EFFECTIVE SAMPLING AREA: A QUANTITATIVE METHOD FOR SAMPLING CRAYFISH POPULATIONS IN FRESHWATER MARSHES

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ABSTRACT

We used mark-recapture techniques to estimate the attraction sampling area of baited traps for monitoring density of the crayfish *Procambarus alleni* in the freshwater marshes of the Florida Everglades. We successfully applied a permanent visible tag suitable for mass or individual marking of crayfish. In laboratory mesocosms, males and females and juveniles and adults entered baited wire traps with similar frequency, reflecting a lack of age- or sex-specific bias in trapping. In flooded marsh habitat, we released marked crayfish among a circular array of baited traps set at a specific radial distance from the release point and determined recapture proportions over 48 h. We then used the methodology of Turchin & Odenaal (1996) to estimate the effective sampling area based on the proportions of crayfish recaptured over various radial distances. In flooded marshes, the mean proportion of recaptures declined from 0.59 in traps with a 1-m sampling radius to 0.02 in traps with a 28-m sampling radius. A log-linear regression model provided the best fit to the capture data, and the effective sampling area of baited traps was estimated as 56.3 m². The effective sampling area serves as a translation coefficient that can be used to calculate actual density of crayfish in a given area from mark-recapture trapping data.

RÉSUMÉ

Nous avons utilisés des techniques de recapture d'individus marquées pour estimer les zones d'attraction de pièges appâtés pour le controle de densité de l'écrevisse *Procambarus alleni* dans les marais d'eau douce des Everglades de Floride. Nous avons appliqué avec succès une marque permanente, visible, convenant pour le marquage en masse ou individuel des écrevisses. En laboratoire, mâles et femelles, juvéniles et adultes, pénétraient dans les pièges en grillage appâtés avec la même fréquence, reflétant l'absence de biais relatif au sexe ou à l'âge dans la capture. Dans l'habitat de marais inondé, nous avons relâché les écrevisses marquées parmi une aire circulaire de pièges appâtés placés à une distance radiale spécifique du point de relâchement, et déterminé les proportions de recapture sur 48 h. Nous avons alors utilisé la méthodologie de Turchin & Odenaal (1996) pour estimer la zone d'échantillonnage effectif d'après les proportions d'écrevisses recapturés sur diverses distances radiales. Dans les marais inondés, la proportion moyenne de recaptures

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décroissait de 0,59, pour les pièges dans un rayon d'échantillonnage de 1 m, à 0,02 pour les pièges dans un rayon de 28 m. Un modèle de régression logarithmique linéaire fournissait le meilleur ajustement aux données de capture, et la zone d'échantillonnage effectif des pièges appâtés était estimée à 56,3 m². La zone d'échantillonnage effectif sert comme un coefficient de traduction qui peut être utilisé pour calculer la densité réelle d'écrevisses dans une zone déterminée d'après les données de recapture d'individus marqués.

INTRODUCTION

A variety of methods have been used to sample crayfish populations in freshwater habitats, but most have a number of associated logistical and quantitative problems. Collection by hand (e.g., Hazlett et al., 1979) is generally not practical in heavily vegetated habitats. Throw traps (Jordan et al., 1996) and pull traps (Kushlan & Kushlan, 1979) have been used to sample crayfish Procambarus alleni (Faxon, 1884) in freshwater marl prairie and slough habitats in the Florida Everglades. These methods are labor intensive, do not sample animals in burrows, and are difficult to use in heavily vegetated habitats. Because these methods sample only a relatively small area, crayfish density may be underestimated or overestimated if sampling is not intensive and the spatial distribution of animals is not uniform. Baited traps have been used for sampling crayfish populations in several studies (e.g., cylindrical wire minnow traps, Momot & Gowing, 1972). The disadvantage of this method is that it samples an unknown area and gives a relative estimate of density with an unknown standard error. However, if the attraction area that a trap effectively samples is known, trap sampling can yield quantitative estimates for monitoring density fluctuations or for analyzing dispersal patterns.

Mark-recapture methods have been widely used to quantify animal abundance in the field (Seber, 1982). However, it is not known if traps that work by attraction (e.g., containing bait, pheromones) conform to the restrictive assumptions of mark-recapture models. For example, most mark-recapture models assume that all animals are equally likely to be captured within a given area (Cormack, 1969), but this assumption cannot be tested until the sampled area is known. Turchin & Odenaal (1996) developed a method of quantifying attraction sampling for insects using mark-recapture experiments with pheromone traps. They used the relationship between the proportions of recaptured insects over all distances from the release point to estimate the effective sampling area. This estimate can be used as a translation coefficient to link recapture probabilities to actual density per unit area (Turchin & Odenaal, 1996). Their method does not assume that all animals within a given area are equally likely to be recaptured or that the estimate is affected by animal distribution as long as traps are located randomly with respect to distribution (Elkinton & Cardé, 1980).

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We used the approach of Turchin & Odenaal (1996) to estimate the effective sampling area of baited traps for sampling crayfish in flooded freshwater marl prairie. We first tested for possible age- or sex-biases associated with trap sampling. We then released marked crayfish in marshes and measured recapture rates at various distances from release to determine the effective attraction area of a baited trap. We assess the effectiveness of this technique for sampling crayfish populations in flooded marsh habitat.

METHODS

To conduct field mark-recapture experiments, we first tested a permanent visible tag for crayfish using colored elastomer plugs that are implanted under the clear exoskeleton of the ventral abdomen. In this procedure, a small amount of liquid elastomer (< 0.01 ml) is injected under the exoskeleton, and this hardens as a plug within 24 h (Northwest Marine Technology, Inc., Shaw Island, Washington, U.S.A.). This method has been used for mass or individual marking of aquatic organisms (e.g., shrimp, Godin et al., 1995). We held tagged crayfish in the laboratory to observe tag loss, mortality rates, and molting success.

We conducted experiments in laboratory mesocosms to test for sampling bias associated with trapping. Equal numbers of adult and juvenile male and female crayfish Procambarus alleni (Decapoda, Cambaridae) were held for 24 h in 2-m diameter mesocosms. Two wire minnow traps (50 cm long \times 20 cm diameter with 1.5-cm entrances at each end) baited with shrimp or two unbaited traps (with feed shrimp scattered around the tank) were placed in the mesocosms that contained no other shelter structures. Fifteen replicates with 6-12 crayfish each were conducted with baited traps and 11 replicates with unbaited traps (each total 114 crayfish). Trapped crayfish were enumerated daily by sex and maturity. When not being used in experiments, crayfish were held in tanks with opened wire minnow traps that served as shelter and that may prevent the development of trap-shy or trapseeking behavior. Trap data were analyzed using a hierarchical log-linear model for a multidimensional contingency table with crayfish sex (male and female) and maturity (adult and juvenile) nested in trap type (baited or unbaited). We examined the likelihood ratio chi square (G^2) to evaluate the fit of the model and the partial chi squares (χ^2) for effects of the individual terms (Zar, 1998).

In the marl prairie marshes of eastern Everglades National Park, Florida, U.S.A., we conducted mark-recapture experiments with a single-release-multipletrap design to estimate the attraction sampling area of baited traps. Crayfish were marked, held without food for 24 h, and then released in the center of a circular array of baited wire traps that were checked after 48 h. Preliminary sampling showed that crayfish catch in traps is similar after 24 and 48 h but declines after 72 and 144 h. Since the probability of recapture may be underestimated if most animals are taken in the near traps (Elkinton & Cardé, 1980), we conducted separate experiments with a circular array of traps set at one of several experimental sampling radii (1, 4, 8, 16, or 28 m) from the release point and each trap at twice those distances from other traps. Eighteen replicates with 20 crayfish each were conducted for each sampling array.

To estimate the effective sampling area of baited traps, we adopted the approach of Turchin & Odenaal (1996). The proportions of recaptured crayfish P were plotted at each distance r from the release point. Regression analyses were conducted to determine the relationship between the dependent variable (P) and the actual, log-transformed, or square root-transformed independent variable (r). The best transformation of radius r as indicated by the coefficient of determination R^2 was used, and an empirical curve was fitted to the data to describe the functional relationship P(r). This function P(r) was then integrated over all radii to calculate the effective sampling area, α (equation 1 from Turchin & Odenaal, 1996):

$$\alpha = 2\pi \int_{0}^{\infty} \mathbf{r} \, \mathbf{P}(\mathbf{r}) \, \mathrm{d}\mathbf{r}$$

Turchin and Odenaal (1996) point out that the effective sampling area, α , is actually a conversion coefficient for estimating density in a given area.

RESULTS

The tagging procedure did not increase mortality; < 2% mortality occurred in both tagged and untagged crayfish held in the laboratory for thirty days. The elastomer tag remained highly visible in the ventral abdomen, and we observed no loss of tags during one molt in 49 crayfish or during multiple molts in 17 crayfish. Nevertheless, crayfish could be expected to lose the elastomer tag if it is placed too near the exoskeleton. One problem noted with the procedure was that in a few animals the elastomer plug was displaced from the original position in one abdominal segment to other segments, probably due to tail-flipping or other strenuous activity by the animal immediately after tagging. Crayfish marked by this method must be held in the laboratory for 24 h to observe the final position of the tag.

In laboratory mesocosms, the presence of shrimp bait was the only significant factor in crayfish capture (G² 1.846, df 3, P = 0.61). More crayfish entered baited traps (86 crayfish) than unbaited traps (28 crayfish) (partial χ^2 30.93, df 1, P < 0.0001). There were no significant differences in the capture of males and



Fig. 1. Proportions of recaptured crayfish *Procambarus alleni* as a function of distance from the release point. Line shows the fitted function $P(r) = 0.57 - 0.22 \ln r$.

females (partial χ^2 0.035, df 1, P = 0.85) or adults and juveniles (partial χ^2 0.526, df 1, P = 0.45).

The mean proportion of recaptures declined from 0.585 (SE \pm 0.03) in traps 1 m from the release point to 0.016 (SE \pm 0.007) at the 28 m radius. Regression analysis indicated that the ln transformation of the independent variable r provided the best fit (F = 252.2, df 1, *P* < 0.0001; R² 0.74). The most parsimonious model of the functional relationship between crayfish recapture P and trap radius r was (fig. 1):

$$P(r) = 0.57 - 0.22 \ln r.$$

Using this P(r) in the Turchin-Odenaal equation, we estimate the effective sampling area as $\alpha = 56.3 \text{ m}^2$. This suggests that a shrimp-baited wire trap would most efficiently sample crayfish in a 4 m radius over 48 h.

DISCUSSION

The autecology of the target species must be considered in any sampling scheme. A potential problem with trapping is the development of behavioral biases associated with trap avoidance ("trap shy") or trap seeking ("trap happy"). Brown & Brewis (1978) suggested that trap sampling for the crayfish *Austropotamobius pallipes* (Lereboullet, 1858) is strongly biased by "trap happy" male adults when compared with hand collections. In the laboratory, we found no evidence of behavioral bias in *P. alleni* males and females or adults and juveniles. Our field sampling in Everglades marl prairie marshes also indicates a lack of trapping bias for all crayfish except for gravid females that may be inactive in burrows (Acosta & Perry, unpubl.). The coefficient for the effective sampling area may need to be approximated separately for unique field conditions (Turchin & Odenaal, 1996). For example, if water flow is strongly directional, baited traps will sample an elliptical area within the downstream chemical plume.

The probability of recapturing marked crayfish in baited traps decreased with distance as expected. Baited crayfish traps set in shallow freshwater marshes appear to effectively sample about 0.01 ha, suggesting that most crayfish within 6 m receive a strong chemical food signal. Beyond this distance, the probability of capture rapidly declines. The effective sampling area can be used to link mark-recapture measurements to estimates of actual densities, and it provides a means to evaluate sampling designs for field use (Turchin & Odenaal, 1996). A key factor in the success of our experiments was the use of a highly visible tag with little or no tag loss or increase in mortality of crayfish. This method appears to have substantial benefits over other tagging procedures such as those involving mutilation (see Weingartner, 1982).

Direct estimation of the attraction sampling area can be made in some cases (e.g., for insects, Byers et al., 1989; Elkinton & Cardé, 1980; Schlyter, 1992). Other studies have attempted to estimate sampling area from catch statistics (e.g., for lobsters, Evans & Evans, 1996), but such estimates are impossible to verify unless actual densities are known. The methodology of Turchin & Odenaal (1996) uses the empirical relationship between the proportions of recaptured animals and distance to quantify the effective sampling area. This approach gives a quantitative estimate of sampling area without many of the restrictive assumptions associated with mark-recapture models.

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LITERATURE CITED

- BROWN, D. J. & J. M. BREWIS, 1978. A critical look at trapping as a method of sampling a population of *Austropotamobius pallipes* (Lereboullet) in a mark and recapture study. Freshwater Crayfish, 4: 159-164.
- BYERS, J. A., O. ANDERBRANT & J. LOFQVIST, 1989. Effective attraction radius: a method for comparing species attractants and determining densities of flying insects. Journ. Chem. Ecol., 15: 749-765.
- CORMACK, R. M., 1969. The statistics of capture-recapture methods. Oceanogr. mar. Biol. ann. Rev., 6: 455-506.
- ELKINTON, J. S. & R. T. CARDÉ, 1980. Distribution, dispersal and apparent survival of male gypsy moths as determined by capture in pheromone-baited traps. Environ. Entomol., **9**: 729-737.
- EVANS, C. R. & A. J. EVANS, 1996. A practical field technique for the assessment of spiny lobster resources of tropical islands. Fish. Res., 26: 149-169.
- GODIN, D. M., W. H. CARR, G. HAGINO, F. SEGURA, J. N. SWEENEY & L. BLANKENSHIP, 1996. Evaluation of a fluorescent elastomer internal tag in juvenile and adult shrimp *Penaeus* vannamei. Aquaculture, 139: 243-248.
- HAZLETT, B., D. RITTSHOF & C. AMEYAW-AKUMFI, 1979. Factors affecting the daily movements of the crayfish *Orconectes virilis* (Hagen, 1870) (Decapoda, Cambaridae). Crustaceana, (Suppl.) 5: 121-130.
- JORDAN, F., K. J. BABBITT, C. C. MCIVOR & S. J. MILLER, 1996. Spatial ecology of the crayfish *Procambarus alleni* in a Florida wetland mosaic. Wetlands, **16**: 134-142.
- KUSHLAN, J. A. & M. S. KUSHLAN, 1979. Observations on crayfish in the Everglades, Florida, U.S.A. Crustaceana, (Suppl.) 5: 115-120.
- MOMOT, W. T. & H. GOWING, 1972. Differential seasonal migration of the crayfish, *Orconectes virilis* (Hagen), in marl lakes. Ecology, **53**: 479-483.
- SCHLYTER, F., 1992. Sampling range, attraction range, and effective attraction radius: estimates of trap efficiency and communication distance in coleopteran pheromone and host attractant systems. Journ. appl. Entomol., **114**: 439-454.
- SEBER, G. A. F., 1982. The estimation of animal abundance: 1-672. (Griffin, London).
- TURCHIN, P. & F. J. ODENAAL, 1996. Measuring the effective sampling area of a pheromone trap for monitoring population density of southern pine beetles (Coleoptera: Scolytidae). Environ. Entomol., 25: 582-588.
- WEINGARTNER, D. L., 1982. A field-tested internal tag for crayfish (Decapoda, Astacidea). Crustaceana, 43: 181-187.
- ZAR, J. H., 1998. Biostatistical analysis: 1-765. (Prentice-Hall).