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## Sampling Blood from Big Brown Bats (*Eptesicus fuscus*) in the Field with and without Anesthesia: Impacts on Survival

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Blood was collected from wild big ABSTRACT: brown bats (Eptesicus fuscus) with and without anesthesia in Fort Collins, Colorado in 2004 to assess the impacts of these procedures on shortterm survival and 1-yr return rates. Short-term survival and 1-yr return rates after release were passively monitored using PIT tag detection hoops placed at selected buildings. Comparison of 14-day maximum likelihood survival estimates from bats not bled (142 adult females, 62 volant juveniles), and bats sampled for blood with anesthesia (96 adult females, 23 volant juveniles) and without anesthesia (112 adult females, 22 volant juveniles) indicated no adverse effects of either treatment (juveniles:  $\chi^2 = 53.38$ , df=41, P=0.09; adults:  $\chi^2 = 39.09$ , df=44, P=0.68). Return rates of bats one year after sampling were similar among adult female controls (75.4%, n=142, 95%, CI=67.4-82.2%), females sampled for blood with anesthesia (83.0%, n=112, 95% CI=74.8-89.5%), and females sampled without anesthesia (87.5%, n=96, 95% CI=79.2–93.4%). Lack of an effect was also noted in 1-yr return rates of juvenile females. These data suggest that the use of anesthesia during sampling of blood has no advantages in terms of enhancement of survival in big brown bats.

*Key words:* Anesthesia, bats, blood, Cormack-Jolly-Seber, *Eptesicus fuscus*, PIT tags, survival.

Anesthesia is commonly used to restrain animals while blood samples are collected and a variety of methods of anesthesia have been reported for numerous small mammals, including bats (Gustafson and Damassa, 1985; Swann et al., 1997; Mathews et al., 2002). Wimsatt et al. (2005) developed methods for the capture, restraint, and blood sampling of big brown bats (*Eptesicus fuscus*) under anesthesia as part of a study of rabies in bats roosting in buildings. They designed a restraint capsule for bats, which attached to an isoflurane nonrebreathing patient circuit, delivering inhalant isoflurane and oxygen to as many as six bats at a time while blood was drawn from an interfemoral vein. Wimsatt et al. (2005) captured bats at roosts in the early evening and transported them to a nearby laboratory for sample collection prior to release at the roost the same night. Under these conditions, no effect of bleeding under anesthesia was observed on short-term (14-day) survival or on 1-yr return rates of marked bats. However, not all studies of bats that might require sampling of blood can be carried out in as convenient a setting. Work at remote locations might dictate sampling without inhalant anesthesia. The purpose of our study was to compare the effects of sampling bats for blood, with and without anesthesia, on short-term daily survival and 1-yr return rates.

All procedures were approved by the Institutional Animal Care and Use Committee at Colorado State University, Fort Collins, Colorado, USA. Bats were captured as they emerged from roosts used by maternity colonies and hence, the sample consisted largely of adult females and volant juveniles of both sexes. All bats captured were marked by subcutaneous insertion of passive integrated transponders (PIT tags) and subsequently detected passively by PIT readers placed at roost entrances (Wimsatt et al., 2005). Roosts where bats were detected for the shortterm survival analysis were monitored for single 14-day intervals immediately postsampling; blood sampling took place on 16

nights between 21 May and 30 July in 2004 (collection dates varied by roost). Bats used to determine 1-yr return rates were sampled and marked in summer 2004, and detected with PIT readers during summer 2005. All bat captures, sample collection, marking, blood sampling, and delivery of anesthesia are described in detail by Wimsatt et al. (2005). We followed the same protocols except we introduced a second treatment of sampling blood without anesthesia and restraint capsules.

A random number table was used to assign captured bats to one of three treatment groups. Controls received all procedures except blood sampling and anesthesia, whereas the two treatment groups were bats bled under anesthesia and those bled without anesthesia. Fourteen-day apparent short-term survival rates were computed based on Cormack-Jolly-Seber (CJS) models using Program MARK (White and Burnham, 1999), a numerical maximum-likelihood program for estimating population parameters from mark-recapture data. Only the first capture and handling event for each individual was used in this analysis. Short-term survival estimates were reported as apparent survival rates ( $\phi$ ) because subsequent recapture procedures could not distinguish bats that died from bats that emigrated, or detect bats that used alternate exits and entrances to buildings without passing readers, or those that lost PIT tags. Fourteen-day capture histories were combined across nine roosts for adult females and across eight roosts for juveniles of both sexes. Capture histories for the two distinct age groups were analyzed separately. Quasilikelihood model selection methods were used and we estimated overdisperion, or  $\hat{c}$ , using the median c-hat approach in Program MARK. Goodness-of-fit  $(\chi^2)$  testing using Program RELEASE, TEST1 (Burnham et al. 1987) was used to determine differences in apparent survival between treatments and controls for each of the two age groups.

One-year return rates to roosts were calculated for adult and juvenile female bats that were sampled in 2004 to investigate the potential longer-term impact of anesthetic procedures and blood sampling on survival (males are not expected to return to maternity roosts). PIT tag detection records at nine roosts monitored in 2005 were used to determine if bats had survived and returned to the same roost 1 yr after sampling. Bats sampled at these roosts were categorized into three treatment groups as described above. One-year return rates ( $\hat{r}$  = number known alive in 2004/number captured in 2005) were calculated separately for adults and yearling females within each of these groups, with confidence intervals for  $\hat{r}$  calculated based on an estimated binomial variance  $= \hat{r} (1-\hat{r})/n$  (Williams et al., 2002).

We sampled 350 adult females, 59 juvenile females, and 48 juvenile males in 2004. No difficulties were observed while sampling blood, and no bats died during sampling as a result of these procedures. We constructed four models in Program MARK to determine treatment effects on short-term survival and capture probabilities for both adult females and juveniles, and ranked these models using quasi-likelihood model selection procedures. For adult females, the model incorporating a treatment effect on both apparent survival  $(\phi)$  and capture probabilities (p) had the lowest strength of evidence. Further evidence of a lack of an effect of blood sampling either with or without anesthesia on survival and capture probabilities was provided by results calculated in Program RELEASE  $(\chi^2 = 39.09, df = 44, P = 0.68)$ . Estimates of apparent survival ( $\phi$ ) of adult females were 0.967 (n=142, 95% CI=0.954-0.976) for bats not bled, 0.972 (n=112, 95%CI=0.958-0.981) for bats bled without anesthesia, and 0.980 (n=96)95%CI=0.966-0.988) for bats bled with anesthesia; 95% confidence intervals overlapped among the three groups. Estimates of capture probability  $(\hat{p})$  were 0.748 (95%)

CI=0.717-0.777) for bats bled with anesthesia, 0.750 (95% CI=0.720-0.778) for bats bled without anesthesia, and 0.765 (95% CI=0.739–0.790) for bats not bled; also with 95% confidence intervals overlapping among the three treatments. Model selection procedures suggested some evidence of a positive treatment effect on apparent survival of juvenile bats, but survival rates among the three treatment groups were not significant at the 0.05 level using Program RELEASE  $(\chi^2 = 22.2, df = 41, P = 0.09)$ . Confidence intervals around estimates of survival and capture probabilities of juvenile bats overlapped: apparent survival was 0.970 (n=23, 95% CI=0.933-0.987) for juveniles bled with anesthesia, 0.964 (n=22, 95% CI=0.924-0.984) for juveniles bled without anesthesia, and 0.896 (n=62, 95%)CI=0.856-0.926) for juveniles not bled.

One-year return rates were similar among adult female controls ( $\hat{r} = 0.75$ , n=142, 95% CI=0.67-0.82), females sampled for blood with anesthesia ( $\hat{r}$ =0.83, n=112, 95% CI=0.75-0.89), and females sampled without anesthesia ( $\hat{r}$ =0.88, n=96, 95% CI=0.79-0.93). Juvenile female controls returned at a rate of 47.2% (n=36, 95% CI=30.4-64.5%), juveniles bled without anesthesia at a rate of 90.9% (n=11, 95% CI=58.7-99.8%), and juveniles bled with anesthesia returned at a rate of 75.0% (n=12, 95%) CI = 42.8 - 94.5%). All three groups in both age classes had broadly overlapping confidence intervals, suggesting that these procedures also had no measurable effect on longer-term fate, although small sample sizes for juveniles leads to greater uncertainty.

Estimates of short-term daily survival and 1-yr return rates by passive monitoring of PIT-tagged big brown bats clearly indicate that blood sampling has no measurable impact on mortality, and that these rates do not differ between bats that are anesthetized or not anesthetized. Delivery of inhalant anesthesia to wildlife in the field can present significant practical problems. The safe use of isoflurane, for instance, requires a temperature-equilibrated precision vaporizer equipped with an oxygen tank, which can be impractical to handle in the field. Although robust and lightweight equipment has been designed to administer isoflurane in the field without the use of compressed gasses (Lewis 2004), sampling without anesthesia simplifies fieldwork and does not decrease survival in big brown bats. We found it easy to restrain big brown bats without the use of anesthesia and avoided the potential for exposures of personnel to bites by wearing leather gloves. Researchers sampling other species of bats may need to judge the hardiness of these species to sampling blood without anesthesia. However, based on our findings with big brown bats, anesthesia might not be necessary to prevent impacts on mortality.

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