



## Research Article

# Sustainability of Biodiversity: Use of a NetWeaver Tool to Identify Suitable Sites for Reintroduction of Fishes

Clare W. Hanson II<sup>1</sup>, Jay R. Stauffer<sup>2\*</sup>

<sup>1</sup>The Pennsylvania State University, USA.

<sup>2</sup>The Pennsylvania State University, USA and South African Institute of Aquatic Biodiversity, SA.

\*Corresponding author: Jay R. Stauffer, The Pennsylvania State University, USA and South African Institute of Aquatic Biodiversity, SA.

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### Abstract

Attempts have been made to reintroduce extirpated fauna to their native ranges to increase biotic diversity, biotic resistance, and sustainability of aquatic communities. Herein, we use a Net Weaver Model to test the suitability of putative reintroduction sites of *Notropis bifrenatus* (Cope) (Bridle Shiner) and *Notropis chalybaeus* (Cope) (Ironcolor Shiner), which have been extirpated throughout most of their historic range. These two species co-exist in Marshalls Creek (Delaware River drainage), Monroe County, Pennsylvania. Although both species were historically widespread, the only known extant populations where these endangered shiners occur syntopically is in Marshalls Creek. The research goal was to determine the unique aquatic habitat characters of Marshalls Creek that support the syntopic populations of Bridle and Ironcolor shiners. To achieve this goal, research objectives were (1) to examine and discern the aquatic habitat characters (i.e., the combination and interaction of biological, chemical, and physical habitat characters) that are coincident with these shiners in Marshalls Creek; (2) to determine if these aquatic habitat characters are commonly found at other historical sites for these shiners; and (3) to speculate as to why these shiners are syntopic in Marshalls Creek. A NetWeaver model was developed to examine aquatic habitat characters of Marshalls Creek research sites and other historical sites on the Atlantic seaboard. NetWeaver model network components were utilized to produce strength of evidence scores (i.e., trueness levels) to compare, contrast, and evaluate aquatic habitat characters to the reference (i.e., benchmark) aquatic habitat characters in Marshalls Creek. This study provided important data relative to these shiners and methodology for scientists and resource managers to assess biodiversity and evaluate potential sites for reintroductions of fishes.

**Keywords:** Restoration of native fauna; biotic resistance

### Introduction

Two of the greatest threats to the sustainability of the ichthyofauna of aquatic systems are the spread of non-native species [1] and the extirpation of native ones [2]. To sustain health and persistence of fishes in aquatic systems it is necessary to prevent the introduction of invasive species and to restore native fishes that have been extirpated. The tools used to assess the quality and sustainability of ecosystems have evolved over time. The use of species-area curves used by Gleason [3] morphed into the use of diversity indices [4-7], autotrophic-heterotrophic ratios [8], saprobian designations [9, 10], and biotic indices [11].

The concentration of calcium content [12], distribution of fauna [13, 14], water zones [15], gradient [16], and stream order [17] have been used to classify streams. Cairns and Dickson [18] used inertia and elasticity to predict the sustainability of a system when subjected to a stress and its ability to recover once a structural or functional change in the biota occurred [19]. The biotic resistance or ability to impede invasive species of aquatic systems is dependent in part on a highly diverse native fauna [20-23]. Recently, to increase and maintain biodiversity and sustainability in aquatic systems, attempts have been made to reintroduce extirpated fauna to their native ranges and to enhance corridors among populations to increase stability of these native populations [2]. Certainly, the ability to identify suitable habitats within the native ranges of aquatic species in which such reintroductions will

be successful, is difficult. The purpose of this paper is to propose the use of NetWeaver™ to evaluate potential reintroduction sites within the Delaware River Basin, Pennsylvania.

NetWeaver is a knowledge-based development system used to interpret and evaluate data. NetWeaver is a graphical tool used by engineers that design knowledge-based natural resource management software. Because of NetWeaver's graphical interface, overall ease of operation, and real-time interface, it was chosen as a fundamental technology and component of the Ecosystem Management Decision Support (EMDS) system [23-25]. The EMDS system is a Decision Support System (DSS) that integrates multi-taxa inventory data sets for analysis to help researchers and resource managers make sound management decisions more efficiently [23]. The EMDS system with its NetWeaver modeling tool component has also been successfully used in a variety of applications including natural resource condition assessment [26], wetlands management [27], and forest ecosystem sustainability [28]. NetWeaver modeling has also been used to study and classify lake water chemistry [29], to assess natural resources and watershed conditions at the Delaware Water Gap National Recreation Area and Upper Delaware Scenic and Recreational River [30], and to analyze U.S. Forest Service projects in various locations [23].

Over the years, NetWeaver has evolved, and its versatility has increased. The NetWeaver2 Knowledge-Base Model is the most recent version of NetWeaver. This modeling tool can be used to compare, contrast, and evaluate any ecosystem and produce scores by altering habitat character information. NetWeaver is a characterization mechanism; it compares the inventory of characters of a reference site to the inventory of characters from other research sites. Concurrently, NetWeaver can be used to study and document ecosystem integrity [24].

The modularity of NetWeaver allows the evolution of complex knowledge bases from small, incremental steps [24]. Key features of NetWeaver include object-based networks of logical propositions and fuzzy logic that provides a complete calculus for knowledge representation and can easily be used by resource managers [23, 24]. NetWeaver was selected as the modeling tool for this study because NetWeaver can examine and discern various characters of a variety of ecosystems [23, 24]. For this study, the NetWeaver modeling tool focused on the Bridle Shiner, *Notropis bifrenatus* (Cope) and Ironcolor Shiner, *Notropis chalybaeus* (Cope) in various habitats on the Atlantic seaboard.

These cyprinids historically were widespread throughout the Atlantic seaboard. Now both species exist in isolated pockets along the Atlantic seaboard with very limited, if any, gene flow among populations. Marshalls Creek may be the only isolated pocket where both shiner species currently occur sympatrically and syntopically [31, 32]. Marshalls Creek (Delaware River drainage, Monroe County, Pennsylvania) is approximately 42 km long. Marshalls Creek's headwaters flow from the Pocono escarpment,

and the entire drainage area is 69.4 km<sup>2</sup>. Marshalls Creek water quality is designated as a high-quality cold-water fishery by the Pennsylvania Code water quality standards [31]. The Bridle and Ironcolor shiners inhabit a 3.7 km reach of Marshalls Creek.

The Ironcolor Shiner is listed as endangered in Maryland and both shiners are listed as endangered in Pennsylvania [33, 34]. The Bridle Shiner is listed as a special concern species in Massachusetts [35]. Ironcolor Shiners are not listed as an Endangered, Threatened, or Candidate species in Delaware [36]. Neither shiner is listed federally as an Endangered, Threatened, or Candidate species [37].

The native range of the Ironcolor Shiner is highly fragmented and includes the lowlands of the Atlantic Coast, Gulf Coast, and Mississippi River drainages from New York to Florida to Texas mostly south of Pennsylvania [32]. Their native range also includes sporadic areas of the southern Great Lakes region in Wisconsin, Illinois, and Indiana [38]. Except in the Southeast, most populations are disjointed [39]. The Ironcolor Shiner has disappeared from some areas of New Jersey. Populations in Iowa have been reported as extirpated. Ironcolor Shiners historically were widespread throughout the Delaware River drainage, however, their populations in Pennsylvania have significantly decreased [31]. The only known population in Pennsylvania is in Marshalls Creek.

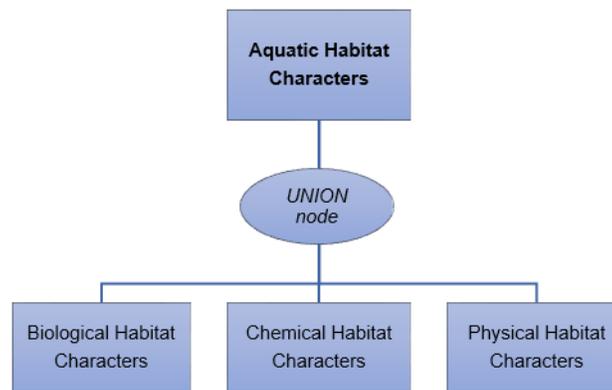
The Bridle Shiner historically inhabited the Susquehanna River drainage and the Delaware River drainage. Populations have significantly decreased and are recently found in one (i.e., Marshalls Creek) of 31 other historical locations in Pennsylvania [40-43]. The Bridle Shiner was once widespread in Maryland. After extensive collection efforts in areas of historical occurrence since 1984 resulted in no Bridle Shiners. The extirpation of the Bridle Shiner in Maryland has now been reported [44].

Several factors including habitat alteration, losses of stream vegetation, urbanization, industrialization, water diversion, industrial and sewage plant discharges, and increased turbidity and sedimentation have all been implicated for these population declines [31,43]. The exact cause of the extirpation was unknown but declines in other regions have been attributed to increased sedimentation, the constant loss of native aquatic vegetation, the increase in non-native plants including *Hydrilla verticillata* (Hydrilla) and *Myriophyllum spicatum* (Eurasian Watermilfoil), and the increase of non-native predators including *Micropterus nigricans* (Largemouth Bass) and *Ictalurus punctatus* (Channel Catfish) [44]. Populations of Bridle Shiners in Virginia have been localized (i.e., the James River drainage) and some populations have been extirpated or nearly so (e.g., the Potomac River drainage and the Rappahannock River drainage) [39, 45]. Habitat alteration (i.e., light reduction that impairs growth of submerged aquatic plants and food-sighting ability of the fish) seems to be the general cause of the localization and population decline [39]. Our purposes

included: (1) documentation of the aquatic habitat characters (i.e., the combination and interaction of biological, chemical, and physical habitat characters) that are coincident with these shiners in Marshalls Creek; (2) determination if these aquatic habitat characters are commonly found at other historical sites for these shiners; and (3) analysis of why these shiners are syntopic only in Marshalls Creek.

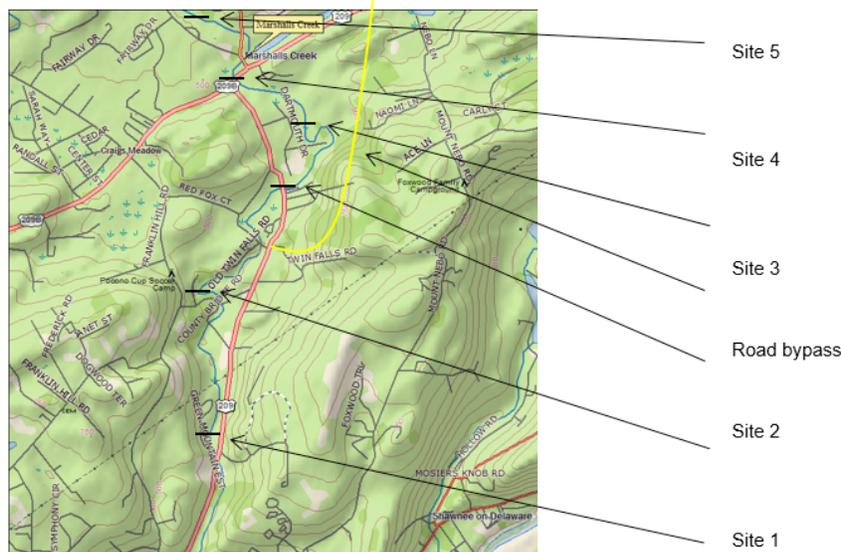
**Materials and Methods**

The modularity of NetWeaver (Fig. 1) allows the evolution of complex knowledge bases from small, incremental steps [24]. Key features of NetWeaver include object-based networks of logical propositions and fuzzy logic that provides a complete calculus for knowledge representation and can easily be used by resource managers [23, 24]. NetWeaver was selected as the modeling tool for this study because NetWeaver can examine and discern various characters of a variety of ecosystems [23, 24].

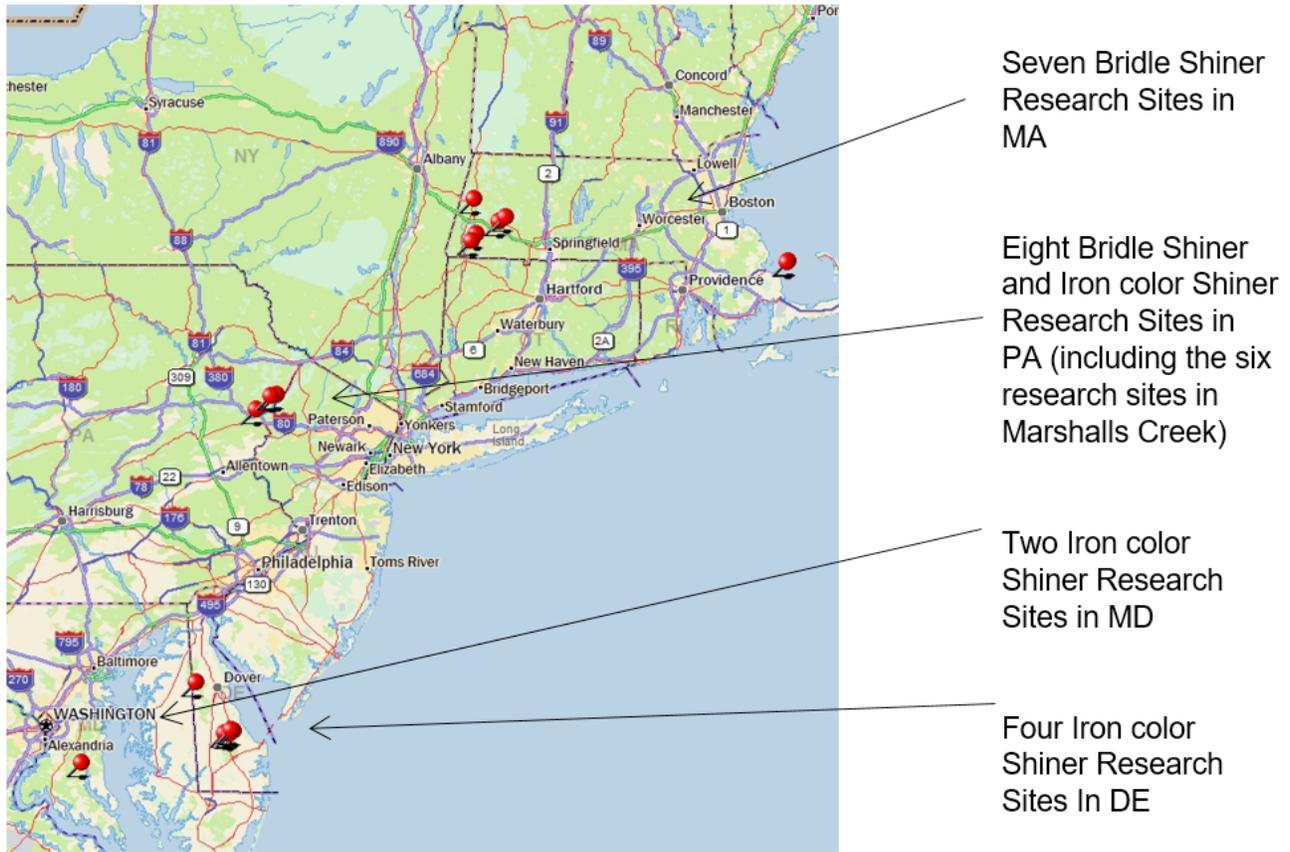


**Figure 1:** Aquatic Habitat Characters Dependency Network for the aquatic habitat scores in the NetWeaver Model [24]

The Bridle and Ironcolor shiners inhabit a 3.7 km reach of Marshalls Creek. Six Marshalls Creek sites (Sites 0–5) were sampled (Fig. 2) and seven sites in Massachusetts, two in Maryland, and four in Delaware (Fig. 3).



**Figure 2:** Research Sites 0-5 (a 3.7 km reach) and Road Bypass Construction at Marshalls Creek, Monroe County, Pennsylvania. Illustration Delorme 2013.



**Figure 3:** Research Sites for the Bridle Shiner in Massachusetts, for the Bridle and Ironcolor Shiner in Pennsylvania, for the Ironcolor Shiner in Maryland, and for the Ironcolor Shiner in Delaware. Illustration Delorme 2013.

We collected fishes with battery powered backpack electrofishing gear (i.e., Smith-Root, LR-24, set at pulsed 300 volts direct current). Fishes were sorted, counted, and identified to species. For all the other historical research sites, pictures were taken of all fishes to document the species collected and all fishes were then released. For Marshall Creek research sites, annual collections have occurred every summer for several years. For all the other historical research sites, collections occurred only once in the summer of 2012.

We collected aquatic macroinvertebrates using a standard D-frame kick net with a 1,200-micrometer mesh [46]. We identified the macroinvertebrates, which were preserved in 70% ethanol, to order, family, or genus depending on specimen type [47, 48]. We stored all specimens in the laboratory at The Pennsylvania State University. For research sites in Marshalls Creek, we had collections for several years every March, August, and December; and we used the same location used for each research site collection. We collected at all the other historical research sites, collections in the summer of 2012.

We collected aquatic plants/algae by hand in 100 m reaches. Due to private property restrictions, we only collected in 50 m reaches at historical research sites (i.e., Eel River, MA; Flat Brook, MA; Schenob Brook, MA; Sambo Creek, PA; and McMichael Creek, PA). We labelled each aquatic plant/algae sample with stream location information and stored in a 2-liter plastic container with freshwater. In the laboratory, we rinsed and drip-dried aquatic plants/algae. We positioned the samples as flat as possible between sheets of newspaper. We used a cardboard drier to separate each specimen. We used a plant press to stack and hold specimens for one month to dry. We identified each plant to genus/species level using 2 references [49, 50]. All specimens were stored at Penn State University. For Marshalls Creek Sites 0–4, PA, we collected aquatic plants/algae in January, March, April, June, August, and October over a period of 2 years. For Marshalls Creek Site 5, PA, we collected aquatic plants/algae in June, August, and October over a period of 1 year. For all the other historical research sites, we collected only once in the summer of 2012.

For all Marshalls Creek sites, we remotely monitored water chemistry at the Marshalls Creek Site 4, PA, (located upstream of the road construction) and Marshalls Creek Site 0, PA, (located downstream of all road construction) in order to determine if road construction was altering the chemical composition of Marshalls Creek. We recorded pH, conductivity (microsiemens/cm), and dissolved oxygen (mg/liter) every 30 minutes on a website using Hach Company sondes (calibrated monthly) and Stevens Company transmission equipment. We used water chemistry data from January 2011–December 2012 to calculate ranges for pH, conductivity, and dissolved oxygen. For all the other historical sites, we measured pH, conductivity (microsiemens/cm), and dissolved oxygen (mg/liter) using a portable chemistry monitoring kit (i.e., YSI Professional Plus, calibrated monthly). For all sites, we determined alkalinity and hardness using commercial aquaria test strips.

We measured water velocity (cm/sec) and depth (cm) measurements with a portable flow meter (i.e., Flo-Mate, Model 2000, Portable Flow Meter (calibrated monthly)) and associated sliding rod. For streams <0.75 m, we took water velocity measurements at 60% of total depth (this approximates mean column velocity). For streams >0.75 m, we measured water velocity at 20% and 80% of total depth (from the top) and we averaged of these velocities to estimate the mean column velocity. For all sites, we used the Rapid Bioassessment Protocols from the United States Environmental Protection Agency to evaluate physical habitat characters including substrate composition (% mud/sand), riparian protection (% erosion), flow status (% flow to both banks), bank stability (% bank erosion), and channel alteration (% altered).

We created NetWeaver model dependency networks, by determining trophic relationships of these shiners with associated fishes. We grouped the fish associates as follows: (1) endangered shiner predator species and (2) endangered shiners non-predator species. Additionally, we determined if a given species was native or introduced.

In order to create NetWeaver dependency networks, we determined trophic relationships of these shiners to the aquatic macroinvertebrates that were present as a given site as follows: (1) edible aquatic macroinvertebrate taxa consumed by endangered shiners and (2) non-edible aquatic macroinvertebrate taxa associated with endangered shiners.

We created NetWeaver dependency networks by assessing spawning relationships of these shiners with aquatic plants/algae present at a given site as follows: (1) spawning plant species used by endangered shiners and (2) non-spawning plant/algae species associated with endangered shiners.

We used NetWeaver software to construct various dependency networks for a reference ecosystem. We developed NetWeaver model as a collection of simple data link questions, fuzzy logic arguments, dependency network (i.e., goal) groups,

and dependency networks (i.e., goals) with their associated logic nodes. For a completed knowledge base, each dependency chain ended with a simple data link question. We used a total of 116 simple data link questions in the NetWeaver model [24]. Hanson [51] dissertation, Chapter 2, pages 37-48, gives a detailed description of this NetWeaver Model. Marshalls Creek Site 1, PA, aquatic (i.e., biological, chemical, and physical) habitat characters were used as the empirical reference data set. Each of the aquatic habitat characters from the other 20 research sites were then entered separately into the model as an empirical input data set. Strength of evidence scores were then produced from the model that compared the aquatic habitat characters from the other 20 research sites to the Marshalls Creek Site 1, PA, reference aquatic habitat characters [24]. A NetWeaver model was constructed in 3 steps. First, the model was built on an empirical reference data set. Second, an empirical input data set was entered into the model. Third, strength of evidence scores (i.e., trueness levels) were produced from the model that compares the empirical input data set to the empirical reference data set [24].

A model was developed that represented the complex aquatic (i.e., biological, chemical, and physical) habitat characters for Marshalls Creek Site 1, PA, reference site. Over 150 biological, chemical, and physical habitat characters were part of this NetWeaver modeling tool. Other historical sites from Maryland, Delaware, Massachusetts, and Pennsylvania, including other Marshalls Creek sites were compared, contrasted, and evaluated using NetWeaver at the Marshalls Creek Site 1, PA. Hanson [51], Appendix A-U, gives detailed, aquatic habitat character collection data for all research sites. All of these network components were utilized to produce strength of evidence scores (i.e., trueness levels) to compare the input aquatic habitat characters of 20 research sites to the reference aquatic habitat characters of Marshalls Creek Site 1, PA [24]. Raw data for all 21 research sites were utilized, and the data resulted from research site collections and measurements were not standardized. All data reported below were used in the NetWeaver dependency networks to produce strength of evidence scores.

For any given empirical input data set, NetWeaver produced strength of evidence scores with a range from -1.0 to +1.0. All strength of evidence scores between -1.0 and +1.0 indicated a trueness level. A score of -1.0 (100% false) was the lowest strength of evidence score possible and a score of +1.0 (100% true) was the highest strength of evidence score possible. When the empirical reference site data (i.e., the Marshalls Creek Site 1, PA, reference aquatic habitat characters) were entered into a NetWeaver, the highest strength of evidence score of +1.0 (100% true) was always achieved because a NetWeaver model was built with the empirical reference data set [24].

To simplify the strength of evidence score, the -1.0 to +1.0 strength of evidence score ranged (a 2.0 difference) was converted to a 0.0 to 200.0 strength of evidence score range, where 0.0 (a -1.0

NetWeaver model strength of evidence score, 100% false) was the lowest converted strength of evidence score possible and 200.0 (a +1.0 NetWeaver model strength of evidence score, 100% true) was the highest converted strength of evidence score possible. A converted strength of evidence score of 200.0 was always achieved for the Marshalls Creek Site 1, PA, reference aquatic habitat characters because it was the empirical reference data set [24].

**Results**

We collected a total of 389 Bridle Shiners and 433 Ironcolor Shiners (Table 1). The only time the presence of Bridle and/or Ironcolor shiners was used in any of the NetWeaver model dependency networks was in the percentage of native species calculation.

	<b>Bridle Shiners</b>	<b>Ironcolor Shiners</b>	<b>Aquatic Habitat</b>
<b>Location</b>	<b>Collected/Observed</b>	<b>Collected/Observed</b>	<b>Characters Score</b>
Marshalls Creek Site 1, PA <sup>1</sup> (model reference site)	261/1000+	307/1000+	200
Marshalls Creek Site 0, PA <sup>1</sup>	8/0	4/0	138
Marshalls Creek Site 2, PA <sup>1</sup>	44/0	35/0	157.9
Marshalls Creek Site 3, PA <sup>1</sup>	50/0	71/0	159.4
Marshalls Creek Site 4, PA <sup>2</sup>	0/0	0/0	141.1
Marshalls Creek Site 5, PA <sup>2</sup>	0/0	0/0	135.9
Long Marsh Ditch, MD <sup>3</sup>	0/0	0/0	130.3
Zekiah Swamp, MD <sup>3</sup>	0/0	0/0	106.8
Nanticoke River, DE <sup>3</sup>	0/0	7/100+	144.6
Gum Branch (main.), DE <sup>3</sup>	0/0	4/50+	137.4
Gum Branch (head.), DE <sup>3</sup>	0/0	5/200+	123.2
West Branch, DE <sup>3</sup>	0/0	0/0	146.4
Clifford Rd Dam Outlet, MA <sup>3</sup>	2/10+	0/0	132.4
Eel Creek, MA <sup>3</sup>	0/0	0/0	108.3
Flat Brook, MA <sup>3</sup>	6/35+	0/0	89.5
Hop Brook, MA <sup>3</sup>	9/200+	0/0	112.2
W. Branch Farm. River, MA <sup>3</sup>	0/0	0/0	117.1
Schenob Brook, MA <sup>3</sup>	0/0	0/0	79.4
Dry Brook, MA <sup>3</sup>	9/20+	0/0	103.4
Sambo Creek, PA <sup>3</sup>	0/0	0/0	96.6
McMichael Creek, PA <sup>3</sup>	0/0	0/0	122.4
<sup>1</sup> 7 Endangered Shiners collections, <sup>2</sup> 2 Endangered Shiners collections, <sup>3</sup> 1 Endangered Shiners collection			

**Table 1:** Bridle Shiners collected/observed, Ironcolor Shiners collected/observed, and strength of evidence scores for aquatic habitat characters for both species at all research sites.

Marshalls Creek Site 1, PA, clearly (by two orders of magnitude) contained the largest populations of Bridle and Ironcolor shiners. Therefore, all aquatic (i.e., biological, chemical, and physical) habitat characters from Marshalls Creek Site 1, PA, were used as a model reference (i.e., benchmark) data set for the NetWeaver model.

Fish associates (31 species) included 16 predator species and 15 non-predator species. Five exotic species and 3 additional (i.e., not found at the model reference site) exotic species were collected. Many of these exotic species had been residents for many years, however they were grouped together for our analysis. Concurrently, seven additional (i.e., not found at the model reference site) predator species were collected. Fish associates included 26 native species and 5 exotic species. Aquatic macroinvertebrates (207 taxa) included 76 edible aquatic macroinvertebrate taxa and 131 non-edible aquatic macroinvertebrate taxa associated with endangered shiners. Aquatic plant/algae (33 species) included 17 species used by for reproduction and 16 species on which the shiners did not spawn.

For Marshalls Creek research sites, a range for pH (i.e., 5.8–8.5), dissolved oxygen (i.e., 7.5 mg/liter–15.0 mg/liter), conductivity (i.e., 80 microsiemens/cm–290 microsiemens/cm), alkalinity (i.e., 20 ppm–50 ppm), and hardness (i.e., 50 ppm–75 ppm) were recorded. For Marshalls Creek research sites, a wide range for water temperature (i.e., 0.5 °C–26.0°C) was recorded. Additionally, the depth, velocity, and substrate composition of mud/sand for Marshalls Creek Site 1, were ideal for both the Bridle and Ironcolor shiner’s spawning and habitat preferences.

This study determined that the aquatic habitat characters found at Marshalls Creek Site 1, PA, were not commonly found at other historical sites for these shiners. The highest strength of evidence score for Marshalls Creek sites (besides the model reference site) was 159.4 and the highest strength of evidence score for other historical sites was 146.4. In all cases research sites with the highest NetWeaver strength of evidence scores for various aquatic habitat characters resulted in a closer similarity to the model reference site.

The unique combination and complex interaction of aquatic habitat characters at Marshalls Creek Site 1, PA, included: (1) an absence of fish associates especially the two exotic/endangered shiners predator species of *Micropterus dolomieu* (Smallmouth Bass) and *M. nigricans* (Largemouth Bass); (2) a presence of aquatic macroinvertebrate taxa, especially the 60 edible aquatic macroinvertebrate taxa consumed by endangered shiners including Ephemeroptera (mayflies) taxa, Trichoptera (caddisflies) taxa, Diptera (true flies) taxa, Amphipoda (scuds) taxa, Gastropoda (snails) taxa and Bivalvia (clams) taxa; (3) a presence of aquatic plant/algae species, especially the 8 spawning plant species used by endangered shiners including feather-leaved submerged aquatic plants (e.g., *Ceratophyllum* spp. (coontail)) and broad-leaved submerged aquatic plants (e.g., *Potamogeton* spp. (pondweed)); (4) satisfactory measurements for chemical habitat characters for these shiners; and (5) satisfactory measurements for physical habitat characters especially depth (>100 cm), velocity (17.6 cm/sec), and substrate composition (80% mud/sand) that are ideal for both the Bridle and Ironcolor shiner’s spawning and habitat preferences.

The aquatic habitat characters dependency network (Fig. 1) was the weighted average (i.e., UNION node) strength of evidence score (i.e., trueness level) for biological habitat characters, chemical habitat characters, and physical habitat characters [24].

Similarly, within the biological habitat characters node there were three dependency network groups (i.e., Fishes, Aquatic Macroinvertebrates, Aquatic Plants/Algae) in the NetWeaver model that we developed. The Fishes dependency network was the weighted average (i.e., UNION node) strength of evidence score (i.e., trueness level) for endangered shiners predator species, additional (i.e., not found at the model reference site) endangered shiners predator species, endangered shiners non-predator species, native species, and additional (i.e., not found at the model reference site) exotic species [24].

Similarly, the aquatic macroinvertebrate dependency network was the weighted average (i.e., UNION node) strength of evidence score (i.e., trueness level) for edible aquatic macroinvertebrate taxa consumed by endangered shiners and non-edible aquatic macroinvertebrate taxa associated with endangered shiners. The aquatic plant/algae dependency network was the weighted average (i.e., UNION node) strength of evidence score (i.e., trueness level) for spawning plant species used by endangered shiners and non-spawning plant/algae species associated with endangered shiners [24].

Similarly, the chemical habitat characters dependency network was the weighted average (i.e., UNION node) strength of evidence score (i.e., trueness level) for pH, dissolved oxygen (mg/liter), conductivity (microsiemens/cm), alkalinity (ppm), and hardness (ppm) [24]. The physical habitat characters dependency network was the weighted average (i.e., UNION node) strength of evidence score (i.e., trueness level) for water temperature (°C), depth (cm), velocity (cm/sec), substrate composition (% mud/sand), riparian protection (% erosion), flow status (% flow to both banks), bank stability (% bank erosion), and channel alteration (% altered) [24].

## Discussion

We developed a NetWeaver modeling tool to assess the biodiversity of the aquatic environment and the habitat variability/characters associated with extirpated species (e.g., Bridle Shiner and Ironcolor shiners) in an attempt to increase sustainability of aquatic faunas through increasing biotic resistance. We demonstrated that the NetWeaver tool was successfully developed to assess the aquatic biodiversity and the habitat variability/characters for these shiners. More importantly, various interacting and discerned aquatic habitat characters data from NetWeaver model dependency networks and their associated strength of evidence scores produced insight that was not evident by univariate examination of the collection and measurement data at research sites.

We postulated that Marshalls Creek (i.e., the Marshalls Creek Site 1, PA) harbored both shiners in relatively large numbers because of a unique combination and complex interaction of aquatic habitat characters; thus, we needed to document (1) the morphological/meristic description, habitat, feeding, and spawning preferences of these shiners; and the native range overlap for these shiners; (2) NetWeaver attributes and dependency networks; (3) collection and measurement data for all research sites; and (4) NetWeaver strength of evidence scores for various aquatic habitat characters at all research sites.

## Conclusions

The loss of biodiversity of natural systems (i.e., the loss of the variety of life forms and processes of natural systems) is directly related to the over-consumptive and overgrown human population [52, 53]. Habitat alteration, overharvesting, pollution, and introduced species are all primary agents for the loss of biodiversity [53]. The loss of species due to introductions has had detrimental impacts on native aquatic faunas throughout the world [54, 55]. Understanding the complex effects of human activities on aquatic ecosystems presents a challenge to ecologists, biologists, and resource managers.

With these attributes, the NetWeaver tool developed for this study produced strength of evidence scores to compare, contrast, and evaluate aquatic habitat character data relative to these shiners. This NetWeaver tool can assist aquatic ecologists, biologists, and resource managers to assess certain biological, chemical, and physical ecosystem elements; the relationships of fishes, aquatic macroinvertebrates, and aquatic plants/algae to endangered and threatened fish species; and the potential uniqueness of aquatic ecosystem biodiversity. Further, this NetWeaver tool may provide a method to predict the effect of biodiversity component changes; and present ecologists, biologists, and resource managers much needed data to make decisions that will protect the biodiversity of a system, increase the potential of that biodiversity to recover should it be impacted, and support reintroductions of fishes.

**Author Contributions:** C. W. Hanson – collection of data, literature review, data analyses, initial preparation of manuscript. J. R. Stauffer Jr. – Initial design of research, aided in collection of data, revision and editing of manuscript.

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were obtained from The Commonwealth of Massachusetts. A Scientific Collection Permit and an Endangered Species Permit were obtained from Maryland. A Scientific Collecting Permit was obtained from Delaware.

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**Conflicts of Interest:** no conflicts of interest.

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