Changes in Structure and Function of Fish Assemblages along Environmental Gradients in an Intensive Agricultural Region of Subtropical Taiwan[1]

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Abstract: Intensive agriculture has degraded streams in subtropical Taiwan, but agricultural impacts on fish assemblages are not well studied. The goal of this study was to understand changes in structure and function of fish assemblages along environmental gradients in an agricultural region of South-central Taiwan. Nineteen sites in the hill and upper plain regions were selected for fish sampling during the base flow period. Water chemistry analyses and rapid habitat assessment were also conducted. Cluster analysis separated fish assemblages into four assemblage groups and a single site. A redundancy analysis (RDA) showed that environmental variables explained 73.9% of species variance. RDA axis 1 represented a habitat-diversity, cover, and nutrient gradient, whereas RDA axis 2 represented a complex riparian condition gradient. Relative abundances of dominant fish species and assemblage groups were related to water and habitat variables. Trophic and tolerance guilds were correlated with RDA axes. Number of fish species increased with decreasing elevation. Both structure and function of fish assemblages changed with water and habitat gradients in these subtropical agricultural streams.

Stream fish assemblages are structured by environmental factors, especially physical habitats and water quality, in agricultural regions (Cooper 1993, Allan 2004, Vondracek et al. 2005, Griffith et al. 2009). Physical disturbances in agricultural watersheds, such as riparian vegetation removal and channelization, can often increase water temperature and sediment input (Nagasaka and Nakamura 1999, Heartsill-Scalley and Aide 2003, Zaimes and Schultz 2011), which can reduce habitat quality and diversity, subsequently changing fish communities (Nerbonne and Vondracek 2001, Vondracek et al. 2005, Mueller et al. 2011). Inputs of excessive nutrients and animal wastes (e.g., from swine) can make streams unsuitable for sensitive fish due to low oxygen levels (Grimvall et al. 2000, Chambers et al. 2006, Sutela and Vehanen 2010). These degraded conditions in agricultural streams occur worldwide, but few examples are known from Taiwan and subtropical East Asia (Wang 1989, Wang et al. 1996).

Few published studies of stream fish in Taiwan describe the relationships between fish assemblages and both water and habitat quality in agricultural watersheds (Wang 1989, Wang et al. 1996) because habitat quality is not required for fish survey programs of water resources and environmental protection agencies. Information about these relationships can yield a more complete picture of agricultural impacts. In general, water and

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habitat conditions deteriorate from high- to low-elevation sites in Taiwan (Wang et al. 1996, Chen 2004). This gradient is associated with increasing agricultural development and urbanization in lower elevations (Lee et al. 2004). Consequently, fish assemblages change with both elevation and environmental degradation (Wang et al. 1996, Yeh et al. 2000, Chen 2004, Guo 2011). Similar patterns of human development along an elevation gradient and related responses of fish assemblages were also observed in Hawaiian (Brasher 2003, Brasher et al. 2006) and Japanese streams (Yoshimura et al. 2005).

In this study, instead of including a large elevation difference, we chose sites at mid and low elevations (hill and upper plain regions), where many fish species are presumed to coexist (Yeh et al. 2000) and stream conditions vary across different disturbance gradients. By sampling across this range, elevation was not the dominant factor controlling all relationships; thus we were able to gain more understanding of the relationships between fish and water and habitat variables. The goal of this study was to understand how structure and function of fish assemblages change along environmental gradients. We first explored the relationships between fish assemblages and environmental variables with multivariate statistics, evaluated the responses of fish guilds to environmental gradients, and then assessed the relationships between fish species richness and elevation. Results of this study can increase our understanding of the extent of degradation, the problems of fish conservation, and the feasibility of bioassessment in agricultural regions of Taiwan.

MATERIALS AND METHODS

Study Area

The study area is located in South-central western Taiwan in subtropical East Asia. The study area, including parts of Bai-Kang, Pu-Tzu, and Ba-Jang River watersheds, is in the hilly and upper plain regions of Chiayi County. The annual mean temperature is 23.0 ± 0.6°C (mean ± 1 SD), and the monthly average temperature ranges from 16.3 ± 1.0 (January) to 28.5 ± 0.5°C (July) (Central Weather Bureau 2012). The region has a monsoon climate, with the wet season from May through September and the dry season from October through April. The mean annual rainfall is approximately 2,300 mm, 80% of which is concentrated in the wet season (Taiwan Institute of Landscape Architects 2006). Streams in this region have steep topography and high rainfall events, which result in a flashy hydrology (Chang et al. 2009). Alluvium is located in the plain region, the Pleistocene Lateritic Terrace deposits and the Toukoshan Formation are located in the hilly region, and the Miocene Kueichulin Formation is located in the mountain region (Ho 2003).

Land use in the study region is primarily agriculture, with towns and small factories dispersed throughout the area (Department of Urban Planning of National Cheng Kung University 1997). The percentage of agricultural land use ranges from 38% to 68% in different townships (Department of Urban Planning of National Cheng Kung University 1997). The major crops include sugarcane, bamboo, pineapple, tea, citrus, longan, yam, pomelo, and persimmon; pig, duck, and chicken feedlots are also distributed in the region (Taiwan Institute of Landscape Architects 2006). The major point sources of wastewater are households, animal feedlots, and factories. Multiple types of pollution sources exist in the watersheds (DHV Planetek Company 2006) and may create complex environmental gradients. No wastewater treatment plants were present in the watersheds. The study area is outside the urban center, Chiayi City. No large forest patch is present, whereas small forest patches are often a mixture of bamboo, old orchard trees, and a few perennial trees (Chen and Chen 2006). The stream channels in this region are characterized by channelization and concrete embankments. This region offers a unique opportunity to explore the relationships between fish assemblages and multiple environmental gradients.

Nineteen study sites on Bei-Kang (BK), Pu-Tzu (PT), and Ba-Chang (BC) Rivers were selected (Figure 1). Three sites in the main stem of Pu-Tzu River are monitoring

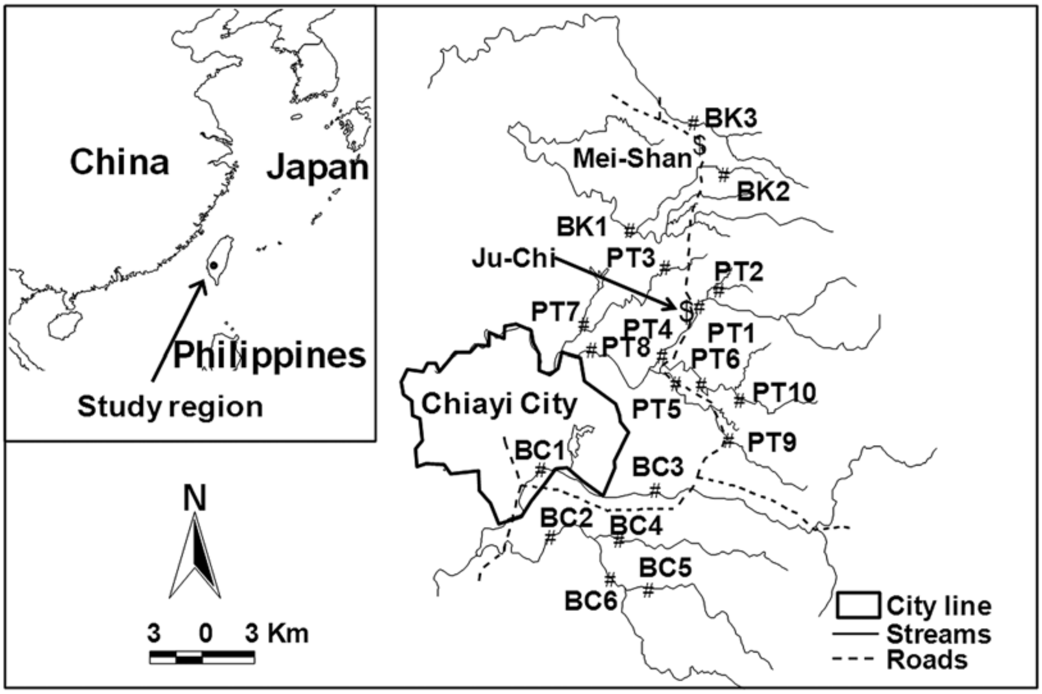

FIGURE 1. Map of study sites in Chiayi County, Taiwan. BK, Bei-Kang River; PT, Pu-Tzu River; BC, Ba-Chang River.

sites of the Environmental Protection Administration. We tried to sample all tributaries in this area. Most sites were in the tributaries or below confluences. Tributaries not selected were either dry or inaccessible for sampling. The sites had low to high levels of agricultural areas based on prior reconnaissance and satellite images (Figure 1) (Liu 2007). According to reconnaissance and satellite images, PT7, BK1, BC4, and BC5 were surrounded by agriculture landscape, whereas BK2, PT1, PT2, PT9, PT10, and BC3 had less agricultural land use. The other sites had medium levels of agricultural land use. However, animal feedlots and a food-processing facility, major sources of nutrients, were not identifiable in satellite images.

Sample Collection

Sampling was conducted in January 2008. Fishes were collected using the electrofishing method with local and national governmental agencies permits. One person operated the backpack electrofisher (Freshwater, Minghsiung, Chiayi, Taiwan) and a fishnet, and two additional people held large fishnets to catch the stunned fish. The length of each sampling site was 100 m (Lee and Liang 2003). Block nets were set at both the upstream and downstream ends of the reach. Single-pass removal sampling was used to estimate fish assemblage compositions (Chen et al. 2004, Han et al. 2007). Fish species, number, and total length to the nearest millimeter were recorded in the field. The high flow in the wet season impeded proper sampling. Voucher specimens were preserved in the laboratory.

Dissolved oxygen (DO), pH, water temperature, and conductivity were measured and recorded in the field (YSI DO200 and YSI model 63 [YSI Inc., Ohio, USA]). The equipment was calibrated before use. One liter of 5-day biological oxygen demand (BOD5) and 500 ml of suspended solids (SS) samples were collected separately. One liter of water samples for NH_4-N, NO_3-N, PO_4-P, and total phosphorus (TP) analyses were acidified with sulfuric acid to $pH < 2$, and 250 ml of NO_2-N samples were not acidified. Water samples were collected in acid-rinsed polyethylene bottles and stored on ice until transported to the laboratory.

Water physical and chemical parameters were analyzed according to Taiwan's National Institute of Environmental Analysis (NIEA) methods (http://www.niea.gov.tw/analysis/epa_www.htm). Water samples were filtered with 33 mm diameter filters of 0.45 μm pore size in the laboratory except for the SS and TP samples. SS samples were filtered on a preweighed 47 mm diameter glass-fiber filter of 1.5 μm pore size, stored in predried and weighed aluminum dishes, then dried in a 105°C oven for 1 hr until weight was constant (precision to 1 mg) (NIEA W210.57A). The weight difference between dried sample plus filter and dried filter divided by filtered water volume is the SS. BOD5 was analyzed according to NIEA methods (W510.55B). NH_4-N was analyzed with the phenol method (NIEA W448.51B), and NO_3-N and NO_2-N were analyzed with spectrophotometric methods (NIEA W419.51A and NIEA W418.51C, respectively). The PO_4-P and TP samples were analyzed with the ascorbic acid method (NIEA W427.53B).

Benthic algal biomass represented by chlorophyll *a* (Chl *a*) was estimated by the following methods. Algae were randomly collected from five rocks in flowing water by using a toothbrush to brush the rock surface inside a rubber circle placed on the rocks and then scraping the sample into a plastic bag. Algal samples were preserved on ice in the field and in a 4°C refrigerator in the laboratory. The inside of the rubber circle has an area of 36 cm^2. Well-mixed known volumes of algal aliquot were filtered through a 47 mm Whatman GF/C filter of 1.2 μm pore size with the assistance of a vacuum pump. Chl *a* was extracted by immersing the filter in 10 ml of 90% aqueous acetone that was then exposed to 30 sec of sonication and kept in a 4°C refrigerator for 24 hr. After 20 min of centrifugation at $500 \times g$, Chl *a* was measured following methods of APHA (1998) with a spectrophotometer (Spectro Instruments) and corrected for pheophytin *a*.

Habitat Assessment

Habitat assessment was conducted following the high-gradient protocols of the U.S. Environmental Protection Agency (Barbour et al. 1999) because the average slope of the study streams is about 1.9% (TIIWE 2005). Ten variables were included in this habitat assessment: available cover (H-AC), embeddedness (H-EB), velocity/depth regime (H-VD), sediment deposition (H-SD), channel flow status (H-CF), channel alteration (H-CA), frequency of riffles (H-FR), bank stability (H-BS), vegetative protection (H-VP), and riparian vegetative zone width (H-RW). The highest score of each variable is 20; higher scores of these variables indicate better habitat conditions. Each variable was assessed according to descriptions of each variable and of score range in Barbour et al. (1999). Scores 0 to 4 indicate poor condition, scores 6 to 10 indicate marginal condition, scores 11 to 15 indicate suboptimal condition, and scores 16 to 20 indicate optimal condition. The total scores of each site were the sum of scores of these 10 variables. Habitat assessment was performed by one person and was calibrated with several on-site visits to ensure the quality of the data.

Data Analyses

The similarity of the fish assemblages from different sites was determined first. The relative abundance data of fish assemblages were subjected to a cluster analysis with the unweighted pair-group method using arithmetic averages (UPGMA). Bray-Curtis dissimilarity was calculated as the distance for clustering. The resulting dendrogram was plotted, and the dominant species (mean relative abundance >10%) was calculated for each cluster group. The differences in environmental variables among cluster groups were compared with one-way analysis of variance (ANOVA). When ANOVA showed a significant result, Student-Newman-Keuls (SNK) post hoc tests were applied to separate different groups. ANOSIM (analysis of similarities) was used to test for significance among cluster groups, and SIMPER (similarity percentage) was used to determine species responsible for cluster groups. ANOSIM is a nonparametric test of significant difference between two or more groups based on Bray-Curtis dissimilarity distance measure, whereas SIMPER is used to assess which taxa are primarily responsible for an observed difference between groups (Clarke 1993).

To assess the relationships between the fish assemblages and the water quality and habitat variables, redundancy analysis (RDA) was employed to explore possible patterns. RDA is a direct extension of multiple regression and models a linear combination of the response variables according to a linear combination of the explanatory variables (Legendre and Legendre 1998). Fish species present at less than three sites were excluded from the RDA. Forward selection was used to select environmental variables. Environmental variables with a high variance inflation factor (VIF) (>20) were removed from analysis. The environmental variables were standardized and centered, and relative abundance data were not transformed.

The relationships between fish guilds and environmental gradients were evaluated with Pearson correlation analysis. Site scores on RDA axes (results of RDA) were used to represent environmental gradients. Fish guilds included trophic groups and tolerance of the fish species to pollution (Table 1). The trophic groups were categorized as herbivores, insectivores, omnivores, and piscivores, and

TABLE 1

Species List, Trophic and Tolerance Guilds of Species, and Nonnative Status (modified from Chang et al. [1999] and Shao [2012]

Family	Species	Abbrev.	Common Name	Trophic Group[a]	Tolerance[b]	Nonnative[c]
Anguillidae	*Anguilla marmorata*	AM	Marbled eel	Pisc./Ins.	H	
Bagridae	*Pseudobagrus adiposalis*	PA	Bagrid catfish	Pisc./Ins.	I	
	Pseudobagrus brevianalis	PB	Bagrid catfish	Pisc./Ins.	I	
Balitoridae	*Hemimyzon formosanus*	HF	Formosan river loach	Herb.	I	
Channidae	*Channa striata*	CS1	Snakehead murrel	Pisc./Ins.	H	V
Cichlidae	*Oreochromis* sp.	OR	Tilapia	Omn.	H	V
	Amphilophus citrinellus	AC	Midas cichlid	Omn.	H	V
Clariidae	*Clarias fuscus*	CF	Walking catfish	Pisc./Ins.	H	
Cobitidae	*Cobitis sinensis*	CS2	Siberian spiny loach	Omn.	M	
	Misgurnus anguillicaudatus	MA	Pond loach	Omn.	H	
Cyprinidae	*Acrossocheilus paradoxus*	AP	Taiwan striped barb	Herb.	M	
	Candidia barbata	CB	Dace	Insect.	I	
	Carassius auratus auratus	CA	Golden carp	Omn.	H	
	Cyprinus carpio carpio	CC	Common carp	Omn.	H	V
	Microphysogobio alticorpus	ML	Deep-body gudgeon	Omn.	H	
	Onychostoma barbatulum	OB	Taiwan shoveljaw carp	Omn.	I	
	Opsariichthys pachycephalus	OP	Freshwater minnow	Insect.	M	
	Pseudorasbora parva	PP	Topmouth minnow	Omn.	M	
	Puntius semifasciolatus	PS	Six-banded barb	Omn.	H	
	Tanakia himantegus	TH	Taiwan bitterling	Omn.	H	
Gobiidae	*Rhinogobius candidianus*	RC	Goby	Insect.	M	
	Rhinogobius giurinus	RG	Goby	Insect.	H	
	Rhinogobius rubromaculatus	RR	Goby	Insect.	M	
Siluridae	*Silurus asotus*	SA	Chinese catfish	Pisc./Ins.	H	V
Poeciliidae	*Gambusia affinis*	GA	Mosquito fish	Insect.	H	V

[a] Herb., herbivores; Insect., insectivores; Omn., omnivores; Pisc./Ins., piscivores/insectivores.

[b] I, intolerant species; M, tolerant species; H, highly tolerant species.

[c] V, nonnative species.

the tolerance to pollution was represented by a classification into intolerant, tolerant, and high-tolerant species. For example, an insectivore/piscivore fish species was classified in both guilds, and percentage of each guild was calculated by dividing its percentage by 2. Trophic groups and tolerance to pollution of fish species were based on the Taiwan fish database (Tzeng 1986, Chang et al. 1999, Shao 2012) and consultation with experts.

The relationships between the number of species (native or nonnative) and elevation and between the numbers of species occupying <50% of sites and elevation were also assessed with Pearson correlation analysis. The relative abundance data were arcsine-square root transformed before analyses, and environmental data were log-transformed except pH. The cluster analysis, ANOVA, RDA, and Pearson correlation analysis were performed with the vegan (Oksanen et al. 2010) and rdaTest packages of the R program (R Development Core Team 2009). ANOSIM and SIMPER were analyzed with the PAST program (Hammer et al. 2001).

RESULTS

Summary of Fish Samples and Environmental Variables

A total of 25 species was collected. The average number of fish species caught at a site was 8 ± 3 (mean ± 1 SD), and the maximum and minimum numbers of fish species caught were 13 and 4, respectively. The average number of fish caught at sites was 236 ± 108. One-third of the species collected occurred at only one site. *Opsariichthys pachycephalus* (Cyprinidae), *Acrossocheilus paradoxus* (Cyprinidae), and *Rhinogobius candidianus* (Gobiidae) were collected from all sites and had average relative abundance of 28.9%, 26.0%, and 14.4%, respectively. Cyprinidae represented 40% of all species collected, followed by Gobiidae (12%). Catadromous *Anguilla marmorata* was the only diadromous migratory species caught. *Rhinogobius giurinus* can have either freshwater or amphidromous populations. Nonnative species were caught at nine sites and had a relative abundance of 7.3% ± 6.7%. Among the five nonnative species collected, *Oreochromis* sp. was the most abundant.

The data of water quality and physical variables are shown in Table 2. The elevation ranged from 33 to 165 m, corresponding to stream orders from 5 to 1. The highest-elevation site was BK2, and the lowest was PT8. Conductivity ranged from 180.1 to 1,249 μS/cm (mean ± 1 SD), and suspended solids (SS) ranged from 0 to 22 mg/liter. The water temperature ranged from 18.5 to 25.4°C, and the pH was from 5.93 to 10.75. The BOD5 ranged from 0 to 20.7 mg/liter, and the dissolved oxygen (DO) was from 7.95 to 13.44 mg/liter. NO_3-N ranged from 0.03 to 3.68 mg/liter, and NH_4-N was from 0.04 to 1.17 mg/liter. Total phosphorus (TP), closely related to PO_4-P concentrations, ranged from 0.01 to 0.89 mg/liter. Nutrients levels were relatively high.

The results of rapid habitat assessment are shown in Table 3. The rapid habitat assessment scores ranged from 93 to 141, with a mean of 112.6. Among the habitat assessment variables, available cover had the highest mean (15.2) because cobbles and boulders were the dominant substrata at most sites. Bank stability and velocity/depth regime had higher scores. Many sites had concrete stream banks, which were stabilized to prevent further erosion. Sediment deposition had the lowest mean because these streams had high sediment loads. Channel alteration and riparian zone width had lower scores, indicating a high degree of human modification of channels and riparian zones.

Relationship between Fish Assemblages and Environmental Variables

The cluster analysis resulted in the dendrogram shown in Figure 2. The cutoff similarity was set at 0.57 based on visual assessment and resulted in four cluster assemblage groups and a single site (BC6). Four groups were not restricted to any drainage basins. ANOSIM showed a significant difference of assemblage groups ($R = 0.82$, $P < .001$). SIMPER indicated that *A. paradoxus*, *O. pachycephalus*, *C. barbata*, and *R. candidianus* contributed to a

TABLE 2

Physical Characteristics and Water Chemistry of Sampling Sites

Sites[a]	Assemblage Groups	Stream Order	Elevation (m)	Canopy (%)	DO (mg/liter)	Conductivity (μS/cm)	Water temp. (°C)	pH	Chl *a* (μg/cm^2)	BOD (mg/liter)	SS (mg/liter)	NO_2-N (mg/liter)	NO_3-N (mg/liter)	NH_3-N (mg/liter)	PO_4-P (mg/liter)	TP (mg/liter)
PT1	2	4	95	14.5	11.09	708.0	21.6	7.23	0.48	3.5	1	0.02	0.14	0.06	N.D.	0.01
PT2	1	4	113	1.0	9.80	483.0	18.5	7.60	2.32	0.8	7	0.02	0.08	0.05	N.D.	0.01
BC2	3	5	43	25.5	12.37	384.6	20.6	7.06	0.92	10.8	0	0.02	2.55	0.09	N.D.	0.01
BK1	1	3	61	41.5	9.51	640.0	20.5	7.07	1.09	7.8	22	0.02	0.38	0.19	N.D.	0.02
PT10	2	3	161	10.0	10.15	319.5	19.7	7.33	0.45	0	2	0.04	0.03	0.04	0.01	0.02
PT9	3	2	151	13.5	9.22	426.5	19.9	7.54	0.84	7.5	1	0.02	0.14	0.08	0.01	0.02
BK2	3	1	165	46.8	9.30	732.0	18.9	7.38	1.69	6.2	0	0.02	0.53	0.11	0.01	0.03
PT6	1	3	111	1.3	10.80	180.1	20.4	7.68	1.52	20.7	0	0.02	0.29	0.15	N.D.	0.05
PT5	2	3	94	5.0	10.53	525.0	20.5	8.00	0.39	7.7	7	0.02	0.82	0.07	0.03	0.06
BC4	2	3	83	13.5	12.51	447.2	22.1	7.19	0.85	5.7	0	0.02	2.94	0.21	0.03	0.06
PT7	2	2	66	15.5	10.30	394.8	22.7	8.12	1.04	5.1	9	0.02	1.29	0.21	0.03	0.06
BC3	3	1	124	34.0	9.41	564.0	22.4	7.49	0.93	1.8	2	0.02	0.34	0.14	0.04	0.06
PT3	3	2	77	57.8	10.68	301.4	19.2	7.38	0.99	5.1	1	0.02	2.15	0.11	0.06	0.08
BK3	2	3	124	0.0	10.12	1,249.0	20.8	5.93	1.57	0	6	0.02	1.93	0.08	0.07	0.10
BC1	1	4	37	0.0	10.98	312.4	25.4	10.75	0.74	0.5	6	0.03	1.01	0.44	0.07	0.16
BC6	5	4	92	45.0	10.96	266.3	20.2	6.83	0.48	1.1	1	0.02	1.18	0.14	0.01	0.20
PT4	2	2	104	67.0	8.73	477.0	22.1	8.31	1.60	2.0	4	0.02	3.68	0.16	0.22	0.26
PT8	4	5	33	10.8	7.95	413.6	19.7	6.91	1.11	0.6	8	0.02	1.46	1.17	0.52	0.63
BC5	4	2	121	9.0	13.44	449.4	21.6	7.18	1.35	12	2	0.03	0.61	0.14	0.82	0.89

Note: Sites are sorted according to TP concentrations. Abbreviations: DO, dissolved oxygen; N.D., not determined; TP, total phosphorus.

[a] BC, Ba-Chang River; BK, Bei-Kang River; PT, Pu-Tzu River.

TABLE 3

Rapid Habitat Assessment Variables and Scores of Sampling Sites

Sites	Available Cover	Embeddedness	Velocity/ Depth Regime	Sediment Deposition	Channel Flow Status	Channel Alteration	Frequency of Riffles	Bank Stability	Riparian Protection	Riparian Zone Width	RHA[a] Scores
BC2	16	4	14	6	9	8	9	17	5	5	93
BC1	14	9	9	9	10	7	10	10	4	14	96
PT8	12	4	10	4	9	15	7	10	13	13	97
PT10	12	4	11	4	16	6	5	15	12	13	98
BC6	16	8	13	8	8	5	11	15	8	7	99
BC3	13	11	13	6	16	4	8	16	12	3	102
PT3	12	7	13	7	16	4	10	16	14	3	102
BK3	16	8	10	7	8	6	13	14	10	12	104
PT6	16	8	14	6	10	14	9	13	10	7	107
PT1	18	8	15	8	16	4	16	18	8	7	118
BC4	16	5	15	5	10	14	15	10	12	16	118
BC5	14	10	13	9	10	15	13	10	13	11	118
PT2	16	10	14	8	10	6	16	17	9	13	119
PT5	16	11	14	8	16	6	15	14	13	6	119
BK2	16	11	14	9	13	12	17	12	16	4	124
PT4	16	9	13	8	15	13	16	12	16	9	127
PT9	16	13	14	9	10	14	16	15	13	8	128
PT7	16	7	14	8	16	15	13	11	15	14	129
BK1	17	11	14	10	16	15	16	12	15	15	141

Note: Sites are sorted according to total RHA score.

[a] Rapid habitat assessment.

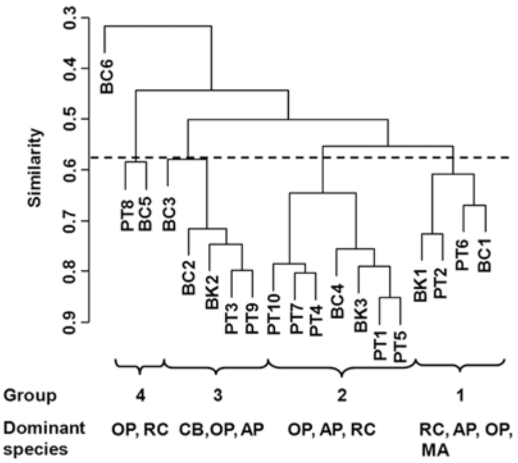

FIGURE 2. Dendrogram of results of cluster analysis of study sites, site groups, and dominant species of each group. Species abbreviations: AP, *Acrossocheilus paradoxus;* CB, *Candidia barbata;* OP, *Opsariichthys pachycephalus;* RC, *Rhinogobius candidianus*.

total of 69.2% of dissimilarity among sites. Group 1 had four sites and four dominant species: *R. candidianus* (goby), *A. paradoxus* (Taiwan striped barb), *O. pachycephalus* (freshwater minnow), and *Misgurnus anguilicaudatus* (pond loach). Group 2 had seven sites and three dominant species: *O. pachycephalus*, *A. paradoxus*, and *R. candidianus*. Group 2 also had two small subgroups: one comprised sites PT4, PT7, and PT10 with *O. pachycephalus* as the most dominant, and the other included PT1, PT5, BK3, and BC4 with *A. paradoxus* as the most dominant. Group 3 had five sites and three dominant species: *Candidia barbata* (dace), *O. pachycephalus*, and *A. paradoxus*. Group 4 had two sites and two dominant species: *O. pachycephalus* and *R. candidianus; O. pachycephalus* had a mean relative abundance of 53.8% in group 4.

Comparison of environmental variables among four assemblage groups showed that channel alteration scores differed significantly among assemblage groups (Table 4), and SNK post hoc tests showed that groups 1 and 4 had higher means than groups 2 and 3. Bank stability differed significantly among assemblage groups, and SNK showed that groups 1, 2, and 3 had higher means than group 4. Riparian vegetation width differed significantly among assemblage groups, and SNK showed that groups 1, 2, and 4 had higher means than group 3. NH$_4$-N and TP were significantly different among assemblage groups, and SNK showed that group 4 had a higher mean NH$_4$-N and TP than the other groups. PO$_4$-P and TP were highly correlated ($r = 0.97$, $P < .001$).

A total of 13 environmental variables was selected based on forward selection for redundancy analysis (RDA) (Table 5). Elevation was added to the RDA because it is an important factor based on literature. Dissolved oxygen was excluded due to high variation inflation factor. Environmental variables explained a total of 73.9% of species variance, and the first two RDA axes explained 48.5% of species variance. The first RDA axis was positively associated with available cover, velocity/depth gradient, bank stability, and conductivity and negatively associated with TP, NH$_4$-N, channel alteration, and SS. The second RDA axis was positively associated with elevation, canopy, and bank stability and negatively associated with riparian zone width, available cover, and SS. RDA axis 1 represented a habitat diversity, cover, and nutrient gradient, whereas RDA axis 2 was more a complex riparian condition gradient.

The RDA biplot showed that dominant fish species scattered apart and were related to different environmental variables (Figure 3). *Opsariichthys pachycephalus* was positively correlated with TP and NH$_4$-N and negatively correlated with available cover, conductivity, and velocity/depth regime. *Rhinogobius candidianus* was positively correlated with SS and riparian width and negatively correlated with elevation and canopy. *Candidia barbata* was positively correlated with elevation and canopy and negatively correlated with SS and riparian width. *Acrossocheilus paradoxus* was positively correlated with available cover, conductivity, and velocity/depth gradient and negatively correlated with TP and NH$_4$-N. The remaining species were near the origins of both axes, indicating that they were relatively unresponsive to environmental gradients in this study.

Group 1 was in the positive direction of riparian width, SS, available cover, and

TABLE 4

Statistical Results of Selected Habitat and Water Quality Variables among Assemblage Groups and the Mean (1 SD) of Assemblage Groups of Each Variable

		Assemblage Groups[a]			
Variables	ANOVA Statistics	1	2	3	4
Elevation (m)	$F = 0.8$; df = 3,14; $P = .53$	80.5 (18.8)	103.9 (11.7)	112.0 (22.9)	77.0 (44.0)
Canopy (%)	$F = 2.6$; df =3,14; $P = .10$	10.94 (10.19)	17.93 (8.45)	35.50 (7.77)	9.88 (0.88)
Stream order	$F = 1.1$; df = 3,14; $P = .37$	3.5 (0.3)	2.9 (0.3)	2.2 (0.7)	3.5 (1.5)
Water temp. (°C)	$F = 0.51$; df = 3,14; $P = .68$	21.20 (2.95)	21.36 (1.06)	20.20 (1.39)	20.65 (1.34)
BOD5 (mg/liter)	$F = 0.48$; df = 3,14; $P = .70$	7.45 (9.46)	3.43 (2.94)	6.28 (3.29)	6.30 (8.06)
Conductivity (µS/cm)	$F = 0.70$; df = 3,14; $P = .57$	403.88 (100.19)	588.64 (119.21)	481.70 (75.63)	431.50 (17.90)
SS (mg/liter)	$F = 1.9$; df = 3,14; $P = .17$	8.75 (4.68)	4.14 (1.26)	0.80 (0.37)	5.00 (3.00)
NH_4-N (mg/liter)	$F = 3.4$; df = 3,14; $P = .048$	0.21 (0.08)B	0.12 (0.03)B	0.11 (0.01)B	0.66 (0.15)A
TP (mg/liter)	$F = 35.2$; df = 3,14; $P < .001$	0.06 (0.04)B	0.08 (0.03)B	0.04 (0.01)B	0.76 (0.19)A
Available cover	$F = 1.62$; df = 3,14; $P = .23$	15.8 (1.3)	15.7 (1.8)	14.6 (1.9)	13.0 (1.4)
Velocity/Depth regime	$F = 0.66$; df = 3,14; $P = .59$	12.8 (1.3)	13.1 (0.7)	13.6 (0.5)	11.5 (2.1)
Channel alteration	$F = 6.4$; df = 3,14; $P = .006$	10.5 (2.3)A	9.1 (1.8)B	8.4 (2.0)B	15.0 (0)A
Bank stability	$F = 6.8$; df = 3,14; $P = .005$	13.0 (1.5)A	13.4 (1.0)A	15.2 (0.9)A	10.0 (0)B
Riparian width	$F = 8.2$; df = 3,14; $P = .002$	12.3 (1.8)A	11.0 (1.4)A	4.6 (0.9)B	12.0 (1.0)A

Note: Variables not listed were not different among assemblage groups.

[a] A and B after values indicate groupings of Student-Newman-Keuls post hoc tests.

TABLE 5

Variance Explained by and Scores of Environmental Variables of RDA Axes

Parameters	RDA Axis 1	RDA Axis 2
Eigenvalues	337.5	302.8
% Variance of species	25.54%	22.91%
Environmental variables		
Elevation	0.05	0.18
Canopy	–0.02	0.19
Conductivity	0.15	–0.03
Water temp.	–0.05	–0.09
TP	–0.36	–0.05
BOD	–0.03	–0.01
NH_4-N	–0.17	–0.07
SS	–0.12	–0.29
Available cover	0.28	–0.18
Bank stability	0.15	0.13
Channel alteration	–0.15	–0.11
Riparian width	–0.05	–0.34
Velocity/depth regime	0.16	0.04

channel alteration and in the negative direction of elevation and canopy (Figure 3), and group 3 was in the opposite direction of group 1. Group 4 was in the positive direction of TP and NH_4-N and in the negative direction of available cover, velocity/depth gradient, bank stability, and conductivity. Group 2 spanned across RDA axis 1.

Relationship between Fish Guilds and Environmental Gradients

The fish guilds defined by trophic and tolerance groups were correlated with environmental gradients (RDA axes). RDA axis 1 was positively correlated with percentage herbivores ($r = 0.78$, $P < .001$) and negatively correlated with percentage insectivores ($r = –0.68$, $P = .001$) (Figure 4). RDA axis 2 was positively correlated with percentage intolerance ($r = 0.87$, $P < 0.001$) and negatively correlated with percentage tolerant and percentage piscivore fish ($r = –0.56$, $P = .01$; $r = –0.74$, $P < .001$, respectively).

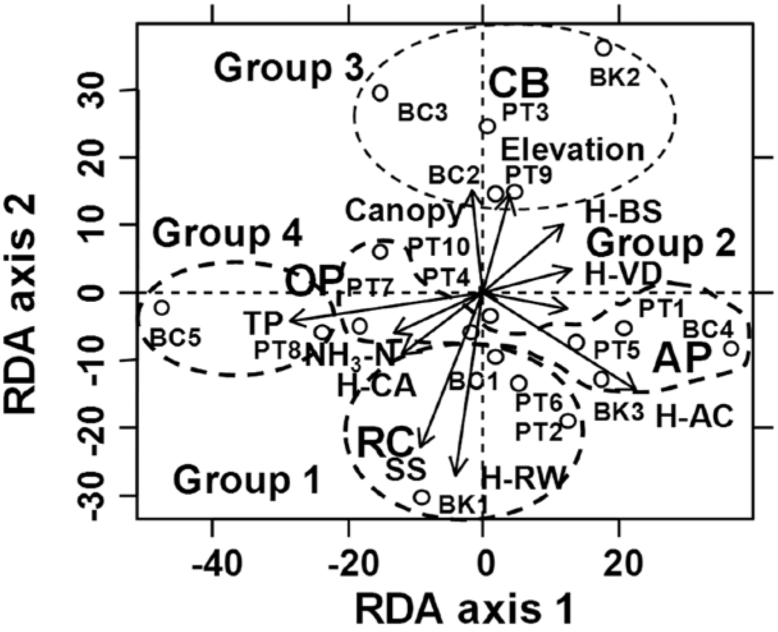

FIGURE 3. Triplot of results of RDA of fish species, sites, assemblage groups, and environmental variables. Abbreviations of environmental variables: H-AC, available cover; H-BS, bank stability; H-CA, channel alteration; H-RW, riparian zone width; H-VD, velocity/depth regimes; Cond, conductivity.

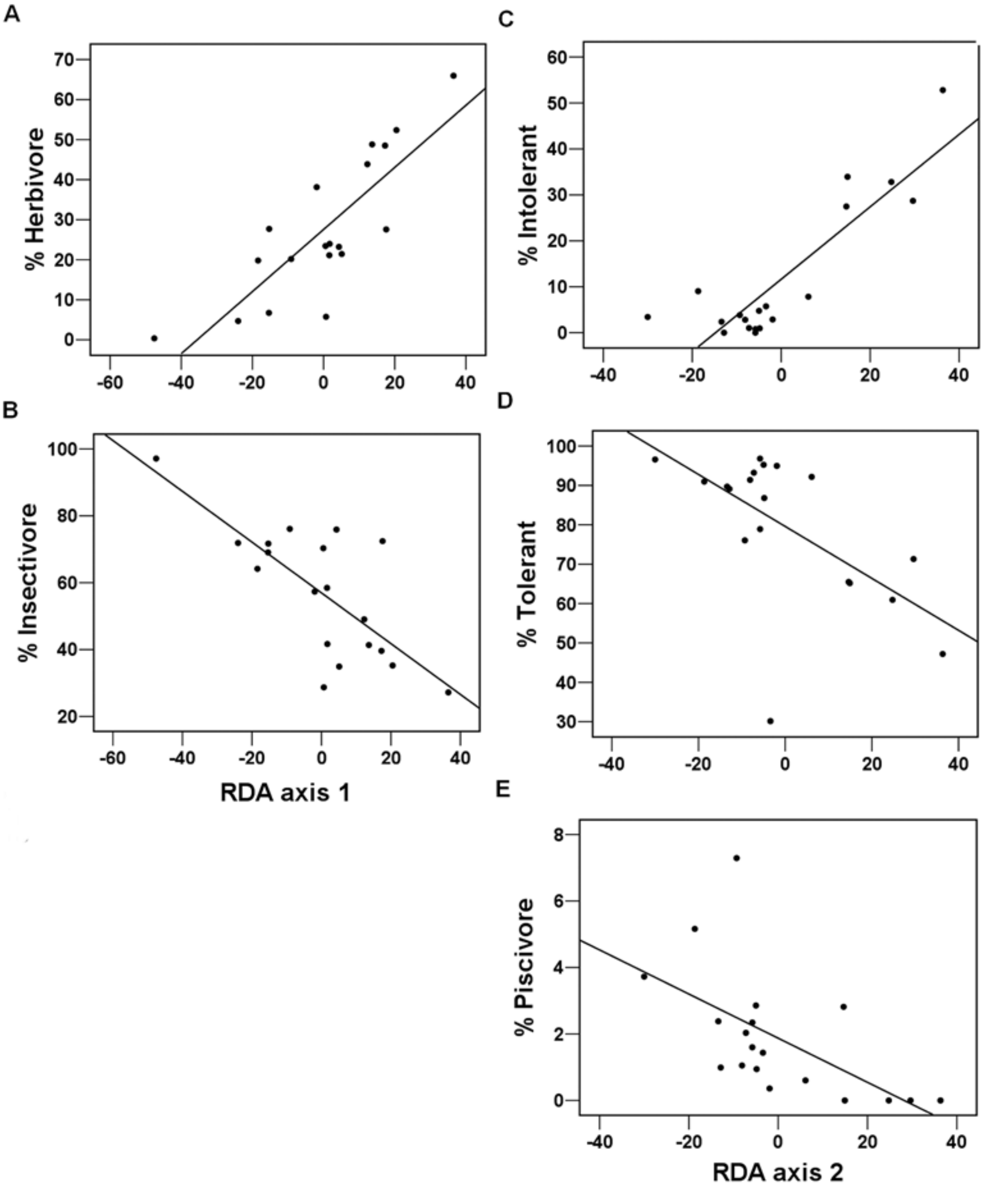

FIGURE 4. Plots of fish guilds with RDA axes and trend lines. *A*, Percentage herbivores; *B*, percentage insectivores; *C*, percentage intolerant; *D*, percentage tolerant; *E*, percentage piscivores. *A–E* were significantly correlated with respective RDA axes.

Elevation Gradient

Despite reduced elevation gradient, the number of species was still negatively correlated with elevation ($r = -0.65$, $P = .003$) (Figure 5) but was not correlated with the RDA axes or the stream order. The number of native species was also correlated with elevation ($r = -0.71$, $P = .001$). Percentage of nonnative species increased with decreasing elevation

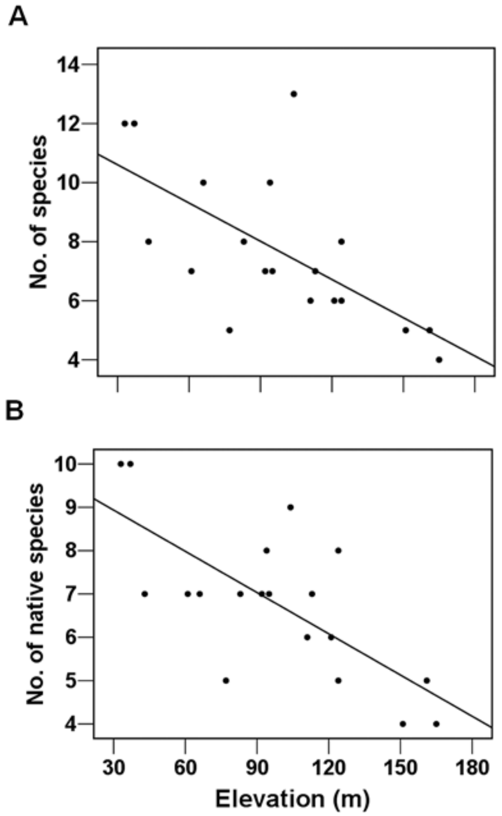

Figure 5. Plots between species richness and elevations and trend lines showing numbers of species that were significantly decreased with increasing elevation. *A*, Total number of species; *B*, number of native species.

($r = -0.58$, $P = .01$), whereas the number of nonnative species was not correlated with elevation. Further analysis showed that the number of species with occurrence frequency <50% was negatively correlated with elevation ($r = -0.51$, $P = .02$), indicating that low-occurrence species increased with decreasing elevation.

DISCUSSION

Assemblages-Environment Relationships

Fish assemblages and water and habitat variables were related as shown by redundancy analysis (RDA). Nutrients, SS, riparian conditions, available cover, and habitat diversity were the major factors affecting fish assemblages. Many sites had NO_3-N > 1 ppm and TP > 50 ppb, indicating widespread nutrient enrichment. Concrete stream banks and grade-control dams were widespread. As in other regions around the world, these water and habitat degradations are commonly observed in agricultural streams (Johnson et al. 1997, Zimmerman et al. 2003, Boody et al. 2005, Chambers et al. 2006, Zaimes et al. 2008). Under intensive agricultural development, native fish assemblages survived under degraded conditions. Dominant species were widely distributed and shared by assemblage groups in this region. However, their relative abundances changed with different environmental conditions as revealed by RDA.

Assemblage groups and related dominant species were related to environmental conditions in this study. Fish assemblage groups were often found to be related to environmental variables in other regions (Brown 2000, Quist et al. 2004, Orrego et al. 2009). Although site assemblages had high similarity and dominant species were widespread, assemblage groups based on assemblages were significantly different as revealed by ANOSIM. SIMPER further indicated that three dominant species contributed to dissimilarity among sites. These results support that dominant fish changed in relative abundance with respect to environmental conditions.

Species-Environment Relationships

The relationships between fish species distributions and environmental conditions in this study provided autecological information that is needed for environmental management (Suen and Herricks 2006). Few studies have documented the relationships between fish distributions, water quality, and habitats in Taiwan (Wang et al. 1996, Chen 2004). For this reason, we could not always find results comparable to ours in the literature.

We classified dominant fish species into groups according to their environmental preference in RDA results. *Candidia barbata* belongs to the higher canopy cover group, whereas *A. paradoxus* and *R. candidianus* belong to the open-canopy group. *Acrossocheilus paradoxus* prefers diverse habitat and high available cover, whereas *O. pachycephalus* has wide distribution and is a tolerant species. In agreement with our results, *C. barbata* has been reported to prefer slow water and canopy cover (Yen 1993, Chuang et al. 2006). *Acrossocheilus paradoxus* was reported to prefer open canopy (Con and Day 2006), but its relationship with habitat diversity was not studied. Also in agreement with our results, Wang et al. (1996) and Chen (2004) reported that *O. pachycephalus* occurred in lower reaches and was more tolerant of pollution.

The correlation between fish guilds and environmental gradients can be explained by the species that occur in the fish guilds. The positive correlation between percentage herbivore and RDA axis 1 reflected the trend of relative abundances of *A. paradoxus* along RDA axis 1 because *A. paradoxus* was the primary herbivore species. The negative correlation between percentage insectivores and RDA axis 1 reflected the trend of relative abundances of *O. pachycephalus* and *R. candidianus* because they were the primary insectivore species. The positive correlation of percentage intolerance with RDA axis 2 reflected that *C. barbata* increased with RDA axis 2. The negative correlation of percentage piscivores with RDA axis 2 was because piscivore/insectivore catfish species were more abundant in lower-elevation sites.

Elevation Pattern of Number of Species

Despite the short elevation gradient, the numbers of native fish species and of all fish species were both negatively related to elevation in this study. Many previous studies have also reported that the number of fish species increased with decreasing elevation (Yeh et al. 2000, Quist et al. 2004, Robinson and Rand 2005, Ibañez et al. 2007, Kang et al. 2009, Li et al. 2012). Lower-elevation sites may provide diverse habitats, less environmental fluctuation, and more resources for more species (Schlosser 1982, Rahel and Hubert 1991). Thus, lower-elevation sites harbored more species than higher-elevation sites.

The number of fish species may increase as an effect of species addition with decreasing elevation in this study. The number of species with occurrence frequency <50% was significantly correlated with the total number of species at each site, and dominant species occurred in most sites. This result may indicate that low-occurrence species were added to sites along the gradient of decreasing elevation. These added species included both native and nonnative species. Our sites were located in the hill and upper plain regions and showed a gradual change in environmental conditions, which is where species addition has been found (Jackson et al. 2001). Hence, species addition, not substitution, may be the reason for the increase in the number of species with decreasing elevation in this study.

Conservation Implications

Fish bioassessment may be feasible in this region. Developing stream bioassessment criteria in this region is important because it will allow us to assess stream conditions and develop management strategies to protect these streams. The relationship between fish assemblages and environmental degradation was revealed by RDA results. In addition, the first two RDA axes were not strongly correlated with natural variation, especially elevation. They can be used as environmental gradients for screening fish bioassessment metrics (Whittier et al. 2007). Furthermore, the correlations of the trophic and tolerance guilds of fish assemblages with environmental gradients imply that the application of rapid bioassessment protocols may be promising (Barbour et al. 1999). However, the fish bioassessment approach needs to be further developed and tested.

Nonnative fish species have spread to almost 50% of the study sites and pose a major threat to native fish. Percentage of nonnative species increased with decreasing elevation. Most sites with nonnative species were below 100 m in elevation, but three sites over 100 m elevation had nonnative species. Introduction by humans is the major source of nonnative fish (Chen et al. 2003). These nonnative species are often tolerant of poor water quality and utilize sites with habitat destruction (Marchetti et al. 2004, Brasher et al. 2006). Therefore, fish communities in lower elevations are often dominated by nonnative fish. This distribution pattern of nonnative species is similar to the pattern observed in Hawai‘i, with greater numbers of introduced species in low-elevation developed sites (Brasher et al. 2006). Several nonnative predators, such as *Channa striata* and *Oreochromis* sp., pose major threats to native fish. *Oreochromis* sp. is the most widespread nonnative fish in Taiwan streams (Chen et al. 2003), and *Channa striata* is a fierce predator. With degrading environmental conditions, these nonnative fish may expand their distribution and increase the threats that they pose to native fish.

Based on results of this study, erosion, nutrients, riparian removal, and in-stream habitat destruction may be the major problems in these agricultural streams in addition to nonnative species invasion. The adoption of management practices for these problems may yield easement of current impacts on stream ecosystems. As revealed by several studies, management practices often improve environmental conditions when the problems are correctly identified (Monaghan et al. 2007, Yates et al. 2007, Gabel et al. 2012).

This study clearly showed that both water quality and habitat variables were related to fish assemblages in this agricultural region. For fish survey programs of water resources and environmental protection agencies in Taiwan, habitat quality should be incorporated to better understand river conditions and their consequent impacts on fish assemblages. Future studies may survey more detailed habitat characteristics to fully assess fish and habitat relationships. Expanding the study region by including different stream basins may yield a broader picture of the relationship between fish assemblages and water and habitat variables in Taiwan.

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