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A comparison of two methods for sampling biomass of aquatic plants

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ABSTRACT

We evaluated the performance of a boat-based vertical rake method for sampling aquatic plant biomass to determine whether it was a suitable alternative to diver quadrat sampling. Boat-based methods are generally considered to be safer and require less collection time than diver quadrat sampling, but they may be less accurate and precise. We compared three aspects of the two sampling methods: detection rates for individual taxa, comparability of biomass estimates, and precision of biomass estimates. Detection rates for the two sampling methods were comparable for all evaluated plant taxa. Similarly, rake and quadrat biomass estimates were comparable in magnitude and precision for most individual plant taxa. However, rake estimates were significantly higher than quadrat estimates for coontail (*Ceratophyllum demersum* L.) and flat-stemmed pondweed (*Potamogeton zosteriformis* Fern.). Linear regression of rake yields against quadrat yields (log-transformed) showed significant positive relationships for all evaluated taxa, and slopes that did not differ significantly from 1 for most taxa. However, rake estimates of total biomass (all taxa combined) were significantly higher and less precise than quadrat estimates, particularly for samples collected from areas with dense plant growth (>200 dry g m^{-2}). Although our results suggested that the rake method was not a perfect surrogate for the quadrat method, the increased sample collection afforded by the vertical rake method may offset the method's inherently lower precision. Consequently, we concluded that the vertical-rake method was a suitable alternative to the diver quadrat method for collection of aquatic plant biomass, particularly in large-scale studies requiring high sampling intensity over expansive areas.

Key words: abundance, macrophyte, quadrat, rake, scuba.

INTRODUCTION

Submersed aquatic plants play an important role in freshwater systems, affecting nutrient dynamics, trophic interactions, biological assemblages, and fish productivity (Jeppeson et al. 1998, Scheffer 2004). Consequently, aquatic plant communities are often monitored as a part of large-scale lake management and restoration projects. Although plant communities can be evaluated in many different ways (e.g., species composition, distribution of plant growth, maximum depth of colonization), plant abundance is generally a key metric in aquatic plant studies. Plant abundance has been evaluated using measures of biovolume, plant height, and density ratings, but biomass per unit area (e.g., dry g/m^2) is the standard measure (Nichols 1984, Madsen 1993, Valley and Drake 2005).

Diver quadrat sampling has been widely accepted as the most accurate and precise method for collecting aquatic plant biomass samples (Sheldon and Boylen 1978, Downing and Anderson 1985, Madsen 1993, Wetzel and Likens 2000); however, diver quadrat sampling does have drawbacks, such as expensive equipment, required scuba certification, and the inherent safety risks of diving. Consequently, boat-based methods that use mechanical dredges, coring devices, or rakes have been adopted by many researchers to address concerns about the safety of diving or to reduce the time and effort required to collect numerous samples (McCauley 1975, Schloesser and Manny 1984, Crowell et al. 1994, Capers 2000, Rodusky et al. 2005, Kenow et al. 2007, Skogerboe et al. 2008).

When designing aquatic vegetation studies, researchers strive to incorporate methods that will provide the most accurate estimates, with the greatest precision, for the least amount of effort (Nichols 1982, Downing and Anderson 1985, Canfield et al. 1990, Madsen 1993). For this reason, diver quadrat sampling has generally been favored in small-scale studies that required high precision from a relatively small number of samples. However, large-scale vegetation studies may achieve better system-wide estimates of plant biomass by using a boat-based method that allows more rapid collection of many samples, provided that the level of bias and loss of precision relative to the quadrat method are sufficiently small.

Previous evaluations of boat-based dredge samplers (Westlake 1969, Schloesser and Manny 1984, Sliger et al. 1990, Rodusky et al. 2005) and large coring devices (McCauley 1975, Madsen et al. 2007) suggested that these samplers could achieve levels of accuracy and precision fairly comparable to the diver quadrat method. However, these boat-based methods generally required cumbersome field equipment and a substantial amount of time and effort to collect each sample (McCauley 1975, Schloesser and Manny 1984, Madsen et al. 2007). Consequently, these mechanical samplers may be suitable alternatives to quadrat sampling in studies where safety concerns far outweigh the need for rapid sample collection but do not provide sufficient reductions in sampling time to be favored in large-scale studies that require the collection of numerous samples.

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Alternatively, boat-based rake methods allow rapid sample collection with simple equipment, making them attractive for large-scale studies that require the collection of many samples. Consequently, several researchers have recently adopted a vertical rake method for sampling aquatic plant biomass in large-scale vegetation management studies (Crowell et al. 1994, Skogerboe et al. 2008). For this method, samples are collected by lowering a long-handled rake vertically to the bottom and turning it 3 times on its axis before retrieving. Biomass is then estimated as the amount of plant material collected from within the circular area sampled by the rotated rake head. This differs from previously used rake methods that collected plant biomass by dragging a rake over a defined sampled area. Although the vertical rake method has been used to conduct more than 150 lake-wide surveys of aquatic plant biomass in Minnesota and Wisconsin, little is known about its comparability to diver quadrat sampling. The aim of our study was to evaluate the comparability of these 2 sampling methods across a wide range of plant taxa and growth densities. The specific objectives were to:

1. Compare the ability of the vertical rake method and diver quadrat method to detect the presence of different plant taxa.
2. Determine whether the vertical rake method produces biomass estimates similar to diver quadrat estimates for individual taxa and total biomass (all taxa combined).
3. Compare the precision of biomass estimates produced by the 2 sampling methods for individual taxa and total biomass.

MATERIALS AND METHODS

Sample collection

We selected 38 vegetated sample sites from 4 lakes in the Twin Cities metropolitan area of Minnesota (Fish Lake, Scott Co.; Otter Lake, Anoka Co.; Rebecca Lake, Hennepin Co.; Weaver Lake, Hennepin Co.). These lakes ranged in size from 60 to 130 ha and from 6 to 16 m deep, with water clarity ranging from 0.5 m to >3.0 m at the time of sample collection. Each lake supported from 11 to 27 aquatic plant taxa. All sampled sites were located in areas between 0.5 and 2.5 m deep with soft sediments and were selected to provide a range of biomass density and plant diversity. At each site, we placed marker buoys at 3 points around the front of an anchored boat (starboard, bow, and port) and collected paired samples (1 rake sample and 1 quadrat sample) at each point from opposite sides but within 0.25 m of the marker buoy's anchor, resulting in 114 samples for each sampling method.

We collected rake samples using a 0.33-m wide, 14-tine, single-headed rake attached to a 3-m long pole. We lowered the rake into the water with the handle perpendicular to the water surface until the rake head reached the sediment. We then rotated the rake on its axis 3 full turns while maintaining contact with the sediment and then retrieved the rake while continuing to turn it slowly to prevent the loss of collected plants. Each sample collected plants from approximately 0.09 m^2 of sediment. Upon retrieval, we placed all collected plants into a labeled plastic bag, drained out excess water, and stored the samples on ice while in the field.

Quadrat samples were collected by divers using scuba equipment and a square PVC quadrat frame (32 by 32 cm; 0.10 m^2) The quadrat frame was placed on the sediment surface immediately adjacent to each marker buoy, and all plant shoots within the frame were removed manually by breaking stems at the sediment. Collected plants were placed into a labeled sealable plastic bag while underwater.

In the lab, all samples were rinsed to remove sediment, and any below-ground plant structures were removed. Remaining above-ground plant structures were sorted by taxonomic group (Crow and Hellquist 2000a, 2000b), dewatered in a salad-spinner to reduce drying time, and dried at 105 C (Wetzel and Likens 2000) for at least 48 h prior to weighing. Results were converted to dry mass per square meter (g/m^2).

Data analysis

To reduce variability due to the inherent patchiness of aquatic plant growth at the sampled scale, we calculated the mean of the triplicate samples collected at each site with each method (site-average yield). These site-average yields (henceforth referred to as "yields") were then $\log_{10}$-transformed to reduce skewness and stabilize variance. All subsequent statistical analyses were conducted using R statistical software version 2.8.1 (R Development Core Team 2008).

We compared the detection of individual plant taxa by each sampling method using McNemar's χ^2 test of symmetry with a continuity correction for small sample size (Agresti 1990, Zar 2010). This test compared the number of sites where only the rake method detected a given taxon to the number of sites where only the quadrat method detected the same taxon, and then tested whether this ratio departed significantly from 1:1. To meet minimum sample size requirements for this test, we only evaluated taxa that occurred at more than 5 sites (rake or quadrat; n = 17 taxa; Table 1).

We assessed the overall comparability of rake and quadrat yields ($\log_{10}$-transformed) by calculating Lin's concordance correlation coefficients (Lin 1989, Zar 2010) for both total biomass and individual taxa. Concordance correlation measures the degree to which pairs of observations fall on the line of perfect concordance (y = x), thus incorporating measures of both bias and precision, with the resulting concordance correlation coefficient (CCC) ranging from 0 (no agreement between observations) to 1 (perfect agreement between observations). To meet minimum sample requirements for this test, we only evaluated taxa encountered by both sampling methods at more than 5 in-common sites (n = 9 taxa; Table 1).

To further evaluate the comparability of the 2 methods, we used linear regression on $\log_{10}$-transformed rake (y) and quadrat yields (x) for individual taxa and total biomass (all taxa combined). To ensure that regressions included sufficient data from each sampling method to produce meaningful results, we only conducted regressions for taxa encountered by both sampling methods (rake and quadrat) at more than 5 in-common sites (n = 9 taxa; Table 1). We chose to exclude sites where only one method detected a given taxon to avoid the strong influence of zeroes upon the regression slopes. We test-

Table 1. Taxa encountered in samples collected from 38 sites across four lakes using the vertical rake and diver quadrat methods. *N* is the number of site occurrences for each taxon using each method.

Taxon	Common Name	N_{Rake}	$N_{Quadrat}$
All Taxa (combined)	—	38	38
Bidens beckii	Water marigold	1	3
Ceratophyllum demersum[abc]	Coontail	31	34
Chara sp.[abc]	Muskgrass	14	9
Eleocharis acicularis	Needle spikerush	1	3
Elodea canadensis [abc]	Canadian waterweed	12	11
Fontinalis antipyretica	Aquatic moss	1	0
Lemna minor	Lesser duckweed	3	2
Lemna trisulca[a]	Star duckweed	4	6
Myriophyllum sibiricum[abc]	Northern watermilfoil	25	25
Myriophyllum spicatum [a]	Eurasian watermilfoil	9	3
Najas flexilis[abc]	Slender water nymph	8	9
Najas guadalupensis [ab]	Southern water nymph	6	10
Nuphar variegata	Spatterdock	0	1
Nymphaea odorata	White waterlily	2	1
Potamogeton amplifolius	Large-leaf pondweed	2	2
Potamogeton crispus (sprouts) [ab]	Curlyleaf pondweed	6	7
Potamogeton foliosus	Leafy pondweed	1	1
Potamogeton gramineus[a]	Variable-leaf pondweed	7	5
Potamogeton nodosus	Long-leaf pondweed	1	1
Potamogeton praelongus [ab]	White-stemmed pondweed	9	7
Potamogeton richardsonii [a]	Clasping-leaf pondweed	3	5
Potamogeton robbinsii [a]	Robbins' pondweed	2	6
Potamogeton zosteriformis [abc]	Flat-stemmed pondweed	14	15
Ranunculus longirostris	Longbeak buttercup	2	1
Stuckenia pectinata[abc]	Sago pondweed	14	15
Utricularia vulgaris	Bladderwort	1	1
Vallisneria americana[abc]	Wild celery	19	18
Zannichellia palustris	Horned pondweed	1	4
Zosterella dubia[abc]	Water stargrass	24	22

[a]Taxa encountered at more than 5 sites in total.
[b]Taxa that occurred at more than 5 sites for each sampling method (not necessarily the same sites).
[c]Taxa that occurred in both rake and quadrat samples at more than 5 in-common sites.

ed each regression using standardized major axis estimation and testing routines (SMATR package; R statistical software) to determine whether relationship slopes deviated from 1, and whether intercepts deviated from zero. We also examined normal quantile-quantile plots and residual plots for all regressions to verify assumptions of linearity, normality, and stable variance. Furthermore, we used mixed-effects models (Pinheiro and Bates 2000) to verify that the nested sample collection in our study (samples clustered within lakes and sites) did not significantly affect the lack of fit or bias the slope of our regression models.

To compare the precision of yields (log_{10}-transformed) from the 2 sampling methods, we tested for equality of variance using Levene's test (Levene 1960, Zar 2010). Only taxa encountered by both sampling methods at more than 5 in-common sites were evaluated using Levene's test (n = 9 taxa; Table 1). To enable easier comparisons to data from previous studies, we also calculated the coefficient of variation (CV) on nontransformed yields for each of the 12 taxa encountered at more than 5 sites for each method (Table 1).

RESULTS AND DISCUSSION

We collected plant biomass at all 38 sample sites and identified 29 aquatic plant taxa (Table 1). Of these 29 taxa, 17 were encountered at more than 5 total sites with either sampling method, 12 were encountered at more than 5 sites for each method, and 9 were encountered by both sampling methods together at more than 5 in-common sites.

Overall, the number of plant taxa retrieved in individual rake samples (4.9 ± 0.28 taxa; $\bar{x}$ ± 1SE) was higher (P = 0.04; Welch's t-test) than the number of taxa retrieved in quadrat samples (4.1 ± 0.21 taxa). However, results from McNemar's tests indicated that the vertical rake method detected all but one of the 17 evaluated taxa at rates that were similar to the quadrat method (Table 2). The only exception was Eurasian watermilfoil (*Myriophyllum spicatum* L.), which was detected significantly more frequently ($P = 0.04$) by the rake method.

Lin's CCC for the 9 evaluated taxa ranged from 0.73 to 0.92, with most taxa exhibiting CCCs between 0.82 and 0.84 (Table 3). This indicates that although there was not perfect agreement between rake and quadrat yields, the level of comparability was generally high and consistent across taxa. Northern watermilfoil (*Myriophyllum sibiricum* Komarov) and wild celery (*Vallisneria americana* Michx.) exhibited the highest CCC (0.92), whereas coontail exhibited the lowest CCC (0.73). However, the CCC for total biomass (0.74) was lower than all individual taxa except coontail.

Linear regressions also indicated that vertical rake yields were comparable to quadrat yields for most of the 9 evaluated taxa (Figure 1). The relationship of rake yield (y) to quadrat yield (x) was significant (all $P < 0.025$) and positive for all of the evaluated taxa and explained 61 to 89% of biomass variability. Furthermore, most of the taxa exhibited regression slopes that did not deviate significantly from 1 and intercepts that did not differ significantly from zero (Figure 1), indicating that rake and quadrat yields (log_{10}-transformed) were statistically equivalent for most taxa. However, there were several notable exceptions. Regression intercepts indicated that flat-stemmed pondweed and sago pondweed (*Stuckenia pectinata* [L.] Börner) were consistently over-sampled by the rake across the measured range of biomass relative to quadrat estimates. By contrast, a slope <1 indicated that water stargrass (*Zosterella dubia* [Jacq.] Small) was under-sampled by the rake, particularly when present at high biomass, and the slope and intercept for northern watermilfoil indicated that it was over-sampled by the rake relative to the quadrat when present at low biomass, but under-sampled by the rake at high biomass (Figure 1).

When all plant taxa were considered collectively (total biomass), the vertical rake method consistently produced higher estimates than the quadrat method at biomass densities > 200 g/m^{-2} (log_{10} = 2.3). Although the relationship between rake yields (y) and quadrat yields (x) for total biomass was significant ($P < 0.001$), positive, and explained 78% of the variability in biomass (Figure 2), the regression slope (1.39) was significantly greater than 1 ($P = 0.003$), and the intercept (-0.66) was significantly less than zero ($P = 0.026$). Closer inspection of the regressions for total biomass and for individual taxa suggested that this over-sampling of total biomass by the rake at high plant density was almost entirely attributable to over-sampling of 2 taxa, coontail and flat-

Table 2. Comparison of detection rates for individual taxa by rake and quadrat biomass sampling methods; McNemar's χ^2 test for symmetry on presence (+) or absence (–) by method (R = rake, Q = quadrat) at the 38 sample sites. *McNemar's* χ^2 is the test statistic for the test of R–/Q+ vs. R+/Q– (df = 1), and *P* is the associated p-value.

Taxon	*R+/Q+*	*R-/Q+*	*R+/Q-*	*R-/Q-*	*McNemar's* χ^2	*P*
All Taxa (combined)	38	0	0	0	0.00	1.00
Ceratophyllum demersum	29	5	2	2	0.57	0.45
Chara sp.	7	2	7	22	1.78	0.18
Elodea canadensis	10	1	2	25	0.00	1.00
Lemna trisulca	2	4	2	30	0.17	0.68
Myriophyllum sibiricum	23	2	2	11	0.00	1.00
Myriophyllum spicatum	3	0	6	29	4.17	0.04
Najas flexilis	6	3	2	27	0.00	1.00
Najas guadalupensis	4	6	2	26	1.13	0.29
Potamogeton crispus	4	3	2	29	0.00	1.00
Potamogeton gramineus	3	2	4	29	0.17	0.68
Potamogeton praelongus	5	2	4	27	0.17	0.68
Potamogeton richardsonii	2	3	1	32	0.25	0.62
Potamogeton robbinsii	1	5	1	31	1.50	0.22
Potamogeton zosteriformis	14	1	0	23	0.00	1.00
Stuckenia pectinata	11	4	3	20	0.00	1.00
Vallisneria americana	16	2	3	17	0.00	1.00
Zosterella dubia	19	3	5	11	0.13	0.72

Table 3. Lin's concordance correlation coefficient (CCC) for paired rake and quadrat site average yields of total biomass and individual taxa. Taxa listed in order of increasing concordance.

Taxon	CCC	95% CI	N (pairs)
All Taxa (total biomass)	0.74	0.62-0.83	38
Ceratophyllum demersum	0.73	0.56-0.84	29
Zosterella dubia	0.78	0.51-0.91	18
Elodea canadensis	0.82	0.52-0.94	10
Potamogeton zosteriformis	0.82	0.61-0.92	14
Chara sp.	0.84	0.40-0.97	7
Najas flexilis	0.84	0.29-0.97	6
Stuckenia pectinata	0.84	0.57-0.95	11
Myriophyllum sibiricum	0.92	0.82-0.96	22
Vallisneria americana	0.92	0.80-0.97	16

stemmed pondweed. Coontail was the most commonly encountered taxon in our study and was the greatest contributor to total biomass at 53% of sites for rake samples (519 ± 138 g/m^{-2}; $\bar{x}$ ± 1SE) and 55% of sites for quadrat samples (134 ± 25 g/m^{-2}). Consequently, the over-sampling of coontail by the rake likely had a substantial effect upon total biomass estimates.

Our evaluations of the precision of rake and quadrat yields indicated that both methods produced estimates of similar precision for most of the evaluated taxa. Tests for equality of variance (Levene's test) indicated that the variance of rake and quadrat yields (log-transformed) were statistically similar ($P > 0.05$) for all evaluated taxa. However, for total biomass (all taxa combined), rake yields were significantly less precise than quadrat yields ($P < 0.008$). Similarly, the coefficient of variation (CV) for rake yields was within 20% of the quadrat CV for 8 of the 12 evaluated taxa (Table 4), with the exceptions being flat-stemmed pondweed (rake = +61%), coontail (+35%), northern watermilfoil (+29%), and southern water nymph (*Najas guadalupensis*; -40%). However, the CV for rake total biomass (1.33) was 1.7 times larger than the CV for quadrat total biomass (0.79; Table 4), roughly mirroring our results from Levene's test.

Our results indicate that the vertical rake method was closely comparable to the diver quadrat method in its ability to detect a wide range of aquatic plant taxa. This finding contrasts with results presented by Capers (2000) and Wood (1963) who reported substantially lower species richness estimates and lower frequency of occurrence for select taxa when using dragged rakes and grapnels relative to diver quadrat sampling. This discrepancy suggests that the vertical rake method we tested was more effective at snagging and retrieving plants than previously used rake and grapnel methods. This enhanced ability may be due to the multiple passes of the rake head (3 rotations) over the defined sample area. These multiple passes may have resulted in more thorough removal and retention of plants than the single pass of a dragged rake or grapnel. Capers (2000) and Wood (1963) both reported that grapnels and rakes often failed to snag and retrieve small or firmly-rooted plants. Because we did not thoroughly assess such plants in our study, caution should be exercised when using the vertical rake method in waters with small, firmly-rooted taxa.

Our study also showed that rake and quadrat estimates of biomass were fairly comparable for individual taxa; however the rake did not sample all taxa equally well. Rodusky et al. (2005) also reported that different taxa showed differing levels of comparability between rake and quadrat biomass estimates. In particular, they found that relative to quadrat sampling, their rake method (hinged oyster-tong rake) under-sampled dense stands of wild celery and *Potamogeton* species and over-sampled moderately dense stands of hydrilla. The differences we observed in rake and quadrat comparability among taxa suggest that the overall comparability of the 2 methods for estimating total biomass is dependent upon the dominant taxa present at the monitored locations.

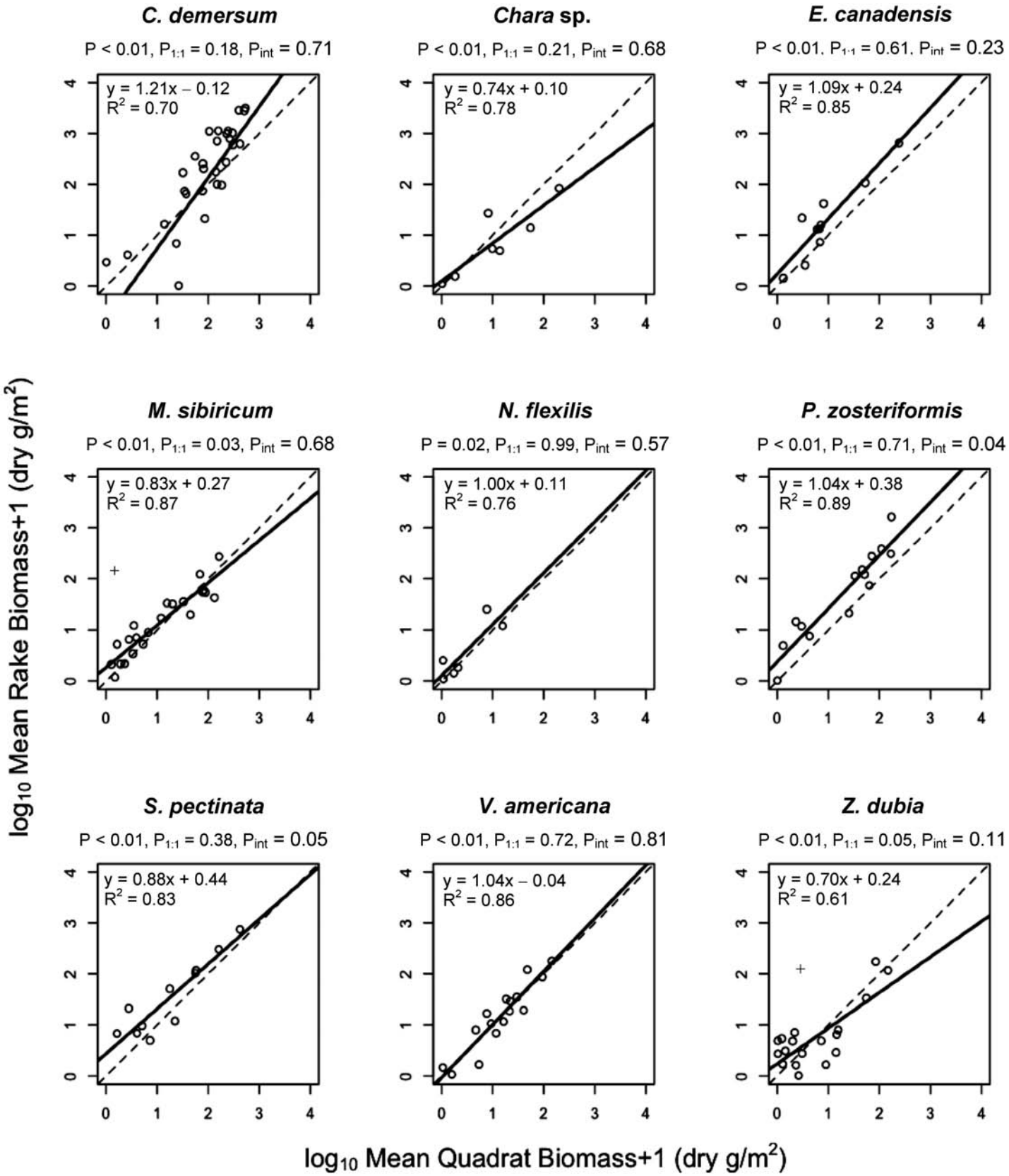

Figure 1. Relationship between rake and quadrat yields (log_{10}-transformed) for individual taxa encountered by both sampling methods together at more than five in-common sites (Table 1). Solid lines indicate fitted regressions and dashed lines indicate 1:1 correspondence. Rejected outliers indicated by "+". P is the p-value associated with the regression, $P_{1:1}$ is the p-value associated with the test for slope = 1, P_{int} is the p-value associated with the test for y-intercept = 0.

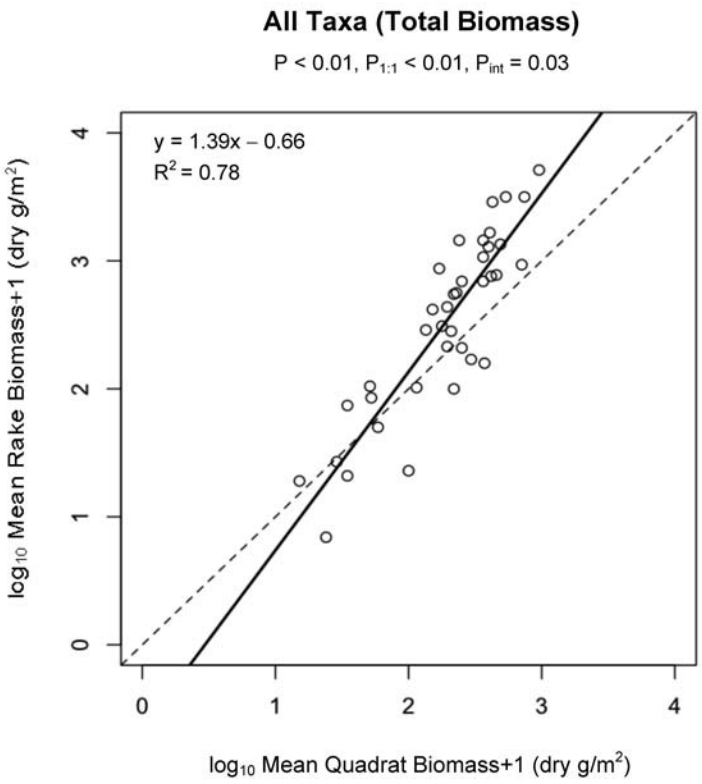

Figure 2. Relationship between rake and quadrat yields (log_{10}-transformed) for total biomass (all plant taxa combined). Solid line indicates fitted regression and dashed line indicates 1:1 correspondence.

Our rake estimates of northern watermilfoil, slender water nymph (*Najas flexilis* Willd.), and wild celery were very similar to quadrat estimates in both magnitude and precision. However, the rake consistently over-sampled flat-stemmed pondweed across the range of sampled growth densities, and severely over-sampled coontail when present at high density.

Over-sampling of coontail by the rake was likely due to coontail's strong intertwining stems and lack of roots, which allowed coontail biomass to be pulled from outside of the intended sample area as the rake was turned. Similar discrepancies may also be expected for taxa that form dense surface canopies, such as Eurasian watermilfoil, curlyleaf pondweed, and hydrilla (*Hydrilla verticillata* [L.f.] Royle). These taxa may be over-sampled by the rake due to the inclusion of canopy growth that extended outside of the intended sample area. The rake may be more prone to snagging this canopy growth as the rake is brought to the surface. Our results did indicate some degree of over-sampling by the rake for canopy-forming taxa (Eurasian watermilfoil, flat-stemmed pondweed, and sago pondweed), but we did not sample dense stands of these taxa; however, Crowell et al. (1994) reported a similar pattern of over-sampling by the rake in dense stands of Eurasian watermilfoil.

For total biomass estimates (all taxa combined), our results indicated that rake estimates were significantly higher and less precise than quadrat estimates, particularly in samples collected from areas with dense plant growth. Crowell et al. (1994) similarly reported substantially higher estimates of total biomass in rake samples (1669 ± 748 g/m²; $\bar{x}$ ± 1SE) than in quadrat samples (799 ± 379 g/m²), but concluded that the precision of the rake method they tested was sufficiently comparable to the quadrat method (coefficient of variation; rake = 116%, quadrat = 110%) to allow meaningful relative comparisons of total biomass (comparing rake samples to previously collected rake samples). Rodusky et al. (2005) similarly reported little difference between the precision of their rake estimates and quadrat estimates, but unlike Crowell et al. (1994), found no consistent pattern to over-sampling or under-sampling of total biomass in rake samples relative to quadrat samples. Both of these previous studies were based on a relatively small number of samples collected from nearly monotypic stands of plants and only compared estimates for a few taxa. However, the reported results further suggest that the vertical rake method may provide substantially more precise estimates of total biomass at sites not dominated by dense coontail growth. Given the differences we observed in rake performance for sampling individual taxa, further evaluation of the vertical rake method should be conducted prior to adoption of this method in waters dominated by plant taxa that were not thoroughly evaluated in our study, particularly for dense stands of Eurasian watermilfoil, curlyleaf pondweed, and hydrilla.

Although our results indicate that the vertical rake method was not a perfect surrogate for diver quadrat sampling, the rake method would allow greater sampling frequency and the collection of substantially more samples than the quadrat method with less effort. This could increase the likelihood of detecting rare plant taxa (Mikulyuk et al. 2010), increase the resolution and geographic coverage of assessments within the study area, and offset the rake method's inherently lower precision relative to the diver quadrat method for estimating total biomass.

To estimate the number of rake samples needed to achieve the same level of precision as the quadrat method, we used a standard formula for estimating sample size based on the anticipated sample variance and mean (Zar 2010).

$$n = \frac{s^2 t^2_{0.05,\, n-1}}{(E \times \bar{x})^2}$$

n = requisite number of samples
s^2 = anticipated sample variance
t = two-tailed critical value of Student's t
E = acceptable % error (e.g. 0.2 = 20%)
$\bar{x}$ = anticipated mean biomass

We used the variances and means observed in our samples for individual taxa to estimate the requisite number of rake (n_{rake}) and quadrat ($n_{quadrat}$) samples. We then calculated the ratio of n_{rake}:$n_{quadrat}$ (rake factor) for each taxon (Table 5). Rake factors for individual taxa were quite variable, ranging from 0.4 to 2.6, but had a median value of 1.0. The rake factor for total biomass was 2.8, indicating that roughly 3 times more rake samples would be needed to achieve the same level of precision as quadrat samples for total biomass estimates in our study lakes. Although we did not record the time needed to collect samples with each method, rake samples required substantially less than one-third the time to collect

TABLE 4. COMPARISON OF COEFFICIENT OF VARIATION (CV; STANDARD DEVIATION ÷ MEAN) FOR SITE-AVERAGED BIOMASS ESTIMATES BY RAKE AND QUADRAT SAMPLING METHODS AT 38 SITES. ONLY TAXA THAT OCCURRED AT MORE THAN FIVE SITES FOR EACH METHOD ARE LISTED. N = NUMBER OF SITES WITH OCCURRENCE OF TAXON FOR EACH SAMPLING METHOD, $\bar{x}$ IS THE MEAN BIOMASS, AND *% DIFFERENCE* IS $[(CV_{RAKE} - CV_{QUADRAT}) \div CV_{QUADRAT}]$.

	Rake			Quadrat			
Taxon	N	$\bar{x}$	CV	N	$\bar{x}$	CV	% Difference
All Taxa (total biomass)	38	215	1.33	38	274	0.79	+69
Potamogeton zosteriformis	14	223	1.91	15	49	1.19	+61
Ceratophyllum demersum	31	602	1.44	34	142	1.07	+35
Myriophyllum sibiricum	25	33	1.74	25	33	1.34	+29
Zosterella dubia	24	17	2.50	22	17	2.11	+18
Vallisneria americana	19	30	1.59	18	26	1.44	+11
Elodea canadensis	12	74	2.43	11	30	2.40	+1
Potamogeton crispus (sprouts)	6	3	1.67	7	2	1.66	+1
Najas flexilis	8	5	1.65	9	3	1.69	-2
Stuckenia pectinata	14	99	2.06	15	50	2.20	-6
Potamogeton praelongus	9	70	1.27	7	56	1.50	-15
Chara sp.	14	13	1.64	9	32	2.03	-19
Najas guadalupensis	6	9	1.00	10	2	1.66	-40

TABLE 5. RAKE FACTORS ($N_{RAKE} \div N_{QUADRAT}$) BASED ON THE REQUISITE NUMBER OF RAKE (N_{RAKE}) AND QUADRAT SAMPLES ($N_{QUADRAT}$) NEEDED TO ACHIEVE THE SAME LEVEL OF PRECISION WHEN ESTIMATING MEAN BIOMASS ($\bar{X}$) OF INDIVIDUAL TAXA. REQUISITE N_{RAKE} *AND* $N_{QUADRAT}$ CALCULATED BASED ON OBSERVED MEAN ($\bar{X}$) AND STANDARD DEVIATION (S) OF BIOMASS FOR INDIVIDUAL TAXA, ASSUMING AN ACCEPTABLE ERROR OF 20% OF THE MEAN (ZAR 2010).

	Rake			Quadrat			*Rake Factor*
Taxon	s	$\bar{x}$	n_{rake}	s	$\bar{x}$	n_{quadrat}	$(n_{\text{rake}} + n_{\text{quadrat}})$
All Taxa (total biomass)	1103	215	173	216	274	62	2.8
Potamogeton zosteriformis	426	223	353	58	49	137	2.6
Ceratophyllum demersum	865	602	200	151	142	112	1.8
Myriophyllum sibiricum	58	33	292	45	33	176	1.7
Zosterella dubia	42	17	604	36	17	433	1.4
Vallisneria americana	47	30	247	37	26	202	1.2
Elodea canadensis	181	74	571	71	30	555	1.0
Potamogeton crispus (sprouts)	5	3	271	3	2	267	1.0
Najas flexilis	9	5	264	5	3	275	1.0
Stuckenia pectinata	203	99	411	109	50	466	0.9
Potamogeton praelongus	89	70	157	84	56	217	0.7
Chara sp.	21	13	261	65	32	400	0.7
Najas guadalupensis	9	9	98	4	2	267	0.4

quadrat samples. This suggests that the vertical rake method could achieve a similar level of precision as the quadrat method while maintaining, or possibly decreasing, the total time required for sampling.

In addition to the time required for sample collection, researchers must also consider the time requirements for sample processing (sorting, drying and weighing of taxa). Although larger samples generally take longer to process than smaller samples, in our experience the diversity and form of plants in each sample also dramatically affect processing time. Many of our largest samples (highest biomass) were nearly monotypic (predominantly coontail) and thus were processed quickly. Conversely, diverse samples were more time-consuming to process. We did not record the time required for sample processing of rake and quadrat samples, so we were not able to evaluate differences in the time required however, the larger number of rake samples required to achieve the same level of precision as the quadrat method would clearly result in an overall increase in sample processing time, particularly if samples need to be sorted by taxa before drying. Before adopting the vertical rake method, researchers should consider this potential need for longer processing time.

Downing and Anderson (1985) reported that the collection of more samples using smaller quadrats was preferable to collecting fewer samples using larger quadrats. Although Downing and Anderson (1985) did not consider sample processing time in their evaluation (they focused on optimizing precision and time for sample collection), they reported that samples from small quadrats (10 × 10 cm) generally required very little processing time (<1 min), while samples from large quadrats (1 × 1 m) took several hours to process. Accordingly, in future studies, it may be worthwhile to evaluate the use of a smaller rake (shorter rake head) to reduce the amount of biomass collected and thus reduce the amount of processing time required.

Despite its shortcomings (e.g., lower precision, potential for increased sample processing time), the vertical rake

method provides key advantages over the diver quadrat method: improved safety, increased sampling intensity, and inexpensive equipment that is simple to use without special training. Given these advantages and the potential to achieve a similar level of precision through the collection of an increased number of samples, the vertical rake method should be considered a suitable alternative to the quadrat method, particularly for large-scale studies requiring the collection of many samples over expansive areas.

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