Ecological Indicators 3 (2003) 93–117

ECOLOGICAL INDICATORS

This article is also available online at:
www.elsevier.com/locate/ecolind

Natural and anthropogenic factors affecting the structure of the benthic macroinvertebrate community in an effluent-dominated reach of the Santa Cruz River, AZ

Terence P. Boyle [a,*], Harold D. Fraleigh Jr. [b]

[a] *US Geological Survey, Fort Collins Science Center, Colorado State University, 335 Aylesworth NW, Fort Collins, CO 80523-9143, USA*
[b] *Natural Resources Ecology Laboratory, Colorado State University, Fort Collins, CO 80523-1499, USA*

Abstract

This study provides an assessment of the ecological conditions of a 46-km effluent-dominated stream section of the Santa Cruz River in the vicinity of the International Waste Water Treatment Plant, Nogales, AZ. We associated changes in the structure of the macroinvertebrate community to natural and anthropogenic chemical and physical variables using multivariate analysis. The analysis shows that biological criteria for effluent-dominated streams can be established using macroinvertebrate community attributes only with an understanding of the contribution of three classes of variables on the community structure: (1) low flow hydrological discharge as affected by groundwater withdrawals, treatment plant discharge, and subsurface geomorphology; (2) chemical composition of the treatment plant discharge and natural dilution; and (3) naturally produced floods resulting from seasonality of precipitation.
© 2003 Elsevier Science Ltd. All rights reserved.

Keywords: Effluent-dominated stream; Santa Cruz River, Arizona, USA; Benthic macroinvertebrate; Multivariate analysis

1. Introduction

Many streams in the arid southwestern US receive a major portion of their base flow from the discharge of industrial or municipal treatment plants (Schmidt, 1993). The flows generated from these sources range from partial to total contribution of stream discharge depending on the season of the year. Streams with this type of hydrology have come to be known as effluent-dominated streams (Schmidt, 1993). While the general concept of an effluent-dominated stream is known, the factors affecting critical ecological components are poorly understood. Effluent-dominated streams present a potential paradox to water quality regulators in that the biological communities they contain are supported, in part or in total, by hydrological discharge from non-natural sources. In spite of the diverse sources of discharge, these streams support aquatic communities whose attributes can be used to establish biological criteria to classify the water body as healthy or impaired.

Section 303d of the Clean Water Act (USEPA, 1994) requires each state to classify streams as impaired or unimpaired based on instream biological criteria. The most common biological criterion used is the status of the benthic macroinvertebrate community. The objective of this paper is to establish critical physical, chemical, and biological parameters associated with the structure of the benthic macroinvertebrate community at four seasons of the year in a southwestern US effluent-dominated stream.

* Corresponding author. Fax: +1-970-491-1511.
E-mail address: tpboyle@cnr.colostate.edu (T.P. Boyle).

1470-160X/03/$ – see front matter © 2003 Elsevier Science Ltd. All rights reserved.
doi:10.1016/S1470-160X(03)00014-1

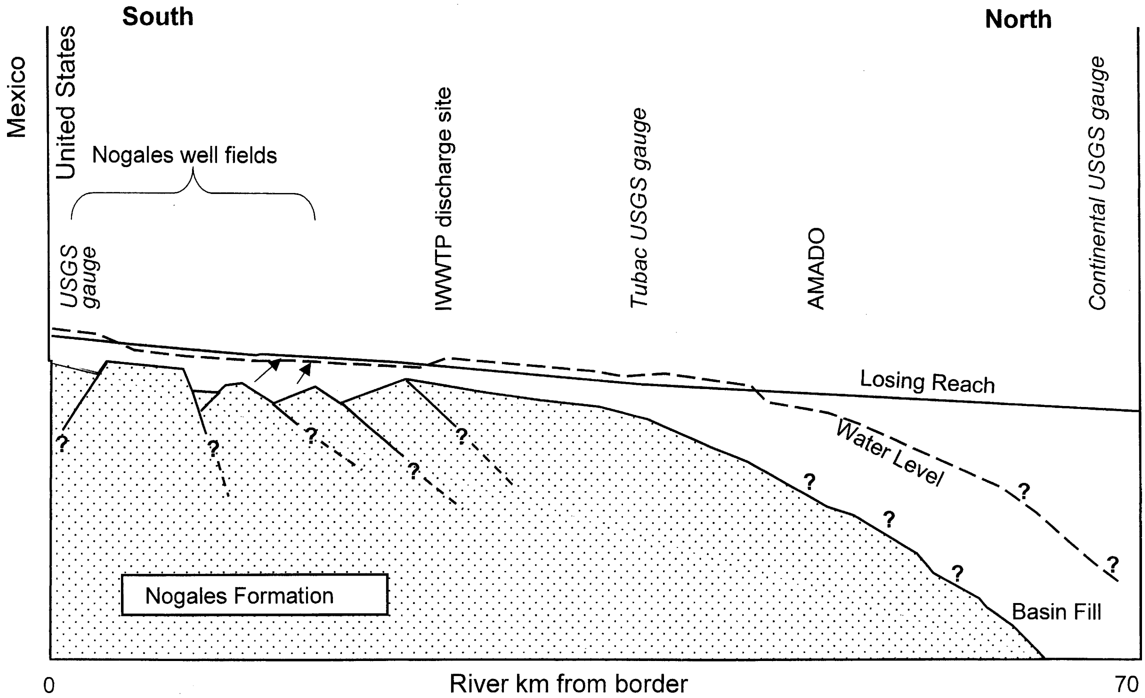

Fig. 1. Generalized long section geomorphic map of the upper Santa Cruz River.

1.1. Santa Cruz River

The Santa Cruz River rises in the Canelo Hills of southern Arizona, flows south into Mexico, then turns north, and reenters the United States east of the cities of Nogales. The Santa Cruz Basin in Mexico is subject to both water withdrawal from well-fields and trans-basin additions. From the Mexico border north there are well-fields along the river that supply the city of Nogales, Arizona. Except during extreme weather events, the upper Santa Cruz River in Arizona has almost no surface flow beyond the water well fields until it reaches the Nogales International Waste Water Treatment Plant (IWWTP), a major input into the Santa Cruz River. The IWWTP is currently the source of a tail-water that reaches to Amado approximately 30-km river downstream from its outfall. The river rarely flows past Amado because the bedrock, which is near the surface upstream of that point, rapidly descends and the valley becomes filled with porous alluvium that only briefly sustains surface flow during periods of intense rainfall (Fig. 1). The upper Santa Cruz River in Arizona is an effluent-dominated stream. Except for storm flow events, the IWWTP outflow has been the major contributor to surface discharge of the river since 1972 (Tellman et al., 1997). This portion of the Santa Cruz River falls within the Southern Deserts Ecoregion (Omernik, 1987).

2. Materials and methods

2.1. Sample sections

Based upon maps and visits to the field, nine sites were selected for water quality sampling and nine sites were selected for benthic macroinvertebrates collection (Table 1; Fig. 2). Given the proximity of Sites 5, 5A, and 5B, the water quality data were the same for all three sites. Effluent discharge measurements provided by the IWWTP were also utilized. The individual sites were chosen to represent a series of sites where water occurred in the river between the US–Mexico border and the river disappeared into the

Table 1
Sample Sites, numeric reference codes, distance from US–Mexico border, and variables collected

(1) USGS gauging station at the US–Mexican border (1.5 km)[a,b,c]
(2) Nogales Wash near the IWWTP (24.7 km)[a,b,c]
(3) Discharge weir at IWWTP (25 km)[d]
(4) Railroad bridge downstream from the IWWTP (25.2 km)[a,b]
(5) In the vicinity of Rio Rico Bridge (26.6 km)
Sites 5[a,b,c], 5A[c,e], and 5B[c,e] proceed downriver within the sample site
(6) North Rio Rico, upstream of Rancho Santa Cruz (37 km)[a,b,c]
(7) Santa Gertrudis Lane (38.4 km)[a,b]
(8) Carmen (41.6 km)[a,b,c]
(9) NAQWA station at Tubac (44.9 km)[a,b,c]
(10) Chavez Siding (47.4 km)[a,b,c]

[a] Hydrological discharge measured.
[b] Temperature, pH, alkalinity, ammonium/ammonia, dissolved oxygen concentration measured.
[c] Benthic macroinvertebrates collected.
[d] Effluent discharge measured.
[e] Physical and chemical data from Sites 5, 5A, and 5B were the same.

alluvium. Accessibility to the river was also factored into the selection process. Sites were sampled on four dates during an annual climate cycle: May, during dry spring; August, during summer monsoons; December, during dry fall; and March, during winter rainy season (Lowe, 1964).

Physical, chemical, and biological measurements and collections made at the sites and standard methodological references are shown in Tables 1 and 2. Temperature and all water samples were collected within 2 h after sunrise on a single day for each sampling period.

Benthic macroinvertebrates were sampled at each site using 10 Surber samples (256 μm mesh) in the various substrate types present (e.g. sand, silt, gravel, cobble) that occurred within the section (Hauer and Resh, 1996). Macroinvertebrate samples were preserved in the field with formalin and later separated from debris in the laboratory. The samples from each site were

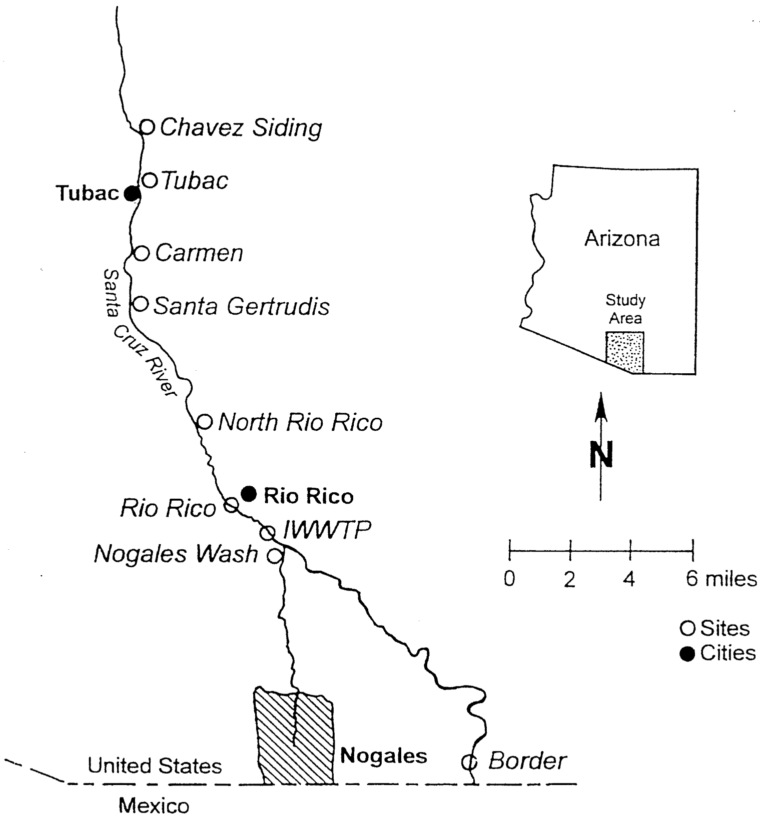

Fig. 2. Sampling site map of the upper Santa Cruz River.

Table 2
Variables collected and references

Variable	Reference
Physical	
Hydrological discharge	USDI (1984)
Temperature	APHA (1980)
Chemical	
pH	APHA (1980)
Alkalinity	APHA (1980)
Ammonium	APHA (1980)
Dissolved oxygen concentration	APHA (1980)
Biological	
Benthic macroinvertebrates	Hauer and Resh (1996)

composited, identified to the lowest possible taxon and counted. A reference collection of the taxa identified and listed at the end of this paper were deposited into the collection of the C.P. Gillette Museum of Arthropod Diversity at Colorado State University.

2.2. Data analysis

We used canonical correlation analysis (CCA) for the integrated analysis of two data sets: (1) benthic macroinvertebrate community structure (species and abundances) and (2) a set of physical and chemical variables, both naturally occurring and anthropogenic. CCA separates the benthic macroinvertebrate communities found at the various sample sites for individual sample dates on the basis of their community structure in two-dimensional ordination, and then tests for significance of association between physical and chemical variables with the variation in community structure (ter Braak and Verdonschot, 1995). Statistically significant gradients of individual environmental variables within the biological community data were shown as vectors with the dimensions of the Pearson product moment correlations on the two axes developed by the correspondence analysis. This allows interpretation of an array of environmental factors associated with variation in the structure of the benthic macroinvertebrate community not possible with univariate statistical analysis.

Selected individual metrics and community attributes were also calculated on the benthic macroinvertebrate data. These individual attributes and parameters of the benthic macroinvertebrate community further interpret the taxonomic and ecological response to changes in the physical and chemical variables in the environment (Barbour et al., 1999; Wallace et al., 1996; Kerans and Karr, 1994; Boyle et al., 1990; Washington, 1984). Specifically, total number of invertebrates, total number of taxa, Shannon–Weiner Diversity Index, Simpson's Diversity Index, number of Chironomidae taxa, number of Oligochaeta taxa, number of Ephemeroptera taxa, and numbers of the most numerous taxa were analyzed.

3. Results

3.1. Discharge

3.1.1. At sample sites

Discharge measurements taken at seven sample sites for three dates (August, December 1997, March 1998) reflected the differences among sites, as well as the seasonal variability (Fig. 3). The March discharge was highest at all sites showing the typical pattern of a characteristic El Niño year. In fact, this was the only sample date where any discharge was observed at the Border site above the IWWTP. The discharge measured in August and December appeared highest at the site closest to the IWWTP (i.e. Rio Rico Bridge) due to freshets entering the main river from thundershowers over local tributaries at the upper sample sites in the vicinity of Rio Rico, which had not reached the lower sites during the time of discharge measurements. The spatial variation in discharge is important in understanding some of the short-term variation in chemical concentrations in the river. Where freshets enter the Santa Cruz from arroyos, they may alter the concentrations of chemical parameters in the main river.

3.1.2. At USGS gauges

The differences of the daily discharge at the three USGS river gauges maintained on the upper Santa Cruz River reflect the effects of the IWWTP discharge and are shown in Fig. 4. The Border site showed brief discharge only during the summer monsoons (August) and winter rainy season (March). Tubac had a base flow present all year and showed the same general pattern of freshets due to seasonal precipitation as at the Border gauge. The extreme events were of comparable magnitude, however, they were more frequent. The discharge at the Continental gauge showed no base

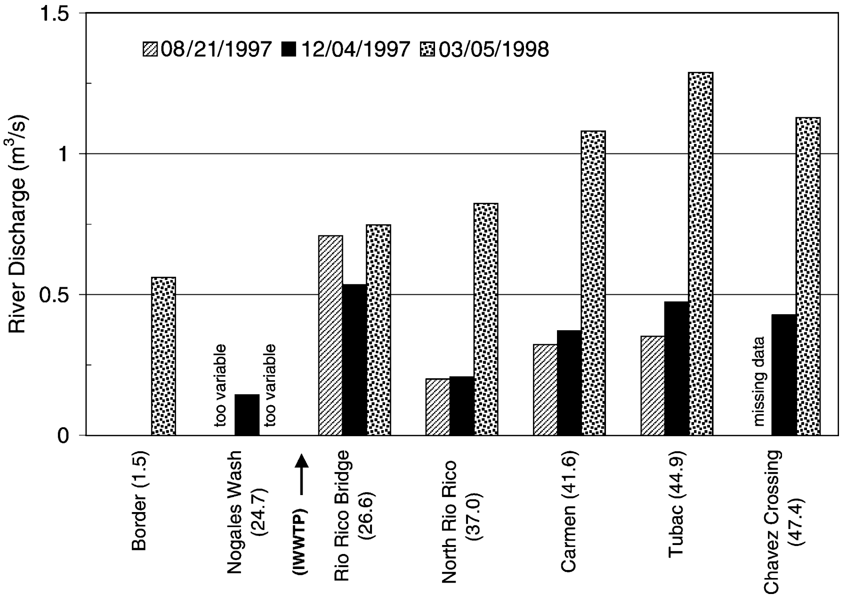

Fig. 3. River discharge measured at seven sampling sites on three dates. Numbers on *x*-axis refer to distance downriver from the US–Mexico border as illustrated in Fig. 2 and indicated in Table 1.

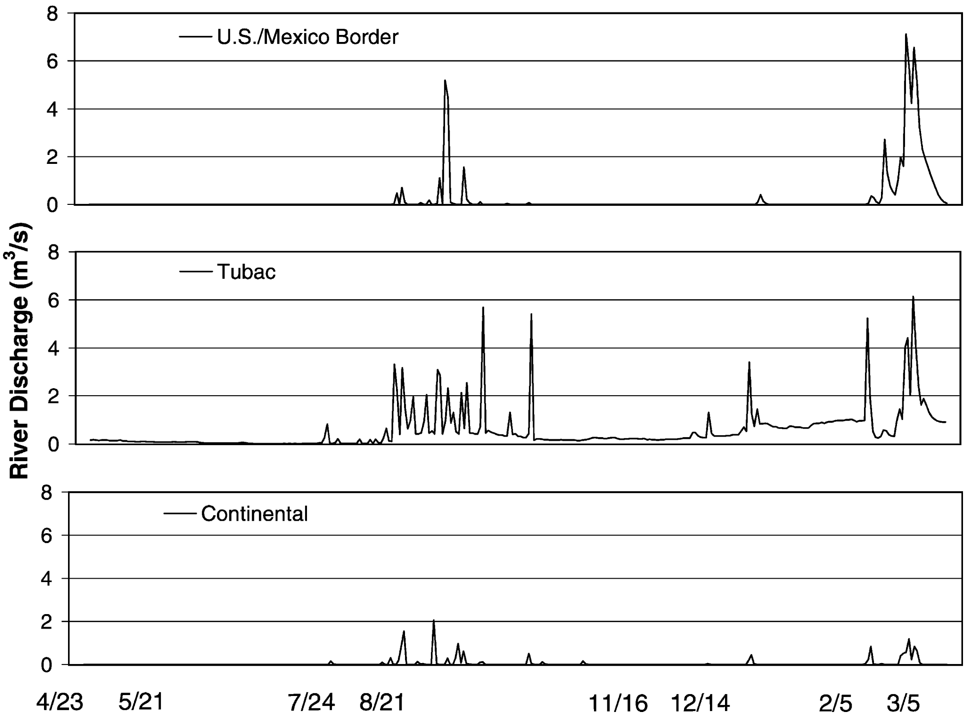

Fig. 4. Santa Cruz River stream flow data at three USGS gauges from 12/04/1997 to 03/05/1998.

flow, and a set of intermittent diminished peak discharges relative to the two stations upstream during the summer monsoons and the March rains.

The large fluctuations in discharge measured at the three USGS gauges are implicated in changes in physical, chemical, and biological qualities in the river (Fig. 4). In April and May 1997, there was no discharge at the Border gauge, low and decreasing flow at the Tubac gauge, and no flow recorded at Continental. In August 1997, there were two small freshets recorded at the Border site prior to the sample time, but it was dry at the time of visit. Rooted submergent vegetation present in the river in May was almost completely scoured and swept away by these freshets at all sites. At Tubac during late July and early August, prior to sampling, there were several substantial flows recorded more than an order of magnitude above the previous base flows. The gauge at Continental recorded three freshets in this time period. In mid-November to mid-December, no flows were recorded by the gauges at the Border or at Continental. However, the gauge at Tubac showed a constant low flow with one small freshet just prior to the sample visit. Finally, the March 1998 discharge record showed flow through the month prior to sampling at the Border site and Tubac with several substantial freshets 2 weeks prior to the sample time. These peak flows were repeated downstream at the Continental gauge but appeared greatly diminished.

3.1.3. At IWWTP

The relation to discharge from the IWWTP to discharge measured at the USGS Tubac gauge 21.2 km downstream can be seen in Fig. 5. Throughout spring and early summer the flow was reduced at Tubac due to ground water withdrawal and evapo-transpiration from the substantial riparian cottonwood–willow gallery forest. In July, August, and September the flows at Tubac exceeded the discharge from the IWWTP due to runoff from the summer monsoonal precipitation. During the autumn dry period the flow at Tubac again was less than the discharge from the IWWTP but greater than in the spring dry period. The winter rains brought the discharge at Tubac to a level exceeding that of the IWWTP outfall. Although this analysis falls short of a real water budget, it does point to the role of the IWWTP in maintaining a certain base flow within the river substantially downstream

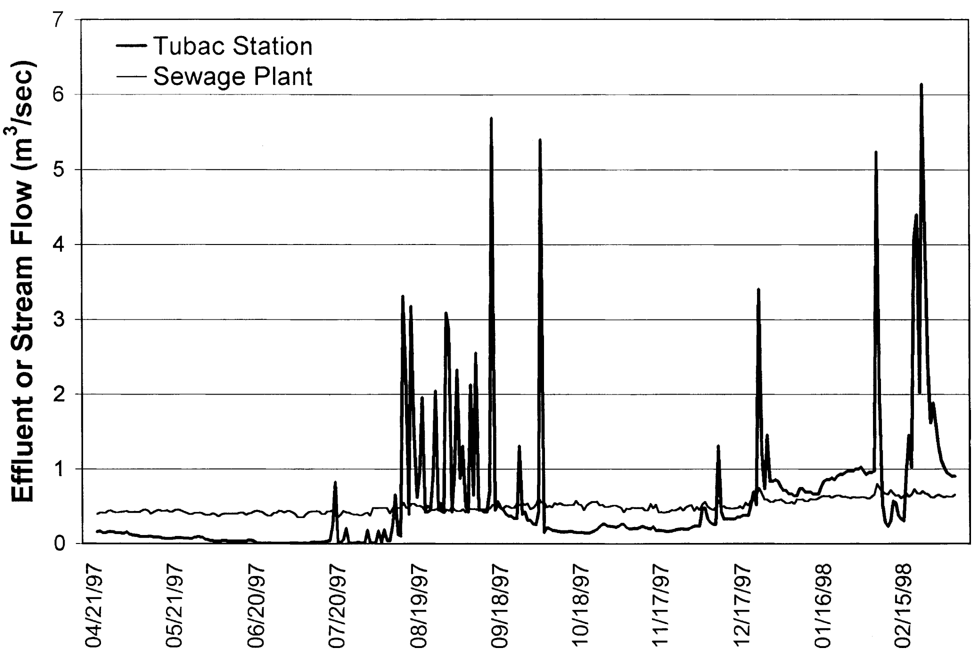

Fig. 5. International Waste Water Treatment Plant effluent and Tubac USGS gauge station stream flow rates, Santa Cruz River, AZ from 12/04/1997 to 03/05/1998.

from its outfall as well as some of the other factors contributing to the variation in discharge in the river.

3.2. Physical and chemical factors

3.2.1. Temperature and oxygen

Dissolved oxygen concentrations were lowest at stations near the discharge of the IWWTP (Fig. 6). Although dissolved oxygen concentrations were recorded as low as 4 mg/l at several stations in the May 1997 samples, these measurements were all taken early in the morning and the levels did not appear to limit aquatic life expected in the river. The water temperature was usually elevated at sites near the IWWTP and decreased with distance downstream, possibly due to equilibration with the air temperature and/or the influence of ground water entering the river. The maximum difference among sites for a given day was 6 °C.

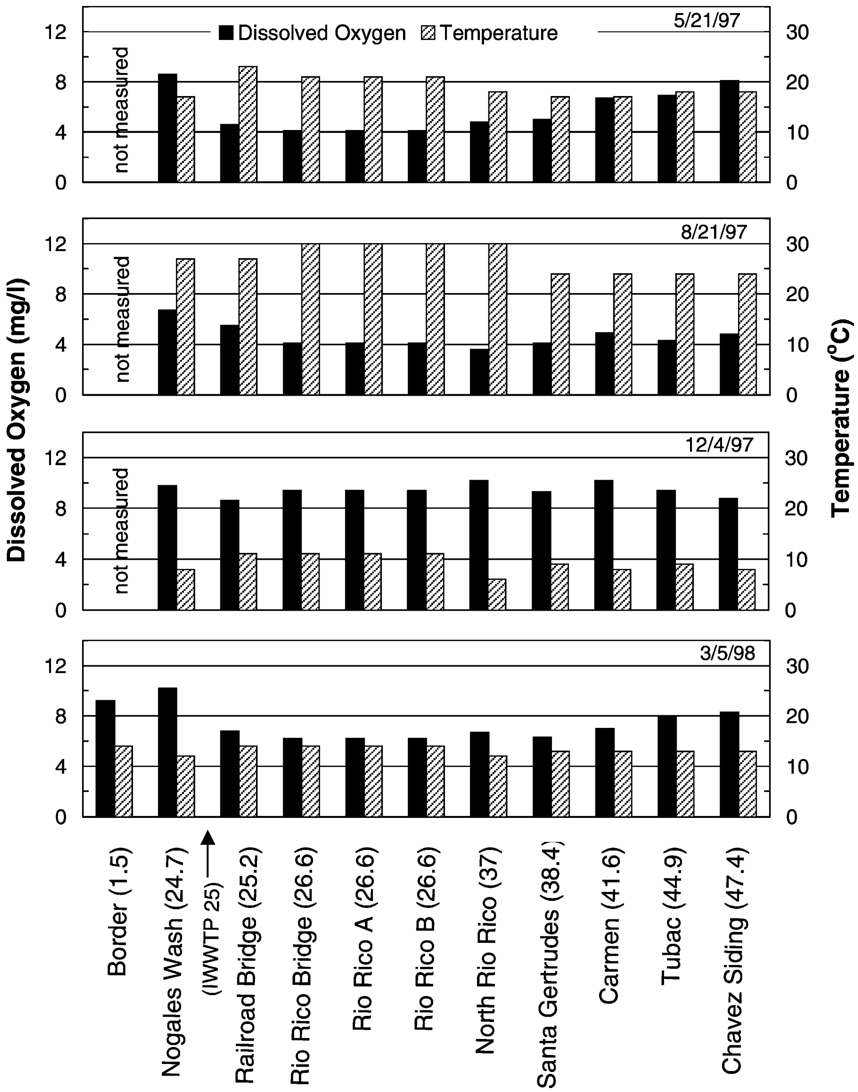

Fig. 6. Dissolved oxygen and temperature on four dates at nine sampling sites along the upper Santa Cruz River. Numbers on *x*-axis refer to distance downriver from the US–Mexico border as illustrated in Fig. 2 and indicated in Table 1.

3.2.2. Nitrogen

In water the ionized form, ammonium, and the un-ionized form, ammonia, are in an equilibrium influenced by both temperature and pH. Ammonia is by far the most toxic of the two forms to aquatic life (USEPA, 1999). The influence of the IWWTP on the concentrations of both forms was evident by comparison with the levels found at the Border, Nogales Wash, Santa Gertrudes, Carmen, Tubac and Chavez Siding sample sites. All were substantially lower than samples taken nearer the IWWTP in the May, August, and March samples (Fig. 7). This pattern was

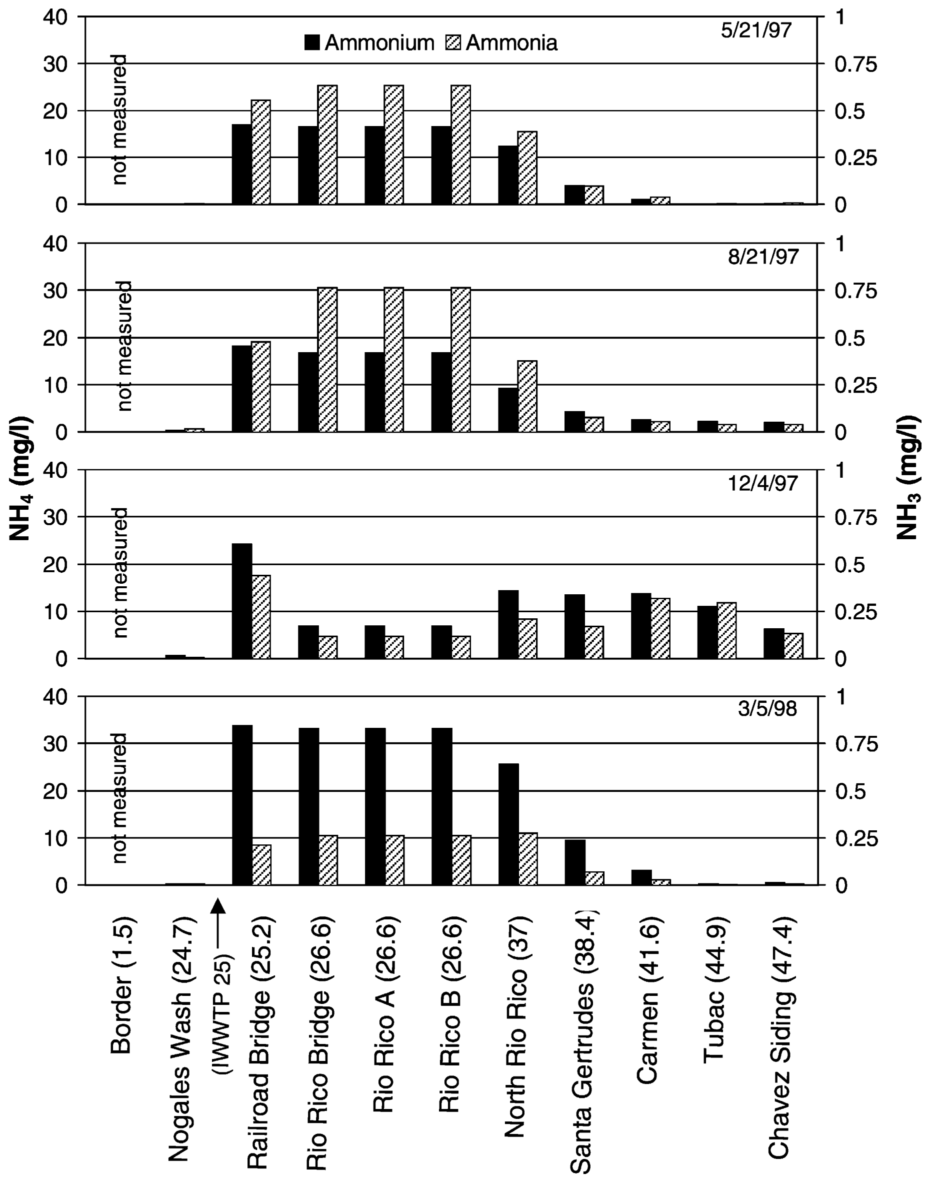

Fig. 7. Ammonium and ammonia on four dates at nine sampling sites along the upper Santa Cruz River. Numbers on *x*-axis refer to distance downriver from the US–Mexico border as illustrated in Fig. 2 and indicated in Table 1.

not apparent in samples taken in December 1997 possibly due to dilution by freshets entering the river from arroyos near Rio Rico, but not yet reaching the downstream sites at the time of sampling. A second pattern was the relatively high concentration of the ammonia form in the May and August samples compared to the December and March samples (Fig. 7). The May and August ammonia levels exceeded those known to be acutely toxic to a number of species of larval fish and aquatic invertebrates listed in the National Toxicity Database AQUIRE maintained by the US Environmental Protection Agency at Duluth (USEPA, 2002). The levels also exceeded the ambient chronic water quality criteria for ammonia on three of the four sampling dates (USEPA, 1999).

3.2.3. Alkalinity and pH

The alkalinity and pH indicated a relatively highly buffered system downstream from the IWWTP that ranged from circumneutral to slightly basic (Fig. 8). However, the alkalinity at the Border site (sample available only in March) and in December at both Nogales Wash and the Rio Rico stations were substantially reduced relative to the other stations.

3.2.4. Macroinvertebrates

A total of 142 taxa were identified from the invertebrate collections (Appendix A). Because of the complexity of analyzing the response of the benthic macroinvertebrate community structure to a number of physical and chemical variables among the various sites, both multivariate and examination of individual qualities or metrics derived from communities were used in tandem.

3.2.5. Multivariate analysis

Canonical correspondence analysis was applied to the community data and associated environmental data as shown in Fig. 9. The axes in the figure have been scaled to emphasize relation of the community data with the environmental variables. The site scores for each sample station for each sample date are displayed as a two-section number (#_#). The first refers to the sample site as previously delineated in Table 1. The second number in the pair is the sample date (1: May 1997; 2: August 1997; 3: December 1997; 4: March 1998). The sample site scores are separated along the two axes that account for the most variability on the basis of community structure information and environmental variables collected at each site. Fig. 9 shows the detail of this separation. The position of the site scores indicates that during May the sites in Nogales Wash (2), Carmen (8), Tubac (9), and Chavez Siding (10) were clearly separated from the other sites (5 and 6) at Rio Rico and closer to the IWWTP. August followed a similar pattern. During December, there was no appreciable separation between any site scores. March 1998 was distinct from the other sample dates in that the position of the site scores, and the structure of the benthic macroinvertebrate community, for all the sample sites were distinctly separated from the site scores of the other dates; however, once again site scores for Sites 5 and 6 were distinctly separated from sites downstream during this sampling period.

Overall, Horizontal Axis 1 clearly separated the sites immediately downstream from the IWWTP and the sites at Nogales Wash (2), Carmen (8), Tubac (9), and Chavez Siding (10). Vertical Axis 2 appeared to further separate Site 2 and one date at Site 10. Vertical Axis 2 also separated Border (1) from all other sites and separated March distinctly from the other sample dates.

The vectors depicted in Fig. 9 are scaled to the coordinates of the Pearson product moment correlations of the various environmental variables calculated individually with the two axes developed by the correspondence analysis. The vectors for those variables determined to be significant in multivariate tests (as shown in Table 3) appear with an asterisk code in Fig. 9. Each vector represents a gradient through the biological data, with the arrow pointing to the area of higher value for the variable, but the gradient extends through the entire biological data set.

The axes represent a composite of the environmental variables included in the analysis. Horizontal Axis

Table 3
Multivariate effects (in order of model selection)

Variable	*P*	*F*
Ammonium (NH_4)	0.005	3.55
pH	0.005	3.16
Temperature	0.005	2.22
Alkalinity	0.005	2.79
Flow variation	0.005	2.26
Ammonia (NH_3)	0.065	1.67

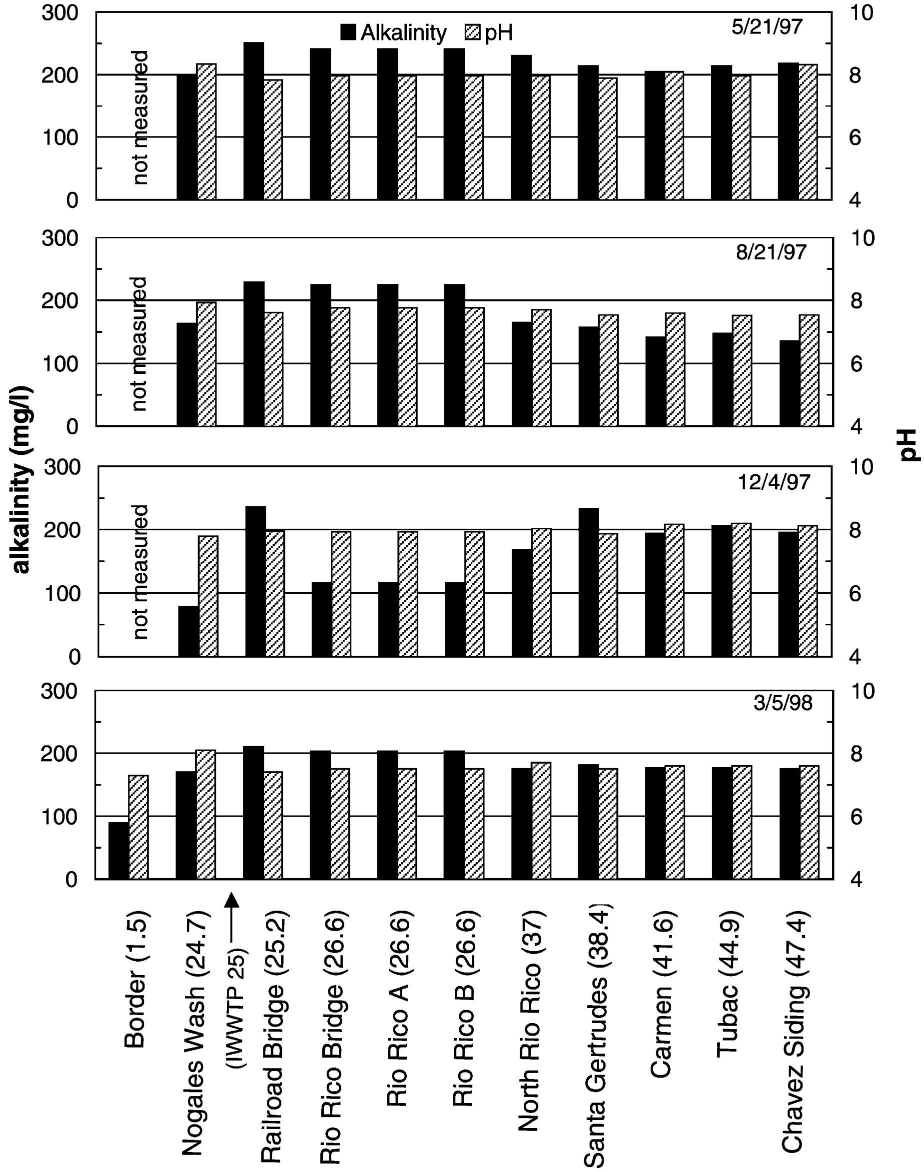

Fig. 8. Alkalinity and pH on four dates at nine sampling sites along the upper Santa Cruz River. Numbers on *x*-axis refer to distance downriver from the US–Mexico border as illustrated in Fig. 2 and indicated in Table 1.

1 was positively correlated with pH and negatively correlated with ammonia and ammonium. The vectors for ammonia and ammonium pointed in the direction of most site scores for Sites 5 and 6 and account for the separation of sites within any given collection date. In general, the greater the distance from (less influence by) the IWWTP, the further right the site scores on the CCA graph. Vertical Axis 2 was positively correlated with the river flow variance (calculated as the coefficient of variation), of the daily discharge at the Tubac USGS gauge for the period of 4 weeks prior to sampling and negatively correlated with pH. Axis 2 was also negatively correlated with ammonia, alkalinity, and temperature. The flow variance and pH

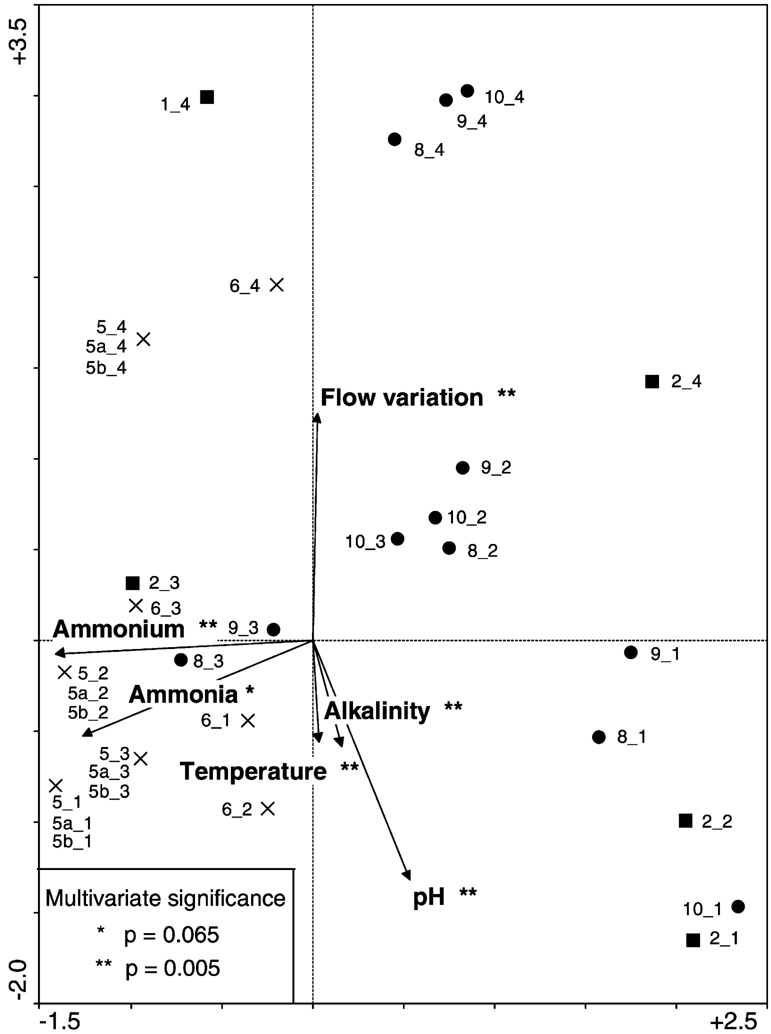

Fig. 9. CCA graph indicating relationships between environmental attributes and seasonal macroinvertibrate communities along the upper Santa Cruz River. The axes in the figure have been scaled to emphasize relation of the community data with the environmental variables. The site scores for each sample station for each sample date are displayed as a two-section number (#_#). The first refers to the sample site as previously delineated in Table 1. The second number in the pair is the sample date (1: May 1997; 2: August 1997; 3: December 1997; 4: March 1998). Vectors represent the Pearson correlation coefficients with the two axes for the environmental variables as labeled. Sites upriver from the IWWTP, near the IWWTP discharge and downriver from IWWTP are plotted as squares, X's, and circles, respectively.

appeared to best explain the separation of the March sample date from other dates. For this analysis 62% of the macroinvertebrate community variability (Table 4) was explained by the environmental variables as represented by the first two axes depicted in Fig. 9. The test of significance for the first axis and overall CCA was highly significant ($P < 0.005$).

3.2.6. *Metric analysis*

Individual parameters of the benthic macroinvertebrate community commonly used as indices include total invertebrate number, taxa richness and two diversity indices: Shannon–Weiner and Simpson's diversity indices. The Shannon–Weiner is more sensitive to changes in dominant species while Simpson's index is

Table 4
Summary of eigen analysis

	Axes				Total inertia
	1	2	3	4	
Eigenvalues	0.368	0.311	0.159	0.108	2.680
Species–environment correlations	0.948	0.963	0.925	0.739	
Cumulative percentage variance					
Species data	13.7	25.3	31.3	35.3	
Species–environment relation	33.8	62.4	77.1	87.0	
Sum of all unconstrained eigenvalues					2.680
Sum of all canonical eigenvalues					1.087

Test of significance of first canonical axis: eigenvalue = 0.368, F-ratio = 4.133, P = 0.0050.
Test of significance of all canonical axes: trace = 1.087, F-ratio = 2.957, P = 0.0050.

more sensitive to changes in rare species (Boyle et al., 1990).

3.2.6.1. Total abundance. The total number of invertebrates found at each site for the May samples appeared to respond to the organic enrichment from the IWWTP in a classic fashion (Fig. 10). The site above the IWWTP had a lower number than the site immediately below the outfall. The numbers below the outfall were reduced further downstream. This was the season of lowest flow and the entire flow of the river was dependent on the discharge from the IWWTP. The

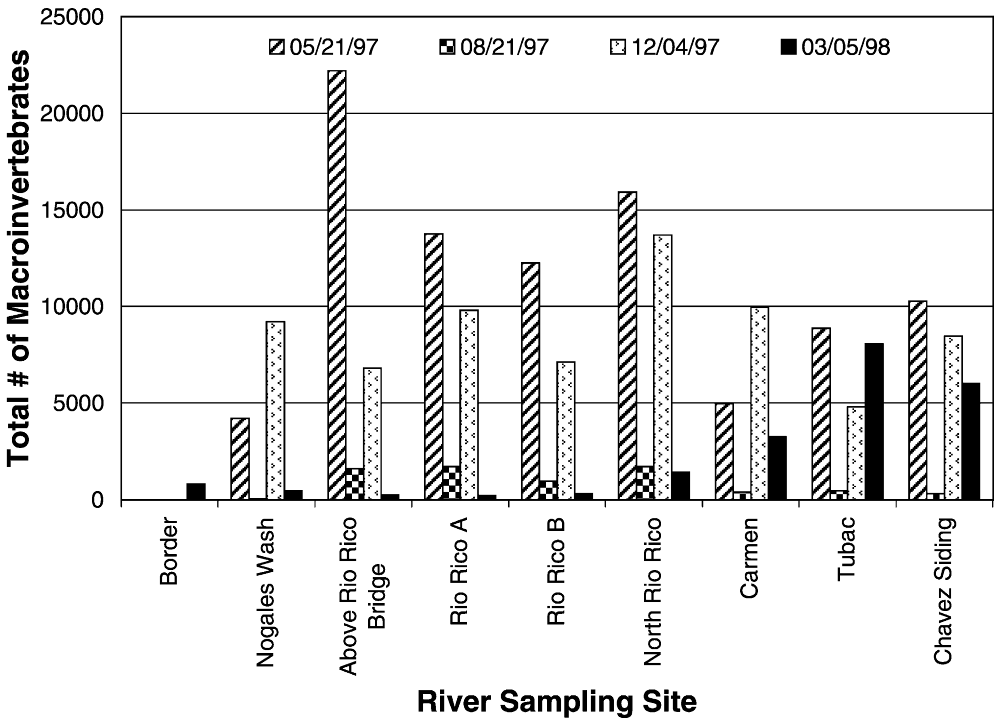

Fig. 10. Total number of macroinvertebrates at nine sampling sites on four dates. Rio Rico Bridge, Rio Rico A, and Rio Rico B were all within the single “Rio Rico Bridge” area sampled for physical and chemical variables shown in Figs. 6–8.

August sample appeared to follow the same pattern; however, the numbers were almost an order of magnitude less that of the previous sample date. The total numbers at the various sites during December did not exhibit any pattern related to the gradient downstream from the IWWTP. The March sample had a reverse relationship compared to the May and August pattern, with the total number increasing with distance downstream from the IWWTP.

3.2.6.2. Taxa richness. The number of taxa present is one measure of biodiversity. High diversity is associated with an unimpacted situation, while a lower biodiversity often signifies environmental stress. In comparing the general pattern of taxonomic abundance at the different sites and sample dates, three trends are apparent: (1) there is a general increase of taxa number at the sites farther below the IWWTP relative to those closer to the discharge within a sample date; (2) at the Border site and two dates at Nogales Wash taxa numbers were much higher than those immediately below the IWWTP; and (3) within a sample site the taxa richness appeared consistently reduced during the August and March sample periods (Fig. 11). The seasonal reduction was in association with high flows and high variability of flows.

3.2.6.3. Diversity indices. The Shannon–Weiner index was usually higher at the downstream stations during May, August, and December (Fig. 12). A similar trend was not apparent in the March samples. This index usually responds only after severe changes in community structure involving gross reduction in density and number and/or replacement of species has occurred (Boyle et al., 1990).

In Simpson's index, a higher value indicates a lower diversity. Simpson's index indicated a similar pattern to Shannon–Weiner (Fig. 13). The minor differences between the responses in the two diversity indices indicated that some taxa were being reduced selectively rather than simple reduction in the relative abundance of all taxa (Boyle et al., 1990). The December-Nogales Wash sample had extremely low diversity as determined by both the Shannon–Weiner and Simpson's

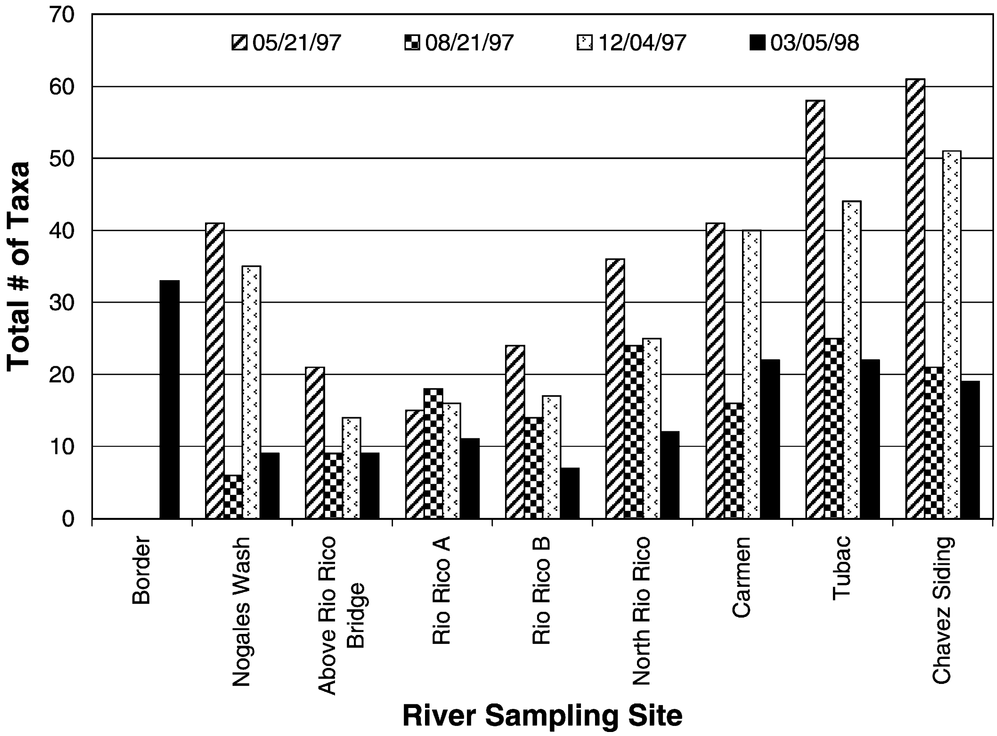

Fig. 11. Total number of macroinvertebrate taxa at nine sampling sites on four dates.

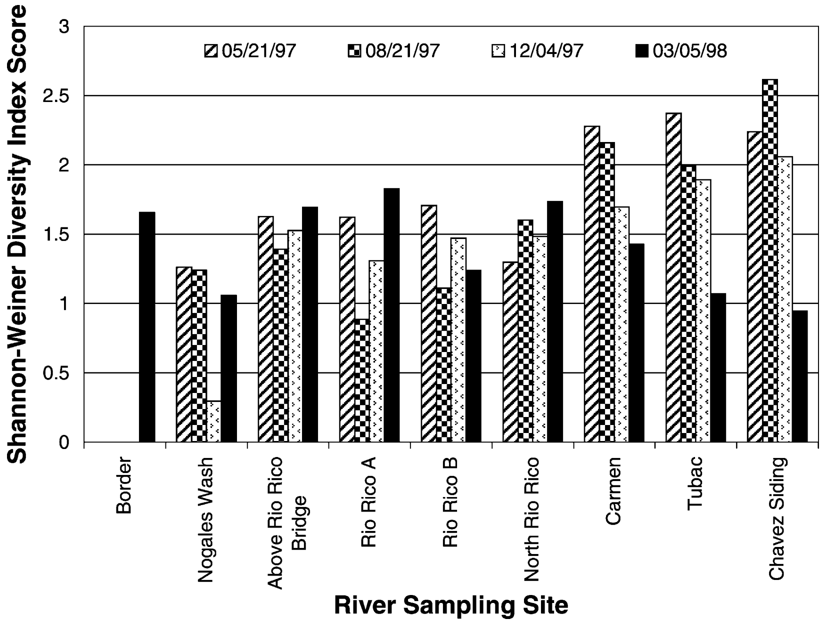

Fig. 12. Shannon–Weiner diversity index scores at nine sampling sites on four dates.

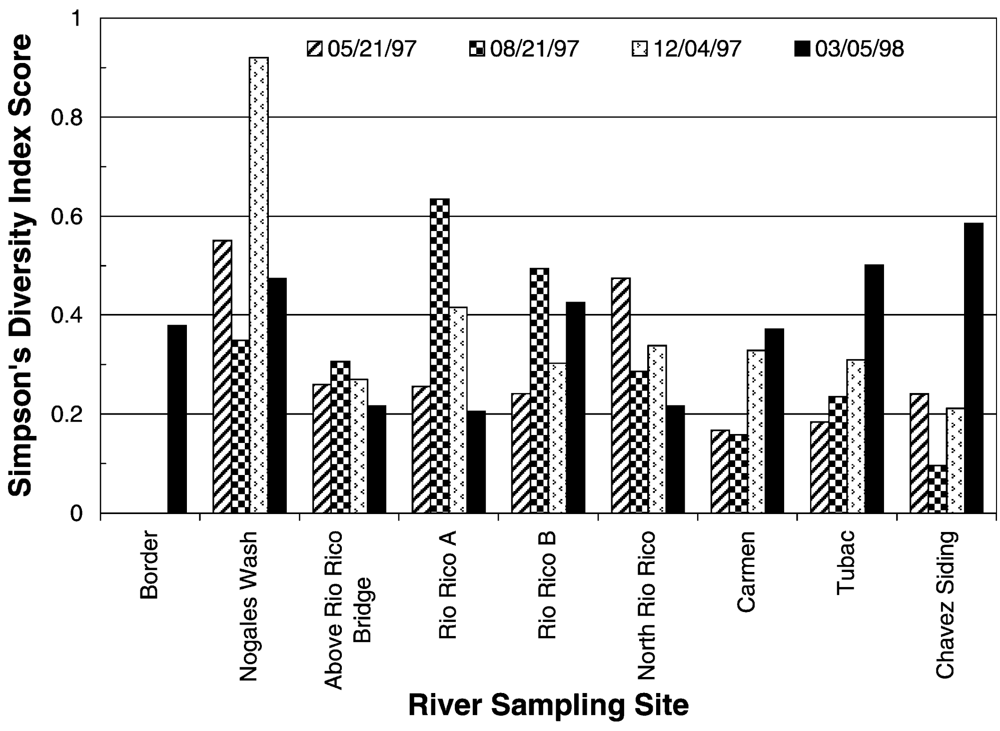

Fig. 13. Simpson's diversity index scores at nine sampling sites on four dates.

indices, and near total domination by Chironomidae (97.4% of total invertebrates, with 8976 individuals in 10 taxa, only 28.6% of total taxa present, 8835 individuals contributed by a single species—*Cricotopus* sp.).

3.2.6.4. Diagnostic taxa. Dense numbers of two groups of invertebrates have been associated with high organic loading, and low dissolved oxygen concentrations have been associated with sewage outfalls. The order Oligochaeta and most members of the insect family Chironomidae have high tolerance to a variety of stresses and when present in dense numbers are a relative indicator of stress (Barbour et al., 1999). However, the number of taxa in each of these categories in the Santa Cruz River is not easily interpreted. Depending on the date, either (1) the number of taxa in each group were high at Border and Nogales Wash, then reduced at sites near the IWWTP discharge, and increased downstream, or (2) they showed no trend (Figs. 14 and 15). Two Chironomidae taxa listed as "tolerant," *Chironomus* sp. and *Cricotopus* sp. (Barbour et al., 1999; LeSage and Harrison, 1980), had high numbers at all sites except the border. On the other hand, two Chironomidae taxa listed as "sensitive," *Rheocricotopus* sp. and *Corynoneura* sp. (Barbour et al., 1999, 1996), were found only at the sites farthest downstream and above the IWWTP.

Members of the order Ephemeroptera are considered to be sensitive to environmental stress and their presence signifies relatively clean conditions (Merritt and Cummins, 1996). The number of Ephemeroptera taxa were highest at the Border and Nogales Wash sites, low or absent below the IWWTP, and relatively higher at the three sites farthest downstream (Fig. 16). While never reaching the high number of Ephemeroptera taxa found in other regions, the relative number does indicate areas of recovery.

We also considered the relative abundance of the dominant (i.e. most numerous) taxon in each sample (Barbour et al., 1996; Kerans et al., 1992) and the population sizes of those taxa in all of the samples. There was no pattern in the proportion of total invertebrate number contributed by the most dominant taxon found in individual samples. There

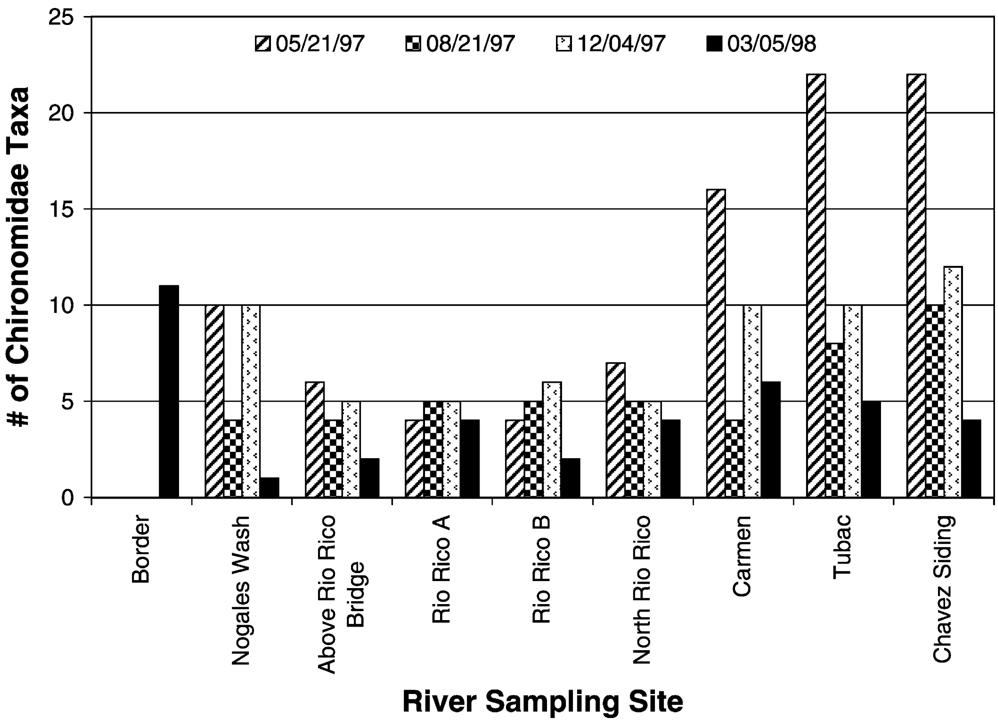

Fig. 14. Number of Chironomidae taxa at nine sampling sites on four dates.

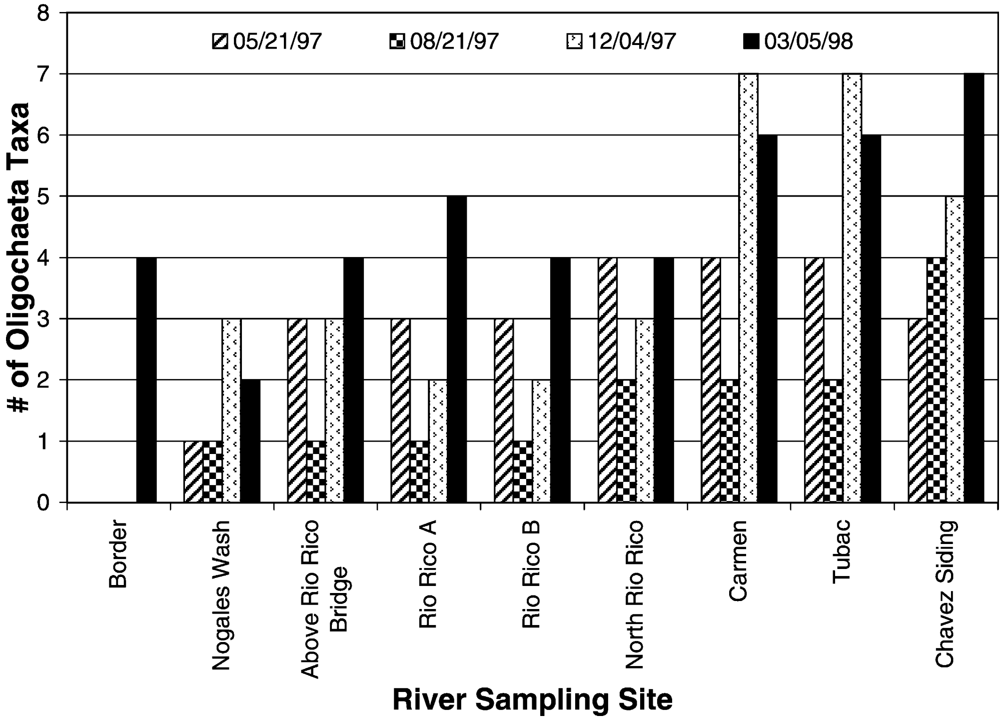

Fig. 15. Number of Oligochaeta taxa at nine sampling sites on four dates.

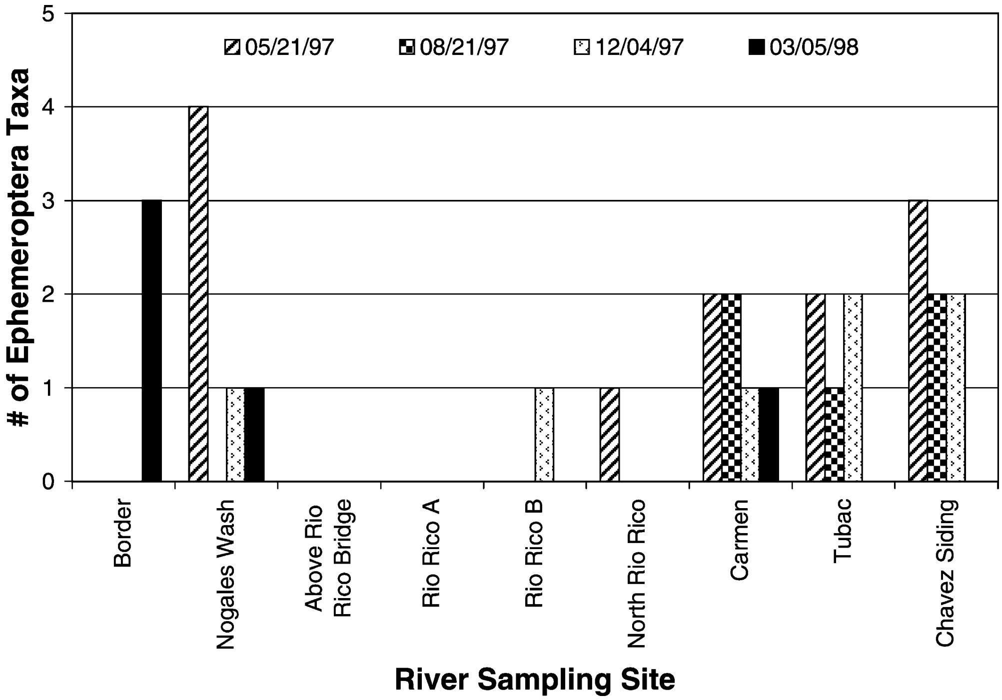

Fig. 16. Number of Ephemeroptera taxa at nine sampling sites on four dates.

were a total of seven taxa that dominated individual samples, two were chironomids (17 sample-dates), two were oligochaetes (11 sample-dates), two were ephemeropterans (3 sample-dates) and one was Collembola (2 sample-dates). *Chironomus* sp., which exploits oxygen-poor habitats because of the presence of haemoglobin (Walsche, 1947), was highest at the sites near the IWWTP (Sites 5 and 6) and relatively low or nonexistent above and downstream from the IWWTP (Fig. 17a). The other dominant chironomid (*Cricotopus* sp.) was highest in the low flow variation periods (May and December) and abundant at all sites during those periods, except at Nogales Wash in May and Border, where it was absent (Fig. 17b). Cricotopus was also absent at the March 1998 Border sample site. While both these genera are indicators of waters with high organic loading, *Cricotopus* is also usually found in waters with naturally high organics (LeSage and Harrison, 1980).

Tubificidae (family within Oligochaeta) also appeared to increase during low flow variation periods, particularly at sites close to the IWWTP, although it was nearly absent above the IWWTP (Fig. 17c). *Nais communis* (Oligochaeta) was highest at the sites

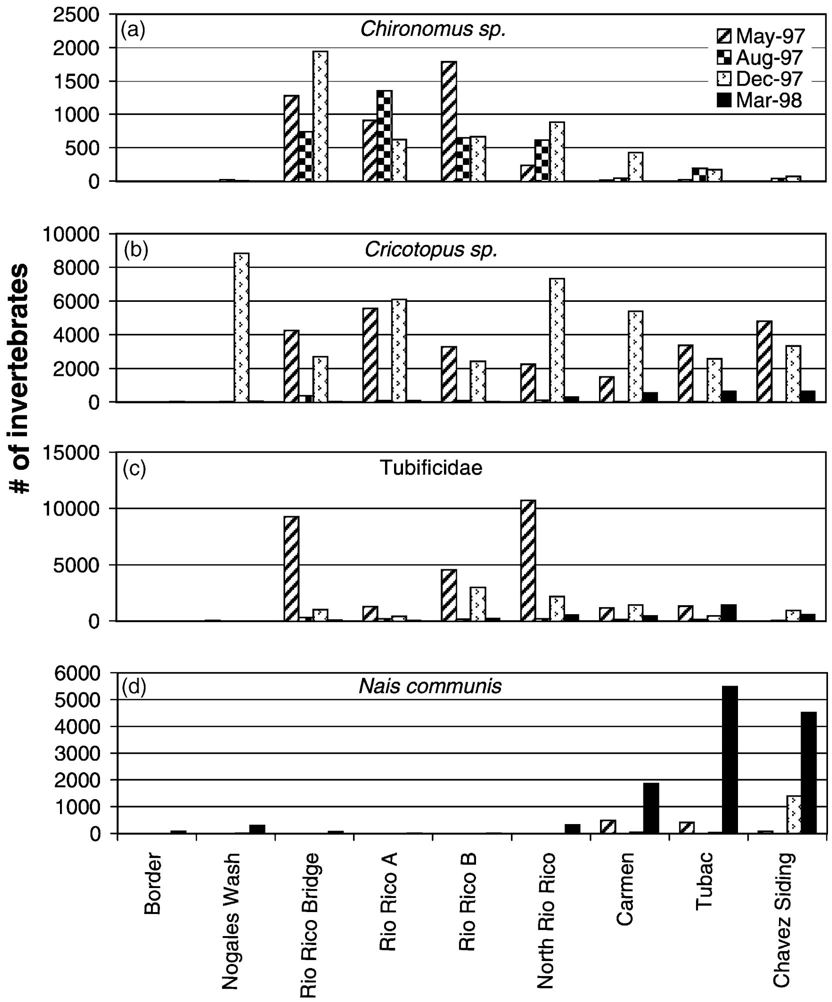

Fig. 17. The taxa that were most numerous in individual samples plotted for nine sampling sites on four dates. *Chironomus* sp. (a) and *Cricotopus* sp. (b) are Chironomidae. Tubificidae (c) and *Nais communis* (d) are Oligochaeta. *Tricorythodes minutus* (e) and *Fallceon quilleri* (f) are Ephemeroptera. Collembola (g) are surface dwellers.

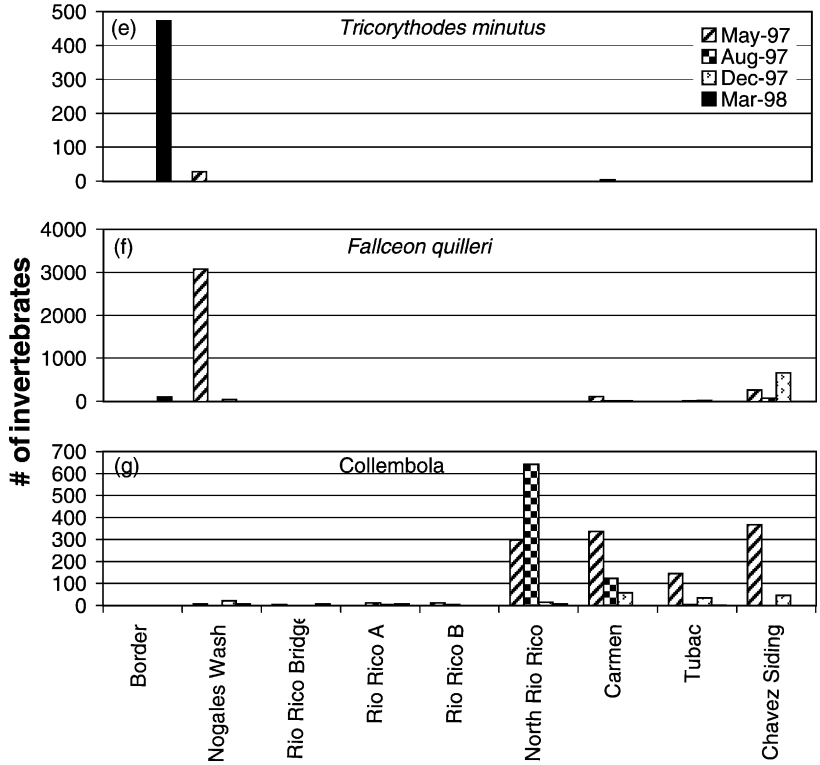

Fig. 17. (*Continued*).

downstream from the IWWTP and relatively low near the treatment plant (Fig. 17d). The two Ephemeroptera taxa (*Tricorythodes minutus* and *Fallceon quilleri*) were never present in samples near the IWWTP and, when present at other sites, had highest densities in samples taken upstream from the IWWTP (Fig. 17e and f). Collembola (identified at order level), low in density above and near the IWWTP, appeared in high numbers during May and August at Sites 6–10 (Fig. 17g). Since Collembola are surface dwellers, their abundance may not be a reflection of water quality.

4. Discussion

4.1. Discharge and vegetation

The status of the biotic system present along the upper Santa Cruz River was a function of a combination of natural and anthropogenic influences. As a result of surface and ground water withdrawals along the upper Santa Cruz River and the increasing depth to bedrock north of Amado, only the reach containing the discharge water from the IWWTP supports a dense riparian woodland comprised of a reproducing willow and cottonwood forest, aquatic resources composed of fish and invertebrate communities, and substantial populations of vertebrates such as birds, reptiles, and amphibians which are directly or indirectly sustained by the waters in the river (Stromberg et al., 1993). The Santa Cruz River upstream from the IWWTP and downstream from Amado was dry for most of the year.

The river discharge measurements varied spatially and temporally due to the discharge from the IWWTP, local freshets entering from various tributaries, and their location on the river. Less discharge in the reach from the border to IWWTP was observed in our study compared to the data in the Patten et al. (1993) report. While we did not investigate the cause of the drying of this stretch of the river, lack of water in the river did prevent collection of some benthic macroinvertebrate community samples in this zone. Discharge was only observed during March 1998 at the Border site. Discharge during the August 1997 sample time varied due to several flash floods observed entering the river from arroyos immediately after brief intense local thundershowers that are typical of the monsoon season in the desert southwest. These freshets and flash floods were not sufficient to change the channel morphology substantially, but did alter substrates, and even habitats supporting benthic macroinvertebrates. Large quantities of sediment could be observed in transport both in suspension and sand in bed load. The floods destroyed large areas of rooted submersed and emergent macrophytes, both in August and March.

Several studies have established that the water provided by the IWWTP effluent supports riparian vegetation along the upper Santa Cruz River (Patten et al., 1993; Stromberg et al., 1993; Tellman et al., 1997). The Arizona Department of Water Resources (1994) emphasized a relationship between historical changes in surface flows and groundwater levels, and Fremont cottonwood–Goodding willow population dynamics as well as other riparian vegetation. The cottonwood–willow forests upstream from the IWWTP were either in poor condition or had been eliminated due to a combination of factors, including grazing by domestic livestock and deepening water-table due to groundwater withdrawal by well fields. Downstream from the IWWTP, effluent release had contributed to the raising of the water table and maintenance of perennial flow, both of which contributed to the support of cottonwood–willow riparian forests.

Patten et al. (1993) attributed effluent water as a major contributor to increasing groundwater levels with the result that perennial flow has been re-established 25 km downstream from the IWWTP. They also conjectured that flow stability plus added nutrients had a positive influence on riparian vegetation. Additional conclusions about the upper Santa Cruz River included that the river section below IWWTP was moderately to heavily polluted and that effluent appeared to have an effect on algal species richness and diversity in the river below the plant (Lawson, 1995; Patten et al., 1993).

4.2. Water chemistry

Previous studies indicated that effluent entering the Santa Cruz River had little effect on metal concentrations and fecal coliform numbers; however, the effluent decreased pH, elevated conductance, organic carbon content, nitrogen, phosphorus, and total petroleum hydrocarbon concentration of sediments (King, 1997; Stromberg et al., 1993). Metal concentrations were low, while nutrients increased as a result of the effluent discharge (Patten et al., 1993). Our results support previous reports with regards to conductance; however, we did not detect any pattern with pH.

The measured levels of ammonium and ammonia in this study were substantially higher than those reported by Patten et al. (1993). In fact, these levels were even higher than the total Kjeldahl nitrogen reported by Patten et al., which includes all of the organic nitrogen as well as ammonia/ammonium present in the sample. This was probably due to an increase in the amount of loading entering the IWWTP from increased populations in both Nogales.

Alkalinity at the Border site and in December at Nogales Wash and Rio Rico stations were substantially reduced relative to the other stations. Currently, water only flows at the Border station during flood events associated with storms (see Fig. 4) and the water may not have had sufficient time to dissolve minerals and gain the water quality characteristics representative of the river channel. The noticeably lower water alkalinity at Nogales Wash, at all sampled dates, relative to that at the Railroad Bridge site just below the IWWTP may have been related to the water quality of runoff from the cities of Nogales.

4.3. Macroinvertebrates

With regard to aquatic macroinvertebrates, we found much higher numbers and diversity throughout the stretch from the US–Mexico border to Chavez Siding (22.4 km downstream from IWWTP) than previously documented (e.g. Lawson, 1995). The species composition varied with river location relative to IWWTP and season. When plotted on a CCA graph, sites for any given date were arranged such that they followed the ammonium and ammonia gradients (vectors) with the site closest to IWWTP (Railroad Bridge, Site 5) at the high concentration. Typically, the site upstream from the IWWTP plotted out towards the low concentration area of the graph. Site scores on the CCA graph that are separated by 2 or more standard deviations are considered to have a nearly complete replacement of the taxonomic composition of the community (Gauch, 1982). During three of the four sampling dates (May, August, and March), there was a complete macroinvertebrate community change between the Rio Rico Bridge and Tubac (18.3 km downstream) sampling sites, near complete or complete at Carmen (15.0 km downstream) sampling site. At the December sampling, there was a near complete community replacement between the Rio Rico and Chavez Siding (20.8 km downstream) sampling sites. This alteration of the macroinvertebrate community would not necessarily be detected by standard biodiversity indices since they are not sensitive to taxa replacements, rather total number of and abundance within taxa.

Community structure frequently changes in response to stress in predictable ways, which is the basis for development of biological criteria to evaluate anthropogenic influences. Gray (1989) summarized the responses into three categories: reduced diversity, increased domination by a single or group of opportunistic (e.g. shorter life-cycle, faster reproducing) species, and reduced individual size. The first two were considered in this study. Near the IWWTP, the total number of benthic macroinvertebrate taxa was reduced relative to both upstream and downstream communities (Fig. 11), although total macroinvertebrate numbers were elevated (Fig. 12). The Shannon–Weiner and Simpson's indices indicated increasing diversity with distance downstream from IWWTP (Figs. 12 and 13). The Ephemeropteran taxonomic richness and population sizes were severely reduced or absent in the vicinity of the IWWTP (Fig. 16). The more opportunistic Chironomidae and Oligochaeta taxa were present throughout the river system, but also increased in taxonomic richness at the downriver sites (Figs. 14 and 15). Chironomidae typically have short life-cycles capable of several generations per year (Tokeshi, 1995).

Although individual species may vary in population density for a wide variety of reasons, examination of key taxa may reveal impacts not otherwise noticed (Barbour et al., 1996; Kerans et al., 1992; Gray, 1989). The variation in population sizes of the seven taxa that dominated the different sites indicated distinct environmental conditions, from highly polluted near the IWWTP to potentially less polluted further downstream (Fig. 17a–g). *Chironomus* sp. and Tubificidae, indicators of high organics and sewage, were densest near the IWWTP and both taxonomic richness and density decreased downstream. *Cricotopus* sp. was common throughout the drainage indicating high organics (LeSage and Harrison, 1980), particularly during periods of low flow variation when freshets did not assist in dilution. *Nais communis* increased in population size with increasing distance from the IWWTP. Numbers of Collembola, which feed on surface water film or bacteria associated with film (Rapoport and Sanchez, 1963), were also reduced near the IWWTP and further upstream, possibly indicating a surfactant in the water. Further downstream, the density of Collembola was higher. *Tricorythodes minutus* and *Fallceon quilleri*, both Ephemeroptera and potentially sensitive to pollution (Barbour et al., 1999) were absent near the IWWTP, but present upstream from and at sites farthest downstream from the plant discharge.

The analysis of both the invertebrates in this study and the algal community in the Patten et al. (1993) report showed a healthier community progressively downstream from the IWWTP. However, a direct comparison of the structure of the benthic macroinvertebrate community with the algal community is difficult in that while ammonia is quite toxic to the invertebrates, it may actually act as a nutrient to algal growth. Moreover, it was difficult to establish an upstream control station for the macroinvertebrates, as was the case for the algal communities because of the lack of discharge at those sites during our study.

Natural variation in river conditions also played a major role in the macroinvertebrate community structure as illustrated by the May 1997 sampling date. While a similar pattern regarding ammonium and ammonia was apparent in the CCA, the samples collected in May were distinctly separated from the others along the vector gradient of the coefficient of variation of river flow rate. All of the sites had nearly complete or complete community replacement relative to the earlier sampling dates.

The elevated ammonia and ammonium concentrations in the river near the IWWTP were implicated in altering the benthic macroinvertebrate community. The absence of sensitive and presence of tolerant indicator taxa supported the classification of the upper Santa Cruz River as polluted.

5. Conclusion

There are questions about the development of biological criteria in streams receiving a major source of their discharge from municipal or industrial effluents. The interpretation of the bioassessment tools derived from the analysis of fish, benthic macroinvertebrate, or algal communities can be affected by these effluents.

This study establishes a number of community level attributes and identifies both naturally occurring variables as well as those associated with effluent that affect the structure of a benthic macroinvertebrate community. Variation in community structure is associated with three classes of variables. First, presence of water in the river is influenced by a combination of groundwater withdrawal, discharge from the IWWTP and the subsurface geomorphology. Secondly, the constituent chemical composition of the discharge from the IWWTP appeared to affect those stations closest to the plant. Finally, community structural changes were associated with naturally produced floods that severely affected both the habitat and the macroinvertebrates. Biological criteria can be established using the qualities of the invertebrate community only by understanding the contribution of these three classes of variables on community structure.

Acknowledgements

Thanks to Boris Kondratieff for his discussions and suggestions as to the response of individual taxa. We also wish to acknowledge the very fine taxonomic identification by Richard Durfee.

Appendix A. List of taxa identified in upper Santa Cruz River

Order	Family	Taxa
Nematoda	Nematoda	Nematoda
	Mermithidae	Mermithidae
Gastropoda	Lymnaeidae	Lymnaeidae
	Physidae	Physidae
Oligochaeta	Enchytraeidae	Enchytraeidae
	Lumbricidae	Lumbricidae
	Lumbriculidae	Lumbriculidae
	Naididae	*Dero* sp.
		Nais communis
		Ophidonais serpentina
		Pristina sp.
	Tubificidae	Tubificidae with hair chaetae
		Tubificidae without hair chaetae
Hirudinea	Erpobdellidae	*Erpobdella punctata*
	Glossiphoniidae	Glossiphoniidae
Acari	Eylaidae	*Eylais* sp.
	Hygrobatidae	*Atractides* sp.
	Lebertiidae	*Lebertia* sp.

Appendix A (*Continued*)

Order	Family	Taxa
	Limnesiidae	*Limnesia* sp.
		Tyrrellia sp.
	Sperchonidae	*Sperchon* sp.
Coleoptera	Dryopidae	*Helichus suturalis*
		Postelichus confluentus
		Postelichus immsi
	Dytiscidae	Dytiscidae
		Hydroporinae
		Copelatus chevrolati renovatus
		Laccophilus fasciatus terminalis
		Laccophilus maculosus shermani
		Laccophilus mexicanus mexicanus
		Laccophilus pictus coccinelloides
		Laccophilus salvini
		Laccophilus sp.
		Liodessus affinis complex
		Rhantus gutticollis
		Rhantus sp.
		Stictotarsus aequinoctialis
		Stictotarsus corpulentus
		Stictotarsus roffi
		Stictotarsus sp.
		Thermonectes nigrofasciatus
	Elmidae	*Microcylloepus pusillus*
	Haliplidae	*Peltodytes* sp.
	Hydraenidae	*Ochthebius* sp.
	Hydrophilidae	*Berosus peregrinus*
		Chaetarthria sp.
		Enochrus carinatus fuscatus
		Enochrus pygmaeus pectoralis
		Enochrus sp.
		Helochares normatus
		Helochares sp. 2
		Helophorus sp.
		Laccobius mexicanus
		Tropisternus ellipticus
		Tropisternus lateralis
		Tropisternus sp. 3
Collembola	Collembola	Collembola

Appendix A (*Continued*)

Order	Family	Taxa
Diptera	Ceratopogonidae	*Atrichopogon* sp.
		Ceratopogonidae genus 1
		Ceratopogonidae genus 2
		Dasyhelea sp.
		Forcipomyia sp.
	Chaoboridae	*Chaoborus* sp.
	Chironomidae	*Ablabesmyia* sp.
		Chironomus sp.
		Corynoneura sp.
		Cricotopus sp.
		Cryptochironomus sp.
		Diamesa sp.
		Dicrotendipes sp.
		Endotribelos sp.
		Euryhapsis sp.
		Goeldichironomus sp.
		Hydrobaenus sp.
		Labrundinia sp.
		Larsia sp.
		Lauterborniella sp.
		Limnophyes sp.
		Mesosmittia sp.
		Microtendipes sp.
		Nanocladius sp.
		Parachironomus sp.
		Parametriocnemus sp.
		Paraphaenocladius sp.
		Pentaneura sp.
		Phaenopsectra sp.
		Polypedilum sp.
		Procladius sp.
		Pseudochironomus sp.
		Pseudosmittia sp.
		Rheocricotopus sp.
		Rheotanytarsus sp.
		Saetheria sp.
		Stictochironomus sp.
		Tanypodinae
		Tanytarsini
		Tanytarsus sp.
		Thienemanniella sp.
		Thienemannimyia group
	Culicidae	*Aedes* sp.
		Anopheles sp.
		Culex sp.

Appendix A (*Continued*)

Order	Family	Taxa
	Dixidae	*Dixella* sp.
	Dolichopodidae	Dolichopodidae
	Empididae	*Hemerodromia* sp.
	Ephydridae	Ephydridae
	Muscidae	Muscidae
	Psychodidae	*Pericoma* sp.
		Psychoda sp.
	Simuliidae	*Simulium vittatum* complex
	Stratiomyidae	*Caloparyphus* sp.
		Euparyphus sp.
		Nemotelus sp.
		Odontomyia sp.
	Tabanidae	*Tabanus* sp.
	Tipulidae	*Erioptera* sp.
		Limonia sp.
		Tipula sp.
Ephemeroptera	Baetidae	*Callibaetis* sp.
		Fallceon quilleri
		Paracloeodes sp.
	Tricorythidae	*Leptohyphes* sp.
		Tricorythodes minutus
Hemiptera	Belostomatidae	*Abedus herberti*
		Abedus sp. 2
		Belostoma flumineum
	Corixidae	Corixidae
		Corisella edulis
		Graptocorixa abdominalis
		Graptocorixa serrulata
	Nepidae	*Ranatra quadridentata*
	Notonectidae	*Notonecta* sp.
	Veliidae	*Microvelia* sp.
Megaloptera	Corydalidae	Corydalidae
Odonata	Calopterygidae	*Hetaerina* sp.
	Coenagrionidae	Coenagrionidae
		Argia sp.
		Hesperagrion heterodoxum
	Gomphidae	*Erpetogomphus* sp.
		Progomphus borealis
	Libellulidae	Libellulidae
		Brechmorhoga mendax

References

Arizona Department of Water Resources, 1994. Arizona Riparian Protection Program Legislative Report.

APHA, 1980. Standard Methods for the Examination of Water and Wastewater. American Public Health Association, Washington, DC, USA.

Barbour, M.T., Gerritsen, J., Griffith, G.E., Frydenborg, R., McCarron, E., White, J.S., Bastian, M.L., 1996. A framework for biological criteria for Florida streams using benthic macroinvertebrates. J. North Am. Benthol. Soc. 15 (2), 185–211.

Barbour, M.T., Gerritsen, J., Snyder, B.D., Stribling, J.B., 1999. Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers: Periphyton, Benthic Macroinvertebrates, and Fish, 2nd ed. Office of Water, US Environmental Protection Agency, Washington, DC, USA, EPA-841-B-99-002.

Boyle, T.P., Smillie, G.M., Anderson, J., Beeson, D.R., 1990. Sensitivity analysis of nine diversity and seven similarity indices used in water quality determinations. Res. J. Water Pollut. Control Federation 62, 749–762.

Gauch, H.A., 1982. Multivariate Analysis in Community Ecology. Cambridge University Press, New York, 298 pp.

Gray, J.S., 1989. Effects of the environmental stress on species rich assemblages. Biol. J. Linn. Soc. 37, 19–32.

Hauer, F.R., Resh, V.H., 1996. Benthic macroinvertebrates. In: Hauer, F.R., Lamberti, G.A. (Eds.), Methods in Stream Ecology. Academic Press, San Diego, CA, pp. 339–370.

Kerans, B.L., Karr, J.R., 1994. A benthic index of biotic integrity (B-IBI) for rivers of the Tennessee Valley. Ecol. Appl. 4, 768–785.

Kerans, B.L., Karr, J.R., Ahlstedt, S.A., 1992. Aquatic invertebrate assemblages: spatial and temporal differences among sampling protocols. J. North Am. Benthol. Soc. 11, 377–390.

King, K.A., 1997. AZ—Contaminants as Limiting Factors of Wildlife Populations in the Santa Cruz River Basin. US Fish and Wildlife Service Region 2.

Lawson, L., 1995. Upper Santa Cruz Intensive Survey: A Volunteer Driven Study of the Water Quality and Biology of an Effluent Dominated Desert Grassland Stream in Southeast Arizona. Arizona Department of Water Quality, 68 pp.

LeSage, L., Harrison, A.D., 1980. The biology of Cricotopus (Chironomidae: Orthocladiinae) in an algal enriched stream: Part I. Normal biology. Arch. Hydrobiol. (Suppl. 4), 375–418.

Merritt, R.W., Cummins, K.W., 1996. An Introduction to the Aquatic Insects of North America, 3rd ed. Kendall/Hunt Publishing Company, Dubuque, IA, USA, 862 pp.

Omernik, J.M., 1987. Ecoregions of the conterminous United States. Ann. Assoc. Am. Geogr. 77 (1), 118–125.

Patten, D.T., Stromberg, J.C., Sommerfeld, M.R., 1993. Final Report. Water and Riparian Resources of the Santa Cruz River Basin: Best Management Practices for Water and Resource Quality. SCERP Project Number WQ93-12. Arizona State University (available from: http://www.scerp.org/scerp/projects/WQ93_12.html).

Rapoport, E.H., Sanchez, L., 1963. On the epineuston or the superaquatic fauna. Oikos 14, 96–109.

Schmidt, K.D., 1993. Proceedings of the Symposium on Effluent Use Management. American Water Resources Association Technical Publication Series, TPS-93-3.

Stromberg, J.C., Sommerfeld, M.R., Patten, D.T., Fry, J., Kramer, C., Amalfi, F., Christian, C., 1993. Release of effluent into the upper Santa Cruz River, Arizona: ecological considerations. In: Schmidt, K.D. (Ed.), Proceedings of the Symposium Effluent Use Management. American Water Resources Association, pp. 81–90.

Tellman, B., Yarde, R., Wallace, M.G., 1997. Arizona's Changing Rivers: How People Have Affected the Rivers. Issue Paper #19. Water Resources Research Center, College of Agriculture, The University of Arizona, 198 pp.

ter Braak, C.J.F., Verdonschot, P.F.M., 1995. Canonical correspondence analysis and related multivariate methods in aquatic ecology. Aquat. Sci. 57, 255–289.

Tokeshi, M., 1995. Life cycles and population dynamics. In: Armitage, P.D., Cranston, P.S., Pinder, L.C.V. (Eds.), The Chironomidae: Biology and Ecology of Non-Biting Midges. Chapman & Hall, New York, pp. 225–268.

USDI (US Department of Interior), 1984. Water Measurement Manual: A Water Resource Technical Publication, 2nd ed. United States Printing Office, Denver, CO, USA, 327 pp.

USEPA (US Environmental Protection Agency), 1994. National Water Quality Inventory: 1992 Report to Congress. Office of Water, US Environmental Protection Agency, Washington, DC, USA, EPA-841-R-94-005a.

USEPA (US Environmental Protection Agency), 1999. Fact Sheet: 1999 Update of Ambient Water Quality Criteria for Ammonia—Technical Version. Office of Water, US Environmental Protection Agency, Washington, DC, USA, EPA-823-F-99-024.

USEPA (US Environmental Protection Agency), 2002. AQUIRE data base (available from: http://www.dnr.state.wi.us/org/water/wm/ww/biomon/_INFO.HTM).

Wallace, J.B., Grubaugh, J.W., Whiles, M.R., 1996. Biotic indices and stream ecosystem processes: results from an experimental study. Ecol. Appl. 6, 140–151.

Walsche, B.M., 1947. On the function of haemoglobin in Chironomus after oxygen lack. J. Exp. Biol. 24, 329–342.

Washington, H.G., 1984. Diversity, biotic, and similarity indices: a review of special relevance to aquatic ecosystems. Water Res. 18, 653–656.