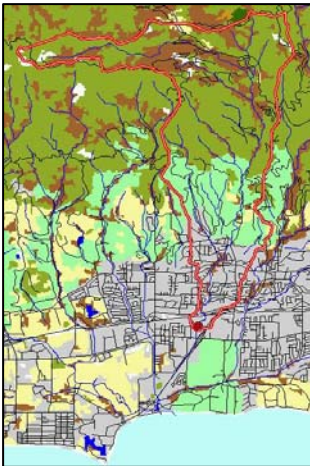


SANTA BARBARA COUNTY CREEKS BIOASSESSMENT PROGRAM

2003 ANNUAL REPORT AND INDEX OF BIOLOGICAL INTEGRITY

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Prepared for:
County of Santa Barbara Project Clean Water
and
City of Santa Barbara

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Executive Summary

Introduction

This report summarizes the results of the 2003 Santa Barbara County Creeks Bioassessment Program, a collaborative effort of County of Santa Barbara Project Clean Water and the City of Santa Barbara. The Program is a long-term effort to assess and monitor the biological integrity of southern Santa Barbara County streams as they respond through time to changing environmental conditions shaped by natural and human influences. The 2003 Program effort represents the fourth consecutive year of rapid bioassessment monitoring in southern Santa Barbara County streams. The Program involves annual collection and analyses of physiochemical and biological data from local streams using standardized methods adapted from the U.S. Environmental Protection Agency's (USEPA's) *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers*.

The objectives of the 2003 Program effort were to (1) continue biomonitoring of streams in the study area, and (2) building on the data collection and analyses conducted thus far, develop a standardized tool known as an Index of Biotic Integrity (IBI) to be used in assessing the biological integrity of study area streams. IBIs are multimetric tools that provide a standardized, integrative, and readily understandable method for measuring the biological integrity of streams and other water bodies. The term "multimetric" refers to the fact that an IBI is built by combining several individual biological metrics into a single index. "Core" metrics included in the IBI all show distinct separation (i.e., are different) between undisturbed "reference" sites, and human-impacted "test" sites. In addition, the core metrics of an IBI collectively represent multiple aspects of biological community structure such as abundance, richness and diversity, composition, disturbance tolerance, and trophic groups. Values for each core metric at a study site are "scored" on a dimensionless scale (e.g., from 0 to 10) in relation to the known distribution among a collection of reference and test sites. Higher scores (e.g., a 10) approach the conditions at the best reference sites, while lower scores indicate greater departure (i.e., degradation) from reference conditions. Scores assigned to the individual core metrics are equally weighted and combined into an overall score, or measure, of biological integrity for the study site. By translating complex biological data into an overall composite measure of biological integrity, an IBI serves as a powerful tool for communicating the biological status of water resources to a wide audience, and an important basis of environmental management decisions.

Study Area

The study area includes approximately 35 miles of the southern Santa Barbara County coast from the Rincon Creek watershed at the Santa Barbara/Ventura County line west to Gaviota Creek. A total of 44 study reaches in 18 coastal streams have been surveyed one or more times during the spring and summer of 2000, 2001, 2002, and 2003.

Methods

Physiochemical and biological data for the study reaches was gathered through a combination of methods including field surveys, laboratory analysis, spatial data analysis using geographic information system (GIS) software, and review of United States Geological Survey (USGS) 7.5-minute quadrangle maps and aerial photographs. Numerous physiochemical and biological parameters were calculated for each study reach based on the data collected. After the data

set was finalized, statistical tests including analysis of variance (ANOVA) and multiple regression analysis were used to evaluate the data, and the IBI was developed.

Results and Discussion

The following core biological metrics were selected for inclusion in the IBI:

- Insect family diversity
- Percent EPT
- Biotic index score
- Percent sensitive BMIs
- Percent non-insects + Diptera
- Percent predators + shredders

Based on the results of statistical testing, core metrics were among the most sensitive to human disturbance, either increasing or decreasing from highly disturbed to moderately disturbed to undisturbed study reach groups. None had significant natural relationships with physiochemical parameters. Collectively, the core metrics are diversified in that they represent different aspects of community structure including diversity, disturbance sensitivity, and trophic structure. The core metrics are also diversified in that some respond positively to human disturbance (biotic index score and percent non-insects + Diptera) while others respond negatively to disturbance (insect family diversity, percent EPT, percent sensitive BMIs, and percent predators + shredders).

The IBI correctly classified 94 percent of moderately disturbed sites (i.e., as Fair or Good) and 85 percent of highly disturbed sites (i.e., and Very Poor or Poor), or 88 percent overall. There were not any gross inaccuracies in classifying sites. No undisturbed sites were classified lower than Fair, and no highly disturbed sites were classified higher than Fair. Biological integrity classifications were in most cases consistent from year to year at stream sites that were surveyed in multiple years.

Statistical results indicate highly significant differences in IBI scores between the undisturbed, moderately disturbed, and highly disturbed groups. This indicates that the IBI is sensitive to changes in the level of human disturbance.

IBI score did not have any significant natural relationships with physiochemical parameters. Therefore, IBI scores do not appear to be significantly influenced by natural physiochemical variability in the study area.

Recommendations

The IBI developed in this study appears to be mostly reliable in properly assessing the biological integrity of study area streams, and does not appear to be strongly influenced by natural physiochemical variability. As such, the IBI appears to be an effective assessment tool for study area streams. The County and City should continue their annual biomonitoring and use the IBI to assess the biological integrity of the study sites. The IBI should be revisited with every two or three years of new data to see if it can be improved by using new core metrics, refining scoring ranges, etc. Another consideration may be to expand the IBI in the future to include core metrics for other assemblages such as aquatic vertebrates and the riparian plant community. Alternatively, separate IBIs could be developed for the other assemblages.

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I. Introduction

A. Overview

This report summarizes the results of the 2003 Santa Barbara County Creeks Bioassessment Program, a collaborative effort of County of Santa Barbara Project Clean Water and the City of Santa Barbara. The Program is a long-term effort to assess and monitor the biological integrity of southern Santa Barbara County streams as they respond through time to changing environmental conditions shaped by natural and human influences. The 2003 Program effort represents the fourth consecutive year of rapid bioassessment monitoring in southern Santa Barbara County streams. The Program involves annual collection and analyses of physiochemical and biological data from local streams using standardized methods adapted from the U.S. Environmental Protection Agency's (USEPA's) *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers* (Barbour et. al., 1999). Bioassessment field surveys include assessment and measurement of physiochemical parameters (e.g., water quality, stream discharge, width, depth, etc.) and the collection of biological data including benthic macroinvertebrate (BMI) samples. BMIs are the main focus of the Program with respect to biological monitoring.

Bioassessment, or biomonitoring as it is also called, is the science of using biological assemblages including BMIs, fish, amphibians, diatoms, etc. to assess and monitor the biological integrity or "health" of aquatic ecosystems. Karr and Dudley (1981) defined "biological integrity" as "the ability to support and maintain a balanced, integrated, adaptive community of organisms having a species composition, diversity, and functional organization comparable to that of natural habitat of the region." (Miller et al., 1988). Accurate assessment of biological integrity requires a method that integrates ecological insights about the structure and dynamics of populations, communities, and ecosystems (Miller, et al., 1988).

The "logic" behind bioassessment is that because different aquatic species have varying habitat requirements and abilities to withstand of water pollution and other types of habitat degradation, one can tell a great deal about the overall condition of a water body in knowing which species are living there. For example, the presence of viable salmonid populations in coastal California streams is generally an indicator of good biological integrity. Salmonids require cool, clean, well-oxygenated waters, and a sufficient prey base of aquatic invertebrates and vertebrates to survive. They are intolerant of increased sedimentation, which can smother their spawning gravels and fill in rearing pools. Declining salmonid populations can be an indicator of several problems including degraded water quality, increased stream temperatures, increased sedimentation, or habitat fragmentation (e.g., fish passage barriers). Beyond individual species, measurements or "metrics" of biological community structure including abundance (i.e., number or biomass of individuals present), diversity (i.e., number of species present), and species composition (i.e., overall proportion of disturbance-sensitive species, trophic group representation, etc.) have proven to be reliable indicators of biological integrity (Rosenberg and Resh, 1993, Barbour et al., 1999).

B. Previous Program Efforts (2000-2002)

The 2002 Annual Report summarizes the findings of the first three years of the Program (2000-2002), and is available at www.countyofsb.org/project_cleanwater/Documents. The past work provided a great deal of insight on (1) relationships between local stream biota, human

disturbance, and natural physiochemical variables, and (2) which metrics are the best indicators of biological integrity in local streams. Key findings from the previous work are as follows:

- About half of the biological metrics evaluated (including individual BMI taxa and community metrics) had strong natural relationships with physiochemical variables. Of the seven physiochemical variables considered, stream temperature and elevation appeared to have the greatest influence on biological metrics.
- Nearly all of the biological community metrics and many individual BMI taxa were strongly related to human disturbance. Human-disturbed study reaches were degraded in terms of ecosystem integrity as evidenced by:
 - ✓ Impaired water quality in the form of higher stream temperature, specific conductance, and nutrient levels;
 - ✓ Lower diversity of BMIs and aquatic vertebrates;
 - ✓ Lower composition of disturbance-sensitive BMIs, and;
 - ✓ Higher composition of disturbance-tolerant BMIs.
- Urban-impacted sites were typically more degraded in terms of water quality and biological community structure compared to agriculture-impacted sites.
- Differences in water quality and biological community structure between undisturbed and lightly disturbed sites were in most cases minor and not statistically significant.

C. 2003 Program Effort

The objectives of the 2003 Program effort were to (1) continue biomonitoring of streams in the study area, and (2) building on the data collection and analyses conducted thus far, develop a standardized tool known as an Index of Biotic Integrity (IBI) to be used in assessing the biological integrity of study area streams. IBIs, popular among water resource agencies in the U.S., are multimetric tools that provide a standardized, integrative, and readily understandable method for measuring the biological integrity of streams and other water bodies. The term "multimetric" refers to the fact that an IBI is built by combining several individual biological metrics into a single index. "Core" metrics included in the IBI all show distinct separation (i.e., are different) between minimally degraded "reference" sites, and degraded "test" sites. In addition, the core metrics of an IBI collectively represent multiple aspects of biological community structure such as abundance, richness and diversity, composition, disturbance tolerance, and trophic groups. Values for each core metric at a study site are "scored" on a dimensionless scale (e.g., from 0 to 10) in relation to the known distribution among a collection of reference and test sites. Higher scores (e.g., a 10) approach the conditions at the best reference sites, while lower scores indicate greater departure (i.e., degradation) from reference conditions. Scores assigned to the individual core metrics are equally weighted and combined into an overall score, or measure, of biological integrity for the study site. By translating complex biological data into an overall composite measure of biological integrity, an IBI serves as a powerful tool for communicating the biological status of water resources to a wide audience, and an important basis of environmental management decisions (Miller et al., 1988, Gerritsen, 1995, Norris, 1995, Barbour, et al., 1996, Reynoldson et al., 1997, Lyons and Wang, 1996, Barbour, et al., 1999, Karr and Chu, 1999, Ode, Rehn, and Harrington, 2002).

Because biological assemblages vary in response to physiochemical gradients that exist through geographic space, IBIs are calculated for specific regions with similar ecological characteristics.

This minimizes the potential for confusion between natural physiochemical and anthropogenic influences. Further, separate IBIs are developed for different classes of water bodies including estuaries, lakes, and streams. If necessary, water body classes can be further classified and partitioned based on physiochemical characteristics such as elevation, stream order, stream width, etc. (Miller et al., 1988, Gerritsen, 1995, Barbour, et al., 1996, Lyons and Wang, 1996, Barbour, et al., 1999, Karr and Chu, 1999, Ode, Rehn, and Harrington, 2002). IBIs are typically (but not always) developed for a single biological assemblage (e.g., BMIs, fish, amphibians, algae, etc.). To provide a more complete assessment of the biological condition of water bodies, U.S. EPA and others recommend developing IBIs for more than one assemblage (Barbour, et al., 1999). Assemblages can respond differently to certain stressors and restoration activities. For example, Mount et al. (1984) found that BMI and fish assemblages responded differently to the same pollution inputs in the Ottawa River in Ohio. BMIs responded negatively to organic loading from a wastewater treatment plant, and exhibited no observable response to chemical input from industrial effluent. Conversely, fish exhibited no response to the organic inputs, and a negative response to metal concentrations in the water (Barbour, et al., 1999).

An alternative to multimetric IBIs are multivariate models, namely the river invertebrate prediction and classification scheme (RIVPACs) and its derivatives. Multivariate models are used extensively in England, Australia, and Canada to assess the biological integrity of streams. RIVPACs models are also being developed in this country (e.g., by the U.S. Geological Survey). Multivariate models such as RIVPACs and its derivatives are empirical (statistical) models that predict which dominant BMI taxa should occur at a site in the absence of environmental stress based on the physiochemical attributes of the site (Gerritsen, 1995, Norris, 1995, Reynoldson et al., 1997, Barbour, et al., 1999, Ode, Rehn, and Harrington, 2002). RIVPACs models are developed for specific habitat types (e.g., riffles, pools, macrophyte stands, etc.) in streams within defined physiochemical classifications and geographic regions. Building a multivariate model requires the collection of data from a large set of randomly selected reference sites to establish which BMI taxa are expected to occur. Multivariate models have been slow to gain acceptance in the U.S. Common explanations for this given in commentaries on the subject include:

- Multivariate models are more complicated and intuitively more difficult to understand compared to IBIs, which makes them difficult to convey to a wide audience (Gerritsen, 1995, Norris, 1995, Reynoldson et al., 1997, Barbour, et al., 1999, Ode, Rehn, and Harrington, 2002).
- Multivariate models consider only the presence or absence of dominant taxa in assessing biological integrity, and do not incorporate important aspects of community structure including density, relative abundances of individual taxa, trophic composition, etc. (Gerritsen, 1995, Norris, 1995).
- Multivariate models require sampling of a relatively large number of randomly selected reference sites (upwards of 300) to yield reliable results (Gerritsen, 1995, Norris, 1995, Barbour, et al., 1999).
- Multivariate models have not been developed thus far for any biological assemblages besides BMIs (Barbour, et al., 1999).
- RIVPACs models have not been tested using multi-habitat BMI sampling approaches such as the one used in this study (Barbour et al., 1999).

- Multivariate models have not been conclusively shown to be more effective than IBIs in measuring biological integrity (Gerritsen, 1995).

Considering the above points and the fact that the scheme (i.e., nonrandom) used for selecting study sites in this Program is incompatible with a RIVPACs model, an IBI is the appropriate stream assessment tool for the study area at this time.

II. Study Area

The study area includes approximately 35 miles of the southern Santa Barbara County coast from the Rincon Creek watershed at the Santa Barbara/Ventura County line west to Gaviota Creek (see Figure 1). There are approximately 40 small 1st-5th order coastal watersheds along this stretch of coast, all of which drain the southern face of the Santa Ynez Mountains. A total of 44 study reaches in 18 coastal watersheds have been surveyed one or more times during the spring and summer of 2000, 2001, 2002, and 2003. Three of the study reaches surveyed over the years are in the Ventura River and Sespe Creek watersheds in Ventura County, or outside of the main study area. These three sites were surveyed in 2002 only. Figures 2, 3, and 4 are maps showing the locations of study streams and reaches. Table 1 lists the study reaches, their locations, and the year(s) in which they were surveyed.

The study reaches range from 1st order mountain tributaries to 5th order lowland streams, and from relatively pristine to severely impaired by human disturbance. The severity of human disturbance in local streams is dictated by the nature and intensity of surrounding land uses. As a general observation, anthropogenic impacts appear to be more pronounced in urbanized areas compared to those in rural areas. Some of the major forms of human disturbance to local streams include: (1) altered hydrology and geomorphology due to water diversions, land clearing and development, and flood control projects, (2) sedimentation of pools and riffle substrates due to increased erosion and deposition of fine sediments from agricultural fields and destabilized creek banks, (3) degraded water quality due to pollution inputs, (4) elevated stream temperatures due to loss of overhanging riparian vegetation and shade, (5) habitat fragmentation due to the construction of in-stream barriers such as dams, road crossings, bridges, and culverts, (6) introductions of invasive, non-native plants (e.g., *Arundo donax*), and wildlife (e.g., bullfrogs and crayfish), and (7) disturbances to vegetation and wildlife associated with trampling (i.e., by cattle, vehicles, bikers, hikers, etc.), noise, lighting, air pollution, and predation by domestic pets.

The study reaches are grouped into four different categories based on the level to which they are subject to human disturbance. Grouping criteria are provided below. Table 1 indicates the category for each study reach.

UNDIST = Undisturbed or minimally disturbed by human activities. Habitat assessment score 150/200 or greater, five percent or less of upstream watershed is disturbed.

MOD DIST = Lightly to moderately disturbed by human activities. Habitat assessment score 120 or greater but less than 150, or if habitat assessment score is greater than 150, greater than five percent of upstream watershed is disturbed.

HIGH DIST= Heavily disturbed by human activities including agricultural and urban/suburban land uses. Habitat assessment score 120 or less, greater than five percent of upstream watershed is disturbed.

FIGURE 1: PROGRAM STUDY AREA

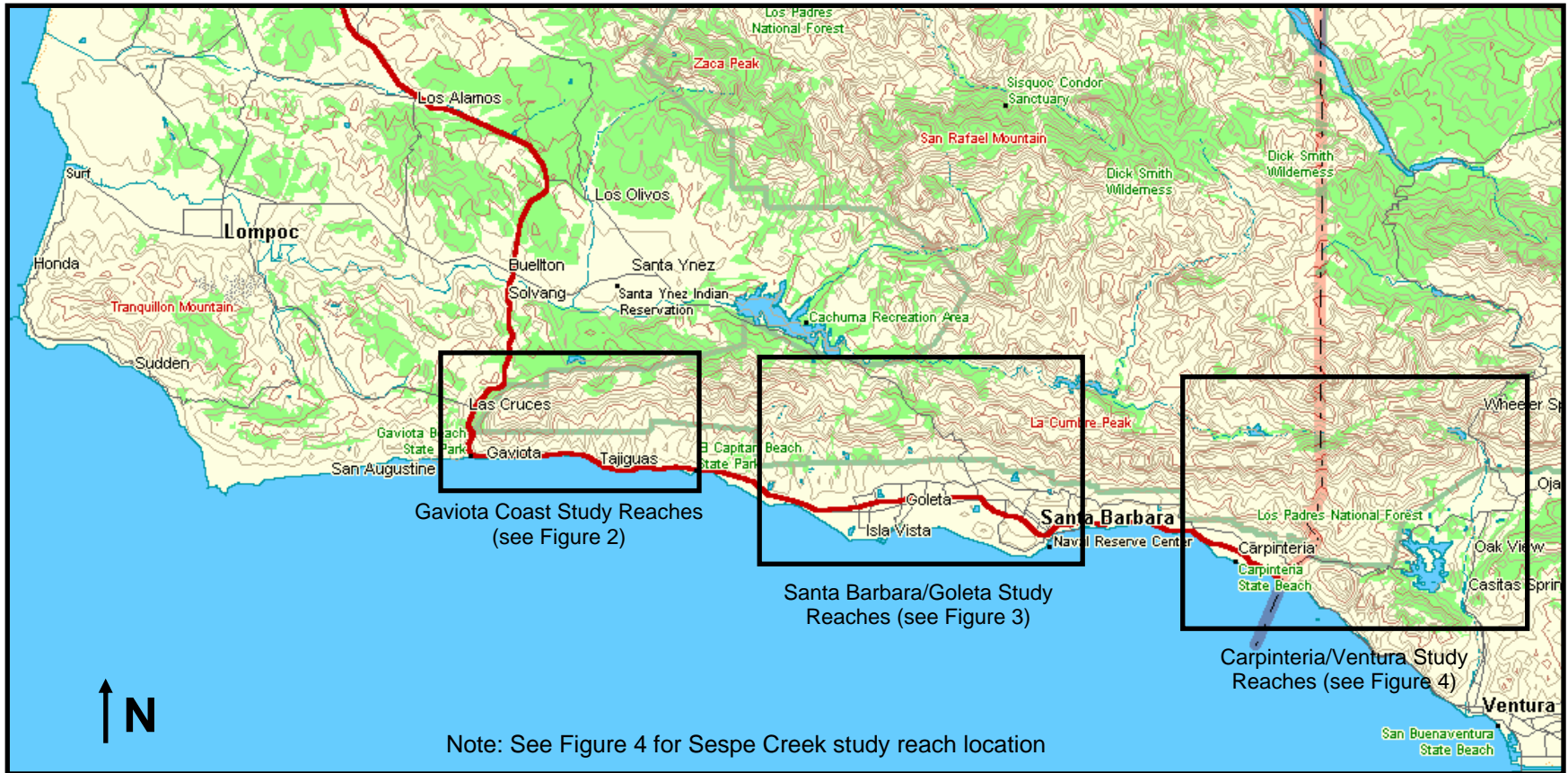


FIGURE 2: GAVIOTA COAST STUDY REACHES

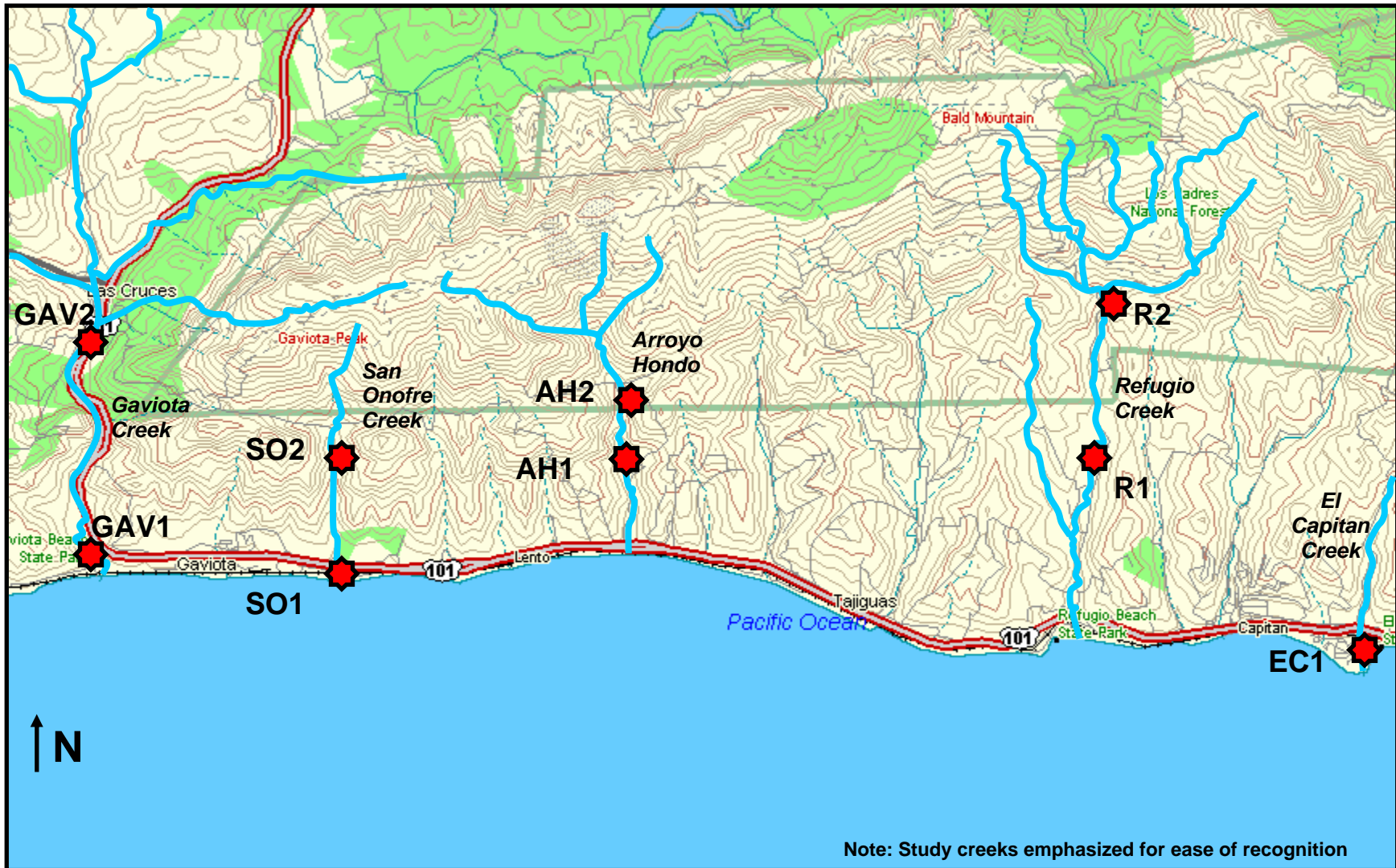


Table 1 Study Reaches			
Study Reach	Location	Disturbance Category	Years Surveyed
SES1	Sespe Creek just below confluence with Little Sespe Creek	UNDIST	2002
MA1	Matilija Creek, approx. 1.25 mi. above Matilija Dam, 0.25 mi. below first residential village	UNDIST	2002
MA2	Matilija Creek, approx. 1 mi. upstream of confluence with Old Man Mtn. Creek	UNDIST	2002
RIN1	Rincon Creek, just upstream of Highway 150 crossing at Gobernador Cyn. Rd.	MOD DIST	2002
C1	Carpinteria Creek, approx. ¼-mi. downstream of Carpinteria Ave.	HIGH DIST	2000, 2001, 2002, 2003
C2	Carpinteria Creek, approx. ¼-mi. upstream of U.S. 101.	HIGH DIST	2000, 2001
C3	Gobernador Creek, approx. ¼-mile upstream of debris basin	UNDIST	2000, 2001, 2002, 2003
F1	Franklin Creek just upstream of entrance into Carpinteria Salt Marsh	HIGH DIST	2000
SM1	Santa Monica Creek just upstream of entrance into Carpinteria Salt Marsh	HIGH DIST	2000
MONT1	Montecito Creek at Val Verde property, just below Hot Springs/Cold Springs confluence.	MOD DIST	2003
SY1	Sycamore Creek just below Mason St. bridge	HIGH DIST	2002, 2003
SY2	Sycamore Creek just below Highway 192 crossing and Coyote Creek/Sycamore Creek confluence.	HIGH DIST	2003
M1	Mission Creek at De la Guerra St.	HIGH DIST	2000, 2002, 2003
M2	Old Mission Creek at Bohnet Park	HIGH DIST	2002
M3	Mission Creek at upstream end of Rocky Nook Park	MOD DIST	2000, 2002, 2003
M4	Rattlesnake Creek, approx. 0.5 mi. upstream of Las Canovas Rd. crossing	UNDIST	2000
M5	Rattlesnake Creek, approx. 0.25 mi. downstream of Gibraltar Rd. crossing	UNDIST	2000
M6	Mission Creek, at falls above Jesuita Trail crossing	UNDIST	2000
AB1	Arroyo Burro at upstream end of Alan Rd.	HIGH DIST	2002, 2003
AB2	Arroyo Burro just downstream of Torino Rd.	HIGH DIST	2000, 2001, 2002, 2003
AB3	San Roque Creek, ¼-mi. upstream of Foothill Rd.	MOD DIST	2000, 2001, 2002, 2003
AT1	Atascadero Creek near Patterson Rd.	HIGH DIST	2001, 2002, 2003
AT2	Atascadero Creek just downstream of Cieneguitas Creek confluence	HIGH DIST	2001, 2002, 2003
SA1	San Antonio Creek, approx. ½ mi. upstream of Tucker's Grove Park	MOD DIST	2000

Table 1 Study Reaches

Study Reach	Location	Disturbance Category	Years Surveyed
SA2	San Antonio Creek, approx. ¼ mi. upstream of Highway 154	MOD DIST	2000, 2003
MY1	Maria Ygnacio Creek, approx. ¼ mi. downstream of San Marcos Rd.	HIGH DIST	2000
MY2	Maria Ygnacio Creek, approx. ¼ mi. upstream of debris basin	UNDIST	2000
MY3	Maria Ygnacio Creek, approx. ¼ mi. upstream of Highway 154	UNDIST	2000
SJ1	San Jose Creek, approx. ¼ mile downstream of U.S. 101.	HIGH DIST	2000, 2001, 2002, 2003
SJ2	San Jose Creek, approx. ½-mile upstream of Patterson Rd. crossing	HIGH DIST	2000, 2001, 2002, 2003
SJ3	San Jose Creek at San Marcos Trout Club	UNDIST	2000, 2001, 2002, 2003
SJ4	San Jose Creek, adjacent to southeast junction of Kinevan Rd. and Hwy 154	UNDIST	2000
T1	Tecolote Creek, approx. 150 ft. upstream of Vereda del Padre	HIGH DIST	2000
T2	Tecolote Creek, adjacent to Vereda Nueva	HIGH DIST	2000
DP1	Dos Pueblos Creek, approx. 150 ft. downstream of U.S. 101.	HIGH DIST	2000
EC1	El Capitan Creek in State Park, approx. 300 ft. upstream of the mouth	MOD DIST	2002
R1	Refugio Creek, approx. 1.5 mi. upstream of U.S. 101	DIST AG	2000
R2	Refugio Creek, approx. ¼ mi. downstream of Circle Barbee Ranch	MOD DIST	2000
AH1	Arroyo Hondo, approx. 1 mi. upstream of U.S. 101.	UNDIST	2000, 2001, 2002, 2003
AH2	Arroyo Hondo, approx. 2 mi. upstream of U.S. 101.	UNDIST	2000
SO1	San Onofre Creek, just below U.S. 101 culvert	UNDIST	2000
SO2	San Onofre Creek, approx. 1 mi. upstream of U.S. 101	UNDIST	2000, 2001, 2002, 2003
GAV1	Gaviota Creek at State Beach, just below entrance road/U.S. 101 junction	MOD DIST	2002, 2003
GAV2	Gaviota Creek, approx. 600 ft. downstream of Las Canovas Creek confluence	MOD DIST	2002

III. Methods

Physiochemical and biological data for the study reaches was gathered through a combination of methods including field surveys, laboratory analysis, spatial data analysis using geographic information system (GIS) software, and review of United States Geological Survey (USGS) 7.5-minute quadrangle maps and aerial photographs. Numerous physiochemical and biological parameters were calculated for each study reach based on the data collected. Table 2 lists each parameter calculated for the study reaches, parameter abbreviations used throughout the remainder of the report, and the method of calculation (e.g., lab, field, etc.). After the data set was finalized, statistical tests including analysis of variance (ANOVA) and multiple regression analysis were used to evaluate the data, and the IBI was developed. Further discussion of methods is provided below.

A. Field Surveys

Field surveys were conducted at a total of 30 study reaches in 2000, 12 in 2001, 23 in 2002, and 20 in 2004. Surveys were conducted in the spring and early summer each year during base stream flow conditions (i.e., low flows) for consistency, as the local stream biota is known to undergo seasonal succession (Cooper et al., 1986). The following was completed during each field survey:

- General observations were recorded on a standard field data sheet, including study reach location, date, time, weather, stream flow conditions, water clarity, and sources of human disturbance.
- A 100-meter study reach was delineated along the stream. Stream habitat units (i.e., riffles, runs, pools, etc.) within the study reach were mapped. Representation of each habitat type was quantified as a percentage of the total reach length.
- Stream widths (wetted perimeter, channel bottom, and bank full) and riparian corridor width were measured at three transects in the study reach. Wetted perimeter width was defined as the cross-sectional distance of streambed that is inundated with surface water. Channel bottom width was defined as the cross-sectional distance between the bottoms of the stream banks. Bank full width was defined as the distance from the ordinary high water mark from one stream bank to the other, as evidenced by visible signs of stream flow such as water marks, stream-carried deposits of sediments and debris, and scour features.
- Plants and wildlife species observed in the stream and riparian zone were recorded.
- Water temperature, specific conductance, and pH were measured in the field using YSI and Oakton handheld meters. Dissolved oxygen concentration was measured in 2000, 2002, and 2003 (not in 2001) using a YSI hand-held meter. Two measurements of each parameter were made, one in a riffle and the other in a pool, and the two values were averaged. Water measurements were made at various times during daylight hours, from mid-morning to late afternoon.
- Stream discharge (Q) was estimated at a selected cross-section in the study reach. Q was estimated by multiplying the measured flow width times the average water depth and velocity, as measured at three to five equally spaced points along the cross-section. Velocity and depth were measured using a Global Water FP101 flow probe.

Table 2
List of Parameters Calculated for Each Study Reach

Parameters	Units of Measurement	Abbreviation	Method of Calculation
PHYSICAL PARAMETERS			
Stream order	None	None	Maps
Elevation	Feet (ft.)	None	Maps
Stream gradient	None	None	Maps
Watershed area	Acres	None	GIS
Percent of watershed area disturbed	None	None	GIS
Wet stream width	Ft.	None	Field
Stream discharge	Cubic feet per second (cfs)	Q	Field
Habitat assessment score	None	None	Field
WATER CHEMISTRY PARAMETERS			
Stream temperature	Degrees Fahrenheit (°F)	None	Field
pH	None	None	Field
Dissolved oxygen concentration	Milligrams per liter (mg/l)	DO	Field
Conductivity	Microsiemens (µS)	None	Field
Specific conductance (corrected to 25° Celsius)	µS	None	Field
BIOLOGICAL PARAMETERS			
BMI density	# per sq. meter (#/m ²)	None	Field/lab
Insect order diversity (field)	None	None	Field
Insect family diversity (field)	None	None	Field
BMI order diversity	None	None	Field/lab
BMI family diversity	None	None	Field/lab
Non-insect order diversity	None	None	Field/lab
Insect order diversity	None	None	Field/lab
Insect family diversity	None	None	Field/lab
Ephemeroptera/Plecoptera/Tricoptera family diversity	None	EPT family diversity	Field/lab
Percent EPT	None	Percent EPT	Field/lab
Percent Plecoptera/Tricoptera	None	Percent PT	Field/lab
Percent sensitive EPT	None	None	Field/lab
Biotic index score	None	None	Field/lab
Percent sensitive BMIs	None	None	Field/lab
Percent tolerant BMIs	None	None	Field/lab
Percent dominant taxon	None	None	Field/lab
Percent two dominant taxa	None	None	Field/lab
Percent Diptera as Chironomidae	None	None	Field/lab
Percent non-insect BMIs	None	None	Field/lab
Percent non-insects + Diptera	None	None	Field/lab
Percent non-insects + Chironomidae	None	None	Field/lab
EPT/Chironomidae ratio	None	None	Field/lab
Percent collector-gatherers	None	None	Field/lab
Percent scrapers	None	None	Field/lab
Percent shredders	None	None	Field/lab
Percent collector-filterers	None	None	Field/lab
Percent predators	None	None	Field/lab
Percent scrapers + shredders	None	None	Field/lab
Percent scrapers + shredders + predators	None	None	Field/lab
Percent collector-gatherers + scrapers + shredders	None	None	Field/lab
Percent collector-gatherers + collector-filterers	None	None	Field/lab
Percent collector-gatherers + predators	None	None	Field/lab
Percent predators + shredders	None	None	Field/lab
Abundances of individual BMI taxa (many)	# individuals/sample	None	Field/lab
Native aquatic vertebrate diversity	None	None	Field
Percent riparian canopy cover	None	None	Field
Percent native riparian plant species	None	None	Field

- BMI samples were collected using a standardized method based on the “multi-habitat” approach described in the EPA’s *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers* (Barbour et al., 1999). In 2001, 2002, and 2003 three samples were collected per study reach: one sample from the downstream third of the reach, one from the middle third, and one from the upstream third. Each sample represents approximately one square meter of stream bottom, collected from 10 individual, 0.1-square meter locations. The 10 locations that constituted each sample were selected based on the relative coverage area of stream habitats (i.e., riffles, pools, falls, etc.) in the section of stream sampled. For example, if a given stream reach contained approximately 50 percent riffles and 50 percent pools, five locations in riffles and five in pools were selected and sampled. Samples were collected using a D-frame net with 250 μm mesh. In locations with flowing water (e.g., riffles and runs), the net was held upright against the stream bottom, and substrata immediately upstream within a defined 0.1-square meter area was scraped and stirred up for approximately 15 seconds using feet and hands. Dislodged BMIs were carried into the net by the stream current. In areas with little or no current (e.g., pools), stream bottom substrata was stirred up by foot, followed by a quick sweep of the net through the water column to capture dislodged BMIs. This was repeated three times in each pool sampling location. In 2000, only one sample was collected per study reach. The method was the same as described above, except that each sample represented two square meters of stream bottom taken from 20 individual 0.1-square meter locations (two square meters total) selected throughout the 100 meter study reach.
 - After each BMI sample was collected, it was rinsed with water in a 500 μm sieve to wash out fine sediments, transferred to a plastic container and preserved in 70 percent ethanol for laboratory analysis.
 - In 2002 and 2003, the first BMI sample collected at each study reach was dumped into a plastic bucket with water and visually screened for five minutes. All BMI orders and families observed were recorded.
 - A semi-quantitative stream habitat assessment was conducted using the protocol provided in the EPA’s *Rapid Bioassessment Protocols for Use in Streams and Wadeable Rivers*. Per this protocol, habitat components were visually assessed and scored, including stream substrate/cover, sediment embeddedness, stream velocity/depth regime, sediment deposition, channel flow status, human alteration, channel sinuosity, habitat complexity/variability, bank stability, vegetative protection, and width and composition of riparian vegetation. Each study reach was assigned a total score of between zero and 200 based on the sum of scores assigned to each habitat component. Criteria from the EPA protocol were used to guide the scoring.
 - Important features in the study reaches were photographed.
- Quality control measures were incorporated into the field surveys to insure accurate and consistent data gathering. Water monitoring equipment was calibrated regularly. Field crew members were trained to properly operate equipment, take measurements, collect BMI samples, and conduct stream habitat assessments. Stream habitat assessment scoring was done as a group by the field crew.

B. Laboratory Analysis

BMI samples were processed in the laboratory to determine BMI community composition (i.e., taxa present and relative abundance) and overall density. Each BMI sample was strained through a 500- μ m mesh sieve and washed with water to remove ethanol and fine sediments. The sample was placed in a plastic tray marked with 25 equally-sized squares in a five by five grid pattern. The entire sample was spread out evenly across the 25 squares. Squares of material were randomly selected using a random number table, and sorted one at a time under a dissecting microscope until a specified number of BMIs were located and picked out. The proportion of the sample sorted was noted, and BMI density was estimated based on the proportion of the one square meter sampling area sorted and the number of specimens picked. 330 BMIs were picked out from each of the 2000 samples, when only one sample was collected per reach. Of the 330 specimens picked from each sample, 300 were randomly selected for identification. Starting in 2001, the sampling strategy was changed to allow the collection of replicate samples (three per study reach) without changing the total number of specimens picked and identified for each study reach. 110 specimens were picked out from each of the samples collected in 2001 and 2002 (i.e., three samples, 330 BMIs per study reach). 100 of the 110 BMIs picked from each sample (300 total per study reach) were randomly selected for identification.

BMI samples were identified under a dissecting microscope using standard taxonomic keys. After processing and identification, sorted BMIs and unsorted sample remnants were bottled separately in 70 percent ethanol for storage. In previous years of the study (2000-2002), BMIs were typically identified to genus, and to species for monotypic genera. Exceptions included Chironomids, which were left at the family level, and some non-insects (e.g., oligochaetes, ostracods, and copepods), which were identified to family, order, or class. Starting this year, we began identifying BMIs to the family level only, forgoing the more detailed genus level identifications. The decision to identify BMIs to family rather than genus was made in response to strong statistical test results (using the first three years of data) which showed that identifying BMIs to genus did not provide any added ability to detect differences in BMI community structure between reference and disturbed study reach groups than did identifying BMIs to family. Identifying BMIs to family rather than genus has resulted in a savings of 30-40 percent of the laboratory costs, and 10-15 percent of the overall costs of the study.

Quality control measures were incorporated into the laboratory analysis to insure random selection and accurate, consistent enumeration and identification of BMIs. BMI sample processing methods were clearly established and strictly followed. Specimens of all identified taxa were sent to another taxonomist for independent identification. The taxonomists compared their results, and together resolved all inconsistencies in identification. All of the sample identifications were then re-examined, and necessary changes were made.

C. GIS Analysis

GIS Arcview software was used to calculate watershed area and watershed land use coverages for each study reach. Watershed area was calculated based on watershed boundaries generated in the GIS with a 30 meter digital elevation model using hydrologic processing tools in Arcview GIS 3.2. Watershed land use coverages for each study reach were calculated in the GIS by superimposing watershed boundaries over a digital land cover GIS layer for the region. The land cover layer was produced by the California Department of Forestry and Fire Protection's (CDF) Fire and Resource Assessment Program (FRAP). The land cover layer is titled LCMMP Vegetation Data 1994 to 1997. The CDF program contains a land use map for the region based

on the following eight land cover categories: urban, agriculture, herbaceous, hardwood, shrub, conifer, water, and barren/other.

The parameter "percent watershed disturbed" was calculated for each study reach by using the following equation:

Percent watershed disturbed = percent urban + percent agriculture + 0.5(percent herbaceous)

Herbaceous areas were counted as partially (i.e., half) disturbed to reflect the fact that much of the herbaceous lands in the region are used for cattle grazing or are previously cleared land.

D. Review of Topographic Maps and Aerial Photographs

USGS 7.5 minute quadrangle topographic maps (1:24,000 scale) for the study area were reviewed to determine stream order, elevation, and gradient for each study reach. Gradient was determined by dividing the elevation change between topographic contours immediately upstream and downstream of the study reach by the stream length between the contours. Stream length was determined by tracing a map wheel over the mapped stream path. Quad maps were also used to check the accuracy of watershed boundaries determined for the study reaches by the GIS digital elevation model. Aerial photographs of the study area from 1999 were reviewed to construct hand-drawn land use maps for the study area watersheds and check the accuracy of the GIS land use mapping layer. The GIS and hand-drawn land use maps were typically in close agreement, and no major adjustments to the GIS calculations were necessary.

E. Calculation of Biological Parameters

Numerous BMI community metrics were calculated for each sample to reflect different aspects of community structure including BMI density, diversity, composition (i.e., taxa present and relative abundances), trophic group representation, and sensitivity to human disturbance. Functional feeding group metrics (e.g., percent predators, shredders, etc.) were determined using functional feeding group designations for individual taxa provided in *An Introduction to the Aquatic Insects of North America* (Merritt and Cummins, 1996). Disturbance-sensitivity metrics (e.g., biotic index score, percent sensitive BMIs, percent tolerant BMIs, etc.) were calculated using disturbance tolerance values for individual BMI taxa provided in CDFG's *List of Californian Macroinvertebrate Taxa and Standard Taxonomic Effort* (2002). This document assigns individual BMI taxa with tolerance values of between 0 and 10 based on their observed ability to withstand human disturbance. A tolerance value of 0 indicates that a BMI is extremely intolerant of human disturbance, with increasing scores indicating greater tolerance. With respect to the calculation of individual metrics, "sensitive" BMIs were those having a tolerance value of two (2) or less, and "tolerant" BMIs were those having a tolerance value of eight (8) or greater. See the 2002 Annual Report for additional details as to how specific metrics were calculated.

Abundances of selected individual BMI taxa were also used as biological metrics. Those selected included insect orders and families that showed especially strong relationships with human disturbance in last year's analyses.

Log (y+1) transformations of several metrics were calculated and used as separate metrics.

F. IBI Development

Developing the IBI required the completion of several distinct steps, including (1) selection of study reaches to be included in the data set, (2) screening and selection of core metrics, (3)

defining scoring ranges for core metrics, (4) defining IBI scoring categories and ranges, and (5) testing the IBI to see how reliable it is in classifying the biological integrity of individual study reaches. Methods used to complete these steps are discussed below.

1. Study Reach Selection and Grouping

Study reaches included in the data set used to develop the IBI were those located in the study area from Rincon Creek to Gaviota Creek. The Matilija Creek and Sespe Creek sites surveyed in 2002 were not included in developing the IBI because these sites possessed some notable differences in physiochemical conditions (e.g., watershed area, width, and geology) and BMI community composition compared to the Santa Barbara coastal streams. There were several BMI taxa found in the Ventura County streams, but not in any of the Santa Barbara coast streams. In addition to the Ventura County streams, study reach M6 (upper Mission Creek) was excluded from the data set. This site was surveyed in late July 2000 at a time when surface water was essentially limited to residual pools (i.e., riffles were dry), which was unlike the conditions found during any of the other surveys (i.e., all other sites had flowing riffle sections).

An important aspect of the study reach groupings (i.e., UNDIST, MOD DIST, and HIGH DIST) is that they were made "a priori" (i.e., before the analyses of biological data) based on physiochemical parameters, namely habitat assessment scores and watershed land use. This avoids the circularity that would exist if the study reaches were grouped using biological criteria.

2. Screening and Selection of Core Metrics

Sensitivity to Human Disturbance

In order to evaluate their sensitivity to human disturbance, all of the biological metrics calculated (see Table 2) were evaluated for differences between the UNDIST, MOD DIST, and HIGH DIST study reach groups using analysis of variance (ANOVA). An ANOVA test compares the means and distributions of a given metric among multiple sampling groups, and indicates the probability that the means for the groups are the same. The probability that the means are the same is expressed as p , which is between 0 and 1. The lower the p , the lower the probability is that the group means are the same. A p of 0.05 or less is generally accepted as indicating a statistically significant difference between group means.

As discussed in the Introduction, selected "core" metrics must distinguish between reference and disturbed sites if the IBI is to be useful in measuring biological integrity. All of the biological metrics tested that significantly (i.e., $p \leq 0.05$) increased or decreased with increasing levels of human disturbance (i.e., from the UNDIST to MOD DIST to HIGH DIST groups) were retained for further screening.

Natural Relationships with Physiochemical Parameters

The next round of screening involved the use of multiple regression analyses to determine the strength and nature of natural relationships (i.e., in the absence of human disturbance) between biological metrics and several physiochemical parameters using data from the UNDIST study reach group ($n=23$). A multiple regression analysis simultaneously evaluates and compares the effects of multiple independent variables (i.e., the physiochemical variables), or "regressors", on a single response variable (i.e., each biological metric). A best-fit equation is calculated that represents the response variable as a function of the independent variables. The correlation coefficient (r^2) and p -value (p) are calculated in regression analyses, and used to interpret the strength of the relationship between the response variable and the regressors.

r^2 is given as a value between 0 and 1, and indicates the how well the equation fits the data. The higher the r^2 , the better the fit of the equation. P indicates the probability that the response variable and regressors are not related as predicted by the best-fit equation, and is given as a value of between 0 and 1. A p of 0.05 or less is generally accepted as indicating a statistically significant relationship between the independent and response variables.

With the exception of dissolved oxygen concentration (not measured at all study reaches), all physiochemical parameters calculated for the study reaches (see Table 2) were considered as potential regressors. First, multivariate correlation analysis was used to determine whether any of the physiochemical parameters were highly correlated among the undisturbed study reaches. High correlation among independent variables (regressors) is termed "collinearity". Strong collinearity among regressors causes multiple regression models to become unstable and sensitive to small changes in the data, thus highly collinear variables must be eliminated to yield reliable results. To accomplish this, where any two or more regressors had a correlation of 0.5 or greater, all but one was eliminated. In these situations, the retained regressor had the fewest correlations above 0.5 and the weakest correlations with remaining regressors. The retained regressors were used in the multiple regression analyses.

Core Metric Selection

Once the above screening analyses were complete, core metrics for inclusion in the IBI were selected. All selected core metrics showed (1) highly significant responses to human disturbance, either increasing or decreasing between UNDIST to MOD DIST to HIGH DIST groups, and (2) weak natural relationships (i.e., not significant) with physiochemical parameters. This in theory at least avoids a situation of confusing biological responses to human disturbance with responses to natural physiochemical gradients. Collectively, core metrics were chosen to represent three major aspects of biological community structure: diversity, disturbance tolerance/sensitivity, and trophic composition (i.e., functional feeding groups).

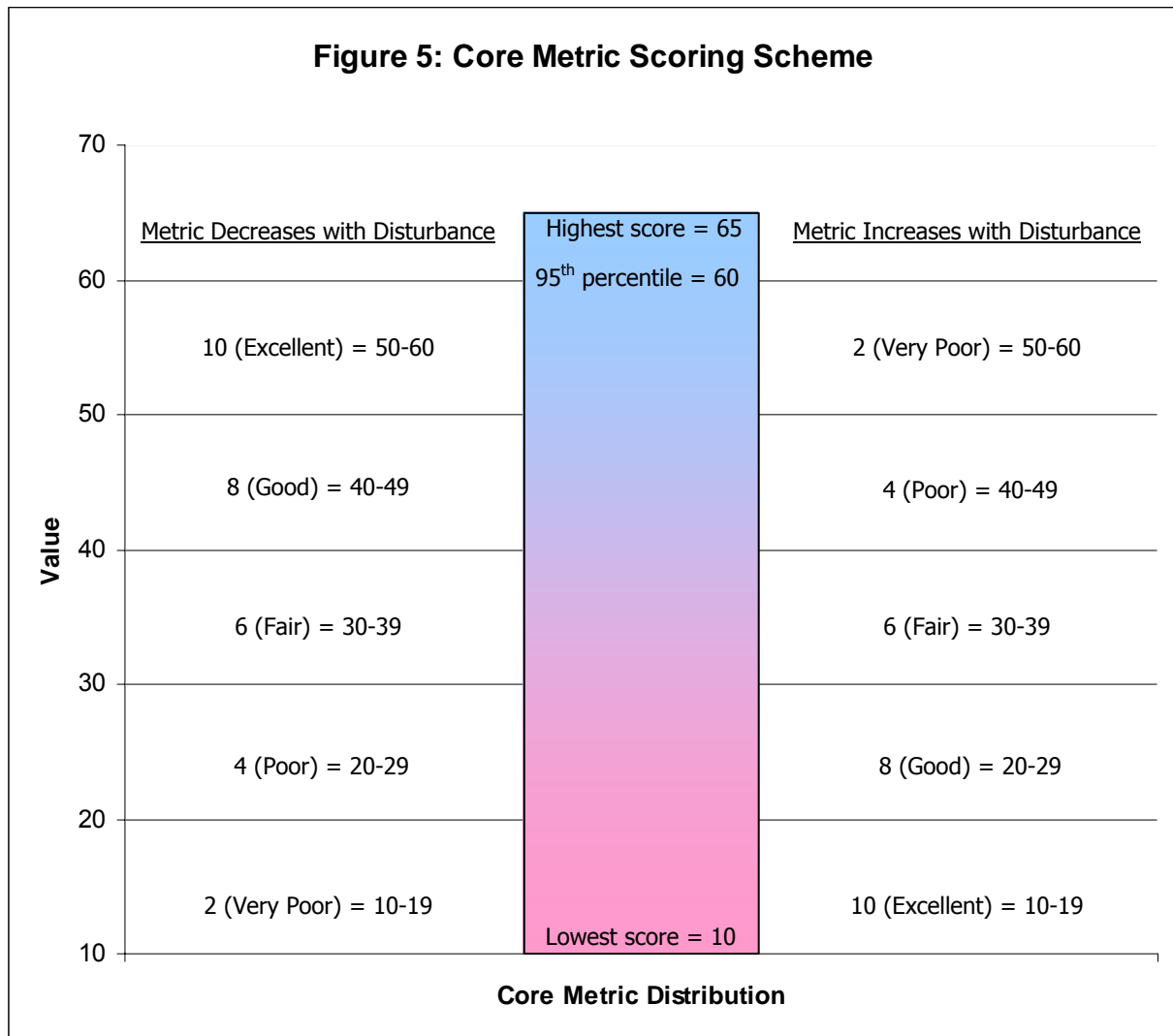
3. Defining Scoring Categories and Ranges for Core Metrics

After the core metrics were selected, scoring ranges of were established for each metric. Two basic metric scoring strategies have commonly been used in developing IBIs. Some have used the distribution of values only from reference (i.e., undisturbed) sites to establish scoring ranges, while others have used the distribution of values from reference and test (i.e., impacted) sites. We chose the latter strategy. Intuitively, this makes sense. If an IBI is intended to assess the integrity of streams having varying levels of human disturbance, metric scoring criteria should be based on the whole range of conditions present, not just reference sites. More specifically, we used the 95th percentile value of the distribution of values in developing metric scoring criteria. This was done to eliminate potential outlier effects (i.e., the highest five percent of scores were eliminated from consideration). This approach to metric scoring is supported by the U.S. EPA Rapid Bioassessment Protocols Guide (Barbour, et al., 1999), which states "more recent studies are finding that a standardization of all metrics as percentages of the 95th percentile yields the most sensitive index...Unpublished data from statewide databases for Idaho, Wyoming, Arizona, and West Virginia are supportive of this alternative for scoring metrics. Ideally, a composite of all sites representing a gradient of conditions is used. This situation is analogous to a determination of a dose/response relationship and depends on the ability of incorporating both reference and non-reference sites."

Figure 5 illustrates how scoring criteria was established for the core metrics. The range between the 95th percentile value (upper end) and the lowest score (lower end) was established and divided into five equal intervals. The five intervals were assigned with the following scores:

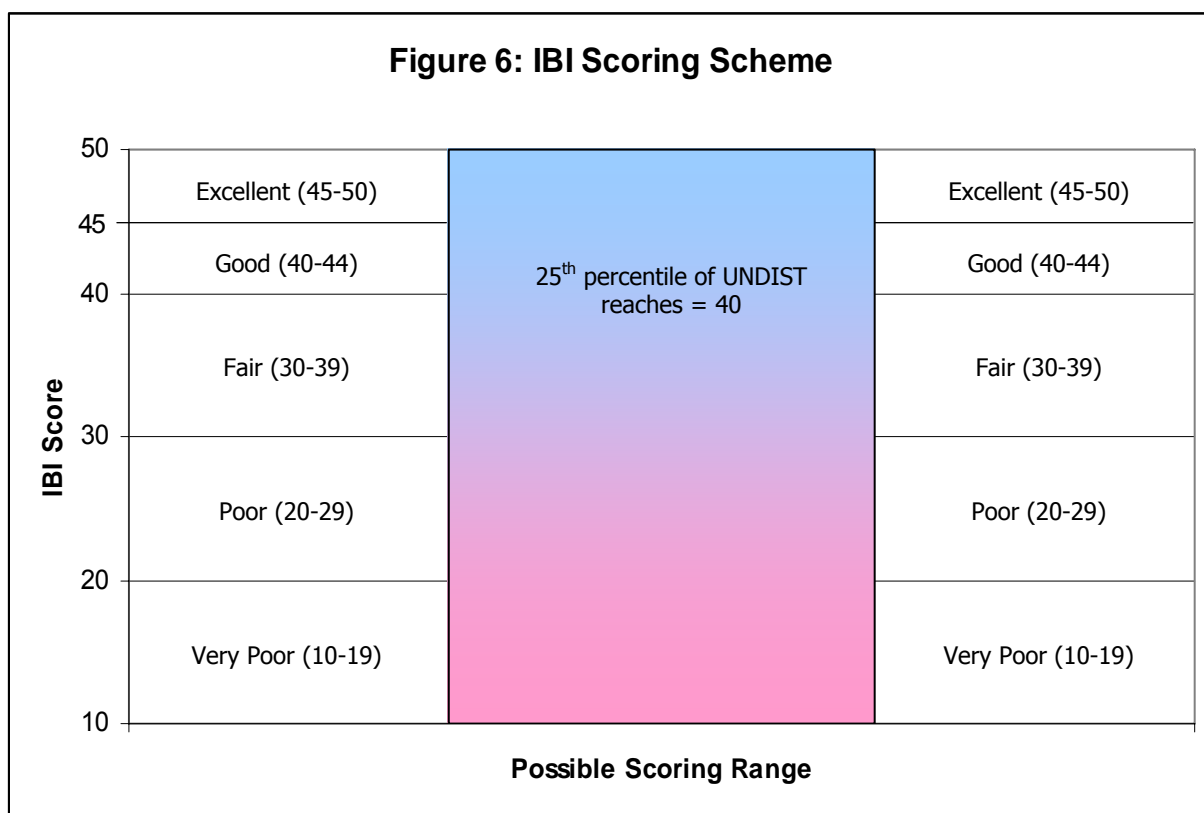
- 10 (Excellent)
- 8 (Good)
- 6 (Fair)
- 4 (Poor)
- 2 (Very Poor)

For metrics that decrease with human disturbance (i.e., highest at reference sites), higher values corresponded with higher scores. For metrics that increase with human disturbance (i.e., lowest at reference sites), lower values corresponded with higher scores (see Figure 5).



4. Defining IBI Scoring Categories and Ranges

After scoring criteria was established for the core metrics, an overall IBI score was tabulated for each study reach by adding the respective scores of the core metrics. Based on the distribution of IBI scores for the study sites, five categories of biological integrity were established: Excellent, Good, Fair, Poor, and Very Poor. Figure 6 illustrates how scoring ranges for the five categories of biological integrity were established. In this illustration, it is assumed that the IBI is composed of five core metrics. Thus, the range of possible scores would be 10 to 50, as possible scores for individual core metrics are from 2 to 10. The 25th percentile score of the UNDIST group distribution (i.e., reference sites) was used as the threshold between the fair and good categories. The score range above the 25th percentile of the UNDIST group distribution was bisected to determine the threshold between the good and excellent categories. The score range below the 25th percentile of the UNDIST group distribution was trisected to determine thresholds between the fair, poor, and very poor categories.



5. Testing the IBI

After the IBI categories and scoring ranges were established, IBI scores were calculated for all of the study reaches to see how reliable the IBI was in classifying sites into appropriate categories of biological integrity, as compared to the a priori classifications based on habitat assessment score and watershed land use. UNDIST sites rated as "Excellent" or "Good", MOD DIST sites rated as "Good" or "Fair", and HIGH DIST sites rated as "Poor" or "Very Poor" by the IBI were considered to be properly classified. The percentage of sites properly classified was calculated based on these criteria.

An ANOVA was completed to compare IBI scores between UNDIST, MOD DIST, and HIGH DIST groups to determine the sensitivity of the IBI to human disturbance.

An ANOVA was completed to evaluate the degree to which IBI scores fluctuated from year to year at the nine study reaches that have been sampled in all four years of the study.

The IBI was also tested using multiple regression analysis for natural relationships between IBI scores at the UNDIST sites and the same physiochemical parameters used in the core metric screening regressions. If the IBI was significantly related to natural physiochemical parameters, its ability to separate the influences of natural physiochemical variability and human disturbance on biological integrity would be questionable.

IV. Results and Discussion

A. Data

Table A-1 in the Appendix presents physiochemical data for the individual study reaches collected in 2003 and previous years of the study. Mean parameter values and ranges among the study reaches are provided at the bottom of the table.

A total of 139 plant species have been observed among all of the study reaches, including 94 native species and 45 introduced (i.e., non-native) species. Table A-2 provides a list of the plant species observed, and a breakdown of their occurrence by study reach. Table A-2 also indicates the number of native and introduced plant species observed at each study reach, and the percentage of plant species observed that are native. The number of years (i.e., 1, 2, 3, or 4) each study reach was surveyed is provided at the top of the table. Plant observations are combined in the table for study reaches that were surveyed in multiple years.

A total of 104 vertebrate species (98 native and six introduced) have been observed among all of the study reaches, including four fish, five amphibians, 11 reptiles, 70 birds, and 14 mammals. Vertebrate species having special regulatory status (i.e., of concern, fully protected, rare, threatened, endangered, etc.) from the state and/or federal government that have been observed include steelhead/rainbow trout, California newt, southwestern pond turtle, two-striped garter snake, coastal western whiptail lizard, coast horned lizard, yellow warbler, and Cooper's hawk. Table A-3 provides a list of observed vertebrate species, and a breakdown of their occurrence by study reach. The number of years each study reach has been surveyed is provided at the top of the table. Vertebrate observations are combined in the table for study reaches surveyed in multiple years.

A total of 10 orders and 61 families of aquatic insects (class Insecta) have been collected and identified among all the study reaches in the four years of study (see the 2002 Annual Report for insect genera data from previous years). Common aquatic insect orders include Ephemeroptera (mayflies, six families), Plecoptera (stoneflies, four families), Trichoptera (caddisflies, 14 families), Coleoptera (beetles, eight families), Diptera (true flies, 13 families), Hemiptera (true bugs, six families), Odonata (dragonflies and damselflies, six families), and Megaloptera (Dobson flies and alder flies, two families). Two semi-aquatic insect orders, Collembola (springtails) and Hymenoptera (wasps), have been found in small numbers. Non-insects found in the study reaches include Gastropoda (snails); several types of crustaceans including Ostracoda, Copepoda, Cladocera, Decapoda, Amphipoda, and Isopoda; Acari (water mites); Turbellaria (flatworms); Oligochaeta (segmented worms); Hirudinea (leeches); and Nematomorpha (horsehair worms). Overall, non-insects composed a small proportion of the

BMI's sampled. Table A-4 provides a list of observed BMI taxa, and a breakdown of their occurrence and abundance by sample and study reach. BMI community parameters are also listed by sample and study reach.

B. IBI Development

1. Screening and Selection of Core Metrics

Sensitivity to Human Disturbance

Table A-5 summarizes the results of the ANOVA tests conducted to evaluate the sensitivity of the biological metrics to human disturbance (i.e., differences between study reach groups). As an example, Figure 7 illustrates the ANOVA for biotic index score, which had a very significant positive relationship with human disturbance ($p < 0.0001$, $r^2 = 0.53$). The illustration shows the distributions (i.e., collection of data points) for each study reach group side by side. The top and bottom points of the diamonds shown for each study reach group indicate the 95 percent confidence limits for the group mean. The center of the diamond is the mean for the group.

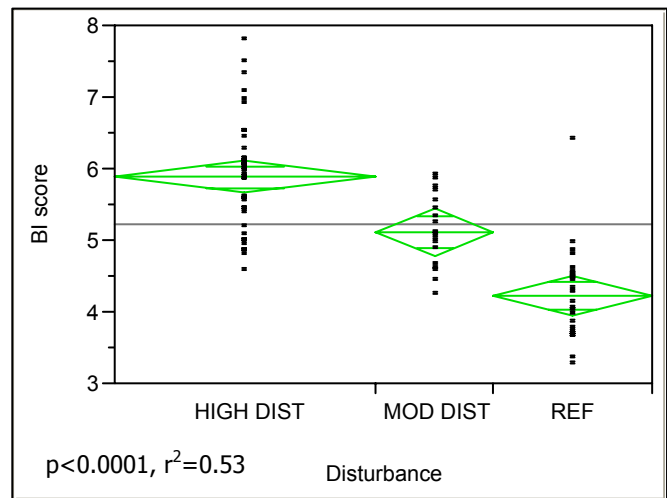


Figure 7: ANOVA Comparison of Biotic Index Score at UNDIST (i.e., REF), MOD DIST, and HIGH DIST Reaches

Overall, 55 of the 59 biological metrics evaluated had significant differences between the UNDIST, MOD DIST, and HIGH DIST groups, many with $p < 0.0001$. The only metrics evaluated that did not have significant differences between study reach groups were BMI density, percent collector-gatherers, percent collector-filterers, and percent collector-gatherers + collector-filterers. Metrics with especially strong negative responses to human disturbance included several diversity metrics, sensitivity metrics such as percent EPT and percent sensitive BMIs, some of the functional feeding group metrics including percent predators, percent shredders, and percent predators + shredders, and individual taxa such as Leptophlebiidae, Heptageniidae, Plecoptera, and Tricoptera. Among the metrics having especially strong positive responses to disturbance were biotic index score, percent dominant taxon, percent two dominant taxa, percent non-insects + Diptera, and percent non-insects + Chironomidae. For some metrics such as Plecoptera and Leptophlebiidae, $\log(y+1)$ transformations of metrics provided better statistical results (i.e., higher r^2 and lower p), while in other cases the opposite was true (e.g., Diptera and Chironomidae) or in still other cases the log transformations had little effect (e.g., percent non-insects + Diptera and percent two dominant taxa). With the exception of field measurements of insect order and family diversity, all metrics with significant responses to human disturbance were included in the multiple regression analyses (see below).

Although not considered for the IBI, insect order and family diversity field metrics (i.e., based on observations made during the field surveys) were among the most sensitive to human disturbance. These field metrics appear to be promising for potential incorporation into an IBI or other assessment method such as the County's Hydrogeomorphic Assessment (HGM) protocol. However, more testing will be necessary to determine whether a protocol can be

established that allows these metrics to be reliably and determined in the field by non-taxonomists. Field identification of BMIs is much more difficult than doing so in the laboratory where one has access to a microscope and detailed taxonomic guides.

Natural Relationships with Physiochemical Parameters

Table 3 provides the results of the physiochemical parameter correlation analysis for the UNDIST sites (n=23). Based on the results, four physiochemical parameters were eliminated from use in the multiple regressions due to high correlations (i.e., above 0.5) with other parameters:

- Gradient (correlated with elevation and watershed area)
- Q (correlated with stream order and watershed area)
- Watershed area (correlated with stream order, gradient, wet width, and Q)
- Conductivity (correlated with specific conductance),

After elimination of these physiochemical parameters, six remained for use in the multiple regression analyses: order, elevation, wet width, temperature, pH, and specific conductance. None of these parameters had correlations of 0.5 or higher between each other.

	Order	Elev. (ft)	Gradient	Wet width (ft.)	Q (cfs)	Wshed area (ac)	Temp. (C)	pH	Cond (microS)	Sp Cond
Order	1	-0.305	-0.3331	0.4611	0.5483	0.5781	-0.0493	0.1778	-0.3599	-0.3556
Elev. (ft)	-0.305	1	0.5088	-0.1185	-0.3674	-0.3832	0.1058	-0.2408	-0.274	-0.3103
Gradient	-0.3331	0.5088	1	0.0341	-0.4855	-0.5579	-0.0004	-0.0354	0.2619	0.2591
Wet width (ft.)	0.4611	-0.1185	0.0341	1	0.4211	0.6782	-0.2327	0.3687	-0.1044	-0.0547
Q (cfs)	0.5483	-0.3674	-0.4855	0.4211	1	0.6064	-0.1531	0.452	-0.3225	-0.2904
Wshed area (ac)	0.5781	-0.3832	-0.5579	0.6782	0.6064	1	-0.3806	0.3308	-0.3262	-0.2243
Temp. (C)	-0.0493	0.1058	-0.0004	-0.2327	-0.1531	-0.3806	1	-0.3236	0.1862	-0.1043
pH	0.1778	-0.2408	-0.0354	0.3687	0.452	0.3308	-0.3236	1	0.2234	0.3185
Cond (microS)	-0.3599	-0.274	0.2619	-0.1044	-0.3225	-0.3262	0.1862	0.2234	1	0.9567
Sp Cond	-0.3556	-0.3103	0.2591	-0.0547	-0.2904	-0.2243	-0.1043	0.3185	0.9567	1

After the physiochemical regressors were selected, the multiple regression analyses were conducted to evaluate the nature and strength of natural relationships between biological metrics and the regressors. Results of the multiple regression analyses are summarized in Table A-6 in the Appendix, which lists r^2 and p for each multiple regression, and equation coefficients for relationships between the biological parameters and each individual regressor. As an example, Figure 8 illustrates the multiple regression for EPT diversity, which was significantly related to the whole model (all regressors, $r^2 = 0.56$, $p=0.0223$) and individually to stream temperature ($p=0.0104$).

As shown in Table A-6, 11 of the 53 biological metrics were significantly related ($p \leq 0.05$) to the whole model. Nine other biological metrics were not significantly related to the whole model, but were significantly related to one or more of the individual regressors. All of these biological metrics were eliminated from further consideration as core metrics.

The strongest physiochemical regressors, as measured by the number of significant relationships with biological metrics were (in order) stream temperature (eight significant relationships), specific conductance (six significant relationships), and elevation (four significant relationships).

Core Metric Selection

Based on the results presented above, six core metrics were selected for inclusion in the IBI:

- Insect family diversity
- Percent EPT
- Biotic index score
- Percent sensitive BMIs
- Percent non-insects + Diptera
- Percent predators + shredders

The core metrics were among the most sensitive to human disturbance among all the metrics tested, either increasing or decreasing from HIGH DIST to MOD DIST to UNDIST groups. None had significant natural relationships with physiochemical gradients among the UNDIST sites. Collectively, the core metrics are diversified in that they represent different aspects of community structure including diversity, disturbance sensitivity, and trophic structure. The core metrics are also diversified in that some respond positively to human disturbance (biotic index score and percent non-insects + Diptera) while others respond negatively to disturbance (insect family diversity, percent EPT, percent sensitive BMIs, and percent predators + shredders).

2. Defining Scoring Categories and Ranges for Core Metrics

Scoring ranges were developed for the core metrics using the criteria presented in Methods. The scoring ranges are provided below in Table 4.

Score	# Insect Families	% EPT	Biotic Index Score	% Sensitive BMIs	% Non-Insects + Diptera	% Shredders + Predators
10 (Excellent)	≥26	≥55	≤4.00	≥28	≤30	≥22
8 (Good)	20-25	41-55	4.01-4.74	21-27	31-47	16-21
6 (Fair)	13-19	28-40	4.75-5.48	14-20	48-63	11-15
4 (Poor)	7-12	14-27	5.49-6.22	7-13	64-80	5-10
2 (Very Poor)	≤6	≤13	≥6.23	≤6	≥81	≤4

3. Defining IBI Classifications and Scoring Ranges

IBI classifications and scoring ranges were developed using the criteria presented in Methods, and are provided below in Table 5.

Category	Score Range
Excellent	54-60
Good	48-53
Fair	36-47
Poor	24-35
Very Poor	12-23

4. Testing the IBI

Accuracy and Consistency of IBI Scores and Classifications

Table A-7 lists IBI scores and biological integrity classifications by year for all of the study reaches surveyed during the past four years. The ranges of scores and classifications are also provided for study reaches that have been sampled in multiple years. Based on the criteria provided in Methods, the IBI correctly classified 94 percent of MOD DIST sites (i.e., Fair or Good) and 85 percent of HIGH DIST sites (i.e., Very Poor or Poor). Overall, 88 percent of MOD DIST and HIGH DIST sites were classified correctly. 78 percent of UNDIST sites were classified correctly (i.e., Good or Excellent). The reliability of the IBI in classifying UNDIST reaches (i.e., reference sites) is set at about 75 percent by design, as the 25th percentile of the UNDIST distribution was used as the threshold between Good and Fair.

Overall, the accuracy of the IBI in classifying sites appears to be quite good. Another good sign is that there were not any gross inaccuracies in classifying sites. No UNDIST sites were classified lower than Fair, and no HIGH DIST sites were classified higher than Fair. The HIGH DIST sites that were classified as Fair (i.e., SJ2, MY1, DP1, T2, and R1) are all subject to agricultural impacts only (i.e., negligible urban impacts), and were close to being placed in the MOD DIST group based on habitat assessment score and watershed land use. Last year's study showed that agriculture-impacted streams in the study area were generally less degraded in terms of biological community structure compared to urban-impacted streams.

Biological integrity classifications were in most cases consistent from year to year at study reaches that were surveyed more than once. In several cases (e.g., C1, M1, M2, AT1, SJ3, and GAV1), the classification was the same for all years surveyed. For other reaches sampled in multiple years (e.g., SY1, M3, AB1, AB2, AT2, SA2, SJ1, SJ2, and SO2), classifications varied across two categories (e.g., Fair to Good). A few reaches (e.g., C2, C3, and AH1) had wider variability, but in no cases did a site vary across more than three categories (e.g., Fair to Excellent). Although infrequent, this variability shows a potential for sampling error in the IBI.

In these cases, replicated or repeated sampling could be conducted at the site to determine the correct classification.

There was very little year to year fluctuation in IBI scores at the nine study reaches surveyed in all four years of the study. Mean IBI scores at these nine reaches ranged from low of 35.1 in 2001 to a high of 39.6 in 2000 and 2003. These differences were not significant ($r^2 = 0.02$, $p=0.917$).

The question of how well this IBI would classify streams outside of the study area is intriguing. To begin exploring this, we calculated IBI score for the three sites surveyed in Matilija Creek (MAT1 and MAT2) and Sespe Creek in 2002, all of which were in the UNDIST group. The IBI scores and classifications for these sites are as follows:

MAT1:	IBI score = 44/60	Biological integrity classification = Fair
MAT2:	IBI score = 46/60	Biological integrity classification = Fair
SES1:	IBI score = 32/60	Biological integrity classification = Poor

The scores and classifications for these sites are lower than what one would expect based on an evaluation of physical habitat conditions and water quality, and the presence of sensitive aquatic vertebrates such as steelhead trout. This indicates that this IBI may not be as effective in assessing the biological integrity of streams outside the study area.

Sensitivity to Human Disturbance

ANOVA results indicate highly significant differences in IBI scores between the UNDIST, MOD DIST, and HIGH DIST groups, with exceptionally strong r^2 of 0.68 and $p<0.0001$. All of the group means were significantly different from one another. This indicates the IBI is highly sensitive to changes in the level of human disturbance. Figure 9 illustrates the ANOVA.

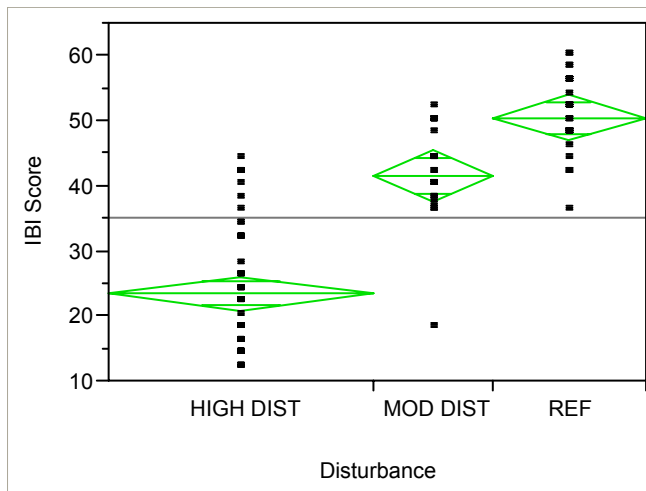


Figure 9: ANOVA Comparison of IBI Score at UNDIST (i.e., REF), MOD DIST, and HIGH DIST Reaches

Natural Relationships with Physiochemical Parameters

Multiple regression analysis results indicate that IBI score was not significantly related to the whole model of physiochemical parameters at the UNDIST reaches ($p=0.1237$). Further, there were not any significant relationships between IBI score and any of the individual regressors. Therefore, IBI scores do not appear to be significantly influenced by natural physiochemical variability in the study area. The consistency of the IBI in equally scoring sites having similar water quality and physical habitat quality but occupying different positions in the landscape is demonstrated by the scoring for SY1 and SY2, both of which are in the HIGH DIST group. SY1 is a low gradient coastal plain reach, while SY2 is a high gradient mountain reach. Despite their different positions in the landscape, both were properly rated in the Poor category by the IBI. Scores for GAV1 and GAV2, both of which are in the MOD DIST category, also demonstrate the consistency of the IBI. GAV1 is a low gradient coastal plain stream, while GAV2 is at higher elevation and gradient in the mountains. Both of these sites were properly rated in the Fair category.

V. Recommendations

The IBI developed in this study appears to be mostly reliable in properly assessing the biological integrity of study area streams, and does not appear to be strongly influenced by natural physiochemical variability. As such, the IBI appears to be an effective assessment tool for study area streams. We recommend that the County and City continue their annual biomonitoring and use the IBI to assess the biological integrity of the study sites. The IBI should be revisited with every two or three years of new data to see if it can be improved by using new core metrics, refining scoring ranges, etc. Another consideration may be to expand the IBI in the future to include core metrics for other assemblages such as aquatic vertebrates (e.g., native vertebrate diversity) and the riparian plant community (e.g., percent native plant species). Alternatively, separate IBIs could be developed for the other assemblages.

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