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**HANKIN AND REEVES' APPROACH TO ESTIMATING
FISH ABUNDANCE IN SMALL STREAMS
LIMITATIONS AND POTENTIAL OPTIONS**



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Hankin and Reeves' Approach to Estimating Fish Abundance in
Small Streams: Limitations and Potential Options

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Abstract: Hankin and Reeves' (1988) approach to estimating fish abundance in small streams has been applied in stream-fish studies across North America. However, as with any method of population estimation, there are important assumptions that must be met for estimates to be minimally biased and reasonably precise. Consequently, I investigated effects of various levels of departure from these assumptions via simulation based on results from an example application in Hankin and Reeves (1988) and a spatially clustered population. Coverage of 95% confidence intervals averaged about 5% less than nominal when removal estimates equaled true numbers within sampling units, but averaged 62% - 86% less than nominal when they did not, with the exception where detection probabilities of individuals were >0.85 and constant across sampling units (95% confidence interval coverage = 90%). True total abundances averaged far (20% - 41%) below the lower confidence limit when not included within intervals, which implies large negative bias. Further, average coefficient of variation was about 1.5 times higher when removal estimates did not equal true numbers within sampling units ($C\bar{V} = 0.27$ [SE = 0.0004]) than when they did ($C\bar{V} = 0.19$ [SE = 0.0002]). A potential modification to Hankin and Reeves' approach is to include environmental covariates that affect detection rates of fish into the removal model or other mark-recapture model. A potential alternative is to use snorkeling in combination with line transect sampling to estimate fish densities. Regardless of the method of population estimation, a pilot study should be conducted to validate the enumeration method, which requires a known (or nearly so) population of fish to serve as a benchmark to evaluate bias and precision of population estimates.

Abundance is the typical parameter estimated to monitor fish populations. The traditional approach to estimating stream-fish abundance is to choose sites (i.e., sampling units) within a stream and then count fish within these sites. For instance, sampling units could be defined as pools, riffles, and glides. Choice of which sampling units to survey is based on either their perceived representativeness of the population of interest or some type of random sampling design. Two of the more widely used methods to obtain within-unit estimates of abundance are snorkeling and electrofishing (Dolloff et al. 1996 and Reynolds 1996, respectively, and references therein).

Building on earlier work by Hankin (1984), Hankin and Reeves (1988) developed a two-stage sampling approach that employed both snorkeling and electrofishing for estimating fish abundance in small streams. A stream first is stratified by habitat type (e.g., riffles, pools, and glides) and reach location (e.g., lower, middle, and upper) and a systematic sample with a single random start is selected within each stratum (first stage). Visual estimates of fish numbers are concurrently obtained by 2 divers snorkeling within each selected unit (second stage). Multiple pass removals using electrofishing are applied within a systematic subsample of the randomly selected units to provide a “true” count of fish based on Zippin’s (1958) estimator. These removal estimates then are used in a ratio estimator (Cochran 1977) to adjust snorkel counts in non-electrofished units for incomplete detection of fish.

Hankin and Reeve’s (1988) approach has been applied in stream-fish projects across North America, including those monitoring threatened and endangered species. A recent search in Science Citation Index (Institute of Scientific Information, Philadelphia, Pa.) identified 45 articles that have cited their paper, although papers actually using their

approach is likely less than this number. As with any method of population estimation, however, there are key assumptions underlying Hankin and Reeves' approach that must be met for abundance estimates to be minimally biased and reasonably precise. Here, I outline these assumptions, describe various factors that may lead to their violation, and use simulation to evaluate the degree to which these assumptions can be violated and still produce minimally biased and reasonably precise results. In addition, I offer a potential modification and an alternative to Hankin and Reeves' approach that represent possible avenues of future research and development. I concentrate on their method of abundance estimation because their method of mapping sampling units was shown to perform reasonably well in field tests by Roper and Scarnecchia (1995).

Key Assumptions and Potential Violations

Key assumptions of the Hankin and Reeve's (1988) approach relate to the ratio estimator used to correct snorkel counts for incomplete detection of fish within snorkeled units. These assumptions include a complete count of fish within electrofished units, a strong linear relationship between abundance estimates and average diver snorkel counts within surveyed units, and constant rates of detection of fish among surveyed units.

Removal Estimates as Complete Counts

Hankin and Reeves (1988) equated Zippin's removal estimator with a complete count of fish within sampled units. Important assumptions underlying this estimator include constant electrofishing effort for each sampling occasion, no births/immigration or deaths/emigration during the sampling period (closure assumption), and identical capture probabilities of fish within and among sampling occasions (Otis et al. 1978, White et al. 1982). Previous studies using known numbers of fish indicated that Zippin

removal estimates underestimated true abundances by 13%– 52.5% (Bohlin and Sundstrom 1977, Peterson and Cederholm 1984, Rodgers et al. 1992, Riley et al. 1993).

Use of standardized protocols for applying equal effort as well as blocknets to ensure population closure may be adequate to reasonably satisfy these two assumptions. However, with respect to population closure, lack of physical barriers to contain fish within each sampled unit during electrofishing will likely lead to movements of fish to areas beyond the unit boundaries. That is, fish in small streams have been shown to exhibit a flight response to electrofishing current, causing them to move outside the sampled area (Nordwall 1999). Violation of the closure assumption of this type will lead to negatively biased removal estimates (White et al. 1982). The magnitude of bias will depend on the relative numbers of fish moving out of the sampling unit. Hankin and Reeves (1988) made no mention of blocknets in their field study.

Identical capture probabilities of fish both within and among sampling occasions will never be exactly met under field situations. That is, capture rates of fish via electrofishing will vary with factors such as: fish density, fish behavior and size; habitat structure; environmental conditions (e.g., stream temperature, turbidity, etc.); sampling gear; and size of sampling unit (e.g., see Northcote and Wilkie 1963, Mesa and Schreck 1989, Rodgers et al 1992, Bayley and Dowling 1993). One or more of these factors can vary both spatially and temporally. Although identical capture probabilities are not achievable under typical field conditions, removal methods may still produce useful results under high capture probabilities and large population sizes (e.g., 2-pass removals: >0.6 for >200 fish and >0.8 for >100 fish; Bohlin 1982).

Linear Relationship Between Removal Estimates and Snorkel Counts

To be unbiased, the ratio estimator requires a straight line relationship between removal estimates (assumed true counts) and snorkel counts that passes through the origin, as well as a proportional relationship between variability in removal estimates and snorkel counts (Cochran 1977). Confidence intervals based on the normal distribution apply for large samples. In practice, one requires at least 30 samples and a coefficient of variation (CV) of less than 0.1 for both removal estimates and snorkel counts for the confidence intervals to be reasonable. Otherwise, variances and confidence interval widths will be underestimated (Cochran 1977:156).

A more or less proportional relationship between variances of removal estimates and snorkel counts seems at least approximately attainable. One would expect an increased variance in counts with increased numbers of fish. Conversely, correlations (r) between snorkel counts and removal estimates will be affected by factors affecting sightability or catchability of fish within and among surveyed units (e.g., habitat structure, fish density, etc.). Therefore, consistently high correlations between snorkel counts and removal estimates are not a certainty, despite the extremely high correlations reported by Hankin and Reeves (1988:840) for juvenile coho salmon (*Oncorhynchus kisutch*; $r = 0.95$ for pools and $r = 0.99$ for riffles) and juvenile steelhead trout (*Oncorhynchus mykiss*; $r = 0.61$ for pools and $r = 0.98$ for riffles). For instance, correlations between snorkel counts and multiple pass removal estimates were low ($r = 0.20-0.44$) for smaller size classes (70-100 mm) of brook trout (*Salvelinus fontinalis*), bull trout (*Salvelinus confluentus*), cutthroat trout (*Oncorhynchus clarkii*) and rainbow

trout (*Oncorhynchus mykiss*) sampled in 35 streams in northcentral Idaho and southwestern Montana (Table 1).

Constant Detection Rates of Fish Among Surveyed Units

Hankin and Reeves' (1988) approach is essentially a double sample in which a "cheap" index (snorkel count) is adjusted for incomplete detection of fish by a more expensive and (presumably) accurate count (removal estimate). Their approach also assumes constant detection or capture probabilities of fish among sampling units (Seber 1982, Pollock and Kendall 1987). Detection rates of fish can vary both temporally and spatially due to factors discussed previously. Thus, detection rates are more likely to vary than remain constant among sampling units in typical field situations.

Simulations

Details

I randomly generated 50,000 simulation runs in SAS (SAS, Inc., 2000) to investigate effects of different types and levels of violations to key assumptions of Hankin and Reeves' (1998) approach on 95% confidence interval coverage and bias of population estimates. Specifically, I investigated effects of different levels of correlation between removal estimates and diver counts (<0.80, 0.80-0.85, 0.85-0.90, 0.90-0.95, and 0.95-1.0) under constant (1.0) and nonconstant (0.1-0.9) detection probabilities among sampling units and with removal estimates equal to (100%) and less than (0-99%) true numbers of fish within sampling units. Further, I incorporated increased variances in removal estimates with larger snorkel counts.

Strata, size of systematic samples, range of diver counts, and abundance were defined based on Hankin and Reeves' (1988:838-842) example application. Detection

rates of fish for diver counts were randomly chosen from 0.2-0.6, which was approximately the range reported by Rodgers et al. (1992). Diver counts also were assumed to be independent as per Hankin and Reeves' example. I used spatial distributions with a standardized Morisita index (Morisita 1962, Smith-Gill 1975) of 0.51 to incorporate population clustering at the 95% confidence level (Krebs 1999), which mimicked spatial clustering of fish populations due to heterogeneity in stream habitats, schooling behavior of fish, and so forth.

Results

When removal estimates equaled true numbers of fish within sampling units, 95% confidence interval coverage averaged about 90% for both constant and nonconstant detection probabilities among units across the range of correlation values (Table 2). These intervals did not reach the nominal rate of coverage because there was a less than perfect linear relationship between removal estimates and diver counts, sample sizes were less than 30 (as per Hankin and Reeves example), coefficients of variation were greater than 0.1 ($C\bar{V} = 0.19$ [SE = 0.0002]; minimum = 0.11 and maximum = 0.47), and spatial clustering lead to an underestimation of the single systematic sample variance based on random sampling (i.e., it assumed a random spatial distribution of individuals; Scheaffer et al 1990). Nonetheless, this rate of coverage is probably close enough to the nominal rate to be suitable for population estimation. When not included within the 95% confidence interval, the true number often was within 8-10% of either end (Table 2).

Hankin and Reeves' (1988) approach performed poorly if a removal estimate did not equal the true number of fish (Table 2), with the exception where detection probabilities were >0.85 and constant across sampling units (95% confidence interval

coverage = 90%). Average confidence interval coverage always was less than 40%, and no more than 20% when correlation values were <0.8 . Further, true values were typically far above or below the confidence limit for noncovered intervals, especially at lower correlation values (Table 2), which implies large bias. Variability in population estimates was greater than those where removal estimates equaled true numbers of fish (true number $>$ removal estimate: $C\bar{V} = 0.27$ [SE = 0.0004]; minimum = 0.13 and maximum = 0.73). Also note that relatively few runs (452/12,500 [3.6%]) were produced with high correlation values (>0.90) under nonconstant detection probabilities among units and where removal estimates did not equal true numbers of fish. This was probably due to additional variability of removal estimates as they represented differing proportions of true numbers of fish among sampling units, which mirrors natural situations.

Potential Modification and Alternative

Simulation results indicated that Hankin and Reeves' (1988) approach may produce misleading results when removal estimates do not equal true numbers of fish within sampling units. Therefore, in this section I discuss a potential modification and alternative to their approach. I emphasize that these suggestions are not offered as definitive remedies, but rather as possible avenues for further research and development. In any event, untested methods of abundance estimation should be field tested and validated using a known population before they are fully implemented.

Mark-Recapture Models and Individual Covariates

A possible modification to Hankin and Reeves' approach is to replace Zippin's removal model, which is a mark-recapture model in which there is trap response (i.e., Model M_b ; Otis et al. 1978, White et al. 1982), with another type of mark-recapture

model. Within the mark-recapture framework, there are a number of models available that relax different assumptions related to capture probabilities of individuals, including a generalized removal model (Model M_{bh}) that accounts for heterogeneity in capture probabilities. Further, one should incorporate individual covariates that represent the most important factors affecting capture probabilities of fish into the mark-recapture model. These covariates would be measured at each surveyed unit. Analogous applications have been suggested for both aquatic (Bayley 1993) and terrestrial (Pollock et al. 1984, Samuel et al. 1987, Steinhorst and Samuel 1989, Manly et al. 1996) environments. Huggins (1989, 1991) developed mark-recapture models that allow individual covariates; these models have been incorporated into program MARK (White and Burnham 1999).

Variance estimates produced by mark-recapture models typically are not corrected for overdispersion so that they underestimate the true variance, perhaps by as much as 2 to 3 times (Bayley 1993). Quasi-likelihood (Wedderburn 1974) theory often is used as a basis for correction of overdispersed data (see Cox and Snell 1989, McCullagh and Nelder 1989). At present, program MARK does not provide a variance inflation factor (Cox and Snell 1989) that corrects for overdispersion in this particular class of models, but it does allow users to specify different values of these factors into these models (G. White, Dept. of Fishery and Wildlife Biology, Colorado State University, Fort Collins, Colo., personal communication).

Line Transect Sampling

Line transect sampling has been broadly applied to both terrestrial and marine species (Buckland et al. 1993), but rarely to freshwater aquatic organisms. Ensign et al.

(1995) applied line transect methods via snorkeling to estimate abundance of benthic stream fishes, but they did not compare their results to a known population and hence the usefulness of this technique within streams remains in question. Nonetheless, line transect sampling potentially offers an alternative method to density estimation that deserves more investigation in stream environments.

In line transect sampling, which is a form of distance sampling (Buckland et al. 1993), the observer moves along a randomly selected line and records the perpendicular distance from the line to every individual (or group of individuals) detected or to the distance category containing it. Then, the detection distances are fitted to a variety of models and the best fitting model is used to generate estimates of density and variance that have been adjusted for visibility bias and overdispersion (Buckland et al. 1993). The critical assumptions for line transect sampling are that every individual on the transect are detected, distances are measured to each detected individual's original location (or distance category), and distances (or distance categories) are measured without error. In addition, reliable estimates generally require high detection rates for individual near the line and a minimum of 80 detections (Buckland et al. 1993). If distance categories are used rather than individual distances, then at least 4-5 of these categories are required to adequately fit the detection function (Buckland et al. 1993). Program DISTANCE (Laake et al. 1993, Thomas 1999) is available to analyze line transect data.

In practice, detecting every fish on a line may be problematic, depending on the size classes that are being sampled. Smaller fish may be hidden in the substrate and hence undetectable to the snorkeler. This may be minimized if a species presence in the water column is strongly correlated with time of day. For instance, bull trout may be

more visible to snorkelers at night than during the day (Peterson 2000). There are methods that correct for incomplete detections on the line, but these typically require independent observers operating simultaneously (see Buckland et al. 1993:200-217 for a review of different methods). However, note that Hankin and Reeves' (1988) approach also requires independence of diver counts.

Movement of fish in response to an observer prior to detection also is a concern in line transect sampling. Violations of this assumption may be minimized via implementation of proper snorkeling protocol. For instance, when moving slowly and carefully, snorkelers can approach close enough to identify individual rainbow and cutthroat trout during daytime without causing a flight response. Further, bull trout typically remain stationary in the water column when spotlighted at night (J. Guzevich, U.S.D.A Forest Service, Rocky Mountain Research Station, Boise, Idaho, personal communication). In addition, bias related to fish movements will be minimized if these movements remain within a given distance category (see below).

Accurate distance measurements to mobile individuals are more likely to be made if distances are recorded in categories. Snorkelers could use a calibrated mask-bar (Swenson et al. 1988) or similar device to estimate distance categories to detected fish, assuming snorkelers could either maintain a constant height above the streambed or record this height with every distance measurement. A somewhat analogous approach was used in aerial line transect surveys by Johnson et al. (1991) where marks on the struts of their airplane corresponded to a given distance from the line to a sighted object on the ground as long as the height above the ground was known.

Discussion

Hankin and Reeves' (1988) approach is particularly sensitive to the assumption that removal estimates are equal to true numbers of fish within surveyed units. Previous studies have indicated that removal estimates can underestimate true abundance by more than 50% (e.g., Riley et al. 1993). Although Hankin and Reeves recognized the shortcomings of using removal estimates in place of complete counts, they still believed their approach to be a practical alternative to estimating fish abundance in small streams. Nonetheless, this assumption should be evaluated before their approach is fully implemented

When reviewing removal estimates, high precision should not be confused with low bias. Low capture probabilities and population sizes (or violation of model assumptions) may yield highly precise abundance estimates that are far from the true population value. This results in what Anderson et al. (1998) described as "highly precise, wrong answers." Estimated capture probabilities can be misleadingly high in these situations (White et al. 1982; see Riley et al. 1993 for empirical evidence) and hence should not be relied upon as an index to validity.

A pilot study should be conducted to ensure that the proposed method for estimating abundance is both reasonable, with respect to its assumptions and feasibility, and cost-efficient (Burnham et al. 1987, Buckland et al. 1993, Thompson et al. 1998). Proper validation of an enumeration method requires a known (or nearly so) population of fish to serve as a benchmark to evaluate bias (e.g., use of a known stocked or marked population of fish by Rodgers et al. 1992). Comparing two index or untested methods (e.g., snorkel counts vs. unverified removal estimates) will only reveal the relative

sampling efficiency between these methods. This comparison is meaningless if the objective is validation, i.e., evaluating magnitude of bias. Usefulness of these abundance estimates depends on how closely they approximate reality, not on how closely they approximate each other.

If neither Hankin and Reeves' (1988) approach nor the suggested alternatives are feasible in a given stream, emphasis should be placed on developing alternatives to estimating fish abundance rather than simply defaulting to an existing approach that is known to be inappropriate. Indeed, too often an existing method is implemented with little thought towards verifying whether it will produce meaningful abundance estimates in the species of interest. I argue that poor data (i.e., seriously biased and imprecise abundance estimates) are worse than no data in this case. Serious biases can lead to misleading interpretations, whereas "the inertia of...imprecise results must be overcome before a legitimate survey can be conducted" (Thompson et al. 1998:xii). Great care and thought must be applied to designing and validating enumeration procedures, for population estimates are only as reliable as the data that generated them.

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Table 1. Estimated correlation coefficient (r) between snorkel counts and multiple pass removal estimates of smaller size classes (70-100 mm) of brook trout, bull trout, cutthroat trout, and rainbow trout in 35 different small streams sampled in northcentral Idaho and southwestern Montana (R. Thurow, U. S. D. A. Forest Service, 316 E. Myrtle St., Boise, ID, unpublished data).

Species	No. of streams	No. of stream sections	r
Brook trout	8	14	0.38
Bull trout	25	65	0.20
Cuttthroat trout	16	25	0.20
Rainbow trout	17	50	0.44

Table 2. Ninety-five percent confidence interval (CI) coverage of true total abundance and relative location of true total abundance and upper or lower 95% confidence limits (CL) from 50,000 simulation runs based on Hankin and Reeves' (1988) approach and example application.

Detection probability among units	Relationship between true unit numbers and removal estimates	Removal estimate and diver count correlation	Percent of 95% CIs containing true total abundance (no./total)	Relationship between true total abundance and lower or upper CL for 95% CI not containing the true total abundance			
				Average % below (SE)	n	Average % above (SE)	n
Same (1.0)	Equal (100%)	<0.80	93 (189/203)	3	4 (1)	11	8 (2)
		0.80-0.85	90 (1024/1138)	38	7 (1)	76	8 (1)
		0.85-0.90	88 (3049/3451)	129	8 (1)	273	8 (<1)
		0.90-0.95	90 (4494/5014)	246	8 (<1)	274	7 (<1)
		0.95-1.0	90 (2431/2694)	255	10 (1)	8	5 (1)
	Unequal (0-99%)	<0.80	14 (58/427)	0	-	369	41(1)
		0.80-0.85	18 (191/1052)	0	-	861	40 (1)
		0.85-0.90	22 (435/2021)	0	-	1586	37 (1)
		0.90-0.95	29 (892/3033)	1	6 (-)	2140	35 (<1)
		0.95-1.0	38 (502/1320)	0	-	818	29 (1)
Different (0.1-0.9)	Equal (100%)	<0.80	94 (190/202)	3	5 (2)	9	5 (1)
		0.80-0.85	90 (991/1105)	39	7 (1)	75	9 (1)
		0.85-0.90	89 (3026/3391)	121	8 (1)	244	8 (<1)
		0.90-0.95	89 (4565/5142)	331	10 (<1)	246	8 (<1)

	0.95-1.0	83 (2200/2660)	451	11 (<1)	9	9 (1)
Unequal						
(0-99%)	<0.80	20 (1540/7806)	0	-	6266	36 (<1)
	0.80-0.85	28 (484/1718)	0	-	1234	31 (1)
	0.85-0.90	35 (398/1123)	0	-	725	30 (1)
	0.90-0.95	37 (158/432)	0	-	274	28 (1)
	0.95-1.0	35 (7/20)	0	-	13	20 (4)
