

Exhibit 46, entered by the California Department of Fish and Game for the State Water Resources Control Board 1987 Water Quality/Water Rights Proceeding on the San Francisco Bay/Sacramento-San Joaquin Delta

The Relationship Between Pollutants and
Striped Bass Health as Indicated
by Variables Measured
from 1978 to 1985

POLLUTANTS AND STRIPED BASS HEALTH

Introduction

The purpose of the Striped Bass Health Monitoring Program is to monitor the condition (as measured by several physical characteristics) of adult striped bass (Morone saxatilis) and to provide baseline data on pollutants in its tissues. The striped bass is a good "indicator species" because it is long lived, resides in the Sacramento-San Joaquin Estuary most of the year throughout its life, and is a top trophic level predator. The program continues work begun by the National Marine Fisheries Service (Whipple 1984) as part of the Cooperative Striped Bass Study (COSBS) (Jung et al. 1984) and uses techniques developed during their work from 1978 to 1983 (Whipple et al. 1984). Knudsen and Kohlhorst (1987) provide a complete account of the most recent years' data as well as a univariate analysis of the cumulative data base.

This report evaluates striped bass health over the first 8 years of the study (1978 to 1985) and looks for consistent relationships between pollutants and health measures that could be used in the development of an annual health index.

Methods

The striped bass health monitoring consists of measuring 17 physical characteristics of mature female bass and concentrations

of potential toxicants in their livers. The toxicants include heavy metals, pesticides, and petroleum by-products (Table 1). Liver concentrations are monitored because pollutants tend to be concentrated by this organ as the result of its function and fat content and yet its fat content is not high enough to interfere with analytical procedures.

Levels of all of the variables being monitored are governed by three factors: first, their response to each other and other persistent features of their environment; second, "capricious" unrelated responses of individual variables to sporadic events that have only temporary effects; and third, random fluctuations in each measure. A technique known as Principal Components Analysis (PCA) (Chatfield & Collins 1980 or Tabachnick & Fidell 1983) was used to help sort the persistent associations which constitute "real patterns" from the sporadic responses which amount to "noise". This technique searches for redundancy in the data (several variables that respond in the same manner) and uses matrix algebra and other mathematical procedures to group closely associated variables into components. The analysis produces as many components as there are original variables used in the data matrix; however, individual variables may appear in more than one component. Each successive component is calculated to remove the maximum remaining variability from the data set while still maximizing the variance of the total component scores. The latter causes each component to be unique and independent. The variables most strongly correlated with each component are those that best

TABLE 1

VARIABLES USED IN THE PRINCIPAL COMPONENT ANALYSES OF POLLUTANT & HEALTH VARIABLES FROM MATURE, FEMALE STRIPED BASS COLLECTED IN THE SACRAMENTO - SAN JOAQUIN DELTA.

Name	Scale	Transformation in PCA on:		Relative Degree of Normality ^{1/}
		All 8 Yrs	1984-85	
Location	0=San Joaquin River 1=Sacramento River	None	None	Binary
Age	Years (interpreted from scales)	Square root	None	Approximately normal or non-normal
STRPtot	Total sum of six sections on one side of the fish for 1=solid & 2=broken striping pattern. The sum of these scores ranged from 6 to 12.	None	None	Approximately normal
RANK2EC	Egg color ranged from yellow to green scored 8 to 16.	Log _e	Log _e	Approximately normal
TAPELARV	Tapeworm larvae abundance ranked from 1 to 5 at each location of occurrence. The variable is the sum of all occurrences. 2=few, 3=average, 4=many, 5=very many/heavily parasitized.	Not Used ^{2/}	Log _e	Approximately normal
TAPERAFI	Tapeworm rafts, scored 2-5 like TAPERARV.	Not Used ^{2/}	Log _e	Non-normal
TAPELESN	Tapeworm induced lesions, scored 2-5 like TAPERARV.	Not Used ^{2/}	Log _e	Non-normal
RNDWLARV	Roundworm larvae, scored 2-5 like TAPERARV.	Not Used ^{2/}	Log _e	Non-normal
TOT_PAR	All parasites combined, scored 2-5 like TAPERARV.	Log _e	Not Used ^{3/}	Approximately normal
MES-FAT	Mesenteric fat abundance rank 1 to 4 where: 1=none, 2=sparse, 3=average, 4=abundant.	None	None	Non-normal
EGGSTAGE	Dominant egg stage in the ovary, 1-11.	None	None	Approximately normal
PERC_RES	% of eggs resorbing, 0-100%	Arcsine-square root	Arcsine-square root	Non-normal
Cd Cr Cu Hg Zn Se	Trace metals in liver in ppm dry weight.	Not Available ^{4/}	Raw, square root, or log _e as needed	Normal or approximately normal
MAH	Monocyclic aromatic hydrocarbons in liver, ppm wet weight.	Log _e	Log _e	Non-normal
AH	Alicyclic hexanes in liver, ppm wet weight.	Log _e	Not used because all values = 0	Non-normal

TABLE 1 (Continued)

Name	Scale	Transformation in PCA on:		Relative Degree of Normality ^{1/}
		All 8 Yrs	1984-85	
LIPID	% Lipid in the liver	Not Available ^{4/}	Arcsine-square root	Approximately normal
TOT_PCB	Total concentration of all forms of PCB in liver, ppm wet weight.	Not Available ^{4/}	Square root	Normal
DDT_MET ^{5/}	The summed concentration of DDT and its metabolic products in liver, ppb wet weight.	Not Available ^{4/}	Log _e	Approximately normal
PESTICID ^{6/}	The summed concentrations of all pesticides in liver, ppb wet weight.	Not Available ^{4/}	Log _e	Approximately normal
TOT_ABN	Sum of all skeletal abnormalities ranked in severity from 1 (least severe) to 5 (most severe) at each location of occurrence.	Log _e	Log _e	Non-normal
KFL	Weight/(fork length) ³ , a standard condition factor for fish.	None	None	Normal
BODYPROP	Body depth behind the operculum divided by fork length.	Square root	None	Normal or approximately normal
IFECUND	Index of fecundity = eggs per fish/weight of the fish.	Square root	None	Normal
GSI	Gonadosomatic index = gonad weight/ fish weight.	None	None	Approximately normal or normal
LSI	Liver somatic index = liver weight/ fish weight.	Arcsine-square root	Arcsine-square root	Approximately normal or normal
TIME	Days since June 1, 1960 a linear time scale over years.	None	Not Used	Non-normal
DAY_IN_Y	Julian day of the year, representing seasonal time trends	None	None	Normal

1/ Relative degree of normality of the raw variable or after the necessary transformation indicated in the two columns at left.

2/ Inconsistently scored over the course of the study.

3/ Redundant with the individual parasite severities used in the 1984/85 analysis.

4/ Not measured in the majority of fish collected between 1978 and 1983.

5/ Includes p,p' -DDT; o,p-, p,p'-DDD; and o,p-, p,p' -DDE.

6/ Includes toxaphene, chlordane, nonachlor, oxychlordane, hexachlorobenzene.

describe it and reflect the most consistent relationships left in the data matrix.

Two issues need to be considered in interpreting the results of PCA: 1) The proportion of the variability in the data matrix explained by each component, represented by its eigenvalue. Components with eigenvalues less than one are usually ignored since they account for less variability in the original data than a single variable. In essence, the object of PCA is to produce a few components that explain most of the variation in the data and certainly more than single variables. Catell's Scree test and an examination of the sorted and shaded correlation matrix are procedures often used to make an even more conservative selection of useful components from those with eigenvalues greater than one. For an explanation of the theory behind these last two techniques and their method of application, see the appendix. 2) The amount of correlation between variables and the most important components. High correlations suggest that a component has adequately reflected persistent associations among the original variables. However, as in any correlation analyses, reasonable explanations for these correlations must exist before a true association (real pattern) is implied. Conversely, high correlations between variables and components with low eigenvalues (components that account for little of the variation in the original data) do not warrant strong conclusions and may only suggest hypotheses for future testing.

RESULTS

Trends for 1978 to 1985

The Principal Components Analysis, over 8 years of data and using 17 variables, exhibited only one strong component. It reflected the sexual maturity of the fish and was unrelated to any pollutant parameters. Hence, no component was strong enough to provide conclusions about the relationship between striped bass health and pollutants. However, seven components each had eigenvalues greater than one and explained more than 5% of the total variance in the data matrix (Table 2). Catell's Scree Test (Figure 1) indicated that three or four of these seven components were important, but the sorted and shaded correlation matrix (Figure 2) indicated that, at most, two strong components existed.

The first component (Table 3) - - the one reflecting sexual maturity - - was highly correlated with the relative size of the ovaries (GSI) and an index of fecundity (IFECUND). A positive, although moderate, association with the variable EGGSTAGE indicates that fishes with greater fecundity and larger ovaries had more mature egg stages. There also is a moderate negative association with the relative size of the liver (LSI) which may represent a normal reduction in the proportional mass of the liver as the fish nears spawning. A negative correlation with the percent of eggs undergoing resorption (PERC_RES) indicates that there is less egg resorption in mature ovaries.

TABLE 2

EIGENVALUES AND PROPORTION OF VARIANCE ASSOCIATED WITH THE UNROTATED COMPONENTS FROM THE PRINCIPAL COMPONENTS ANALYSIS ON POLLUTANT & HEALTH VARIABLES FROM MATURE, FEMALE STRIPED BASS COLLECTED IN THE SACRAMENTO - SAN JOAQUIN DELTA FROM 1978 TO 1985. (N=199)

COMPONENT #	EIGENVALUE	CUMULATIVE PROPORTION OF VARIANCE		ABSOLUTE PROPORTION OF VARIANCE	
		IN DATA SPACE	IN COMPONENT SPACE	IN DATA SPACE	IN COMPONENT SPACE
1	2.9432	0.1731	0.2615	0.1731	0.2615
2	1.9305	0.2867	0.4330	0.1136	0.1715
3	1.5701	0.3790	0.5724	0.0923	0.1394
4	1.3411	0.4579	0.6916	0.0789	0.1192
5	1.2170	0.5295	0.7997	0.0716	0.1081
6	1.1576	0.5976	0.9025	0.0681	0.1028
7	1.0975	0.6622	1.0000	0.0646	0.0975
8	0.8981	0.7150		0.0528	
9	0.8466	0.7648		0.0498	
10	0.7681	0.8100		0.0452	
11	0.6997	0.8511		0.0411	
12	0.6585	0.8899		0.0388	
13	0.5846	0.9243		0.0344	
14	0.4720	0.9520		0.0277	
15	0.3776	0.9742		0.0222	
16	0.2777	0.9906		0.0164	
17	0.1603	1.0000		0.0094	

THE EIGENVALUES ARE FOR EACH COMPONENT BEFORE ROTATION. THE CUMULATIVE OR ABSOLUTE PROPORTION OF VARIANCE IN DATA SPACE IS THE AMOUNT OF VARIABILITY IN THE ORIGINAL DATA ACCOUNTED FOR BY THAT MANY COMPONENTS OR THAT COMPONENT, RESPECTIVELY, AND THE PROPORTION IN COMPONENT SPACE IS THE AMOUNT OF VARIABILITY IN THE PRINCIPAL COMPONENT ANALYSIS SOLUTION ACCOUNTED FOR BY THAT MANY COMPONENTS (CUMULATIVE), OR THAT COMPONENT INDIVIDUALLY (ABSOLUTE).

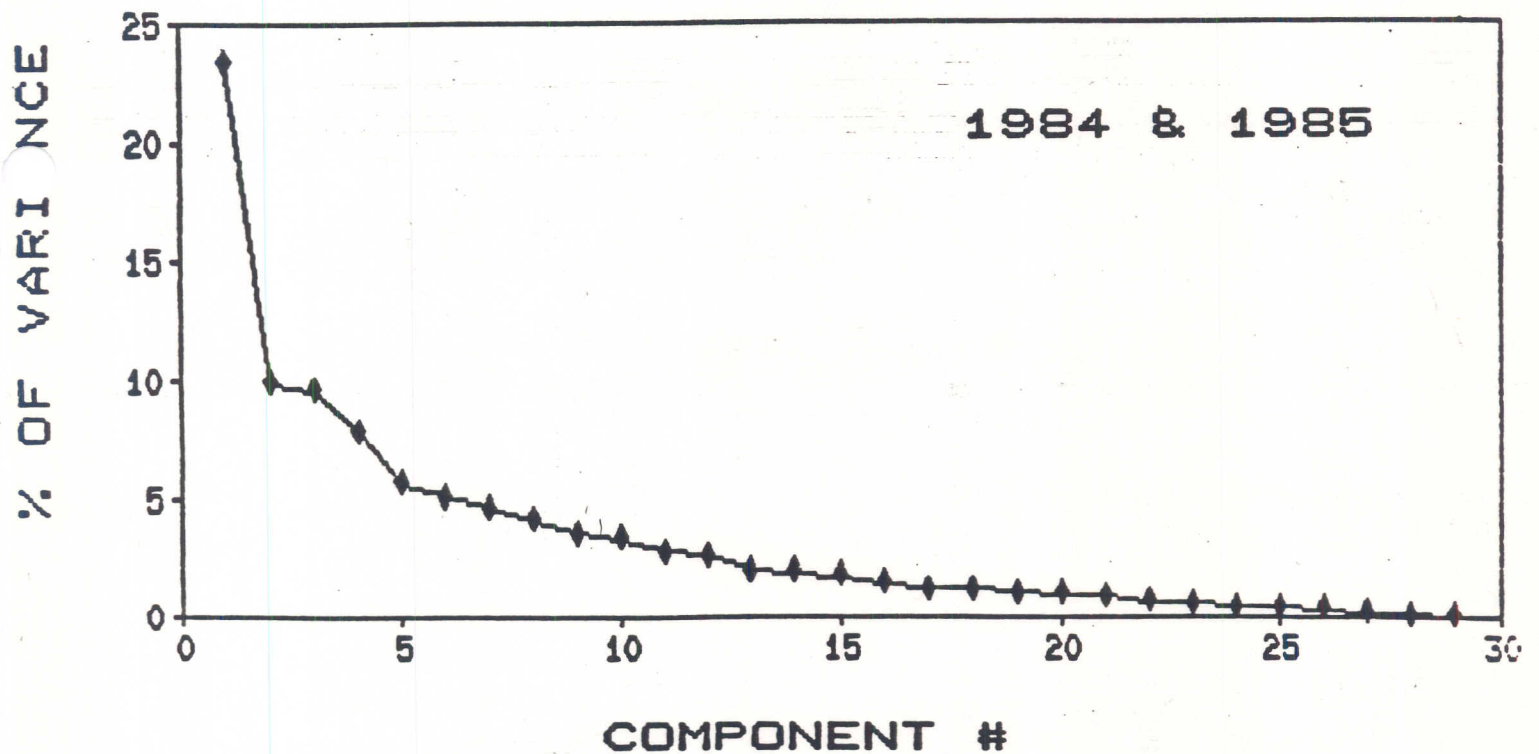
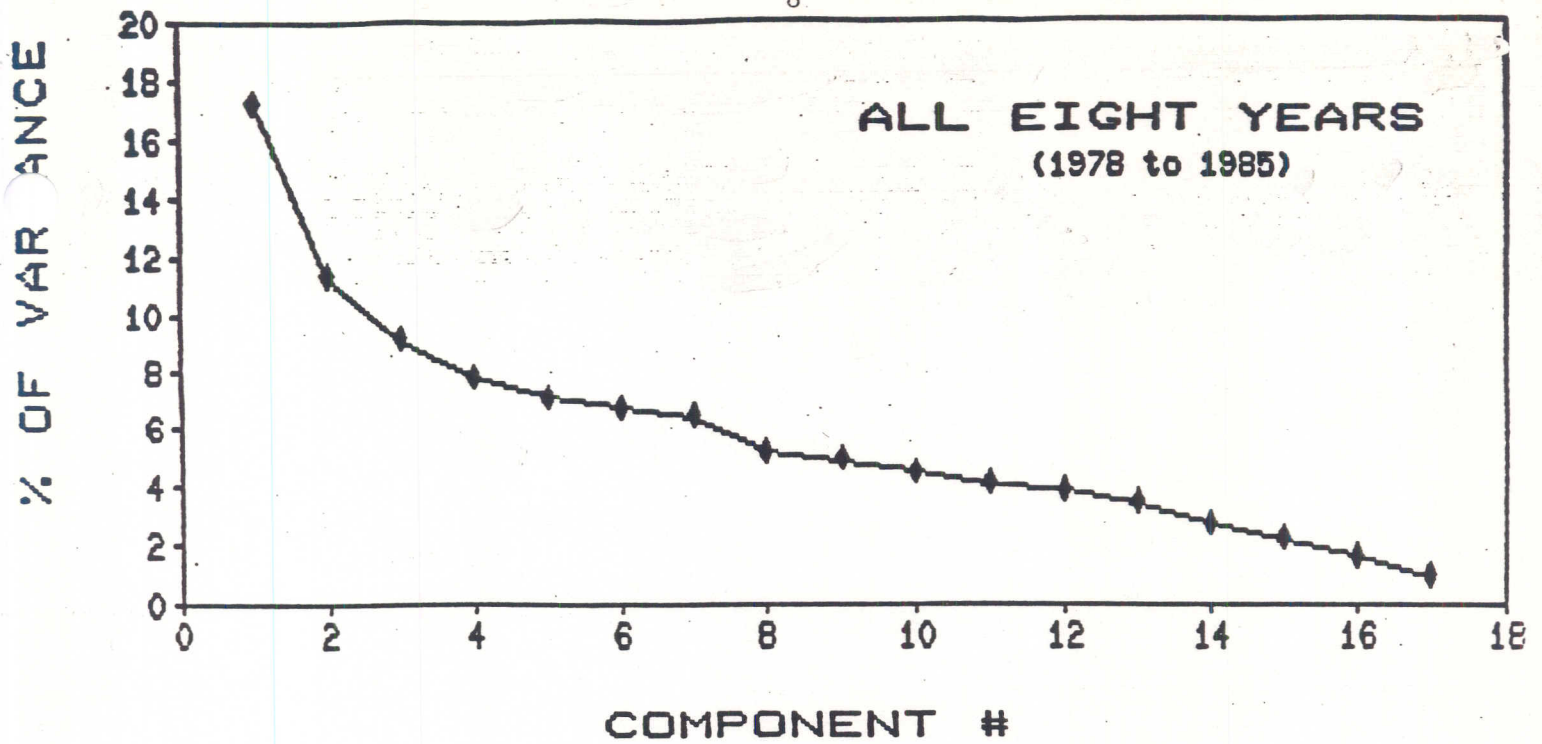


FIGURE 1--CATELL'S SCREE TEST PLOTS OF THE PERCENT OF VARIANCE IN THE RAW DATA ACCOUNTED FOR BY EACH UNROTATED COMPONENT. COMPONENTS ARE FROM ANALYSES ON POLLUTANT AND HEALTH VARIABLES IN MATURE, FEMALE STRIPED BASS FROM THE SACRAMENTO-SAN JOAQUIN DELTA. THE APPROPRIATE NUMBER OF COMPONENTS TO INTERPRET IS LESS THAN OR EQUAL TO THE COMPONENT NUMBER AT WHICH THE CURVE BEGINS AN APPROXIMATELY LINEAR DECLINE (1978-1985: COMPONENT #4; 1984-1985: COMPONENT #5). BELOW THAT POINT, EACH ADDITIONAL COMPONENT SIMPLY ACCOUNTS FOR SIMILAR BUT GRADUALLY DECREASING PROPORTIONS OF THE TOTAL VARIANCE REMAINING IN THE DATA.

TABLE 3
 SORTED ROTATED COMPONENT LOADINGS (PATTERN MATRIX) FOR THE PRINCIPAL COMPONENTS ANALYSIS ON POLLUTANT & HEALTH VARIABLES FROM
 MATURE, FEMALE STRIPED BASS COLLECTED IN THE SACRAMENTO - SAN JOAQUIN DELTA FROM 1978 TO 1985. (N=199)

COMPONENT #	1	2	3	4	5	6	7
COMPONENT NAME	SEXUAL MATURITY	MORPHOLOGY & SEASONAL LIVER CONDITION	SACRAMENTO RIVER FISH	LESS ALYCYCLIC HEXANES THROUGH TIME	OLDER FISH WITH MORE PARASITES	ABNORMALITIES & BROKEN STRIPING PATTERN	MONOCYCLIC AROMATIC HYDROCARBONS
GSI	0.860	0.000	0.000	0.000	0.000	0.000	0.000
IFECUND	0.849	0.000	0.000	0.000	0.000	0.000	0.000
BODYPROP	0.000	0.635	0.000	0.000	0.000	0.000	0.000
KFL	0.000	0.625	0.000	-0.418	0.000	0.318	0.000
DAY_IN_Y	0.000	-0.539	0.000	0.000	0.000	0.000	0.000
LSI	-0.404	0.520	0.416	0.000	0.000	0.000	0.000
LOCATION	0.000	0.000	0.753	0.000	0.000	0.000	0.000
EGGSTAGE	0.433	0.000	-0.664	0.000	0.000	0.000	0.000
TIME	0.000	0.000	0.000	0.769	0.000	0.000	0.000
AH	0.000	0.000	0.000	-0.696	0.000	0.000	0.000
TOT_PAR	0.000	0.000	0.000	0.000	0.764	0.000	0.000
AGE	0.000	0.000	0.000	0.000	0.682	0.000	0.000
TOT_ABN	0.000	0.000	0.000	0.000	0.000	0.792	0.000
STRPtot	0.000	0.000	0.000	0.000	0.000	0.581	-0.447
MAH	0.000	0.000	0.000	0.000	0.000	0.000	0.781
RANK2EC	0.000	-0.487	0.000	0.000	0.000	0.000	0.000
PERC_RES	-0.475	0.000	0.000	-0.334	0.000	0.000	0.444
% OF VARIANCE	13.52	9.87	9.45	9.41	8.14	7.94	7.88

THE ABOVE COMPONENT LOADING MATRIX HAS BEEN REARRANGED SO THAT THE COLUMNS APPEAR IN DECREASING ORDER OF VARIANCE EXPLAINED BY COMPONENTS. THE ROWS HAVE BEEN REARRANGED SO THAT FOR EACH SUCCESSIVE COMPONENT, LOADINGS GREATER THAN 0.5000 APPEAR FIRST. LOADINGS LESS THAN 0.3160 HAVE BEEN REPLACED BY ZERO AS THEY WERE NOT INTERPRETED. THE "% OF VARIANCE" EQUALS THE AMOUNT OF VARIABILITY (VARIANCE) IN THE ORIGINAL DATA ACCOUNTED FOR BY THAT SPECIFIC COMPONENT. A "NAME" WAS ASSIGNED TO EACH COMPONENT BASED ON THE VARIABLES THAT WERE MOST HIGHLY CORRELATED WITH IT, TO HELP THE READER INTERPRET AND IDENTIFY INDIVIDUAL COMPONENTS.

The remaining six components each explain less than 10% of the variance in the data matrix (Table 3), are highly correlated with only a few variables, and are derived from at most one strong and a few weak bivariate correlations. For these reasons, any associations between pollutants and striped bass health exhibited by these components must, at best, be considered as no more than working hypotheses.

Five intermediately correlated variables representing body proportion, condition factor, time in days into the spawning season, relative liver size, and egg color (BODYPROP, KFL, DAY_IN_Y, LSI, RANK2EL) make up the second component (Table 3). However, an examination of the sorted and shaded correlation matrix (Figure 2) and the bivariate correlation matrix (Table 4) indicates that body proportion (BODYPROP) and condition factor (KFL) are not very well related to the others. The two associations between 1) deep body proportion and condition factor (Table 3 and 4), and 2) proportional liver size (LSI), which tends to be greater early in the season, and yellowish egg color (low RANK2EC), which is an indication of immature eggs, are logical redundancies and provide no indication of the relationship between pollutants and fish health.

The third component, like the first, reflects the relationship between larger proportional liver size (LSI) and a less mature dominant eggstage in the ovary (EGGSTAGE) and also indicates that sampling in the Sacramento River tended to collect fish with earlier egg stages.

TABLE 4

CORRELATION MATRIX FOR THE PRINCIPAL COMPONENTS ANALYSIS ON POLLUTANT & HEALTH VARIABLES FROM MATURE, FEMALE STRIPED BASS COLLECTED IN THE SACRAMENTO - SAN JOAQUIN DELTA FROM 1978 TO 1985. (N=199)

	LOCATION	AGE	STRPtot	RANK2EC	EGGSTAGE	PERC_RES	MAH	AH	TOT_PAR	TOT_ABN	KFL	BODYPROP	IFECUND
LOCATION	1.000												
AGE	0.137	1.000											
STRPtot	-0.049	-0.131	1.000										
RANK2EC	0.040	0.051	0.075	1.000									
EGGSTAGE	-0.221	0.029	0.122	0.034	1.000								
PERC_RES	-0.090	-0.062	-0.110	-0.142	-0.302	1.000							
MAH	-0.129	0.128	-0.070	-0.067	-0.023	0.192	1.000						
AH	0.046	-0.042	-0.079	0.010	-0.002	0.153	0.025	1.000					
TOT_PAR	0.050	0.207	-0.010	-0.037	0.016	-0.250	-0.049	-0.136	1.000				
TOT_ABN	-0.016	0.183	0.144	0.059	0.116	0.108	0.152	0.015	-0.088	1.000			
KFL	-0.146	0.026	0.016	-0.115	0.163	0.131	-0.010	0.117	-0.044	0.169	1.000		
BODYPROP	0.005	0.150	-0.079	0.123	-0.043	-0.013	0.086	0.029	0.053	0.067	0.301	1.000	
IFECUND	0.148	0.109	-0.037	0.049	0.110	-0.366	-0.087	-0.070	0.031	0.009	-0.004	0.026	1.000
GSI	-0.122	0.250	-0.047	0.135	0.546	-0.350	0.020	-0.053	0.030	0.105	0.127	0.015	0.643
LSI	0.104	-0.145	-0.114	-0.195	-0.554	0.357	-0.006	0.082	-0.122	-0.074	0.225	0.046	-0.207
TIME	0.152	-0.032	0.055	0.062	-0.119	-0.335	-0.022	-0.296	0.114	0.022	-0.339	0.035	0.356
DAY_IN_Y	0.032	0.049	0.077	0.118	0.206	-0.087	0.079	-0.056	-0.023	0.156	-0.079	-0.137	0.157
	GSI	LSI	TIME	DAY_IN_Y									
GSI	1.000												
LSI	-0.447	1.000											
TIME	0.062	-0.115	1.000										
DAY_IN_Y	0.196	-0.405	0.018	1.000									

Components 4 and 7, respectively, show associations between alicyclic hexanes or monocyclic aromatic hydrocarbons and increased egg resorption. However, because the correlations among the individual variables are so small (Table 4) and the components account for such a small proportion of the total variability in the data (Table 3), a conclusion that the accumulation of these compounds in fish affects resorption is not warranted. These results only point to hypotheses for future testing.

The fifth component shows an increase in total parasites with the age of the fish, but doesn't associate this increase with any pollutant variables.

Component number six indicates that the total number of skeletal abnormalities, composed mostly of abnormalities in the gill rakers, is associated with fishes having a more broken striping pattern and that there is a slight positive relationship between more robust fish (large KFL) and skeletal abnormalities. There is no indication of pollutant effects.

Trends for 1984 and 1985

In the most recent 2 years of the health monitoring, many more pollutant variables were measured in all fish sampled than in the first 6 years. Hence, the more recent data yield more insight about the relationship between pollution and fish health. Of course, the conclusions drawn from these 2 years of data reflect only what happened in 1984 and 1985. It remains to be

seen whether the 1986 and 1987 data, which we have collected and are processing, support these trends.

Eight components were interpretable and had eigenvalues greater than one (Table 5) and each explained more than 5% of the total variance after rotation (Table 6). According to Catell's Scree Test (Figure 1), the first four or five components should be interpreted and an inspection of the sorted and shaded correlation matrix confirmed that, at most, there were five strong components (Figure 3). The first five components had three or more correlations with the original variables that were greater than 0.50 (Table 6). Three of the first five components demonstrated some degree of pollutant-fish health relationships. The sixth, seventh, and eighth components represented very weak relationships that also are presented here simply as potential working hypotheses.

As in the analysis based on all 8 years, the first component (Table 6) is a sexual maturity component, and again, there is no evidence of pathological problems caused by pollutants despite associations with zinc, copper, cadmium and selenium in the liver. Percent egg resorption is now positively associated with sexual maturity rather than negatively as in the analysis over 8 years, but the relationship is still weak. There is a slight tendency for a decrease in percent lipid in the liver with increasing maturity of the ovaries. Variables representing increasing sexual maturity are also slightly associated with collections made later in the season. The second component (Table 6) represents a relationship between pesticides, DDT and its metabolites, total

TABLE 5

EIGENVALUES AND PROPORTION OF VARIANCE ASSOCIATED WITH THE UNROTATED COMPONENTS FROM THE PRINCIPAL COMPONENTS ANALYSIS ON POLLUTANT & HEALTH VARIABLES FROM MATURE, FEMALE STRIPED BASS COLLECTED IN THE SACRAMENTO - SAN JOAQUIN DELTA IN 1984 & 1985. (N=74)

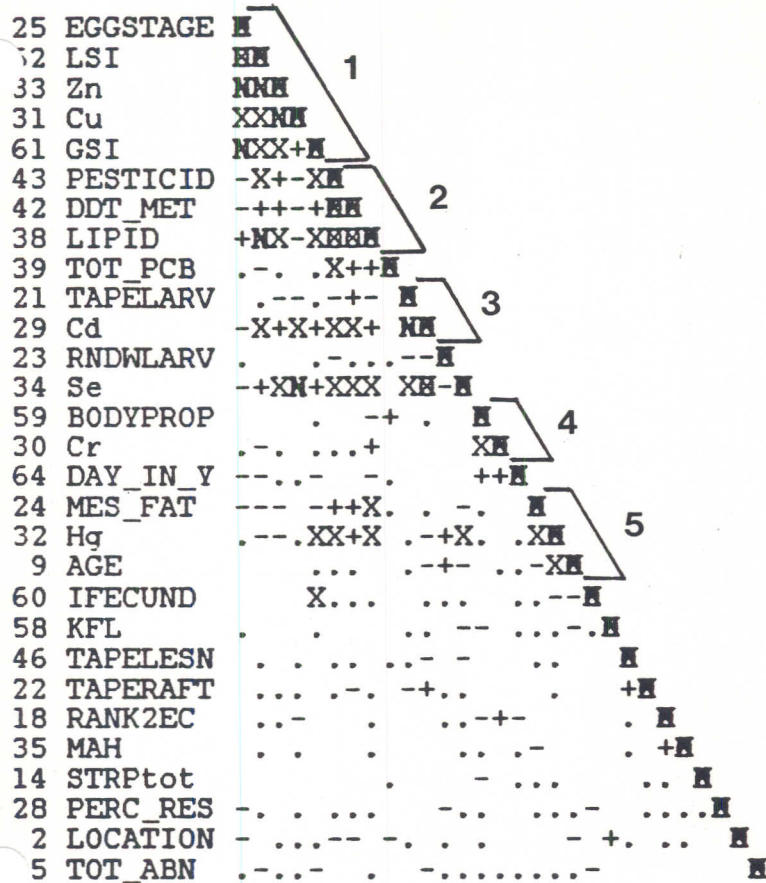
COMPONENT #	EIGENVALUE	CUMULATIVE PROPORTION OF VARIANCE		ABSOLUTE PROPORTION OF VARIANCE	
		IN DATA SPACE	IN COMPONENT SPACE	IN DATA SPACE	IN COMPONENT SPACE
1	6.8108	0.2349	0.3309	0.2349	0.3309
2	2.8981	0.3348	0.4716	0.0999	0.1407
3	2.7935	0.4311	0.6073	0.0963	0.1357
4	2.3164	0.5110	0.7199	0.0799	0.1126
5	1.6768	0.5688	0.8013	0.0578	0.0814
6	1.5009	0.6206	0.8742	0.0518	0.0729
7	1.3681	0.6677	0.9407	0.0471	0.0665
8	1.2208	0.7098	1.0000	0.0421	0.0593
9	1.0591	0.7464		0.0366	
10	0.9937	0.7806		0.0342	
11	0.8161	0.8088		0.0282	
12	0.7763	0.8355		0.0267	
13	0.5948	0.8561		0.0206	
14	0.5741	0.8759		0.0198	
15	0.5262	0.8940		0.0181	
16	0.4341	0.9090		0.0150	
17	0.3802	0.9221		0.0131	
18	0.3702	0.9348		0.0127	
19	0.3174	0.9458		0.0110	
20	0.2994	0.9561		0.0103	
21	0.2808	0.9658		0.0097	
22	0.2146	0.9732		0.0074	
23	0.1948	0.9799		0.0067	
24	0.1540	0.9852		0.0053	
25	0.1479	0.9903		0.0051	
26	0.1196	0.9944		0.0041	
27	0.0861	0.9974		0.0030	
28	0.0475	0.9990		0.0016	
29	0.0276	1.0000		0.0010	

THE EIGENVALUES ARE FOR EACH COMPONENT BEFORE ROTATION. THE CUMULATIVE OR ABSOLUTE PROPORTION OF VARIANCE IN DATA SPACE IS THE AMOUNT OF VARIABILITY IN THE ORIGINAL DATA ACCOUNTED FOR BY THAT MANY COMPONENTS OR THAT COMPONENT, RESPECTIVELY, AND THE PROPORTION IN COMPONENT SPACE IS THE AMOUNT OF VARIABILITY IN THE PRINCIPAL COMPONENT ANALYSIS SOLUTION ACCOUNTED FOR BY THAT MANY COMPONENTS (CUMULATIVE), OR THAT COMPONENT INDIVIDUALLY (ABSOLUTE).

TABLE 6
 SORTED ROTATED COMPONENT LOADINGS (PATTERN MATRIX) FOR THE PRINCIPAL COMPONENTS ANALYSIS ON POLLUTANT & HEALTH VARIABLES FROM
 MATURE, FEMALE STRIPED BASS COLLECTED IN THE SACRAMENTO - SAN JOAQUIN DELTA IN 1984 & 1985. (N=74)

COMPONENT #	1	2	3	4	5	6	7	8
COMPONENT NAME	SEXUAL MATURITY	FAT SOLUBLE POLLUTANTS	PARASITES & TRACE METALS	MORPHOLOGY, SEASON & Cr	LEAN OLDER FISH & Hg	FECUNDITY & FISH CONDITION	TAPEWORM LESIONS & RAFTS	YELLOW EGG COLOR & MAH's
EGGSTAGE	0.857	0.000	0.000	0.000	0.000	0.000	0.000	0.000
LSI	-0.806	0.000	0.000	0.000	0.000	0.000	0.000	0.000
Zn	0.742	0.000	0.000	0.000	0.000	0.000	0.000	0.000
Cu	0.685	0.000	0.000	0.000	0.000	0.000	0.000	0.418
GSI	0.638	0.000	0.000	0.000	0.000	0.540	0.000	0.000
PESTICID	0.000	0.854	0.000	0.000	0.000	0.000	0.000	0.000
DDT_MET	0.000	0.842	0.000	0.000	0.000	0.000	0.000	0.000
LIPID	-0.399	0.707	0.000	0.358	0.000	0.000	0.000	0.000
TOT_PCB	0.000	0.688	0.000	-0.351	0.000	0.000	0.000	0.000
TAPELARV	0.000	0.000	0.765	0.000	0.000	0.000	0.000	0.000
Cd	0.455	0.000	0.623	0.000	0.000	0.000	0.000	0.000
RNDWLARV	0.000	0.000	0.592	0.000	0.000	0.000	0.000	0.000
Se	0.383	0.000	0.544	0.000	0.347	0.000	0.000	0.371
BODYPROP	0.000	0.000	0.000	0.872	0.000	0.000	0.000	0.000
Cr	0.000	0.000	0.000	-0.678	0.000	0.000	0.000	0.000
DAY_IN_Y	0.354	0.000	0.000	-0.613	0.000	0.000	0.000	0.000
MES_FAT	0.000	0.322	0.000	0.000	-0.740	0.000	0.000	0.000
Hg	0.000	0.000	0.000	0.000	0.723	0.000	0.000	0.000
AGE	0.000	0.000	0.497	0.000	0.574	0.000	0.000	0.000
IFECUND	0.000	0.000	0.000	0.000	0.000	0.810	0.000	0.000
KFL	0.000	0.000	-0.430	0.000	-0.356	0.573	0.000	0.000
TAPELESN	0.000	0.000	0.000	0.000	0.000	0.000	0.788	0.000
TAPERAFT	0.000	0.000	0.000	0.000	0.000	0.000	0.744	0.000
RANK2EC	0.000	0.000	0.000	-0.370	0.000	0.000	0.000	-0.704
MAH	0.000	0.000	0.000	0.000	0.000	0.000	0.000	0.689
STRPtot	0.000	0.000	0.000	0.000	0.413	0.000	0.359	0.000
PERC_RES	0.429	0.319	0.000	0.000	0.000	-0.462	0.000	0.000
LOCATION	0.000	0.429	0.449	0.000	0.000	0.000	-0.349	0.000
TOT_ABN	0.354	0.000	0.000	0.000	0.000	0.000	0.000	0.000
% OF VARIANCE	14.14	11.86	9.73	8.18	7.99	6.62	6.40	6.06

THE ABOVE COMPONENT LOADING MATRIX HAS BEEN REARRANGED SO THAT THE COLUMNS APPEAR IN DECREASING ORDER OF VARIANCE EXPLAINED BY COMPONENTS. THE ROWS HAVE BEEN REARRANGED SO THAT FOR EACH SUCCESSIVE COMPONENT, LOADINGS GREATER THAN 0.5000 APPEAR FIRST. LOADINGS LESS THAN 0.3160 HAVE BEEN REPLACED BY ZERO AS THEY WERE NOT INTERPRETED. THE "% OF VARIANCE" EQUALS THE AMOUNT OF VARIABILITY (VARIANCE) IN THE ORIGINAL DATA ACCOUNTED FOR BY THAT SPECIFIC COMPONENT. A "NAME" WAS ASSIGNED TO EACH COMPONENT BASED ON THE VARIABLES THAT WERE MOST HIGHLY CORRELATED WITH IT, TO HELP THE READER INTERPRET AND IDENTIFY INDIVIDUAL COMPONENTS.



THE ABSOLUTE VALUES OF
THE MATRIX ENTRIES HAVE BEEN PRINTED ABOVE IN SHADED FORM
ACCORDING TO THE FOLLOWING SCHEME

.	LESS THAN OR EQUAL TO	0.115
-	0.115 TO AND INCLUDING	0.230
+	0.230 TO AND INCLUDING	0.345
X	0.345 TO AND INCLUDING	0.460
N	0.460 TO AND INCLUDING	0.575
H	0.575 TO AND INCLUDING	0.690
H	0.690 TO AND INCLUDING	0.805
H	GREATER THAN	0.805

FIGURE 3.-- SORTED AND SHADED MATRIX REPRESENTING CORRELATIONS BETWEEN POLLUTANT AND HEALTH VARIABLES MEASURED IN MATURE, FEMALE STRIPED BASS FROM THE SACRAMENTO - SAN JOAQUIN DELTA, 1984 - 1985. THIS MATRIX PROVIDES A MEANS OF CONFIRMING THAT THE COMPONENTS CALCULATED BY PCA REPRESENT REAL ASSOCIATIONS BETWEEN VARIABLES. THE HEAVILY SHADED SQUARES SHOW WHICH VARIABLES ARE MOST HIGHLY CORRELATED WITH EACH OTHER. THE HIGHLY CORRELATED VARIABLES TEND TO BE THE SAME AS THOSE COMBINED INTO THE STRONGEST COMPONENTS BY PCA, AND ARE DELINEATED BY THE NUMBERED BRACKETS AT THE RIGHT EDGE OF THE FIGURE. THE LAST SQUARE IN EACH ROW IS ALWAYS THE DARKEST SYMBOL AS IT REPRESENTS A CORRELATION OF 1.0 (CORRELATION BETWEEN VARIABLE LISTED AT START OF ROW AND ITSELF). PROCEEDING ACROSS ANY ROW, THE FIRST SYMBOL INDICATES THE CORRELATION OF THAT VARIABLE WITH THE FIRST VARIABLE IN THE LIST, THE SECOND SYMBOL REPRESENTS ITS CORRELATION WITH THE SECOND VARIABLE IN THE LIST, ETC. SEE THE APPENDIX FOR A COMPLETE EXPLANATION.

PCBs and body fat represented by percent fat in the liver and a slight correlation with mesenteric fat. These relationships are logical as all of these toxicants are fat soluble. The correlations also suggested that these pollutants are weakly associated with an increase in egg resorption. These toxicants were slightly more prevalent in fish from the Sacramento River.

An association between tapeworm larvae, roundworm larvae, and the trace metals cadmium and selenium was found in component three. Tapeworms are more strongly associated with the concentration of the two metals than are roundworms (Table 7). This condition is slightly associated with older fish and fish with a lower condition factor (KFL). This association of tapeworms and trace metals tends to be slightly more common in fish from the Sacramento River. These associations suggest either that increased levels of cadmium and selenium may affect a fishes susceptibility to parasitization or that parasitization may affect uptake or depuration of trace metals. Laboratory experiments are needed to determine which, if either, of these alternatives is correct. The association among the variables also could be by chance.

Component number four indicates that fish with a deeper body form (BODYPROP) have lower concentrations of chromium in their tissues and are more abundant early in the season. Fish with these traits also tended to have slightly more lipid in the liver, slightly lower total PCBs, and slightly more yellowish or immature egg color. Although this component represents associations

between fish health and pollutant variables, there are no obvious functional relationships among the variables.

The fifth component represents an association between lower mesenteric fat abundance and greater concentrations of mercury in older fish. Fish with these characteristics also tended to have slightly higher selenium concentrations, slightly lower condition factors (KFL) and slightly more broken striping pattern. This component suggests that mercury accumulation in older fish may lead to poor physical condition as represented by low mesenteric fat and slightly lower than average weight proportional to their length (KFL).

The remaining components represent very weak trends in the data (Table 6 & 7). However, they present interesting associations among the variables that should be considered as working hypotheses. Component six indicates that fish with greater body-weight-corrected fecundity tend to have slightly larger gonads (GSI), as would be expected; are in slightly better condition (KFL); and have slightly less egg resorption. Component seven shows an expected association between tapeworm lesions and rafts, and indicates that they are slightly more common in fish with a broken striping pattern from the San Joaquin River. In component eight, fish with yellower egg color are slightly associated with monocyclic aromatic hydrocarbons and the trace metals copper and selenium. This last component is the only one of the final three that suggests a relationship between pollutants and fish health, implying that trace metals and monocyclic

aromatic hydrocarbons may be related to more immature (yellow) egg color. Possibly, these compounds cause a delay in or retardation of egg maturation.

SUMMARY AND DISCUSSION

It is unfortunate that stronger relationships among variables could not be demonstrated with the 8-year data set, but the low number of correlations exceeding 0.30 (Table 4) indicates that there were not many strong, consistent relationships among variables. Also, with only two pollutant variables (MAH and AH) monitored consistently in all fish throughout all 8 years of the study, there was not much potential for drawing conclusions about toxicants and fish health. The last 2 years of the study show some trends which, with 4 or 5 years of consistent results might lead to a striped bass "health index" based on egg resorption, condition factor (KFL), trace metal and pesticide concentrations, and degree of parasitization.

Most fish collected after 1978 did not contain either monocyclic aromatic hydrocarbons or alicyclic hexanes above the limits of analytical detection (79% had no MAH, 84%, no AH); therefore, there was little potential for measuring the effects of these chemicals. Considering their high volatility and the rapid depuration rates of monocyclic aromatic hydrocarbons by striped bass (Korn et al. 1976 and Whipple et al. 1981), there may not be much potential for measuring the effects of these compounds in

field-sampled fish. Effects of monocyclic aromatic hydrocarbons and alicyclic hexanes may not be traceable because these chemicals are depurated before the fish are sampled.

Actually, the problem of tracing effects may be common to many toxic compounds. Fish may be exposed to pollutants during any phase of their migration, experience chronic toxic effects, and then excrete most or all of these compounds prior to our sampling.

Egg resorption, a variable likely to reflect the impacts of pollutants on reproduction, was not strongly related to any chemical compound, although it was slightly correlated with the fat soluble pesticides (PCB's and DDT and its metabolites). Resorption was only vaguely related to monocyclic aromatic hydrocarbons over the years (component #7, Table 3), and to general condition of the fish (component #6, Table 6).

With the exception of component 2 from the 1984-85 data set that accounted for 11.86%, the rest of the pollutant-health components, individually, accounted for less than 10% of the variability in the data matrix. While the amount of data available normally would be sufficient to demonstrate strong relationships, perhaps the observed concentrations of pollutants had only very minor or subtle effects, or pollutants were no longer present in fish tissues often enough or in concentrations high enough to relate them to the physical condition of the fish at the time of collection. These effects may not be obvious without more data. Principal Components Analysis is more robust with more than 500 cases, but we only have 74 fish with a full

complement of pollutant measurements and 199 fish with just two pollutant measures. We plan to examine effects of annual sample size through various statistical and boot-strapping methods later this fall.

Our sampling also may fail to detect toxic effects for several other reasons: 1) Measurements may not be precise enough. Many of the variables were non-normally distributed which might degrade the quality of the results of the Principal Component Analysis. Some of the measures are based on ranks which, perhaps, could be better defined or measured differently. 2) Pollutant concentrations in the majority of the fish collected may be below some, as yet, undefined threshold for chronic impacts on striped bass health. 3) We may have failed to measure the most important pollutants since we do not test for all possible compounds. 4) We may have failed to select the best measures of striped bass condition to reflect pollutant impacts.

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APPENDIX 1

Detailed Description of Analytical Methods

Principal Components Analysis uses matrix algebra and eigen analysis to summarize all of the variability in the original data into independent dimensions called components. Principal Components Analysis is used to summarize patterns of variability in the data and not to test for statistical differences. The goal of Principal Components Analysis is to extract the maximum variance from the data set with each component. The first principal component is the linear combination of observed variables that maximally separates cases (data points) by maximizing the variance of their component scores. The second component is formed from variability remaining in the data set after the variance associated with the first component is removed; it is the linear combination of observed variables that extracts maximum variability from the data uncorrelated with the first component. Each component is uncorrelated with every other component and there are as many of them computed as there are original variables used in the analyses (i.e., if eight variables are analyzed, eight components are produced). Since each component is statistically independent, it represents a unique and independent trend among the variables represented in the original data. For a complete explanation of the computations involved in Principal Components Analysis see Chatfield & Collins 1980 or

Tabachnick and Fidell 1983, and refer to Gauch 1982, Pielou 1984, or Pimentel 1979 for its application to biological data.

The use of Principal Components Analysis assumes that most of the variability in the data will be accounted for by relatively few independent trends (components) common to the variables measured, so not all of the components extracted from the data are rotated to enhance their interpretation. Rotation is the process of adjusting the fit of those components chosen for interpretation to the data in order to maximize the correlations between the original variables and these components. The process of rotation also simplifies the interpretation of the analyses by making each variable correlate with as few components as possible. The correlations between the original variables and these new rotated components computed from them are found in what is called the "Pattern Matrix" or the "Loading Matrix" for the rotated components. The interpretation of this matrix allows the statistical analyst to decide which of the original variables were intercorrelated and have common patterns of variability. Matrices for our two analyses are found in Tables 3 and 6. The components were rotated orthogonally to retain the statistical independence of each component and because oblique rotations, which allow components to be intercorrelated, are difficult to analyze and explain. The techniques for choosing which components to rotate and interpret are discussed after the following section on the nature of the raw variables and their transformations to meet the assumptions of Principal Components Analysis.

Transformation to Meet Assumptions

Two different Principal Components Analysis's were examined: one for the data in 1984 and 1985 where all variables were measured on the majority of fish collected, and one over all 8 years of the study using the 17 variables that were measured in all fish in all years. Data screening was performed on each variable using the whole data set if it was to be used in the analysis of the whole study, and separately using only the 1984-85 data if it was to be used in the analysis of those years.

Since Principal Components Analysis does not test for statistical significance, its assumptions are less restrictive than those of most other parametric, multivariate techniques (see Tabachnick & Fidell, 1983). If the technique is being used to simply describe the data, as in this study, and not to test assumptions about the number of components, then even the assumptions of multivariate normality and linear relationships among variables can be relaxed. If variables are univariate and multivariate normal in distribution with linear relationships between them, then the outcome and fit of the analysis is enhanced. To the extent that normality and linearity fail, the result is degraded, but still may be worthwhile.

The assumptions of multivariate normality and collinearity among variables cannot be tested directly. Using variables that are univariate normal (or transformed to approximate it) and eliminating multivariate outliers from the data will probably

assure multivariate normality. If variables were not normally distributed, their scales were transformed to meet or approximate normality. The variables used in the analyses, their scale, and transformations, if any, are shown in Table 1, which also indicates their relative degree of normality and in which Principal Components Analysis they were used. Multivariate outliers were eliminated based on their Mahalanobis distances from the multivariate mean (centroid) of all the data. Nine data cases were eliminated from the analysis of 8 years of data because they had less than one chance in a thousand of being representative of the data as a whole ($p < 0.001$). A stricter criteria ($p < 0.01$) was applied to the smaller data set for 1984-1985 and two cases were eliminated. Bivariate scatterplots of selected variables (transformed if necessary) were used to assess their bivariate normality and implied colinearity. No departure from linearity was obvious so all variables were assumed to be linearly related.

It would be best to use only variables which were continuous and had interval or ratio scales of measurement for Principal Components Analysis, since they provide the most precise measures of variability. However, some of the variables were ranked variables with ordinal scales of measurement. Using these ranked variables is valid, especially when Principal Components Analysis is used to summarize the patterns in the data. Ranked variables are commonly used by psychometricians in Principal Components Analysis and Factor Analysis to test hypotheses and develop theory. Ranked variables were used where it was impractical to

measure on a finer scale or where there was no accepted standard scale of measurement for the feature which we wished to quantify.

The Principal Components Analyses were done on the correlation matrix instead of the sum of squares and cross products matrix because the former is equivalent to analyzing Z-transformed variables. This approach was necessary because all the variables were not in the same units (i.e. counts) and did not have similar variances.

Choosing Components

There are three methods of selecting how many components to extract from the data and rotate for best fit prior to interpretation. 1) At most, only those unrotated components with an eigenvalue ≥ 1 would be selected, because those with eigenvalues < 1 represent less of the variability in the data than a single original variable. 2) Evaluate those components screened by the first method for the percent of total variance extracted by each of them. Components that extract less than 5% of the total variance in the data, especially after rotation, obviously do not contain much meaningful information and could be eliminated from further consideration. Another way of assessing the value of each unrotated component is to plot the percent of variance extracted by each component from the first to the last component as a curve and to use only those components before the point at which the curve levels off. This is known as Catell's Scree Test and is

based on the fact that, after the first few meaningful components have been computed, the rest of the components will tend to account for similar but gradually decreasing proportions of the total variance. 3) A final and conservative evaluation is to examine the sorted and shaded correlation matrix among the variables sorted by classification analysis. This matrix tends to group variables according to their associations and graphically demonstrate strong trends. This matrix is interpreted by qualitatively examining it for the number of strongly related groups of variables. Figure 3 is the best example included in this report and I have labeled the five groups I was able to differentiate in the matrix. The first two groups are obvious and strongly interrelated, with many variables correlated at $r \geq 0.690$. The next three groups are less apparent and based on weaker correlations, often with $r \leq 0.575$. The number of visible groups, based on a qualitative inspection, should equal the number of strong components, and one should not interpret too many more components than this since the standard matrix indicates those groups for which there are very strong bivariate correlations.

Even after selecting components for extraction and rotation with these techniques, it is necessary to evaluate the reliability of each component to ensure a parsimonious interpretation of the results. Components should have three or more variables correlated with them at levels >0.50 for them to be considered reliable. A component with only two variables strongly correlated with it is no better than a bivariate correlation and when only one variable is strongly correlated with it, may simply represent

a variable which is unrelated to any other in the data set. The interpretation of components correlated with less than three variables (at $r > 0.50$) should be done cautiously and be supported by the interpretation of the bivariate correlation matrix. Variables which correlate with components at < 0.316 should probably not be interpreted except with much larger data sets (300-500+ cases).

Reliable components also are most likely to be produced when an analysis has many more data points than variables used in the analysis or components interpreted. Since both data sets are small (8 yrs=199 cases, 1984-85=74 cases), it is important not to interpret too many components or use too many variables. This study has restricted the analyses to the smallest set of meaningful variables possible by combining all monocyclic aromatic hydrocarbons into one variable, and doing the same for pesticides. This method is especially appropriate for the pesticides since they are all intercorrelated and tend to be associated with a similar set of variables.