accurate in locating carcasses placed the day of the search (20/21 carcasses ex-)pected to be found were found), and the estimated daily mortality, based on extrapolation from the sampled areas to the total, ranged from 40 to 70 birds/ha. Remains were found of only 5 (21%) of 24 chicks that had been in the field for 24 hr (i.e., birds placed the previous morning). One of these was a partial carcass, the other remains consisted of feathers. Remains were found on three occasions in locations where no chick had been placed, indicating that some carcasses were moved within the area by scavengers. The estimated cumulative mortality on units A and B always was less than the actual cumulative mortality (Table 1). The effect of a delay of 24 hr between the placement of carcasses and a carcass search also is evident. Estimated cumulative mortality increased during the first 4 days of the trial, but based on the estimate on day 5 (16 June), there appeared to be fewer carcasses present, even though the cumulative number of carcasses had increased. Results thereafter were erratic (Table 1).

None of the 25 chicks placed in trial 2 was found intact after 24 hr. No detectable remains were found of 19 (76%) of these birds. Feathers remained from the other six birds 1 day after placement, feathers from three of these persisted to the second day and feathers from one bird were still present on the third day after placement. All remains found in unit C were within 1 m of a site where a chick had been placed.

The proportion of carcasses expected to be found that were detected in trial 3 was 94% in unit A and 96% in unit B. The number of carcasses detected during the ten replicate surveys, each of which included an area equal to 10% of the unit, ranged from one to five ($\bar{x} = 3.20$, SD = 1.40) in unit A and from zero to five ($\bar{x} =$ 2.20, SD = 1.48) in unit B. Based on a *t*-test (Brockett and Levine, 1984), the means were not significantly different (at $\alpha = 0.05$) (t = 1.56, 18 df). By extrapolation, the estimated density of carcasses based on individual surveys ranged from 0 to 100/ha. The precision of these estimates, for surveys in which at least one carcass was found, ranged from $\pm 24\%$ to $\pm 165\%$ of the mean at the 90% confidence level (Brockett and Levine, 1984).

Potential scavengers seen in the pasture about the study site included at least one coyote (*Canis latrans*), several American crows (*Corvus brachyrhynchos*), two Swainson's hawks (*Buteo swainsoni*), a redtailed hawk (*B. jamaicensis*) and a blackbilled magpie (*Pica pica*). No specific attempt was made to observe the activity of scavengers on the units; however, crows, flying over the area, were seen on two occasions to drop to the ground and then fly off carrying a chick. A western kingbird (*Tyrannus verticalis*) was seen collecting chick feathers on the study site.

DISCUSSION

Results of this study support the conclusion by Stutzenbaker et al. (1986) that: "carcasses are quickly assimilated into the environment;" however, our findings cannot be extrapolated directly to any actual epizootic. While the study area and the scavengers present were representative of pastures in this area of Saskatchewan during late spring, the birds used were not indigenous and were more conspicuous than most wild species. This may have accelerated scavenging. The density of carcasses on the study area was greater than in most natural events and this also may have increased the rate of removal by scavengers (Linz et al., 1991). Accumulations of carcasses at similar density do occur in nature but are unusual; for example, we observed a similar density of carcasses when migrating Lapland longspurs (Calcarius lapponicus) were poisoned by consuming granules containing the pesticide carbofuran (unpubl.). Our presence in the area daily and handling of the carcasses at the time of placement also may have influenced the results by either attracting or deterring scavengers. It appeared that there was adequate scavenging capacity in the area to remove most of the chicks rapidly throughout the study. All carcasses placed in unit C disappeared on the first day of the study and we did not detect more rapid disappearance as the study progressed. Linz et al. (1991) suggested that at some density of carcasses the scavenging system would become satiated and the scavenging rate would fall. No evidence was seen of this phenomenon, perhaps because the density of carcasses was less than that required to satisfy scavengers.

Despite limitations of the simulation. some conclusions can be drawn that apply to natural situations. One is that small bird carcasses disappear rapidly under some circumstances. In trials 1 and 2, remains of only about 20% of birds could be found 24 hr after placement. This is similar to the fate of songbird carcasses in Maryland cornfields where 77% of birds disappeared during the first day (Balcomb, 1986). Carcass disappearance in these two studies was more rapid than that reported in studies by Woronecki et al. (1979) and Tobin and Dolbeer (1990), perhaps because the carcasses in this and Balcomb's study were in open fields where scavenging birds may be active (Linz et al., 1991). Variability in the rate at which carcasses disappear in different situations must be considered in assessing the validity of carcass searches as an indicator of mortality. For example, Pain (1991) found that duck carcasses in exposed positions on land persisted an average of 1.5 days, whereas carcasses concealed by vegetation on land, and carcasses in water, persisted an average of 3.3 and 7.6 days, respectively. Tobin and Dolbeer (1990) concluded that careful searches conducted within 2 days of pesticide application "should usually suffice for detecting significant songbird mortality" in New York orchards. However, even within their study, the rate at which carcasses disappeared varied markedly and none of 25 carcasses placed in one orchard could be found the following day. In contrast, Stutzenbaker et al. (1986) concluded that failure to find carcasses during intensive

searches was not reliable evidence to rule out extensive mortality of ducks in a Texas marsh. Any delay between deaths and a search for dead birds exacerbates the problem. In our study or that described by Balcomb (1986), a 1 day interval resulted in serious underestimation of the extent of mortality. Because the rate of carcass removal is variable and site specific, it should be measured during outbreaks by observing the rate at which marked, freshly dead carcasses disappear.

Another conclusion is that most small bird carcasses disappear without leaving readily observed remains. In trial 2, 76% of carcasses were gone completely within 1 day and no carcass remained intact after 24 hr. In trial 1, only two intact carcasses in place for ≥ 24 hr were found. Balcomb (1986) reported that 58% of songbird carcasses disappeared without leaving observable remains and Tobin and Dolbeer (1990) found that 75% of bird carcasses were removed completely. The fact that few intact carcasses persist is important because it limits the opportunity to diagnose the cause of an epizootic, or to assess its extent. Of 275 chicks placed in trials 1 and 2, only two carcasses that had been in place for \geq 24 hr were found. These were the only specimens that would have been suitable for necropsy. This emphasizes the need to investigate outbreaks as soon as possible after their discovery and the value of every specimen found during an investigation.

In using quadrat sampling, decisions must be made about the size and shape of individual quadrats and the total area to be sampled. Seber (1982) noted that an estimate from a large number of small quadrats usually has smaller variance than one from a small number of large quadrats and recommended individual quadrats be as small as possible. Although the plots in our study were small and we considered our searches to be thorough, approximately 5% of chicks placed just prior to searches went undetected in trials 1 and 3. The vegetation was relatively open and short and searches were done by the same peo-

ple who had placed the birds less than 1 hr earlier, suggesting that accuracy in locating cryptic species in dense cover would probably be much lower. Use of even smaller plots might be advantageous in such circumstances but Seber (1982) suggested that quadrats should not be so small that the majority are found to be empty. This suggestion was violated in our study. We used circular plots because it was easier to locate a central point and define the perimeter of a circle than to locate and measure a square or rectangular quadrat. About 15 circular plots were located and searched per hour by two people after the baseline was established. The decision to search 10% of the area each day was based on our experience that sampling at about this level of intensity is the maximum feasible in many field situations. It complies with what Seber (1982) described as a "general rule often stated" that about 5 to 10% of the total area should be sampled in estimating population density and compares favorably with some attempts to estimate mortality, such as that of Bartmann (1984) in which 1.05% of a winter range was searched for dead deer. Sampling at this level was reliable for detecting the presence of dead birds on the area (remains of at least one bird were found in 19/20searches in trial 1) and provided a reasonably accurate estimate of daily mortality in trial 1. Accuracy of the search technique for detecting partial carcasses and feathers was not assessed. The accuracy of a search could be assessed in an actual epizootic by marking a known number of carcasses prior to the search and measuring the detection rate of marked carcasses. Extrapolation from the 10% sample underestimated the cumulative mortality and did not reflect the course of the epizootic accurately in trial 1. In trial 3, the same area was sampled repeatedly with overlap among some plots and some carcasses were found more than once. This might have biased the results because of a greater probability of detecting carcasses that were found more than once; however, the accuracy of the

TABLE 2. Relationship between the number of plots searched and the confidence interval of the estimated number of carcasses in area A, trial 3. Twenty-five intact carcasses were present on the area and each plot was equal to 1% of the total area.

Number	Number of	95% confidence interval	
of plots searched	Standard Mean deviation		
10	0.20	0.42	0 to 0.46
25	0.24	0.44	0.07 to 0.41
50	0.22	0.42	0.10 to 0.34
75	0.29	0.48	0.18 to 0.40

search technique was 95% in this trial compared to 95.2% in trial 1, suggesting that any such bias was minor. The high degree of variability among repeated samples from the same area in trial 3 supports the conclusion by Seber (1982) that "it is not possible to obtain reasonably precise estimates for small populations without sampling a very high proportion of the population area." The effect of the number of plots sampled in unit A on the confidence interval of the estimate is shown in Table 2. Based on these calculations, it is evident that sampling at a rate feasible in the field may result in estimates with such wide confidence limits that they are of only limited value.

MANAGEMENT IMPLICATIONS

At best, a carcass count provides an estimate of the number of carcasses present at the time the count is made. Such counts are of limited use for estimating cumulative mortality and, hence, for assessing the significance of a disease occurrence. Delay between death of the animals and the survey increases the probability of misinterpretation. Persistence of carcasses is highly variable and should be measured during outbreaks by observing marked carcasses to determine the rate of disappearance. Carcasses marked prior to searches can be used to assess the accuracy of the search procedure and allow estimation of the number of carcasses present using capture/recapture calculations. Preliminary sampling should be done to determine the type of sampling appropriate for the situation and to predict the level of sampling necessary to yield estimates with a defined level of confidence.

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