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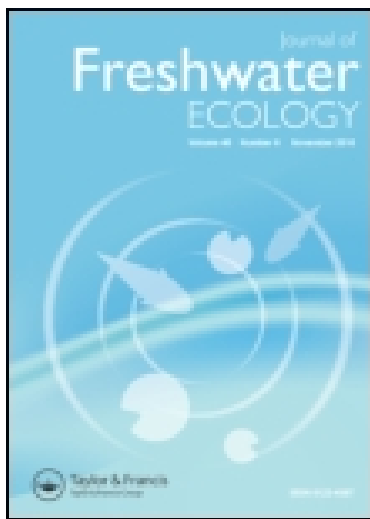
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## Land use and surface water withdrawal effects on fish and macroinvertebrate assemblages in the Susquehanna River basin, USA

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Water withdrawals in the Susquehanna River basin, USA, are increasing due to burgeoning shale gas extraction activities. In order to determine if flow alteration resulting from shale gas industry surface water withdrawals impacts fish and macroinvertebrate assemblages in lotic habitats, data were collected upstream and downstream of 12 withdrawal and three reference sites in headwater, cold water, and large warm water streams. Watershed size ranged from 4 to 517 km<sup>2</sup> and average daily withdrawals ranged from 0.05 to 1.4 million liters. Analysis of withdrawal data indicated that approved withdrawals far exceeded actual withdrawals across all stream types. The largest withdrawals relative to stream size were from headwater streams, where on average 6.8% of average daily flow was withdrawn daily. Fish and macroinvertebrate assemblage similarity at study sites depended largely on stream sampled, rather than position upstream or downstream of withdrawals. Regression techniques were employed to determine if catchment-level variables or withdrawal metrics best described variation in fish and macroinvertebrate metrics shown to be sensitive to flow alteration. The catchment-level variables were responsible for the majority of observed variation in fish metrics. Macroinvertebrate models performed poorly, indicating that the stream sampled or variables not included in the analyses were responsible for the majority of variation. Overall, evidence suggests impacts of shale gas withdrawals within the Susquehanna basin are limited at the present state of flow alteration. Potential reasons include protective measures such as pass-by flow restrictions, which require withdrawals to cease when flows drop below a predetermined low flow threshold, maximum instantaneous and daily withdrawal limits, and recent initiation of withdrawals (1–3 years of operation).

**Keywords:** fish assemblages; macroinvertebrate assemblages; water withdrawals; shale gas extraction; flow alteration; lotic habitats

### Introduction

The extensive influence of the natural flow regime on ecological processes in lotic habitats has been well documented (Poff et al. 1997). Stream flow creates and maintains physical habitat, which in turn influences biological communities in stream ecosystems that have adapted to natural flow regimes (Bunn & Arthington 2002; Power et al. 2008). Unaltered flow regimes are becoming less common as anthropogenic water use continues to increase (Jackson et al. 2001; Baron et al. 2002). Consequently, conflicts between human use and ecosystems arise as flow alteration resulting from impoundments, diversions, and

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water withdrawals can impact biological communities (Freeman & Marcinek 2006; Kanno & Vokoun 2010; Carlisle et al. 2010).

The Susquehanna River basin (herein referred to as Basin) is a 71,251 km<sup>2</sup> (27,510 mi<sup>2</sup>) watershed covering portions of New York, Pennsylvania, and Maryland (Figure 1). The Basin comprises about 43% of the Chesapeake Bay watershed area and the Susquehanna River supplies approximately 50% of freshwater inflow to the Chesapeake Bay (Seitz 1971); thus, the two systems are inextricably linked (Schubel & Pritchard 1986). Many existing public, industrial, commercial, and private/residential users currently withdraw water from the Basin (SRBC 2013). The shale gas extraction industry has recently begun targeting natural gas reserves in the Marcellus and Utica shale formations in the Mid-Atlantic and northeastern USA. The shale gas industry represents an increasingly large source of water withdrawals and consumptive use, thereby increasing the potential for flow alteration of lotic systems in the Basin. Unconventional shale gas extraction methods require large quantities of water to hydraulically fracture horizontal wells in order to stimulate gas production. For an in-depth explanation of unconventional gas extraction, see Ground Water Protection Council (GWPC) (2009). In the Basin, an average of 17 million liters (4.5 million gallons, Mgal), of water are used to hydraulically fracture one unconventional gas well. Although the shale gas industry often reuses water from previous fracture events, freshwater on an average comprises approximately 84% (14.4 million L; 3.8 Mgal) of the water used for each fracture event (SRBC 2012a). Since large-scale drilling operations began in 2008, 3515 wells have been drilled in the Basin through June 2012 (PADEP 2012). Although other industries withdraw greater quantities of water, the shale gas industry's water use is substantial when compared with water users that have historically operated in the Basin (Table 1). Also, the withdrawals included in

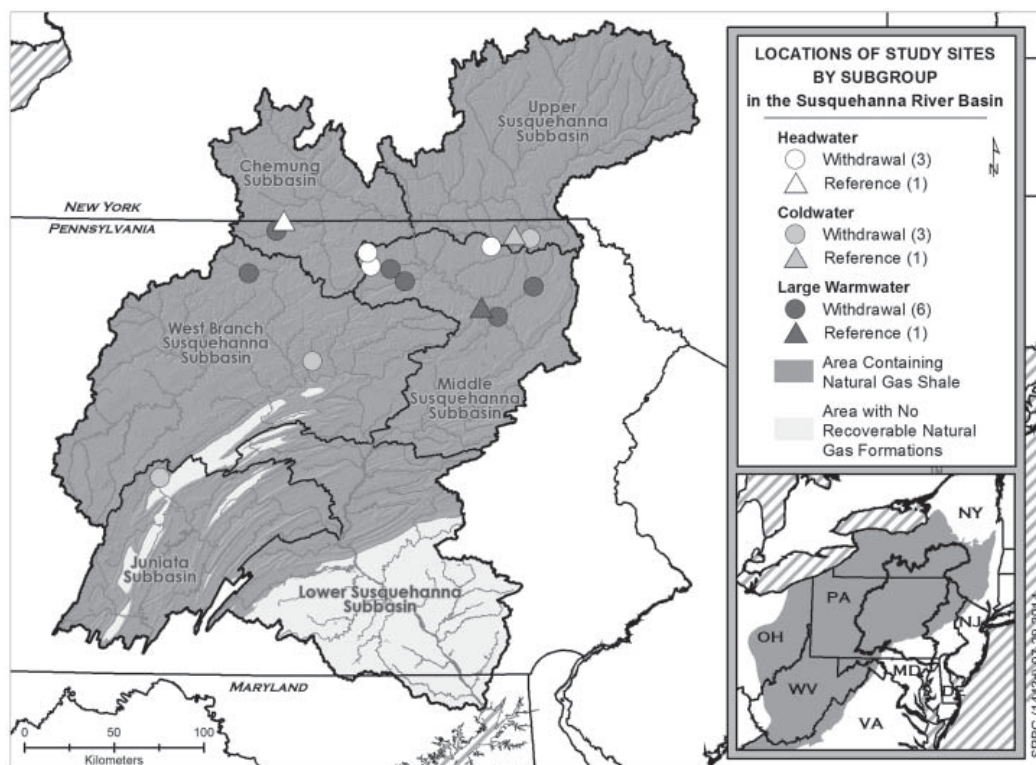


Figure 1. Locations of study sites, by stream type, in the Susquehanna River basin.

Table 1. Reported total groundwater and surface water withdrawal quantities of major water use industries within the Susquehanna River basin during 2011. Values are presented in million liters per day (million gallons per day in parentheses) in descending order of total water withdrawal quantity (SRBC 2013).

Water use industry	Ground water withdrawals	Surface water withdrawals	Total water withdrawals
Electric generation	21.2 (5.6)	10,406.1 (2749.0)	10,427.3 (2754.6)
Water supply	193.1 (51.0)	215.8 (57.0)	408.8 (108.0)
Mining	174.1 (46.0)	5.7 (1.5)	179.8 (47.5)
Manufacturing	64.4 (17.0)	94.6 (25.0)	159.0 (42.0)
Other	37.9 (10.0)	5.3 (1.4)	43.2 (11.4)
Natural gas extraction	3.4 (0.9)	31.4 (8.3)	34.8 (9.2)

this study operated intermittently, which differs from most other established users that withdraw steadily for at least a portion of the year. Furthermore, the shale gas industry's water use often occurs in undeveloped watersheds and is expected to increase over time due to large estimated quantities of recoverable natural gas resources in the shale layers underlying 85% of the Basin (Figure 1) (Coleman et al. 2011).

There are many concerns focused on water quality impacts associated with unconventional shale gas extraction and hydraulic fracturing (Orsborn et al. 2011; Olmstead et al. 2013; Weltman-Fahs & Taylor 2013). This study examined another major concern: the ability of watersheds to accommodate a relatively new industry that requires large volumes of fresh water. Although the Basin is considered relatively water rich with an average annual precipitation of 102 cm (40 inches), surface water yield is highly dependent on spatiotemporal variables including stream size, underlying geology, season, and hydrologic condition (DePhilip & Moberg 2010). The Susquehanna River Basin Commission (SRBC) regulates water withdrawals and consumptive uses within the Basin's boundaries, including regulations that require approval for water withdrawn in any amount for unconventional shale gas development. SRBC regulates shale gas withdrawals with restrictions that are intended to be protective of aquatic ecosystems (SRBC 2002, 2012b). There is a paucity of regionally specific data available to determine the efficacy of these withdrawal regulations. Concerns arise regarding not only the current level of flow alteration, but also the magnitude of potential flow alteration caused by increasing water use in the Basin (DePhilip & Moberg 2010).

Stressor–response relationships (e.g., Davies & Jackson 2006) have proven valuable for the management of aquatic ecosystems. Analyses completed herein follow the same premise of establishing flow alteration–ecological response curves. Although difficult to establish, flow alteration–ecological response curves have encouraged prudent management of water resources (Poff & Zimmerman 2010). In this study, responses of flow preference guilds and other ecological, habitat, trophic, life history, and general assemblage metrics of fish and macroinvertebrate assemblages previously shown to be sensitive to flow alteration were examined. This study was intended to characterize the initial response of fish and macroinvertebrate assemblages to shale gas withdrawals and to assess the similarity of impacts across headwater, cold water, and large warm water stream types. Regression techniques were used to determine whether catchment-level variables (e.g., drainage area (DA), land use) or withdrawal metrics best explained variation in fish and macroinvertebrate assemblage metrics.

## Methods

### *Study area and site selection*

This study was conducted in the Pennsylvania portion of the Susquehanna River basin in the eastern USA (Figure 1). Mixed forested and agricultural land uses are found in this relatively undeveloped, rural watershed (DePhilip & Moberg 2010). A total of 15 sites were included in this study. Twelve withdrawal sites were selected based on the presence of shale gas withdrawals that had been operating for approximately one year or more. Three reference sites were also included. In order to examine impacts of withdrawals in different stream types that commonly occur in the Basin, withdrawal sites were grouped into three stream types. Three sites were located in headwater streams with a small DA (4.4–23.1 km<sup>2</sup>) and history of glaciation. Three sites were located in intermediate-sized (85.3–91.7 km<sup>2</sup>) cold water streams with the potential to support natural trout reproduction. Six sites were located in large (243.5–516.9 km<sup>2</sup>) warm water streams. Three reference sites were selected, one in each stream-type category, and placed where no withdrawals were present upstream (US) in the watershed (Figure 1 and Table 2). Reference sites were selected that were similar to withdrawal sites in DA and land use, and were not intended to represent minimally altered conditions with maximum biological integrity (Stoddard et al. 2006).

### *Field sampling*

All sites included in this study were sampled between 1 May 2012 and 25 June 2012. An upstream–downstream (US–DS) (i.e., control–impact) methodology was utilized to collect data from two stream reaches at all withdrawal and reference sites, for a total of 30 stream reaches. Stream reaches were positioned US and DS of withdrawal intake structures at withdrawal sites, while reaches were positioned around an arbitrary point at reference sites. Reach length was determined by multiplying the average wetted stream width by 10 (minimum length 100 m, maximum 400 m). Effort was made to capture similar physical habitat features in US and DS reaches (Table 2), avoid influence of tributaries, and keep riparian land use consistent.

Fish sampling was conducted by pulsed direct current electrofishing using either a battery backpack unit (Appalachian Aquatics, Inc.) or a tote barge unit (Smith-Root, Inc.) controlled by a 1.5 KVA electrofisher and powered by a 2000 W generator, depending on stream size. Three electrofishing passes were conducted in order to sample all available habitats. If the stream was too large to be covered completely by each pass, then passes were coordinated so that the entire stream width was covered throughout the three passes. All fishes captured were identified in the field when possible and released or preserved in buffered formalin and returned to the laboratory for identification. Stocked salmonids were not included in analysis. Due to difficulty with positive identification and similar ecological traits between the two species of sculpin collected (*Cottus bairdii* and *C. cognatus*), individuals were identified to the genus level.

Benthic macroinvertebrate samples were collected and subsampled according to Pennsylvania Department of Environmental Protection (PADEP) protocol (2009). A D-frame net with 500-micron mesh was used to collect macroinvertebrate samples. Samples consisted of a composite of six kicks from best available riffle and run habitat in the stream reach, with each kick disturbing approximately 1 m<sup>2</sup> of substrate immediately upstream of the net for approximately 1 minute. The composite sample was preserved in 95% ethanol in the field and returned to the laboratory for processing. Organisms were

Table 2. Summary of catchment-level variables of study sites, including withdrawal sites and reference sites where no withdrawals were present (mean  $\pm$  standard deviation). HW = headwater, CW = cold water and WW = warm water.

	Drainage area (km <sup>2</sup> )	% glaciated	% forest	% agricultural	% urban	Mean width US (m)*	Mean width DS (m)*	Discharge (m <sup>3</sup> s <sup>-1</sup> )*
HW group ( <i>n</i> = 4)	13.4 $\pm$ 7.6	100 $\pm$ 0.0	66.3 $\pm$ 12.3	32.9 $\pm$ 12.1	0.4 $\pm$ 0.2	3.5 $\pm$ 1.2	3.7 $\pm$ 1.3	0.1 $\pm$ 0.04
CW group ( <i>n</i> = 4)	94.8 $\pm$ 15.2	51.1 $\pm$ 56.5	78.6 $\pm$ 9.1	18.0 $\pm$ 9.4	1.3 $\pm$ 1.0	12.8 $\pm$ 2.2	12.6 $\pm$ 3.2	1.2 $\pm$ 0.7
Large WW group ( <i>n</i> = 7)	341.9 $\pm$ 104.5	91.9 $\pm$ 21.4	69.4 $\pm$ 18.7	28.5 $\pm$ 18.7	0.8 $\pm$ 0.5	20.0 $\pm$ 8.0	17.6 $\pm$ 5.5	2.3 $\pm$ 1.1

\*Mean width and discharge were instantaneous values measured during field sampling.

Table 3. Withdrawal metrics of withdrawal sites (mean  $\pm$  standard deviation). Withdrawal and flow quantities shown in million liters per day (mLd).

	Maximum approved daily withdrawal (mLd)	Mean daily withdrawal (mLd)	Average daily flow (mLd)	WI*	Period of operation (years)	% operating days	No. with pass-by	Range of pass-by thresholds
HW group ( <i>n</i> = 3)	4.7 $\pm$ 2.5	0.9 $\pm$ 0.5	15.2 $\pm$ 9.2	6.8 $\pm$ 2.7	1.0 $\pm$ 0.4	58.9 $\pm$ 8.1	3	20% ADF
CW group ( <i>n</i> = 3)	0.4 $\pm$ 0.2	0.06 $\pm$ 0.02	148.0 $\pm$ 20.7	0.04 $\pm$ 0.01	1.4 $\pm$ 0.8	32.3 $\pm$ 14.9	1	15% ADF
Large WW group ( <i>n</i> = 6)	3.6 $\pm$ 3.2	0.4 $\pm$ 0.2	444.4 $\pm$ 127.2	0.1 $\pm$ 0.05	2.8 $\pm$ 0.3	56.7 $\pm$ 21.6	4	20%–25% ADF

\*WI (withdrawal index) represents the average percentage of average daily flow withdrawn from the stream over the entire period of the withdrawal, on a daily basis.



randomly picked and identified until a 200-organism ( $\pm 40$ ) subsample was obtained. Organisms in the subsample were identified to genus level, when possible, and enumerated, with few exceptions (PADEP 2009).

### Data analysis

Catchment-level variables examined included land use characteristics (% forest, % urban, and % agriculture), which were calculated using ArcGIS version 10.1 (ESRI, Inc.; <http://www.esri.com>). DA and percent of watershed historically covered by glaciers were calculated using Pennsylvania (PA) StreamStats (USGS 2010). Percent forest in watersheds was highly correlated with % agriculture ( $r = -0.997$ ,  $p < 0.001$ ); therefore, only % forest was used in analyses. Assignment of sites into stream-type categories was verified by evaluating catchment-level variables to ensure similarity among physical characteristics (Table 2).

Reported daily withdrawal data were used to construct water withdrawal metrics. Period of operation, defined as the length of time, since the withdrawal was initiated until the date of sampling, was calculated. Average daily flow (ADF) for each study site was obtained by using the DA ratio method (Equation (1)), using data from the representative US Geological Survey gages (Stuckey & Roland 2011). A withdrawal index (WI) was calculated for each site (Equation (2)), which allowed a comparison of withdrawal quantities relative to stream flow. The WIs represented on a daily basis the average percentage of ADF withdrawn from the stream over the entire period of the withdrawal:

$$\text{ADF(ungaged site)} = \text{DA ratio} \times \text{ADF(reference gage)}, \quad (1)$$

$$\text{WI} = [\text{mean withdrawal quantity(million L/d)}/\text{ADF(million L/d)}] \times 100\%. \quad (2)$$

Previous studies have most often used a low flow statistic to calculate WIs. We instead chose to use ADF values to standardize withdrawal size relative to stream flow, due to pass-by flow restrictions at 8 out of 12 withdrawal sites (66.7%) in this study that prevented withdrawals from operating during periods of low flow. Also, withdrawal sites included in this study had pass-by thresholds set at a percentage of ADF (15%–25%, Table 3).

Non-metric multidimensional scaling (NMDS), using Bray–Curtis dissimilarity (Bray & Curtis 1957) was used to ordinate samples DS and US of water withdrawals and at reference sites. NMDS allowed visualization of the relative similarity among fish and macroinvertebrate assemblages. Stress values obtained ranged from  $<0.01$  to 0.16, indicating good representation of the data with little chance of misinterpretation (Field et al. 1982; Clarke 1993). The metaMDS function in the vegan package of the R software environment was used to complete the NMDS analyses (R Development Core Team 2006; Oksanen et al. 2011).

Subsequently, the information theoretic approach (Burnham & Anderson 2002) was used to determine if null models, catchment-level variables, or withdrawal metrics best explained variation in fish and macroinvertebrate assemblage metrics of interest. Twelve fish assemblage metrics were modeled representing six ecological traits: flow preference (% fluvial specialists, % fluvial dependents, and % macrohabitat generalists), trophic guild (% benthic invertivores), origin (% native and % non-native), indicator taxa (% *Cottus*, % *Catostomus commersoni*, % *Rhinichthys cataractae*, and % Centrarchidae), habitat preference (% benthic fishes), and Shannon diversity index. Assignment of flow

preference guild traits was generally consistent with Kanno and Vokoun (2010). Fluvial specialist species require lotic habitats for a majority of their life history, while fluvial dependents require lotic habitats for only a small portion of their life history. Macrohabitat generalists are commonly present in lentic and lotic systems. Ecological traits were assigned to fishes based upon information in references from the Mid-Atlantic region (Cooper 1983; Smith 1985; Jenkins & Burkhead 1993; Stauffer et al. 1995; Snyder 2005; Supplemental data Appendix 1). Indicator fish taxa were chosen based upon previous research that grouped fishes that share similar life history strategies, habitat preferences, or other characteristics that make them sensitive to flow alteration (Frimpong & Angermeier 2009; DePhilip & Moberg 2010; Mims & Olden 2012).

Ten macroinvertebrate assemblage metrics were selected that represented four functional traits identified by Poff et al. (2006): life history (% multivoltine), mobility (% with high crawling rate), morphology (% small body size and % free ranging), and ecology (% depositional, % rheophilic, % scrapers and shredders, % collector–filterers, % predators, and % burrowers). In addition, 10 general assemblage metrics were modeled: taxa richness, % Chironomidae, % tolerant individuals, Hilsenhoff biotic index (HBI), % Ephemeroptera, % Plecoptera, % Trichoptera (EPT), PA benthic index of biotic integrity (PA IBI) scores, and Shannon diversity. Functional traits identified by Poff et al. (2006) were available for 73 out of 109 taxa (67%) collected during this research. The remaining aquatic insect taxa were assigned traits from Merritt et al. (2008), when available. The majority of these metrics were among those (DePhilip and Moberg 2010) expected to be most sensitive to flow alteration within the Basin. Aforementioned fish and macroinvertebrate metrics preceded by ‘%’ indicate relative abundance (e.g., [Ephemeroptera individuals/total individuals in the sample]  $\times$  100).

Eight linear mixed effect models including different combinations of random and fixed effects were constructed to examine variation in fish and macroinvertebrate metrics. These candidate models reflected a-priori hypotheses intended to characterize whether catchment-level variables or withdrawal metrics best explained the variation. A random stream intercept was included in all models in order to avoid pseudoreplication, due to multiple observations from the same stream (Hurlbert 1984). A null model containing only a random stream intercept and fixed intercept was intended to characterize the variation explained by stream membership and/or variables not included in this study. All additional models contained a DA fixed effect, as study sites spanned a wide range of DA (4–517 km<sup>2</sup>). Stream size is one of the most important factors influencing fish and macroinvertebrate assemblage composition globally (Vannote et al. 1980; Oberdorff et al. 1995), which means any credible model could not ignore the explanatory potential of DA (Freeman & Marcinek 2006). Three models contained combinations of catchment-level variables, two models contained withdrawal metrics, and one model contained a combination of catchment-level and withdrawal variables (Table 4). One additional categorical model was included to quantify variation described by position US or DS of an intake.

Akaike’s information criterion corrected for small sample size ( $AIC_c$ ) was used to determine the level of support for the eight competing models. The model with the lowest  $AIC_c$  value was determined to have the most support, while competing models had a  $\Delta_i$  ( $AIC_i - AIC_{\min}$ )  $<$  2. All models were compared using maximum likelihood estimation due to differing combinations of fixed effects contained in each model. The response variable was natural log (ln) transformed whenever necessary in order to ensure homoscedasticity was attained, residuals were approximately normally distributed, and individual observations did not have undue influence on the relationship. In order to aid interpretation and improve numerical stability of the model, the catchment-level variables were

Table 4. Different combinations of fixed effects included in models tested reflecting eight a-priori hypotheses. Random and fixed effects included in each model are denoted by an 'x'.

Model	Random effects		Fixed effects								Categorical position
	Stream intercept	Intercept	DA	% forest	% glaciated	% urban	WI	Operation			
Null	x	x									
DA	x	x	x								
Reduced catchment-level	x	x	x	x							
Catchment-level	x	x	x	x	x						
Reduced withdrawal	x	x	x				x				
Withdrawal	x	x	x				x		x		
Full	x	x	x	x	x		x		x		
Categorical	x	x	x							x	

Note: DA = drainage area; WI = withdrawal index.

centered as follows:  $(\beta_{ji} - \beta_{j\text{mean}})$ , where  $\beta_{ji}$  is observation  $i$  for catchment-level variable  $j$  and  $\beta_{j\text{mean}}$  is the mean of catchment-level variable  $j$ . Competing models were then presented and validated using reduced maximum likelihood estimation. All analyses were performed using the lme4 package in R (R Development Core Team 2006; Bates et al. 2012).

## Results

Land use was similar across headwater, cold water, and large warm water groups. The majority of land use in each group was forested. Urban land use was minimal across all stream types. Mean width and discharge increased with increasing DA of stream types (Table 2).

Mean daily withdrawals were far less than maximum approved daily withdrawals for all groups. On average, withdrawals had been operating longest in large warm water streams, while withdrawals in headwater streams were most recently initiated. Withdrawals in headwater streams and large warm water streams generally were active for a larger percentage of time compared to withdrawals in cold water streams. WI values indicate that on average a larger percentage of ADF was withdrawn from headwater streams compared with cold water and large warm water streams (Table 3). Withdrawal metrics from all stream types are shown in Figure 2 for calendar year 2011, the last full year preceding data collection, as withdrawal activity during that year was representative of the full period of operation of withdrawals. Withdrawal activity was variable between and within stream types and seasons. Notably, withdrawals were reduced in the summer months in terms of mean percent of days with withdrawal activity (Figure 2(a)) and mean daily withdrawal quantity (Figure 2(b)). Withdrawal activity and total withdrawals operated intermittently, especially in the cold water stream type, where all withdrawals were inactive for the first half of 2011. Withdrawals from two cold water sites were again inactive for a period of months prior to the sampling period.

Thirty-seven fish taxa were collected during electrofishing (Supplemental data Appendix 1). *Rhinichthys cataractae*, *Semotilus atromaculatus*, *R. atratulus*, and *Catostomus commersoni* were the most widespread species, respectively, occurring at 26–30 of the 30 stream reaches. Fish species richness ranged from 4 to 26 across study sites, with median richness lowest in the headwater streams and highest in large warm water streams (Figure 3(a)).

One hundred and nine macroinvertebrate taxa were collected during field sampling in 2012, including 83 genus level and 26 higher level identifications (Supplemental data Appendix 2). Chironomidae, Oligochaeta, and *Acentrella* mayflies were the most common taxa collected, occurring at 30, 27, and 25 stream reaches, respectively. Macroinvertebrate taxa richness ranged from 6 to 36, with median richness lowest in large warm water streams and highest in cold water streams (Figure 3(b)). Median fish and macroinvertebrate richness were similar US and DS of withdrawal intakes within each stream type (Figure 3).

NMDS ordination of fish and macroinvertebrate abundance data from all study sites indicated that unique assemblages were generally present in each stream type, as study sites within the same stream type appear grouped (Figure 4). For both fish and macroinvertebrates, sites on the same stream were very similar in terms of assemblage composition. The similarity of assemblages on the same stream appeared greater than the similarity of assemblages at sites located US or DS of withdrawal intakes within the same stream type. Furthermore, similarity of withdrawal sites on the same stream approximated

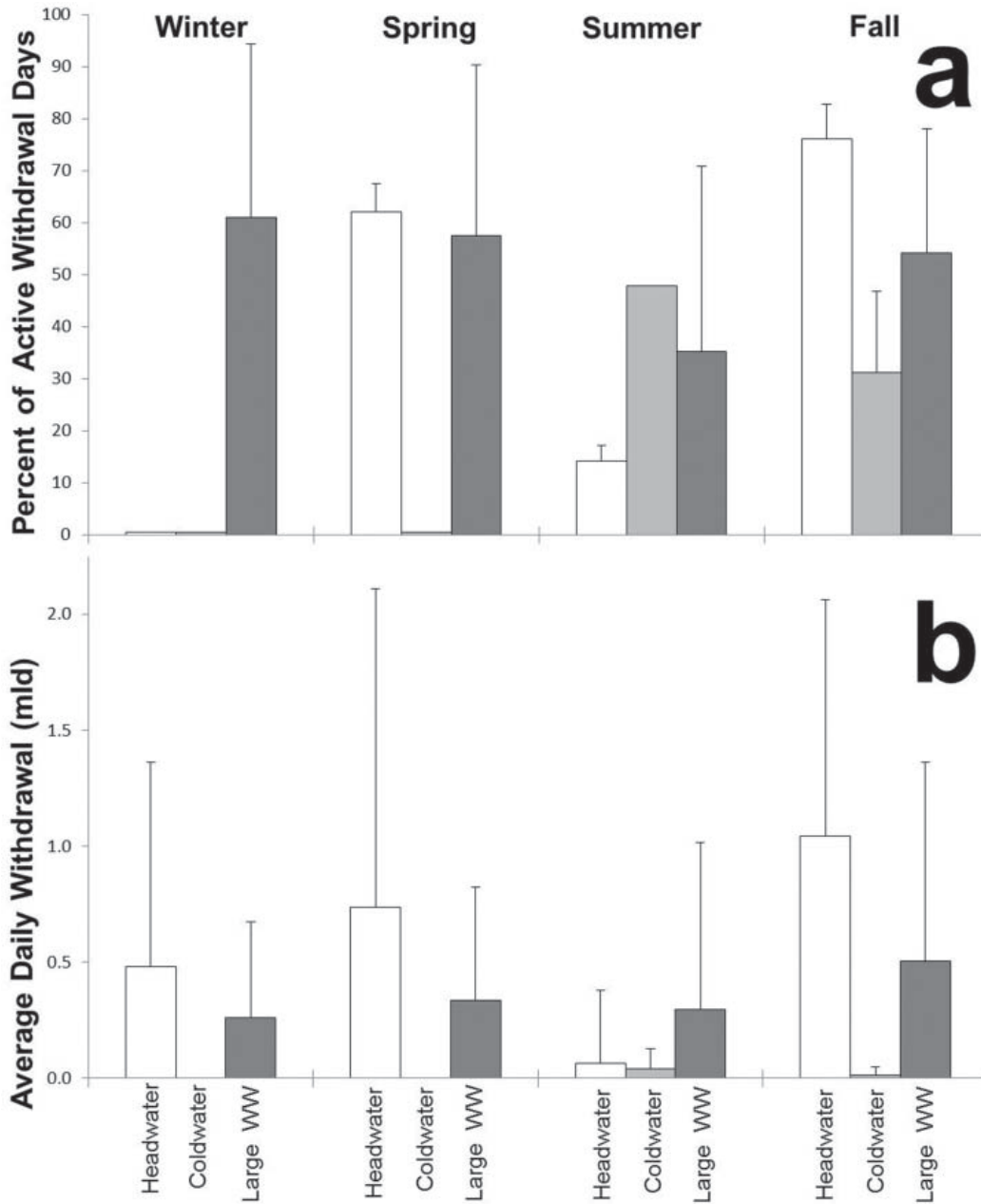


Figure 2. Percent of active withdrawal days (a) and average daily withdrawals in million liters per day (b) at study sites, by stream type and season during 2011.

the similarity between the reference sites, which were located on streams with no water withdrawals present (Figure 4).

Models containing only a random stream intercept and a fixed intercept and some combination of DA and % forested fixed effects received the most support (highest  $w_i$ ) for explaining variation in fish metrics. The DA model received the most support for 6 out of 12 metrics tested (Table 5). These metrics included  $\ln(\% \text{ macrohabitat generalists})$ ,  $\ln(\% \text{ benthic invertivores})$ ,  $\ln(\% \text{ non-native})$ ,  $\ln(\% \text{ } R. \text{ cataractae})$ ,  $\ln(\% \text{ Centrarchidae})$ , and Shannon diversity. The reduced catchment-level model received the most support for three metrics, including  $\ln(\% \text{ fluvial dependents})$ ,  $\ln(\% \text{ native})$ , and  $\ln(\% \text{ } Cottus)$ . The

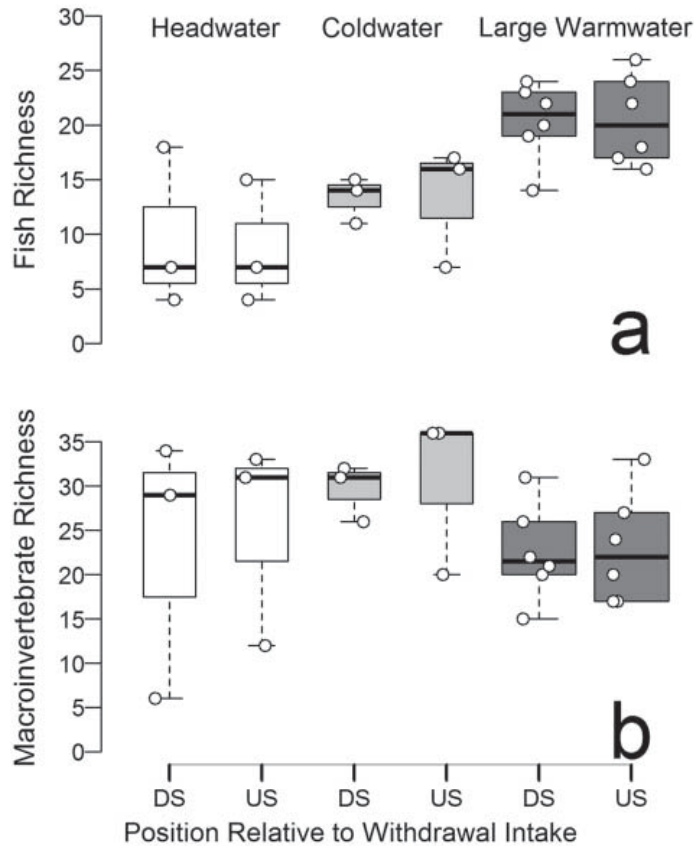


Figure 3. Fish (a) and macroinvertebrate (b) taxa richness US and DS of water withdrawals at study sites, by stream type. Bold lines indicate median values.

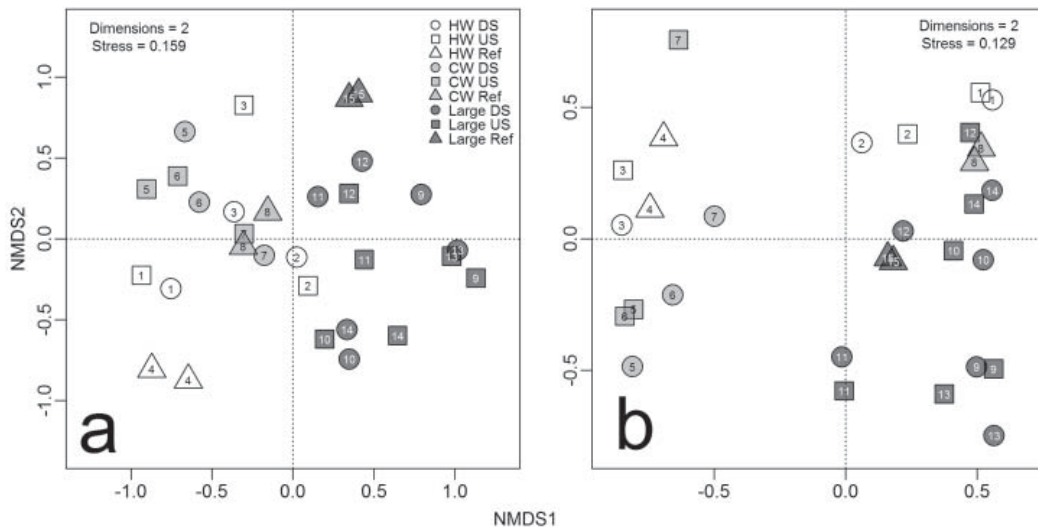


Figure 4. Non-metric multidimensional scaling ordination of fish (a) and macroinvertebrate (b) assemblage abundance data from all study sites. Symbology indicates position US or DS of withdrawal intake or reference site (Ref) and stream type. Points with identical numbers are located on same stream.

Table 5. Parameter estimates  $\pm$  standard error for fish models with most support, defined as model with largest  $AIC_c$  weight of evidence ( $w_i$ ). Blanks indicate parameter was not included in model with most support. Parameter estimate for intercept only indicates null model received most support.

Trait	Response variable	Intercept	Catchment-level variables				Withdrawal metrics			
			DA	% forest	% glacial	% urban	Period	WI	Categorical	
Flow preference	In% fluvial specialist	4.45 $\pm$ 0.02								
	In% fluvial dependent	1.63 $\pm$ 0.20	0.003 $\pm$ 0.001	-0.02 $\pm$ 0.01						
	In% macrohabitat generalist	1.50 $\pm$ 0.17	-0.002 $\pm$ 0.001							
Trophic Origin	In% benthic invertivore	3.63 $\pm$ 0.18	0.003 $\pm$ 0.001							
	In% native	4.51 $\pm$ 0.03	-0.0003 $\pm$ 0.0001	0.05 $\pm$ 0.002						
	In% non-native	1.54 $\pm$ 0.28	0.004 $\pm$ 0.002							
Indicator taxa	In% <i>Cottus</i>	2.19 $\pm$ 0.29	-0.005 $\pm$ 0.002	0.04 $\pm$ 0.02						
	In% <i>C. Commersoni</i>	0.39 $\pm$ 0.23								
	In% <i>R. cataractae</i>	2.14 $\pm$ 0.20	0.002 $\pm$ 0.001							
	In% Centrarchidae	0.68 $\pm$ 0.14	0.002 $\pm$ 0.001							
Habitat	In% benthic	3.14 $\pm$ 0.22								
Diversity	Shannon diversity	1.81 $\pm$ 0.11	0.002 $\pm$ 0.001							

null model received the most support for three additional metrics, including ln(% fluvial specialists), ln(% *C. commersoni*), and ln(% benthic individuals). No models containing withdrawal metrics received the most support. Similarly, null models or models containing only catchment-level variables as fixed effects were most often considered competing ( $\Delta_i < 2$ ). Only one model that contained withdrawal metrics was considered competing, which was the categorical position withdrawal model describing variation in ln(% macrohabitat generalists). The supported models suggest that DA is positively correlated with ln(% fluvial dependents), ln(% benthic invertivores), ln(% non-native), ln(% *R. cataractae*), ln(% Centrarchidae), and Shannon diversity. Conversely, DA was negatively correlated with ln(% macrohabitat generalists), ln(% native), and ln(% *Cottus*). Forested land use was positively correlated with ln(% native) and ln(% *Cottus*) and negatively correlated with ln(% fluvial dependents, Table 5).

Null models containing only a random intercept and fixed intercept terms and no catchment-level or withdrawal-fixed effects received the most support for explaining variation in 16 out of 20 macroinvertebrate metrics. The reduced catchment-level model received the most support for two metrics, while the DA and withdrawal models each received the most support for one metric (Table 6). Models including catchment-level variables were more often considered competing compared to models including withdrawal metrics. Models including withdrawal metrics were, however, more often considered competing for macroinvertebrate metrics than fish metrics. The null models that received the most support were ln(% multivoltine), ln(% highly mobile), ln(% small body size), ln(% free ranging), ln(% rheophilic), ln(% scrapers and shredders), ln(% predators), ln(% burrowers), ln(taxa richness), ln(% Chironomidae), ln(% tolerant individuals), HBI, ln(% Ephemeroptera), ln(%EPT), ln(PA IBI) score, and Shannon diversity. The models receiving the most support that include a DA fixed effect indicate that DA is positively correlated with ln(% depositional individuals), ln(% collector–filterers), and ln(% Trichoptera) and negatively correlated with ln(% Plecoptera). Forested land use was negatively correlated with ln(% depositional individuals) and ln(% collector–filterers). The ln(% Trichoptera) increased with lengthening period of withdrawal operation and decreased with increasing WI (Table 6).

## Discussion

Water withdrawals at sites included in this study were conditioned with restrictions intended to be protective of the natural flow regime. Instantaneous and maximum daily withdrawals were limited to prevent withdrawals from diminishing seasonal or high flow events, which have significant ecological value (Poff et al. 1997). Pass-by flows were required to prohibit withdrawals from operating when stream discharge drops below a predetermined low flow threshold, which was essentially a safeguard intended to ensure flow alteration did not continue during critical low flow periods. Pass-by thresholds were required at 8 out of 12 withdrawal sites and ranged from 15% to 25% of ADF (SRBC 2002). The remaining four withdrawal sites that were not conditioned with pass-by flows had maximum daily withdrawal limits that were less than 10% of  $Q_{7,10}$  flow values. A  $Q_{7,10}$  flow is defined as the lowest average, consecutive seven-day flow that would occur with a frequency of 1 in 10 years (Brandes et al. 2005). Analysis of water withdrawal data suggests that shale gas withdrawals included in this study were operationally driven within the confines of their permits. As evidenced by withdrawal operations at study sites in 2011, average water use was least during summer low flow months, when pass-by thresholds were most often met (Figure 2). During higher flow seasons, however, water



Table 6. Parameter estimates  $\pm$  standard error for macroinvertebrate models with most support, defined as model with largest AICc weight of evidence ( $w_i$ ). Blanks indicate parameter was not included in model with most support. Parameter estimate for intercept only indicates null model received most support.

Trait	Response variable	Intercept	Catchment-level variables				Withdrawal metrics			
			DA	% forest	% glacial	% urban	Period	WI	Categorical	
Life History	In% multivoltine	3.82 $\pm$ 0.10								
Mobility	In% highly mobile	0.88 $\pm$ 0.20								
Morphology	In% small body size	3.96 $\pm$ 0.08								
	In% free ranging	4.23 $\pm$ 0.06								
Ecology	In% depositional	0.92 $\pm$ 0.15	0.003 $\pm$ 0.001	-0.01 $\pm$ 0.1						
	In% rheophilic	2.50 $\pm$ 0.25								
	In% scrapers and shredders	2.10 $\pm$ 0.23								
	In% collector-filterers	2.01 $\pm$ 0.24	0.003 $\pm$ 0.001	-0.03 $\pm$ 0.02						
General	In% predators	0.46 $\pm$ 0.09								
	In% burrowers	3.40 $\pm$ 0.16								
	In taxa richness	3.12 $\pm$ 0.10								
Assemblage	In% Chironomidae	3.35 $\pm$ 0.17								
	In% tolerant individuals	1.72 $\pm$ 0.13								
Metrics	HBI	4.75 $\pm$ 0.26								
	In% Ephemeroptera	3.18 $\pm$ 0.25								
	In% Plecoptera	1.25 $\pm$ 0.18	-0.003 $\pm$ 0.001							
	In% Trichoptera	1.87 $\pm$ 0.22	0.003 $\pm$ 0.001							
	In% EPT	3.63 $\pm$ 0.23								
	In PA IBI score	3.94 $\pm$ 0.11								
	Shannon diversity	0.65 $\pm$ 0.09							0.18 $\pm$ 0.06	-0.07 $\pm$ 0.04

use was greater but more variable and was likely driven by shale gas industry operations. Since the dependent variable was average withdrawals over the entire operating period, the variability of withdrawals was not considered statistically in this study. Further research should be directed at discerning potential impacts of more intermittent withdrawals versus withdrawals that operate steadily.

Fish and macroinvertebrate assemblage similarity at study sites depended largely on stream sampled, rather than position US or DS of withdrawals. This serves as evidence that withdrawals included in this study are not causing structural shifts to assemblages at this early stage of withdrawal operation. Regression techniques used to establish flow alteration—ecological response curves were included in analyses to examine specific metrics shown to be sensitive to flow alteration. Difficulties were encountered while attempting to establish these curves, including variation in observed fish and macroinvertebrate metrics, limited gradient of withdrawal size, small sample size, and other interacting variables (i.e., land use) that exert substantial control over fish and macroinvertebrate metrics. Important information was gained, however, as modeling results suggest catchment-level variables are an important control on variation.

Fluvial specialist fishes are of considerable interest in flow alteration studies. Previous studies have documented decreases in fluvial specialists as a result of flow alteration (Armstrong et al. 2001; Freeman & Marcinek 2006; Kanno & Vokoun 2010). Conversely, this study found that withdrawal metrics were not responsible for considerable variation in ln% fluvial specialists. The only model containing withdrawal metrics to be considered competing among fish metrics was the categorical position model describing ln(% macrohabitat generalists). The parameter estimates indicate that higher proportions of macrohabitat generalists were present US of water withdrawals compared with sites DS (Table 5). The finding confounds a-priori hypotheses, which predict the proportion of fishes that can tolerate lentic habitats to be higher DS of withdrawals, where the potential for flow alteration exists. This suggests withdrawals at present may not be impacting macrohabitat generalist fishes. Instead, macrohabitat generalist abundance at sites in this study may be controlled by localized habitat suitability, which is not determined by the presence or absence of a withdrawal (Montgomery 1999; Walters et al. 2003). The only metric to receive the most support from a model containing withdrawal metrics was ln(% Trichoptera), which suggested that Trichoptera could be sensitive to larger withdrawals and riparian disturbance that often coincided the initial stage of withdrawal construction at study sites. At sites where withdrawals had been operating longer ln(% Trichoptera) increased, indicating possible recovery as disturbed riparian habitats were recolonized by vegetation. The ln(% Trichoptera) decreased with increasing WI values, indicating possible impacts of withdrawals on Trichoptera taxa. Dewson et al. (2007) observed that certain Trichoptera, such as Hydropsychidae taxa, which are ubiquitous in the Basin (*Hydropsyche*, *Cheumatopsyche*, and *Diplectrona* were collected at 24, 21, and 5 study sites, respectively), could be vulnerable to decreased water velocities due to filter-feeding techniques that depend on flow to deliver food items. Due to pass-by flow restrictions that prohibit withdrawals during critical low flow periods and smaller WIs in this study compared with the previous studies that observed impacts, it is unlikely that withdrawals in this study impacted flow velocities.

One potential reason for the lack of flow alteration impacts detected in this study is that the magnitude of flow alteration was less in the Basin streams compared with previous studies that clearly detected impacts. Also, the previous studies often examined impacts of water supply or irrigation withdrawals that operated steadily for at least a portion of the year. Conversely, shale gas industry withdrawals included in this study operate

intermittently during non-summer, higher flow periods, potentially lessening impacts to biota. Two previous studies that documented detrimental impacts to fluvial specialist fishes in Georgia Piedmont and southern New England streams observed maximum WI values of 13.3 and 120, respectively (Freeman & Marcinek 2006; Kanno & Vokoun 2010). These studies used  $Q_{7,10}$  values to standardize WI values. When WI values from this study are calculated using identical methods (using  $Q_{7,10}$  values instead of ADF), the median value is 0.09 with a maximum of 14.3. These values are still inflated, as large withdrawals are not permitted to operate below a pass-by threshold, which far exceeds  $Q_{7,10}$  values, at 8 out of 12 withdrawal sites in the Basin. Additionally, the impacts of water withdrawals to biota are thought to be greatest during low flow periods (DePhilip & Moberg 2010) due in part to reductions of riffle habitat in far greater proportion than pool habitat (Armstrong et al. 2001; Hakala & Hartman 2004). Previous studies that did not examine withdrawals with pass-by flow restrictions potentially observed a greater degree of flow alteration and critical habitat loss especially during stressful conditions. Furthermore, the documented resilience of macroinvertebrates to low flow conditions may allow biotic integrity to endure the relatively low levels of flow alteration observed in this study. For instance, Wills et al. (2006) observed reductions in % filter feeding, erosional, and EPT macroinvertebrate taxa only when 90% of summer flow was diverted from a Michigan trout stream.

Null models overwhelmingly received the most support for macroinvertebrate metrics of interest, indicating stream membership or variables not included in this study, were responsible for a large portion of variation. Conversely, the models containing catchment-level variables most often received support for fish metrics. It is not surprising that fish and macroinvertebrate assemblage metrics responded differently to uniform models, as the previous studies have shown fish and macroinvertebrates respond differently when faced with similar factors. Macroinvertebrate assemblages are more sensitive to localized habitat and land use (Richards et al. 1997; Lammert & Allan 1999; Sponseller et al. 2001), while fishes are influenced more by conditions on broad spatial (catchment) scales (Flinders et al. 2008). Catchment-level variables in this study (e.g., % forest) represent watershed-level conditions and do not reflect more localized conditions to which macroinvertebrates are more responsive.

Aside from flow alteration, other factors associated with withdrawals could potentially impact fish and macroinvertebrate assemblages. For instance, the riparian disturbance required to accommodate withdrawal infrastructure has the potential to cause sedimentation and removal of canopy, which could affect fish and macroinvertebrate assemblage composition. Previous studies have identified the potential for sedimentation to coincide widespread natural gas development activities (Entrekin et al. 2011; Drohan et al. 2012; Weltman-Fahs & Taylor 2013). Berkman and Rabeni (1987) found certain fish taxa in trophic and reproductive guilds decreased with increasing fine sediment cover and Larsen et al. (2009) observed decreases in EPT richness as a result of increasing sedimentation. In this study, stream reaches were positioned such that US reaches were located up-gradient from withdrawal intakes and infrastructure, which were accompanied by varying degrees of riparian disturbance. Downstream reaches were located down-gradient from disturbances and thus would receive sediment inputs from disturbed areas. In order to determine whether position US or DS of withdrawal intakes and associated infrastructure explained variation in metrics of interest, the categorical position model was included in the model selection process. The lack of support for this model suggests that the best management practices (e.g., erosion control) may be effective at preventing impacts other than those caused by flow alteration. However, the widespread land

clearing and riparian disturbance required to accommodate industry infrastructure presents a unique monitoring challenge as impacts may be cumulative and difficult to detect within relatively small stream reaches. Consequently, the design employed for this study may not effectively measure sedimentation impacts, which may best explain the lack of support for the categorical model.

## Conclusions

The modeling results of this study do not unequivocally indicate that withdrawals are not impacting fish and macroinvertebrate assemblages at sites included in this study. Rather, evidence suggests that withdrawals are generally not impacting fish and macroinvertebrate assemblages to a greater magnitude than river continuum and landscape controls (e.g., DA, land use). To further examine whether withdrawals are impacting communities and to better quantify impacts, larger data-sets should be compiled for withdrawal sites that have been operating for a longer duration of time in each of the major groups of streams in this analysis. In addition, a broader gradient of withdrawal size should be examined within each stream type. Future research should include more novel biotic indicators such as total biomass and change-point metrics that may be more sensitive to slight changes (Walters & Post 2008; Baker & King 2010). SRBC has since updated its pass-by guidance to a seasonally based pass-by threshold paradigm (SRBC 2012b), which replaces the percent ADF pass-by threshold that was required year round for withdrawal sites in this study. The efficacy of this new policy should be also investigated in future studies.

It is unlikely that withdrawals in cold water and large warm water streams included in this study, which averaged 0.04% and 0.10% of ADF daily, have the potential to impact biological communities due to the small amounts of water withdrawn relative to stream flow. Conversely, the largest withdrawals relative to stream size observed in this study were from headwater streams, which averaged 6.8% of ADF daily. These large withdrawals in small watersheds have a greater potential for impacts. Further research should therefore be focused on withdrawals in small watersheds and should not be limited to shale gas industry withdrawals. Local site-level variables such as riparian land use, substrate characterization, and channel morphology characteristics should be included in future research efforts to improve modeling performance. Measures of flow alteration on different time scales, (e.g., seasonal) also warrant consideration for inclusion in future studies.

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**Supplemental data**

Supplemental data for this article can be accessed here.

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