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**CHIRONOMIDAE (DIPTERA: INSECTA)
GENERA ACCUMULATION CURVES FOR STREAMS
OF THE LOWER CHEYENNE RIVER WATERSHED**

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ABSTRACT

The objective of this analysis was to determine the number of Chironomidae (Diptera) larvae needed to achieve 90% of the genera in fixed-count subsamples. Composite macroinvertebrate samples from 12 sites with three sites being sampled twice were collected during the summer of 2007 and processed using U.S. EPA lab protocols (U.S. EPA 2004). Chironomidae were mounted and identified to genus following Merritt et al. (2008). For each sample, number of genera and percent of Chironomidae needed to reach 90% of genera in the subsample were determined with a genera-accumulation curve. The number of Chironomidae individuals (range = 5-269) and genera (range = 2-14) were highly variable among the 15 samples. Percent of subsample needed to reach 90% of the composite generic richness averaged 25.3% (range = 10-70%). The large range of individuals needed to reach 90% of total genera lead to the conclusion that site-specific curves would be necessary to determine subsampling effort.

Keywords

Chironomidae, subsampling, accumulation curve, stream, river

INTRODUCTION

The Chironomidae are the most widely distributed and frequently the most abundant insects in freshwater. Approximately 15,000 species are thought to exist in aquatic and semi-aquatic habitats (Cranston 1995). Chironomidae encompass a wide array of functional feeding and habit guilds (Berg 1995, Pinder 1995). They are one of the most frequently investigated macroinvertebrate groups in freshwater productivity studies and serve to link primary producers to higher consumers (Tokeshi 1995).

Chironomidae collected for water quality assessment efforts are often identified only to family level due to significant laboratory preparation needed to identify specimens (Rabeni and Wang 2001). The large number of larvae often collected results in significant commitment of time and labor before data can be generated. Given these considerations, one must carefully weigh the costs of identifying chironomids to a lower taxonomic level.

Species-area curves have been commonly used in the field to determine the sampling area required to adequately describe plant communities (Cain 1938). The species-area relationship is generally asymptotic, demonstrating reduced returns of new species with additional effort. Thus, effort in identifying individuals beyond the inflection point of the curve would normally involve considerable sampling effort with little return in new taxa. These ideas have been extended to biological monitoring of invertebrates in water resource studies to determine the minimum total counts necessary to estimate richness measures (Vinson and Hawkins 1996). Genera accumulation curves provide a means of reducing laboratory costs by quantifying the level of effort required to reach a target percentage of the total number of genera within a sample.

The objective of this effort was to evaluate subsample sizes of Chironomidae needed from composite stream samples to reach 90% of the Chironomidae genera found within the original sample. We hypothesized a similar asymptotic relationship among multiple stream samples collected from tributaries within the same river basin.

STUDY AREA

Invertebrate samples were collected from 12 sites within the Lower Cheyenne River basin. The Cheyenne River watershed (both the upper and the lower reaches) encompasses 24,240 mi² (Foreman et al. 2007). The Lower Cheyenne River Watershed (LCRW) is defined as the drainage area of the Cheyenne River

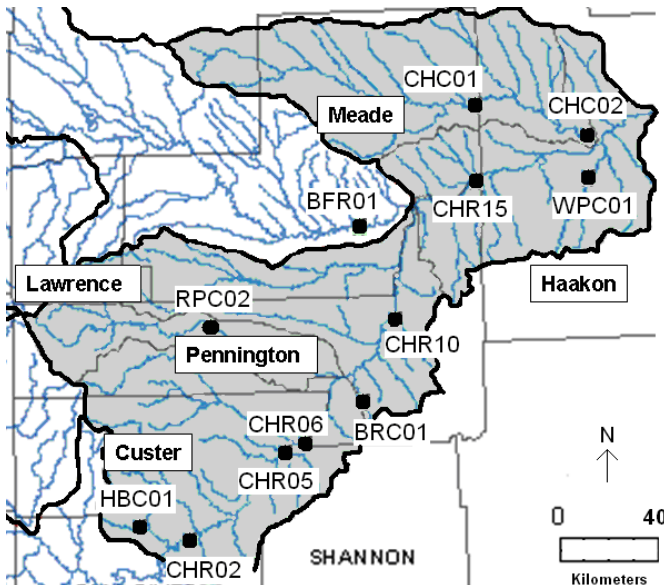


Figure 1. Lower Cheyenne River Watershed and location of sites sampled for Chironomidae larvae.

between the discharge of Angostura Reservoir and the confluence of the Cheyenne River with Lake Oahe (Figure 1).

The landscape within the LRCW is diverse. Portions of the Black Hills National Forest and Badlands National Park are contained within this area. Ranching activities constitute the largest portion of land use. Other management practices include irrigated farming, dry-land farming, urban, and mining activities. Beneficial uses of the waterways within this watershed include domestic water supply, marginal to permanent warm water fish propagation, immersion recreation, limited contact recreation, wildlife and fisheries recreation, livestock watering, and irrigation (Foreman et al. 2007).

METHODS

Twelve sites were sampled during the summer of 2007 with three being sampled twice (Figure 1). Samples were collected from streams from May through August 2007 following U.S. EPA Western Pilot Protocols (Peck et al. 2006). These samples were brought back to the laboratory and were processed following U.S. EPA (2004) protocols. Chironomidae were mounted on slides using CMCP-10 and identified using Merritt et al. (2008).

Ten replicate genera lists were created for each sample. Random numbers were then assigned to each genus, and each list was sorted from lowest to highest. For each composite sample, consecutive groups of 10% of the individuals present were subsampled until all individuals within the composite sample had been sampled. Subsamples that contained abundances that were not divisible by ten were multiplied by ten, and the relative abundances across taxa were adjusted accordingly. The number of genera within each percentage category was counted, and the average and standard deviation for generic richness were determined. Genera-accumulation plots were then used to determine the minimum percentage category needed to reach 90% of the total genera within each composite sample.

RESULTS

Four genera accumulation patterns were observed from the 15 samples processed in this effort. Five samples displayed little or no variation in percent genera with increasing subsample size above 10% (Figure 2). A second group of five samples demonstrated accumulation above 90% of total genera after 20% of the subsample was identified. A third group of three samples required between 30%-40% of midges be identified, and the final group of two samples required 70% or more of midges being subsampled and identified to reach 90% of the genera in their respective composite samples.

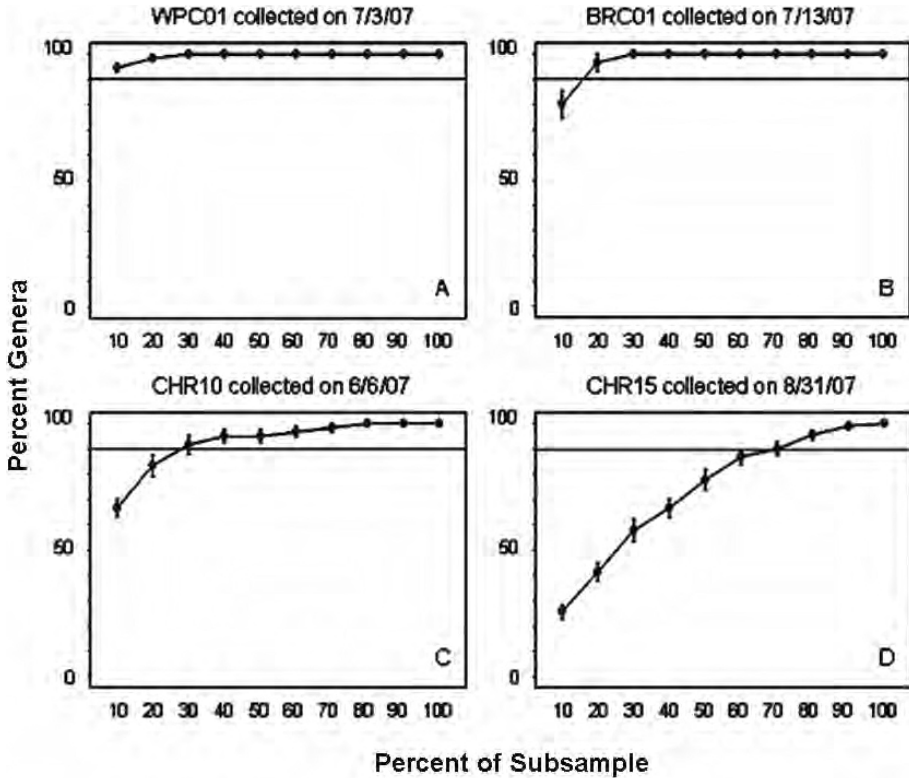


Figure 2. Genera-accumulation patterns observed from Lower Cheyenne River tributary streams (A: fewer than 10% of individuals must be identified to detect 90% of genera; B: 90% of genera detected by identifying 20% of individuals; C: 90% of genera detected by identifying 30%-40% of individuals, D: 90% of genera detected only after >70% of individuals are identified). 90% of taxa on the y-axis marked with a horizontal line.

Chironomidae abundance (range = 7-269, average = 73.5) and total generic composite richness (range = 2-14 genera, average = 8.3 genera) varied widely among the 15 samples (Table 1). In general, those composite samples with fewer total midge genera required lower subsampling effort to reach 90% of the total generic richness. Across all 15 samples, subsampling effort required to reach 90% of total composite richness was highly variable (range = 10-70%, average = 25.3%).

Table 1. Chironomidae generic richness, total abundance, and the percent subsample needed to reach 90% of composite sample generic richness.

Sampling Location and Date	Composite Subsample Richness	Composite Subsample Abundance	Percent of Subsample Needed to Reach 90% Composite
BFR01 3Aug07	12	37	20
BRC01 13Jul07	3	26	20
BRC01 18Jul07	3	19	10
CHC01 20Jun07	4	23	10
CHC02 6Jul07	14	269	10
CHR02 15Jun07	12	40	70
CHR05 10Aug07	5	5	20
CHR06 30Jul07	2	8	10
CHR10 6Jun07	6	55	30
CHR10 15Aug07	7	7	30
CHR15 31Aug07	9	30	70
HBC01 5Jun07	10	143	20
RPC02 23May07	10	36	30
WPC01 3Jul07	13	202	10
WPC01 10Jul07	14	202	20

DISCUSSION

Results of this effort suggest large variation in effort required to adequately describe patterns in Chironomidae richness among tributaries of the same large river system. Samples displayed the characteristic asymptotic relationship between genera accumulation and subsample size (Soberon and Llorente 1993, Willott 2001, Thompson and Thompson 2007). However, we hypothesized that similar genera-accumulation curves existed among the sampled tributaries. Furthermore, we expected that if these genera accumulation curves were similar, a consistently sized subsample might be taken which would adequately describe Chironomidae richness within these streams.

Generally, we found that lower subsampling effort would be required to reach 90% of total sample richness from composites with fewer total midge genera. However, subsampling effort also depended on the relative abundances (evenness) of different midge genera among all genera within the sample. Samples with many midge genera (high total richness) but low evenness (dominance of 1-2 genera) would require greater subsampling effort than those with even apportioning of individuals among genera.

On average, 25% of larvae would need to be identified and enumerated from a 500-count subsample to account for 90% of total generic richness. However, this subsample percentage might range to 70% when total richness is high

and evenness is low. Because richness and evenness components of community structure are linked to seasonal life-history characteristics and the effects of disturbance, genera-accumulation curves are likely to differ spatially and temporally within a watershed. Thus, the use of taxon-accumulation curves might serve only as a guide to better understand the quality of the data generated from analyses that are often limited by available funds and sampling effort.

Processing macroinvertebrate samples is a time-consuming process. Attempts have been made to reduce the effort in processing while not compromising results; some examples include fix-counts and volumetric sub-sampling (Vinson and Hawkins 1996, Barbour and Gerritson 1996). A 200-fixed subcount was deemed sufficient to distinguish sites when using metrics that concerned relative taxa richness; however it was suggested that large or rare organisms be removed followed by a minimum 300-count subsample when using metrics that relied on specific taxa or similarity indices (Vinson and Hawkins 1996). Chironomidae often comprise a large portion of invertebrate taxa richness within a site and thus their use in monitoring cannot be overstated. However, they require more effort to correctly identify due to the requirement that they be mounted on glass slides and viewed under high magnification. Genera accumulation patterns appear to vary widely among sites. However, intra-site similarity seen among duplicate samples collected during this effort was high. Variation in community composition among years should also be considered, particularly since many state agencies conduct watershed assessments on a 5-year rotation.

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