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Title: Taxonomic Analyses of the Oregon Chub (Oregonichthys crameri)

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The Oregon chub, Oregonichthys crameri, one of Oregon's endemic freshwater fishes, is restricted to the Willamette and Umpqua rivers and their tributaries. Specimens from the two rivers have traditionally been considered the same species. The goal of this study was to re-examine that assumption and determine if the Willamette Oregon chub and the Umpqua Oregon chub represent different taxa.

The Oregon chub were examined through three sets of data: meristic, morphometric, and electrophoretic. Univariate (General Linear Model) and multivariate (Principal Components Analysis and Discriminant Function Analysis) analyses were performed.

Univariate analyses resulted in significant differences between the Willamette and the Umpqua in the meristic characters: precaudal vertebrae, caudal vertebrae, the difference between precaudal and caudal vertebrae,

pelvic fin rays, rudimentary dorsal rays, total dorsal rays, upper procurrent caudal rays, total caudal rays, maxillary barbels, and cephalic lateral line counts; in the morphometric characters: body width, head depth, eye length, and anal base length; and in the electrophoretic character, lactate dehydrogenase in muscle tissue.

Discriminant function analysis based on meristic characters correctly classifies 90% of the Willamette and 65% of the Umpqua specimens. Discriminant function analysis based on morphometric characters correctly classifies over 89% of the Willamette and over 84% of the Umpqua specimens. Descriptions of the Willamette and Umpqua Oregon chub subspecies are given.

Taxonomic Analyses of the Oregon Chub
(*Oregonichthys crameri*)

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INTRODUCTION

The Oregon chub, *Oregonichthys crameri* (Snyder, 1908), is one of Oregon's endemic freshwater fishes and is restricted to the Willamette and Umpqua rivers and their tributaries. Specimens in the two rivers have traditionally been considered the same species. The goal of this study is to re-examine that assumption and determine if the Willamette Oregon chub and the Umpqua Oregon chub represent different taxa. If corroboration of meristic, morphometric, and electrophoretic characters indicate sufficient differences to recognize the Willamette Oregon chub and the Umpqua Oregon chub as different taxa, a strong case could be made for federal and state protection of the rapidly decreasing Willamette Oregon chub.

Although the Oregon chub has been the subject of few studies, the decline of Willamette populations has been known for years (Bond, 1974). The Willamette Oregon chub has been eliminated from most of its historical range with remnant groups currently restricted to approximately 15 miles on the Middle Fork of the Willamette River (Pearsons, 1989). Dimick and Merryfield (1945) suggested that the decline in the numbers of several Willamette fishes may have been due to pollution from industrial and domestic wastes. Pearsons (1989) suggests a number of biological, chemical, and physical causes are responsible for the decline of the Willamette Oregon chub.

Snyder's (1908) original description of O. crameri was based on specimens collected around the turn of the century from Oregon City on the Willamette River (type locality) and from Elk Creek (paratypes) on the Umpqua River, by Frank Cramer and Keinosuke Otaki. Originally placed in the genus Hybopsis, the Oregon chub was the only member known to occur west of the Rocky Mountains and was even then considered very different from the other members of Hybopsis (Snyder, 1908).

Ichthyologists, particularly those who study cyprinids, have debated the position of the Oregon chub relative to other species of Hybopsis for years. Hubbs (In Schultz, 1929) established name the Oregonichthys as a subgenus of Hybopsis. Schultz and Hubbs (1961) treated Oregonichthys as a synonym of Hybopsis, but indicated that it could be ranked as a subgenus. Reno (1969) referred the Oregon chub to the subgenus Oregonichthys and added that "there is little doubt ...that it could be a separate genus." Unpublished osteological work of Cavender (Hubbs, et al. 1974) supported the segregation of H. crameri into a separate genus, Oregonichthys. The Oregon chub is now generally accepted as a single species of the monotypic genus Oregonichthys.

Four previous studies from the literature indicate the possibility of different taxa of Oregon chub in the two river systems. First, biological and geological evidence indicate that other Columbia basin fishes may have been isolated in the Umpqua. It is believed that during the Pleistocene, the Umpqua River captured a portion of the Willamette River (Diller, 1915). The terraced slopes at a low pass across Calapooya Mountain suggest that once the Umpqua flowed through the area making the North Umpqua a branch of the Willamette. This stream capture may have resulted in an exchange and/or isolation of Willamette and Umpqua river fishes, including the Oregon chub. Geographic isolation is considered a necessary step in speciation (Mayr, 1982). Currently, the Umpqua drains from the Cascades through the Coast Range to the Pacific Ocean, and the two river systems continue to remain separate. However, it appears that Pass Creek, of the Umpqua, could flow into the Willamette drainage at the town of Divide before the creek continues its westward flow, providing an avenue of exchange.

Currently, two cyprinid species are thought to have evolved as Umpqua River endemics: the Umpqua dace, *Rhinichthys evermanni* Snyder, 1908, which is similar to *Rhinichthys cataractae* (Valenciennes *In* Cuvier and Valenciennes, 1842), and the Umpqua squawfish, *Ptychocheilus umpquae* Snyder, 1908 which is similar to the northern squawfish, *Ptychocheilus oregonensis* (Richardson, 1836). If the Oregon chub experienced the same history of isolation and degree of selective adaptation as these species pairs, a similar level of taxonomic divergence might be expected.

Second, Bond and Long (Long, 1980) mentioned possible subspecific differentiation of the Willamette and Umpqua populations, although no evidence was provided.

Third, Bond (1974) noted that Umpqua populations of Oregon chub were generally found in faster waters, whereas Willamette populations tend to be found only in slackwaters, an observation that suggests differences in habitat selection in the two rivers.

Finally, Pearsons (1989) in an ecological and behavioral study of Oregon chub kept in aquaria, noted behavioral differences between the Umpqua and Willamette Oregon chubs. Umpqua Oregon chubs tend to stay close to the bottom, rising only to feed. Willamette Oregon chubs, however, are readily found throughout the water column. In the lab, differences in spawning behavior were also noted; Willamette Oregon chub spawned in aquatic vegetation, whereas the Umpqua Oregon chub spawned on or over gravel or other hard substrates.

MATERIALS AND METHODS

This project is designed to evaluate the similarities and differences of Oregonichthys crameri within and between the Willamette and Umpqua River systems based on three data sets: meristic, morphometric, and electrophoretic characteristics; and to determine if the differences merit separate taxonomic status.

Meristic and Morphometric characters

Standard counts and measurements (Hubbs and Lagler, 1964) (with exceptions listed below) were examined on 312 specimens (181 Willamette and 131 Umpqua) (Appendix 1) for meristic and morphometric characters. The specimens were further divided into six areas for within and and between group analyses: a northern, middle, and southern area for each river system was defined (Table 1). The lateralis system was examined on 66 specimens and followed the methods of Illick (1956) (with exceptions listed below). All specimens were fixed in 10% formalin and later stored in 45% isopropanol.

The following meristic characters were counted from radiographs: caudal vertebrae; precaudal vertebrae; total vertebrae; principal dorsal fin rays; rudimentary dorsal rays; total dorsal rays; principal anal fin rays; rudimentary anal rays; total anal rays; upper procurrent caudal rays; upper primary caudal rays; lower primary caudal rays; lower procurrent caudal rays; and total caudal rays. For vertebral counts, the Weberian apparatus was counted as four (4), and the urostyle as one (1). A variable defining the difference between precaudal vertebrae and caudal vertebrae (precaudal minus caudal vertebrae) was also calculated. In the multivariate analyses, this character was coded in the data as precaudal vertebrae minus caudal vertebrae plus one.

Counts of the number of barbels, number of left pectoral fin ray counts, number of left pelvic fin ray, presence of breeding tubercules, and number of pharyngeal teeth were made on the actual specimen.

Pharyngeal teeth counts are listed for each arch (left or the right arch) not the set of teeth. Pharyngeal teeth were collected from the specimens used for electrophoresis (Table 2, Appendix 1).

In the results, barbel counts are given as 0 equals no barbels, 1 equals either a left or a right barbel, and 2 equals both a left and a right barbel.

The numbers of pores in four cephalic lateral line canals were examined (N=66). The canals were the number of supraorbital, infraorbital, preoperculomandibular, and supratemporal. All counts were made on the left side of the head and includes pores at the center of the head.

Morphometric characters measured include: standard length (SL); predorsal length; maximum body depth; maximum body width; caudal peduncle length; least caudal peduncle depth; length of dorsal fin base; length of anal fin base; dorsal fin length; anal fin length; left pectoral fin length; left pectoral fin base length; left pelvic fin length; left pelvic fin base length; head depth; head length; maximum head width; snout length; eye length.

All morphometric characters were measured in millimeters (mm). The characteristics were measured to the nearest .05 mm with dial calipers. Only specimens over 18.0 mm SL were included.

Quantitative characters were analyzed with the General Linear Model (GLM) univariate test which is appropriate for analysis of variance when sample sizes are unequal and compared with Tukey's Studentized Range Test. Tukey's test is also known as the "honestly significant difference test" and provides a means of conducting "multiple comparison" tests. One disadvantage in using multiple comparison tests is that if one of the groups in the comparison has a much smaller sample size (as in the between river analysis in this data set), the analysis may fail to reject the null hypothesis (SAS Institute, 1988). This occurred in the upper procurrent caudal count, rudimentary dorsal ray count, and total dorsal counts analyses. Significance levels are assumed at $P < 0.05$.

Principal components and discriminant function analyses contained in the SAS and Statgraphics software packages were used for multivariate analysis. The meristic and morphometric data were analyzed separately because the morphometric measurements covary with body size and the meristic variables are assumed to be static after a threshold standard length.

Electrophoretic characters

Starch gel electrophoresis, the process of separating proteins with electric currents, is widely used as a means of detecting protein variation. The variation is expressed through polymorphic allozymes and indicates changes in the amino acid sequence of a protein, but the variation does not necessarily produce obvious morphological or behavioral differences.

In addition, electrophoretic results are often verified through genetic breeding experiments. As the Oregon chub have not yet been bred in captivity, breeding experiments were beyond the scope of this study. Instead the purpose of this portion of the study is to determine if the electrophoretic results echo the pattern of variation found in the meristic and morphometric data.

A second purpose for this portion of the study was to estimate the time of separation of the two rivers by calculating the allelic frequency, genetic identity, genetic distance, and time of separation based on the theory of a molecular clock (Nei, 1972).

With the exception of some senescent individuals collected from aquaria, the specimens for this portion of the analysis were collected in July and August, 1987. Approximately 100 specimens were collected by dip nets and push nets, and either immediately frozen on dry ice or transported live to OSU and later frozen at -80 degrees Celsius. Only 88 specimens were actually used in the analysis (Table 2, Appendix 1)). The remainder were damaged, too small to obtain adequate samples, or simply did not produce distinguishable results. Because of the methods of preservation and the decreased quality of the specimens after collecting tissue samples, these specimens were kept separate and not used for any other portion of the study except pharyngeal teeth extraction.

Standard starch gel electrophoresis procedures of Abersold *et al.*, (1987) and Utter *et al.*, (1974) were used. Initially nineteen enzymes were stained (Table 3) and four tissues types were collected: liver, eye, gill, and muscle. Three different buffer stains were used: 1) Ridgeway (RW), a tris, citric acid gel buffer with a pH of 8.5 and a lithium hydroxide boric acid tray buffer pH 8.5 (Ridgeway et al. 1970); 2) MF, a tris, boric acid, EDTA gel and tray buffer, pH 8.5 (Markert and Faulhaber, 1965); 3) AC, an amine citrate gel and tray buffer, pH 6.1 or 7.0. Nomenclature follows The Enzyme Nomenclature (1979).

Some of the loci known to show differential mobilities in other cyprinid species (Ferguson *et al.*, 1980) and included in this study are, acid phosphatase-1 (ACP), alcohol dehydrogenase (ADH); phosphoglucomutase (PGM) and peptidases (TAPEP and DPEP) (Dowling and Moore, 1984).

The presence of significant variation in an enzyme system at a locus can be determined by calculating the allelic frequency in that system. The allelic frequency at a given locus is equal to the proportion all alleles of its electrophoretic mobility and is calculated from the genotype and allele counts.

RESULTS AND DISCUSSION

Meristic characters

Univariate Analysis

Precaudal Vertebrae

Significant differences were noted in the number of precaudal vertebrae between specimens of the Oregon chub in the Willamette and Umpqua Rivers ($P < 0.001$). Frequency percentages for the groups and rivers for this variable are listed in Table 4. The Willamette Oregon chub possess fewer precaudal vertebra (18.36) than the Umpqua Oregon chub (18.86). The mode for Willamette specimens was 18 and the mode for the Umpqua specimens was 19.

In comparing the variation within the rivers, all three Willamette groups are significantly different from all three Umpqua groups. The Willamette group means range from 18.31 in the middle Willamette area (MW), to 18.42 in the northern Willamette area (NW). The Umpqua groups range from 18.82 in the northern Umpqua area to 18.93 in the southern Umpqua area (SU).

Caudal Vertebrae

Significant differences were noted in the number of caudal vertebra between specimens of the Oregon chub in the Willamette and Umpqua Rivers ($P < 0.001$). The Willamette mean is 17.41, and the Umpqua mean is 16.83 (Table 5). The Willamette group means range from 17.39 in the northern Willamette to 17.58 in the southern Willamette. The Umpqua group means range from 16.77 in the middle Umpqua to 16.87

in the northern Umpqua area. All three Willamette groups are significantly different from the three Umpqua groups.

Total Vertebrae

No significant differences exist in the total number of vertebra ($P > 0.65$). The Willamette mean is 35.78; Umpqua mean is 35.69. The range for both groups is 34-37.

Precaudal minus Caudal Vertebrae

The artificial variable which expresses the difference between the number of precaudal vertebrae minus caudal vertebrae is significantly different between the two river systems ($P < 0.001$) (Table 6). The mean for the Willamette specimens is 0.95 with a mode of 0. The Willamette specimens have approximately the same number of precaudal vertebrae and caudal vertebrae; over 68% of the Willamette specimens have a difference of one or less precaudal minus caudal vertebrae. The mean for the Umpqua Oregon chub is 2.03 with a mode of 2. The Umpqua specimens have a longer trunk and possess a greater difference between the number of precaudal vertebrae than caudal; over 80% possess 2 or more precaudal vertebrae than caudal.

Pectoral Fin Rays

No significant differences exist in the number of pectoral fin ray counts between rivers ($P > 0.64$). The Willamette mean is 12.83, and the Umpqua mean is 12.85.

Pelvic Fin Rays

Significant differences exist in the number of pelvic fin rays between rivers ($P < 0.04$). The Willamette Oregon chub have slightly more pelvic fin rays, 7.99, than the Umpqua 7.92 (Table 7). Both rivers have a mode of eight rays. However, when the groups are examined, the only groups which differ significantly are the northern and middle Willamette. The NW group has the lowest mean, 7.73, possibly because it also contains the smallest specimen. The MW group possesses the highest mean, 8.03, because of a few specimens which contain 9 pelvic rays. This is one case in which, at least for the Willamette, the within variation is greater than the between river variation.

Upper Procurrent Caudal Rays

Significant differences exist in the number of upper procurrent caudal rays between the two river systems ($P < 0.0001$). The mode of both rivers is seven (7) and both rivers range from six (6) to eight (8) rays (Table 8). However, upper procurrent caudal rays in the Willamette Oregon chub average 6.99; the specimens have a higher frequency with specimens with seven and eight rays. The Umpqua Oregon chub possess an average 6.65 rays and have a higher frequency of specimens with six rays.

The variation between groups is composed of the middle Umpqua group with the lowest mean, 6.56, is significantly different from the three Willamette groups. The northern Umpqua group with the second lowest mean 6.65, is also significantly different from the southern Willamette group with a mean of 7.03, but not from the NW group which actually has a higher mean, 7.10.

Upper Primary Caudal Rays

No significant differences exist in this character ($P > 0.75$). Both groups possess a mean of 9.97, and a median of ten upper primary caudal rays.

Lower Primary Caudal Rays

No significant differences exist in this character ($P > 0.72$). All Umpqua specimens have nine lower primary caudal rays. The Willamette specimens have an average of nine and a mode of nine rays, but a few specimens have eight or ten rays.

Lower Procurrent Caudal Rays

No significant differences exist in this character ($P > 0.87$). The mean of the Willamette groups is 6.38 and the mean of the Umpqua groups is 6.39. Both river systems range from five to eight rays.

Total Caudal Rays

Significant differences exist in this character between rivers ($P < 0.005$). The Willamette sample mean is 32.35 with a range of 29 to 35 rays. The Umpqua sample mean is 32.01 with a range of 30 to 34 rays (Table 9).

In comparing the groups, there are no significant differences either within the rivers or between. The northern Willamette group has the largest mean, 32.50, but perhaps the sample size of 10 is insufficient to reject it from the northern and middle Umpqua groups which have the lowest means 32.03 and 31.87, respectively.

Rudimentary Dorsal Rays

Significant differences exist in this character between rivers ($P < 0.001$). Both rivers have a range of one to two rays with a mode of one (Table 10). The Willamette average is 1.06 with very few specimens (6.7%) possessing two rudimentary dorsal rays. The Umpqua average is 1.28 with over 39% of the specimens possessing two rudimentary dorsal rays.

The group comparisons showed that middle (mean 1.03) and southern Willamette areas (mean 1.08) are significantly different from the northern (mean 1.31) and southern Umpqua (mean 1.32) groups. The northern Willamette group has the smallest mean, all 11 specimens possess only one rudimentary dorsal rays, but it is not significantly different from the Umpqua groups.

Principal Dorsal Rays

No significant differences exist in this character between rivers ($P > 0.76$). The mean is 9.01 with a range of 9 to 10 rays for both river systems.

Total Dorsal Rays

Significant differences exist in the number of total dorsal rays between the Willamette and the Umpqua Oregon chub ($P < 0.001$). The mean for the Willamette specimens is 10.07, and the mean for the Umpqua specimens is 10.28. The Willamette range for total dorsal rays is 10 to 11, and the Umpqua range is from 10 to 12 rays (Table 11).

The northern Umpqua (mean 10.31) and southern Umpqua (10.35) groups are significantly different from the middle (10.10) and southern Willamette groups. The northern Willamette has the smallest mean, 10.0, but Tukey's test failed to reject it as significantly different from the Umpqua groups.

Rudimentary Anal Rays

No significant differences exist in this character between river systems ($P > 0.13$). The average of the Willamette groups is 1.18 and the average of the Umpqua groups is 1.25. Both groups have a range of 1 to 2 rays and a mean of 1.

Principal Anal Rays

No significant differences exist in this character ($P > 0.17$). The Willamette sample mean is 8.02 and the Umpqua sample mean is 8.00. The Willamette specimens range from 8 to 9 rays and the Umpqua range from 7 to 9 rays.

Total Anal Rays

No significant differences exist in this character ($P > 0.28$). The Willamette mean is 9.22 with a range of 9 to 11 rays. The Umpqua mean is 9.27 with a range of 8 to 10 rays.

Maxillary Barbels

Significant variation occurs with this character between the two river systems ($P < 0.0004$). The sample mean for the Willamette specimens is 1.10; the Umpqua specimens mean is 1.45 (Table 12). Approximately 33% of the

Willamette Oregon chub lack barbels, about 23% have only one barbel, and about 43% have both the left and the right barbel. Approximately 20% of the Umpqua Oregon chub are missing barbels, 15% have only one barbel, and the other 65% have both the left and the right barbel.

The northern (mean 0.60) and middle Willamette (0.94) area groups are significantly different from the middle (1.63) and southern (1.67) Umpqua area groups.

A summary of the group means and ranges for the previous meristic variables examined between the Willamette and Umpqua River systems are included in Table 13. In general, the within river variation is less than the between river variation.

Cephalic Lateral Line Canals

The lateralis system is known to be unique in the Oregon chub (Illick, 1956; Reno, 1969). The superficial neuromasts of *O. crameri* have a green, extracellular structure common in protozoans but unique among teleosts. The extreme variation in the patterns of canal neuromasts did not allow *O. crameri* to be compared with members of the *Hybópsis* genus in Illick's (1956) study. However, the variation suggests that it might be useful for species level differentiation.

The cephalic lateral line canals were analyzed as a separate data set (N=66). Both the Willamette and the Umpqua Oregon chub have the same basic canal structure. The differences occur in the number of pores per canal and in the frequency of whole versus broken canals in the supratemporal canal (Tables 14 & 15).

The supraorbital canal begins ventral to the anterior portion of the nare. After one to three pores, the canal curves and extends posteriorly and beyond the eye. It is not uncommon for the last one or two pores to leave the general curve of the canal and extend laterally, often towards the midline. The Willamette Oregon chub tend to have more supraorbital canal pores (9.4) than the Umpqua Oregon chub (8.8). The Willamette Oregon chub range from 8 to 11 pores and the Umpqua Oregon chub range from 7 to 11. The Willamette Oregon chub also tend to have a broken supratemporal canal more often than Umpqua specimens.

The infraorbital canal begins ventral and anterior to the nare. After the first two or three pores the canal curves posteriorly and extends below and past the eye where the canal then makes a sharp dorsal curve, paralleling the preopercle and joining the postocular commissure. The Willamette Oregon chub have more infraorbital canal pores (12.7) than the Umpqua (10.9). The Willamette Oregon chub range from 11 to 15 pores, and the Umpqua Oregon chub range from 9 to 13. There does not appear to be any pattern in breakage in the infraorbital canal in either river system.

The preoperculomandibular canal begins posterior and lateral to the symphysis of the lower jaw. The canal follows the curve of the preopercle to its end. The Willamette and Umpqua Oregon chub have about the same number of preoperculomandibular canal pores 12.5 and 12.3, respectively. Preoperculomandibular canal pores in the Willamette Oregon chub range from 10 to 15, and in the Umpqua Oregon chub from 10 to 14. In both rivers the Oregon chub tend to have this canal broken with the same frequency.

The left supratemporal pore counts were frequently broken in the Willamette Oregon chub (10-36% per group), but was never broken in the Umpqua Oregon chub. Both rivers had from 3 to 5 supratemporal pores. The Willamette mean is 3.9 and the Umpqua mean is 3.7.

Pharyngeal Teeth

Most of the Willamette and Umpqua Oregon chub possess pharyngeal teeth counts of 1,4 or 4,1 per arch. In the Willamette Oregon chub, the Shady Dell specimens possess the highest frequency of pharyngeal teeth counts of 0,4 or 4,0, (4 of 14). One of the 14 possess a count of 2,4 and the other 9 possess a count of 1,4 or 4,1. In the other Willamette group, Dexter Reservoir, 11 of the 16 specimens possess a count of 1,4 or 4,1. The other 5 specimens possess a count of 2.4 or 4,2.

The Umpqua Oregon chub have less variation in this character. The Umpqua specimens tend to be 1,4-4,1 (39 of the 46 Umpqua counts). Four of the counts were 0,4 or 4,0 and three were 2,4 or 4,2. Even though the Willamette Oregon chub have a higher percentage of specimens with pharyngeal teeth counts not equal to 1,4 or 4,1, a chisquare analysis between rivers show that the calculated X2 value, 3.92, is less than the tabulated value of a X2 corresponding to 2 degrees of freedom, 5.99.

Multivariate Results

In a discriminant analysis of meristic characters, over 90% of the Willamette specimens are correctly classified (152/168) but only 65% of the Umpqua specimens (65/100) are correctly classified (Table 16). When the test is repeated with only the univariate significant variables, the results are about the same (Table 17), suggesting that it is primarily those variables that contain discriminating information.

In a principal components (PC) analysis on the Willamette and Umpqua Oregon chub (N=270), the first principal component accounts for 18% of the total variation and the second PC accounts for 16% of the variation. The other principal components drop off considerably, requiring 11 PCs to account for over 90% of the total variation (Table 18). Figure 1 is a plot of the first two component weights. Caudal vertebrae is heavily weighted on the negative end of PC1, while precaudal vertebrae and the difference between precaudal and caudal vertebrae are at the opposite end. In a scatterplot of the first two principal components (Figure 2), the Willamette Oregon chub generally fall to the left of zero on PC1 while the Umpqua Oregon chub fall to the right of zero, following the univariate pattern of the Willamette specimens possessing more caudal vertebrae and the Umpqua specimens possessing more precaudal vertebrae. Three of the four caudal meristic parameters are clustered together on the positive end of PC2 (Figure 1). Caudal vertebrae and total vertebrae are the only variables with negative loadings on PC2, and they are both fairly close to the origin, indicating low contributions to the discrimination process. As a result, separation along the second principal component is less clear in the scatterplot (Figure 2).

In an attempt to determine further that the Willamette and the Umpqua Oregon chub compose different principal components, a separate analysis was run on the meristic variables for each river system. In separate principal component analyses on the Willamette and the Umpqua Oregon chub, the pattern of variation is similar; PC1 explains 17% of the total variation in the Willamette (Tables 19) and 19% of the variation in the Umpqua (Table 20). In the Willamette all variables load positively on the the first principal component (PC) except caudal vertebrae. Total caudal rays has the highest loading in both river systems. In the Umpqua three of the variables load negatively with caudal vertebrae possessing the highest negative loading and precaudal minus caudal vertebrae possessing the highest positive loading. The second principal component accounts for 14% of the total variation in both the Willamette and the Umpqua. Although the magnitude differs, both rivers have the highest negative loading on precaudal minus caudal counts and the highest positive loading on caudal vertebrae in PC2. The third principal component accounts for approximately 12% of the variation in the Willamette specimens and 13% in the Umpqua. In PC3, the Willamette specimens possess the highest positive loading on rudimentary dorsal rays and the highest negative loading on total caudal rays; the Umpqua specimens have the highest positive loading on the total dorsal rays and the highest negative loading on the total anal rays. The fourth principal component accounts for 10% of the variation in the Willamette and 11% in the Umpqua. In the Willamette the highest positive loading is on total vertebrae and the highest negative loading is on total dorsal rays. In the Umpqua, the highest positive loading on total anal rays and the highest negative loading on precaudal vertebrae. The fifth principal component accounts for 8% of the variation in the Willamette and 9% in the Umpqua. The Willamette specimens possess the highest positive loading on total vertebrae and the highest negative loading on precaudal vertebrae. The Umpqua specimens possess the highest positive loading on total vertebrae counts and the highest negative loading on total caudal followed closely by upper procurrent caudal rays. The sixth principal component accounts for 6% of the variation in the Willamette and 7% in the Umpqua. The Willamette specimens possess the highest positive loading on principal dorsal rays and the highest negative loading on pelvic rays. The Umpqua specimens possess the highest principal anal rays and the highest negative loading on pectoral fin rays. The first six principal components account for only 67% of the total variation in the Willamette River and 72% in the Umpqua River. In both systems, ten principal components are required to account for over approximately 90% of the total variation (Tables 19 & 20).

Morphometric characters

Univariate Analyses

Standard Length

Univariate analyses between rivers show that the Willamette groups are significantly larger than the Umpqua ($P < 0.004$). The exception is the northern Willamette group, with an average standard length of 26.69 mm, and a range of 19.00 mm to 47.40 mm. It is statistically smaller than all other groups. This group, however, has the fewest (12) specimens, and the overall Willamette mean length is still larger, 39.04, than the Umpqua, 36.04, when these small specimens are included. The northern Umpqua group has the second smallest mean, 33.74, with a range of 21.40 to 49.50, is also significantly different from the middle and southern Willamette groups, but also from the southern Umpqua group.

Results of univariate analyses of predorsal length, body depth, head length, head width, caudal peduncle depth, dorsal length, pectoral length, and pelvic length, followed the pattern established by standard length. All morphometric characters are influenced by allometry, as is evident in the principal component analysis (see next section). The variables which did not follow the size-based trend should contain useful shape information.

One way to reduce allometric effects in a univariate analysis is to restrict it to a narrow size range. A subset of specimens 25 - 45 mm standard length were examined (138 Willamette Oregon chub and 108 Umpqua Oregon chub) (Table 21). In this restricted sample size, Willamette specimens still tend to be somewhat larger (37.71) than the Umpqua specimens (36.68), but not significantly so ($P > 0.09$). Unlike the meristic data, the morphometric variables in both the complete data set and the size restricted data set show considerable overlap both within and between river systems.

The results of univariate analyses of specimens from 25 to 45 mm are included below. The variables are divided into four sections: 1.) variables which are the same length in both rivers or insignificantly larger in the Willamette Oregon chub; 2.) variables which are significantly larger in the Willamette Oregon chub; 3.) variables which are insignificantly larger in the Umpqua Oregon chub; and 4.) variables which are significantly larger in the Umpqua Oregon chub.

<u>Variables insignificantly larger in the Willamette Oregon chub</u>

Most of the variables fall into the first category. Given the high correlation of standard length with the other variables (79-94%), most of the variables tend to be larger in the Willamette Oregon chub, but not significantly so.

Predorsal Length

No significant differences exist in predorsal length between the Willamette and the Umpqua Oregon chub ($P > 0.29$). The Willamette mean is 20.23 mm and the Umpqua mean is 19.83 mm.

Body Depth

No significant differences exist in body depth between the Willamette and the Umpqua Oregon chub ($P > 0.09$). The Willamette mean is 9.84 mm and the Umpqua mean is 9.52 mm.

Head Length

No significant differences exist in head length between the Willamette and the Umpqua Oregon chub ($P > 0.89$). The Willamette mean is 9.17 mm and the Umpqua mean is 9.15 mm.

Head Width

No significant differences exist in head width between the Willamette and the Umpqua Oregon chub ($P > 0.90$). The Willamette mean is 5.35 mm and the Umpqua mean is 5.34 mm.

Caudal Peduncle Depth

No significant differences exist in caudal peduncle depth between the Willamette and the Umpqua Oregon chub (P

> 0.99). The mean for both the Willamette and the Umpqua Oregon chub is 3.25 mm.

Pectoral Fin Length

No significant differences exist in pectoral fin length between the Willamette and the Umpqua Oregon chub ($P > 0.67$). The Willamette mean is 6.77 mm and the Umpqua mean is 6.72 mm.

Pelvic Fin Length

No significant differences exist in pelvic fin lengths between the Willamette and the Umpqua Oregon chub ($P > 0.33$). The Willamette mean is 6.01 mm and the Umpqua mean is 5.90 mm.

Pelvic Fin Base Length

No significant differences exist in pelvic fin base lengths between the Willamette and the Umpqua Oregon chub ($P > 0.76$). The Willamette mean is 1.47 mm and the Umpqua mean is 1.46 mm.

Dorsal Fin Length

No significant differences exist in dorsal fin lengths between the Willamette and the Umpqua Oregon chub ($P > 0.99$ mm). The mean for both the Willamette and the Umpqua Oregon chub is 8.66 mm.

Variables significantly larger in the Willamette Oregon chub

The second group, characters which are significantly larger in the Willamette Oregon chub, includes three variables.

Head Depth

Significant differences exist in head depth between the Willamette and the Umpqua Oregon chub. The Willamette Oregon chub have a significantly deeper heads, 5.02 mm, than the Umpqua Oregon chub 4.80 mm ($P < 0.008$).

Eye Length

Significant differences exist in eye length between the Willamette and the Umpqua Oregon chub ($P < 0.004$). The Willamette Oregon chub have larger eyes, 2.76 mm than the Umpqua Oregon chub 2.65 mm.

Caudal Peduncle Length

Significant differences exist in caudal peduncle length between the Willamette and the Umpqua Oregon chub ($P < 0.0001$). The Willamette Oregon chub have significantly longer caudal peduncle lengths, 10.53 mm, than the Umpqua Oregon chub, 9.74 mm.

Variables insignificantly larger in the Umpqua Oregon chub

Snout Length

No significant differences exist in snout length between the Willamette and the Umpqua Oregon chub ($P > 0.10$). However, the snouts of the Willamette Oregon chub, 2.27, tend to be smaller than the snouts of the Umpqua Oregon chub 2.36 mm.

Dorsal Fin Base Length

No significant differences exist in dorsal fin base length between the Willamette and the Umpqua Oregon chub ($P > 0.15$). The Willamette Oregon chub tend to have a slightly shorter dorsal fin base, 5.67 mm, than the Umpqua Oregon chub, 5.85 mm.

Pectoral Fin Base Length

No significant differences exist in pectoral fin base length between the Willamette and the Umpqua Oregon chub ($P > 0.53$). The Willamette chub tend to have a slightly shorter pectoral fin base, 1.63 mm, than the Umpqua Oregon chub of the same general size, 1.65 mm.

Anal Fin Length

No significant differences exist in anal fin lengths between the Willamette and the Umpqua Oregon chub ($P > 0.16$). Although the Willamette Oregon chub, 7.60 mm, tend to have a slightly shorter anal fin, 7.79 mm, than the Umpqua Oregon chub.

Variables significantly larger in the Umpqua Oregon chub:

The fourth and final group is perhaps the most relevant group, variables which are significantly larger in the Umpqua Oregon chub, despite their overall smaller size.

Body Width

Body width is significantly larger in the Umpqua Oregon chub, despite their overall smaller size ($P < 0.01$). The Willamette mean is 5.24 mm and the Umpqua mean is 5.54 mm.

Anal Fin Base Length

Anal fin base length is significantly larger in the Willamette Oregon chub than in the Umpqua Oregon chub ($P <$

0.02). The Willamette mean is 4.31 mm and the Umpqua mean is 4.50 mm.

Multivariate Analyses

In a principal component analysis on all Willamette and Umpqua Oregon chub (N=311), the first PC accounts for over 87% of the total variation but the second PC accounts for only 3% of the total variation (Table 22). The plot of the first two components weights (Figure 3) shows positive loadings for all variables on the first principal component, indicating that the most influencial factor of the morphometric variables is size. In a scatterplot of the first two principal components (Figure 4), there is no clear separation of Willamette and Umpqua Oregon chub. A separate summary analyses PC scores, PC2 shows that the Willamette mean is 0.037 and the Umpqua mean is -0.052. Most of the observations simply fall around those means.

The third PC accounts for 2.10%, and the fourth PC accounts for 1.55% of the total variation (Table 23). PC3 was not very discriminating, but a summary of PC4 scores show better separation with a Willamette mean of 0.215 and a Umpqua mean of -.297. Consequently, a scatterplot of PC4 and PC1 (Figure 5) and a scatterplot of PC4 and PC2 (Figure 6), generally group the Willamette specimens on the positive end and the Umpqua specimens on the negative end of the scale.

In separate principal component analyses on the Willamette and the Umpqua Oregon chub from 25 to 45 mm, the correlation matrices show that standard length is highly correlated to all of the morphometric characters (66 - 94% in the Willamette and 73 - 96% in the Umpqua). Again, the first principal component clearly represents a trend based on size. The eigenvalue for this first principal component accounts for over 76% of the variation in the Willamette groups and over 85% of the variation in the Umpqua groups: the greatest variation of all of other principal components (Tables 23 & 24). The second principal component accounts for about 4% of the variation in the Willamette, and 3% of the variation in the Umpqua. In the Willamette, PC2 has a has high positive loadings on pectoral base length and a high negative loading on body width. In the Umpqua, PC2 has a high positive loading on pelvic base length and a high negative loading on eye length. The PC3 accounts for about 3% of the variation in both the Willamette and the Umpqua. In the Willamette, PC3 has the highest positive loading on pelvic fin base length, and in the highest negative loading is on eye length. In the Umpqua, PC3 has the highest positive loading is on snout length and the highest negative loading is on body width. PC4 accounts for approximately 3% of the variation in the Willamette and 2% in the Umpqua. In the Willamette the highest positive loading is on pelvic base length and the highest negative loading is on pectoral base length. In the Umpqua, PC4 has the highest positive loading on snout length and the highest negative loading on anal fin base length. PC5 accounts for 2% of the variation in 2% of the variation in the Willamette and 1.5% in the Umpqua. In the Willamette, PC5 has the highest positive loading on snout length and the highest negative loading on pelvic base length. In the Umpqua, PC5 has the highest positive loading on eye length and the highest negative loading on anal base length. PC6 accounts for approximately 2% of the variation in the Willamette and 1% in the Umpqua. In the Willamette, PC6 has the highest positive loading is on eye length and the highest negative loading is on anal base length. In the Umpqua, PC6 has the highest positive loading on pelvic base length and the highest negative loading on pectoral base length.

Although the univariate results show considerable overlap in many of the characters, and the principal component plots do not clearly distinguish the populations, the morphometric variables do contain useful discriminating information. In a discriminant analysis of morphological characters for specimens from 25 to 45 mm, over 90% of the Willamette and almost 88% of the Umpqua specimens are correctly classified (Table 25). When the analysis is repeated with all of the specimens (N=310) the results are about the same indicating that it is not the small (or larger) specimens which are consistently misclassified (Table 26).

Electrophoretic Analysis

The interpretation of electrophoretic results is problematic, mainly because only a small number of loci, those with an overall ionic charge, can be examined. Consequently, while the presence of allelic variation may indicate genetic differences, the absence of variation does not signify genetic identity (Ryman and Utter, 1987). In addition, many of the runs were experimental since there is not an established system for conducting electrophoresis on the Oregon chub.

In preliminary runs, gill and eye tissues did not produce any results with any of the buffer systems. It seemed that the mass of those samples was simply too small, even though both eyes and both sets of gills from each specimen were usually used. Occasionally, liver samples would produce resolution, but the results were too irregular to analyze with confidence. Muscle tissue consistently produced good results with all buffers. Since the degree of variation did not vary significantly between buffer systems, the analyses focused on Ridgeway buffer system which consistently produced excellent resolution.

Of the 19 enzymes examined with muscle tissue, nine produced no resolution and, therefore, no apparent variation. Six of the enzymes produced fair resolution, but still no apparent variation. Three of the enzymes produced good resolution and possible rare variation. And in one enzyme there was good resolution with repeatable variation (Table 27).

Lactate dehydrogenase was the only obvious variant in the Oregon chub. Table 28 lists the genotype, allele counts, and allele frequencies in LDH for 88 specimens. LDH is a known tetramer and exists in at least five forms (isozymes) in most animal tissues (Markert and Faulhhaber, 1965). Two of those forms are known to occur in cyprinids: LDH-A is known to occur in muscle tissue; LDH-B is known to occur in brain, heart, and muscle (Buth, 1984). All of the variation in this study occurred at the most cathodal isozyme, designated LDH-A. Both river systems are dominated by a common homozygote (AA) at LDH-A, 31 of 33 in the Willamette, and 35 of 55 in the Umpqua. Heterozygotes in the Umpqua populations (AB) occur toward the anodal end of the gel. The Willamette Ospecimens have fewer heterozygotes (AC), 2/33 than the Umpqua (AB) 13/55. Moreover, the Willamette and Umpqua heterozygotes are phenotypically different from each other, indicating the presence of a different allele (Figure 7). The heterozygote in the Willamettte populations occur toward the cathodal end. As might be expected in a small population with a low frequency of heterozygotes, the Willamettte group lacks alternate homozygotes (CC), while Umpqua group possess seven alternate homozygotes (BB).

Rare variants may exist in PGI, 6PG, and MDH for muscle tissues in the Ridgeway buffer system, but the results are inconclusive due to a small sample size.

Genetic Distance

The genetic distance between two groups of organisms expresses the average number of codon or nucleotide differences per gene. Gene frequency data generated from protein electrophoresis is one method of determining genetic distance. Nei, the founder of the genetic distance measure, suggests that "many" proteins be examined for accurate results (Nei, 1987). Many is not defined, but it is certainly more than one. Avise (1975) analyzed 24 enzymes to calculate genetic distance in minnows. Nevertheless, since only one loci in LDH was the only consistent variant, it is the only one included in the analysis. Another possible problem with the molecular clock hypothesis is that the genetic distance may not be linear, particularly when a bottleneck in population size has occurred (Grant, 1987), as may have occurred in the Willamette Oregon chub.

The genetic distance is calculated by first determining the allele frequencies (Table 28) and the genetic identity (I), where $I = Jxy/(JxJy)1/2$; and Jxy equals the genotype for the common allele for both the Willamette and the Umpqua groups; Jx equals the allele frequency for Willamette genotypes; Jy equals the allele frequency for Umpqua genotypes; JxJy equals the allele frequency for Willamette genotypes times the allele frequency for Umpqua genotypes.

In this case I = .7275/(.9418 x .625)1/2 = 0.9482. The genetic distance then is D = -ln I or D = -ln (.9482) = 0.0532.

Therefore, the time of separation (t) is defined as t = 5 X 106 X D or a time of separation of 266,000 years ago, certainly within the realm of possibility since the Pleistocene began over a million years ago. However, this figure should be viewed with some skepticism, since it is based on limited data.

SUMMARY AND CONCLUSIONS

A problem in determining if speciation has occurred in allopatric forms, is that reproductive isolation, the primary criterion for a species, cannot be tested (Mayr, 1982). A continuum exists from population to race to subspecies to species without a nonarbitrary criteria which would enable a taxonomist to determine if a particular level of difference in a phenetic character was indicative of differentiation at one level or another. For example, in the two Umpqua cyprinid species previously mentioned: R. evermanni is distinguished from R. cataractae because R. evermanni possesses more dorsal rays, more lateral line scales, a longer relative snout length, anal fin height, pectoral fin length, and a shorter caudal peduncle length than R. cataractae. Whereas, P. umpquae differs from P. oregonensis in only one character, scale counts (Snyder, 1908). However, Bond (1973) suggests that the Ptychocheilus are only subspecies.

In general, species status is recognized when the overlap in the range of taxonomics character is small, and the means are statistically different (Mayr, 1969). If the variation between the two river systems is small, with overlapping ranges, the Willamette and Umpqua chubs should remain a single allopatric species. If variation exists but it does not clearly designate species status, the allopatric populations of questionable status should be considered separate subspecies (Mayr, 1982).

Pattern of differentiation of Umpqua endemics

There are five primary fish genera encompassing seven endemic species in the Umpqua River (Minckley et al., 1986). Two of those species are known to exist only in the Umpqua River: *Rhinichthys evermanni* Snyder, and *Ptychochelilus umpqua* (Snyder). With the historical geological evidence of a possible stream capture of part of the Willamette by the Umpqua. the ecological differences between the Oregon chub in the two rivers (Pearsons. 1989), and the apparent success and divergence of the Umpqua chub, I believe the case has been made to recognize a third Umpqua endemic, *Oregonichthys crameri* subspecies.

Formal Description of the Willamette Oregon chub

WILLAMETTE OREGON CHUB

See Figure 8.

Holotype: U. S. National Museum. (US) 61574. 65 mm, Willamette River at Oregon City, F. Cramer and K. Otaki.

Materials examined: See Appendix 1.

Description: Generally less than 60 mm SL. body deep and compressed. caudal peduncle slender. eye round, snout blunt, mouth oblique; golden color with black-speckled scales.

Scales usually present on the ventral surface anterior to the insertion of the pectoral fins (Markle, pers. comm.).

Primary dorsal rays 9; total dorsal counts 9 to 11, usually 10, primary anal rays 8; maxillary barbel often absent; teeth in the minor row sometimes 1, but often absent. Area anterior and posterior to the posterior end of the anal fin about the same.

Diagnosis: Generally longer and more slender than the Umpqua Oregon chub. In the Willamette subspecies, anal fin base length tends to be shorter but eye length, caudal peduncle length, and head depth tend to be larger than Umpqua Oregon chub of the same size.

Usually two, or fewer precaudal vertebrae than caudal vertebrae, (95.6%) (Table 6). Precaudal vertebrae often 19 (51% of the specimens) or 20 (29%) (Table 4). Caudal vertebrae usually 16 (38%) or 17 (40%) (Table 6). Upper procurrent caudal rays usually 7 (71%), rarely 6 (13%) or 8 (15%). Rudimentary dorsal rays usually 1 (92%), rarely 2 (8%) (Table 10). Pectoral fin rays 11-14, usually 13 (55%) but often 12 (33%), 14 (10%), or 11 (2%). Pelvic fin rays usually 8 (79%) but sometimes 7 (11%) or 9 (10%) (Table 7). Willamette Oregon chub lack maxillary barbels in 35% of the specimens; 22% have only one barbel and 43% have both a left and right barbel (Table 12).

The cephalic lateral line canals are generally more developed in the Willamette Oregon chub. Pores in the supraorbital canals range from 8-11 with an average of 9.4. Infraorbital canals range from 11-15 with an average of 12.7. Preoperculomandibular canals range from 10-15 with an average of 12.5. Supratemporal canal pores range from 3 to 5, with an average pf 3.9, and is usually broken (Table 14).

A phenotypically distinct form of lactate dehydrogenase (LDH-A) in muscle tissute differentiates the Willamette Oregon chub from the Umpqua Oregon chub. Also a high level of homozygosity exists for the Willamette Oregon chub with this enzyme.

Distribution: Remnant populations restricted to 18% of its historical sites, approximately 15 miles of the Middle Fork of the Willamette River near Dexter Reservoir (Pearsons, 1989).

Formal Description of the Umpqua Oregon chub

See Figure 9.

Holotype: Oregon State University (OS) 5494; a female; 42.00 mm SL; Cow Creek, 1 mile below Polan Creek., Douglas Co., Oregon, 7 August 1970, Bond and Juntunen.

Materials Examined: See Appendix 1.

Description: The Umpqua Oregon chub is generally similar to the description of the Willamette Oregon chub.

Scales usually absent on the ventral surface anterior to insertion of the pectoral fins (Markle, pers. comm.).

Primary dorsal rays 9, total dorsal count 10 to 12, usually 10, but often 11; primary anal rays 8. Pharyngeal teeth in the minor row usually 1, but often sometimes 2 or absent. Area anterior to the posterior end of the anal fin generally longer than the area posterior to the posterior end of the anal fin.

Diagnosis: Generally shorter standard length but more robust than the Willamette subspecies. Anal fin base length tends to be longer than in the Willamette chub. Eye length, caudal peduncle length, and head depth tend to be smaller in the Umpqua Oregon chub than Willamette Oregon chub specimens of the same (Table 21).

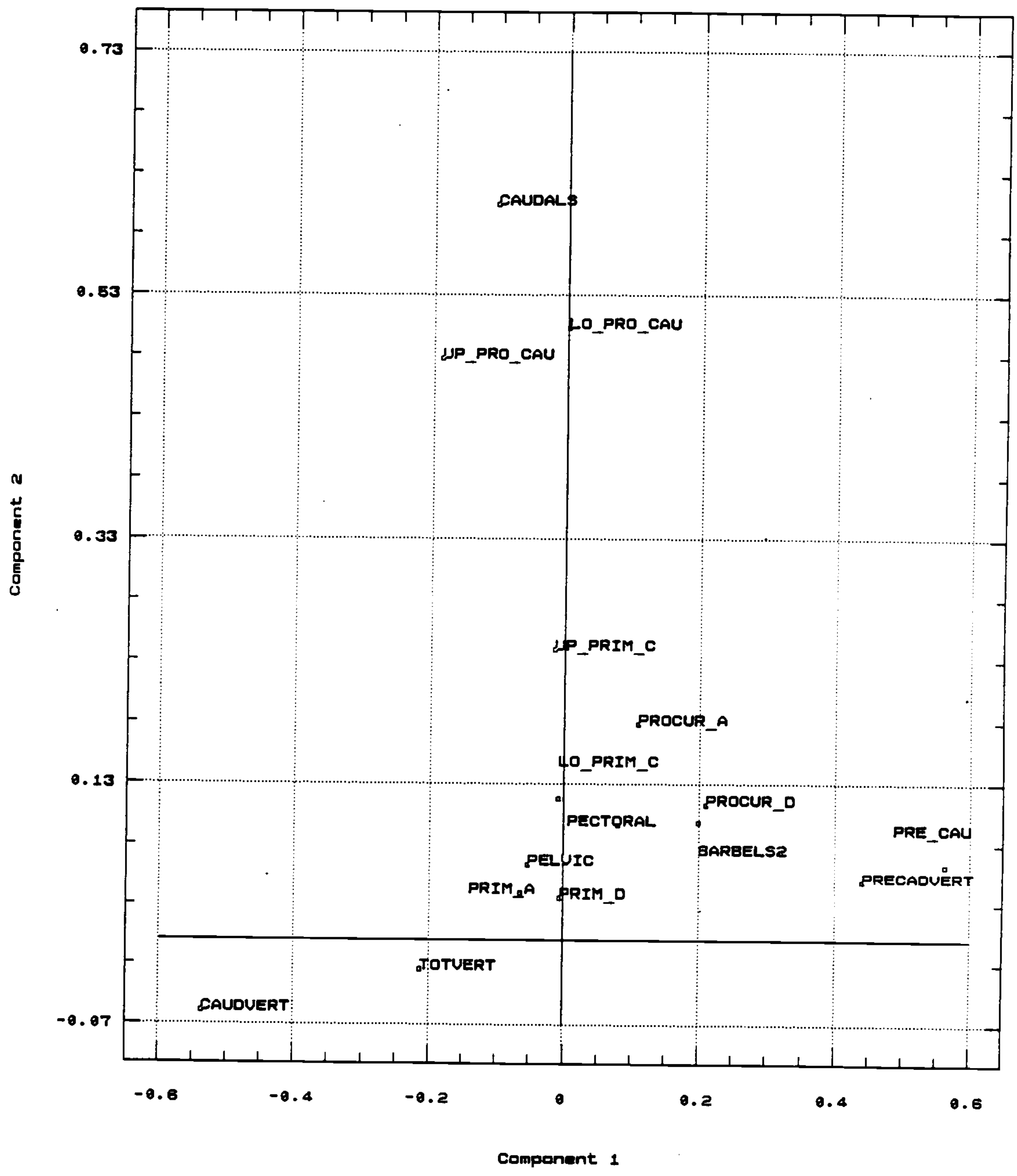
Usually 2 or more precaudal vertebrae than caudal vertebrae, (80.9%) (Table 6). Precaudal vertebrae usually 20 (80%) but sometimes 19 (14%). Caudal vertebrae usually 16 (63%) but sometimes 15 (26%). Upper procurrent caudal rays usually 7 (62%) but sometimes 6 (37%), rarely 8 (0.9%). Procurrent dorsal rays usually 1 (61%) but often 2 (39%). Pectoral rays 12-14, usually 13 (66%) but often 12 (20%) or 14 (14%). Pelvic fin rays 6-8, usually 7 (52%) but often 8 (47%), rarely 6 (0.8%). Most of the Umpqua Oregon chub have both a left and right maxillary barbel, 63%, 16% have only one barbel, and 21% lack barbels (Table 12).

The cephalic lateral line canals are generally not as developed in the Umpqua Oregon chub. Supraorbital canal pores range from 7-11 with an average of 8.8. Infraorbital canal pores range from 9-13 with an average of 10.9. Preoperculomandibular canal pores range from 10-14 with an average of 12.3. Supratemporal canal pores range from 3-5 with an average of 3.7, and is not broken (Table 15).

A phenotypically distinct form of lactate dehydrogenase (LDH-A) in muscle tissue differentiates the Willamette Oregon chub from the Umpqua Oregon chub.
Distribution: Throughout the Umpqua River system, except the North Umpqua. Common in 70% of its historical sites (Pearsons, 1989).

In conclusion, if the Willamette Oregon chub and the Umpqua Oregon chub are to be recognized as different subspecies, immediate steps should be taken to secure state and federal protection of the Willamette Oregon chub and its habitat. In addition, close attention should be given to the Umpqua Oregon chub, to ensure that it does not undergo the same fate as the Willamette Oregon chub.

Figure 1. Plot of first two components weights (PC 1 and PC2) of meristic variables for the Willamette and Umpqua Oregon chub (CAUDALS = total caudal rays; LO_PRO_CAU = lower procurrent caudal rays; UP_PRO_CAU = upper procurrent caudal rays; UP_PRIM_C = upper primary caudal rays; LO_PRIM_C = lower primary caudal rays; PROCUR_A = rudimentary anal rays; PROCUR_D = rudimentary dorsal rays; PECTORAL = pectoral rays; PELVIC = pelvic rays; BARBELS2 = maxillary barbels; PRE_CAU = precaudal minus caudal vertebrae; PRECADVERT = precaudal vertebra; CAUDVERT= caudal vertebrae; TOTVERT = total vertebra; PRIM_A = primary anal rays; PRIM_D = primary dorsal rays).



0.73
0.53
0.33
0.13
-0.07
Component 2
CAUDALS
LO_PRO_CAU
UP_PRO_CAU
UP_PRIM_C
PROCUR_A
LO_PRIM_C
PROCUR_D
PECTORAL
PRE_CAU
BARBELS2
PELVIC
PRECADVERT
PRIM_A
PRIM_D
TOTVERT
CAUDVERT
-0.6
-0.4
-0.2
0
0.2
0.4
0.6
Component 1

Figure 1.

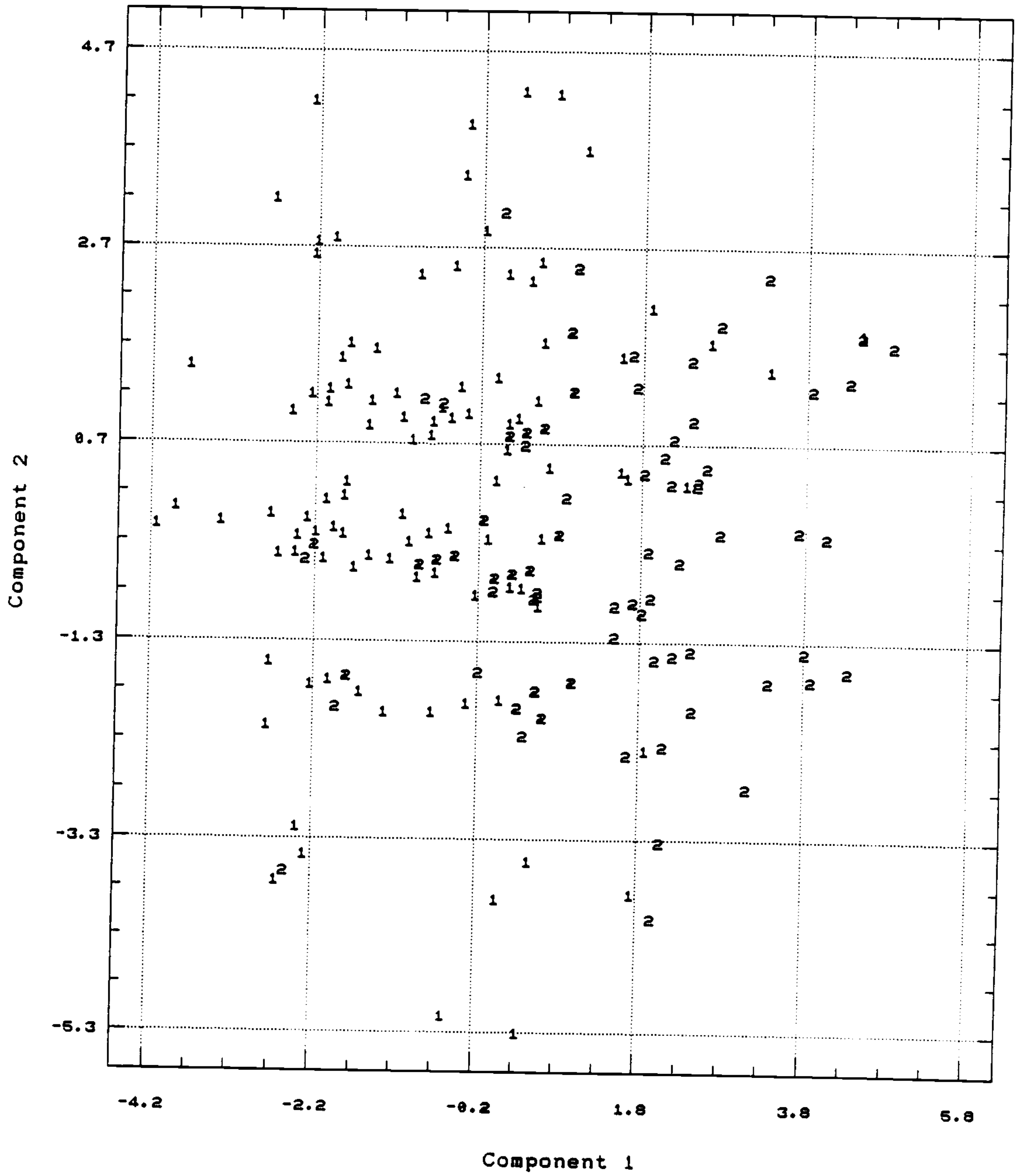


Figure 2. Scatterplot of the first two principal components of meristic variables for the Willamette and the Umpqua Oregon chub (1 = Willamette, N=168; 2 = Umpqua, N=102).

Figure 3. Plot of the first two component weights (PC2 and PC1) of morphometric variables for the Willamette and the Umpqua Oregon chub (PECT_L=pectoral length; P_BASE_L= pectoral base length; PELV_L=pelvic length; PELV_B_L=pelvic base length; DORSAL_L=dorsal length; D_BASE_L=dorsal base length; ANAL_L=anal length; A_BASE_L=anal base length; HEAD_L=head length; HEAD_W=head width; HEAD_D=head_d; SL=standard length; PREDOR_L=predorsal length; BODY_W= body width; BODY_D= body depth).

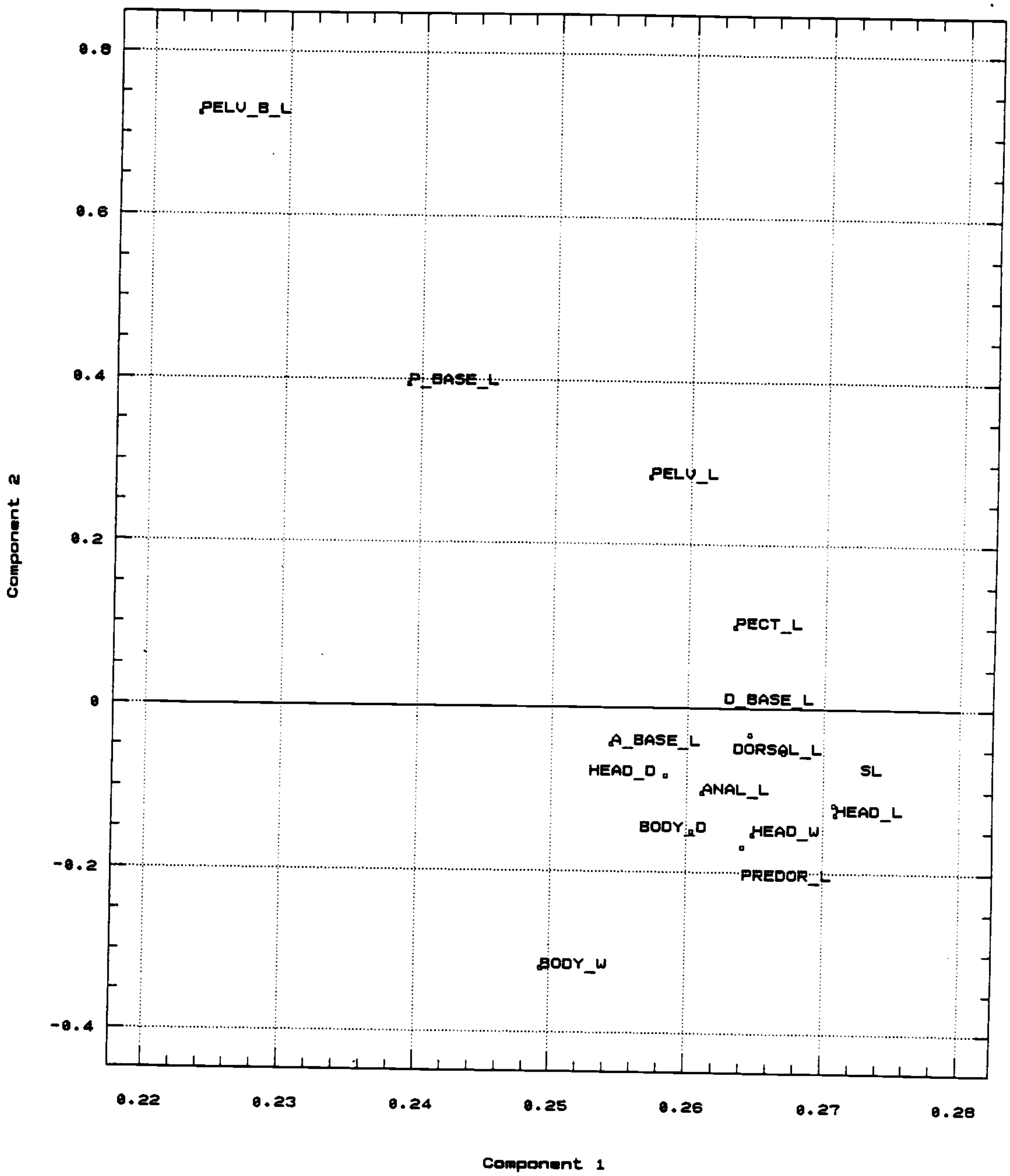

PELV_B_L
P_BASE_L
PELV_L
PECT_L
D_BASE_L
A_BASE_L
DORSAL_L
HEAD_D
SL
ANAL_L
HEAD_L
BODY_D
HEAD_W
PREDOR_L
BODY_W
0.8
0.6
0.4
0.2
0
-0.2
-0.4
Component 2
0.22
0.23
0.24
0.25
0.26
0.27
0.28
Component 1

Figure 3.

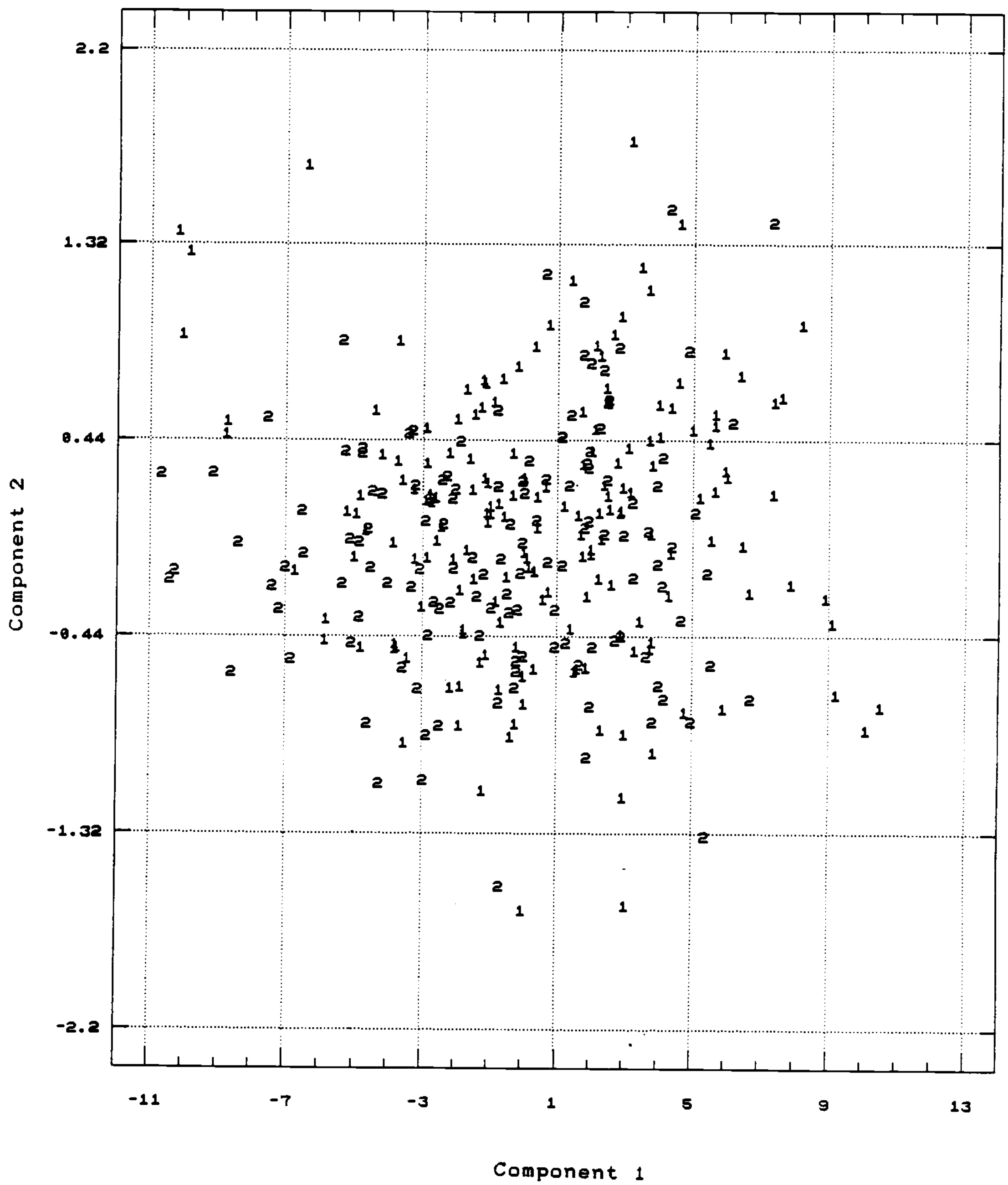


Figure 4. Scatterplot of the first two principal components (PC2 and PC1) of morphometric variables for the Willamette and the Umpqua Oregon chub (1 = Willamette, N=181; 2 = Umpqua, N=131).

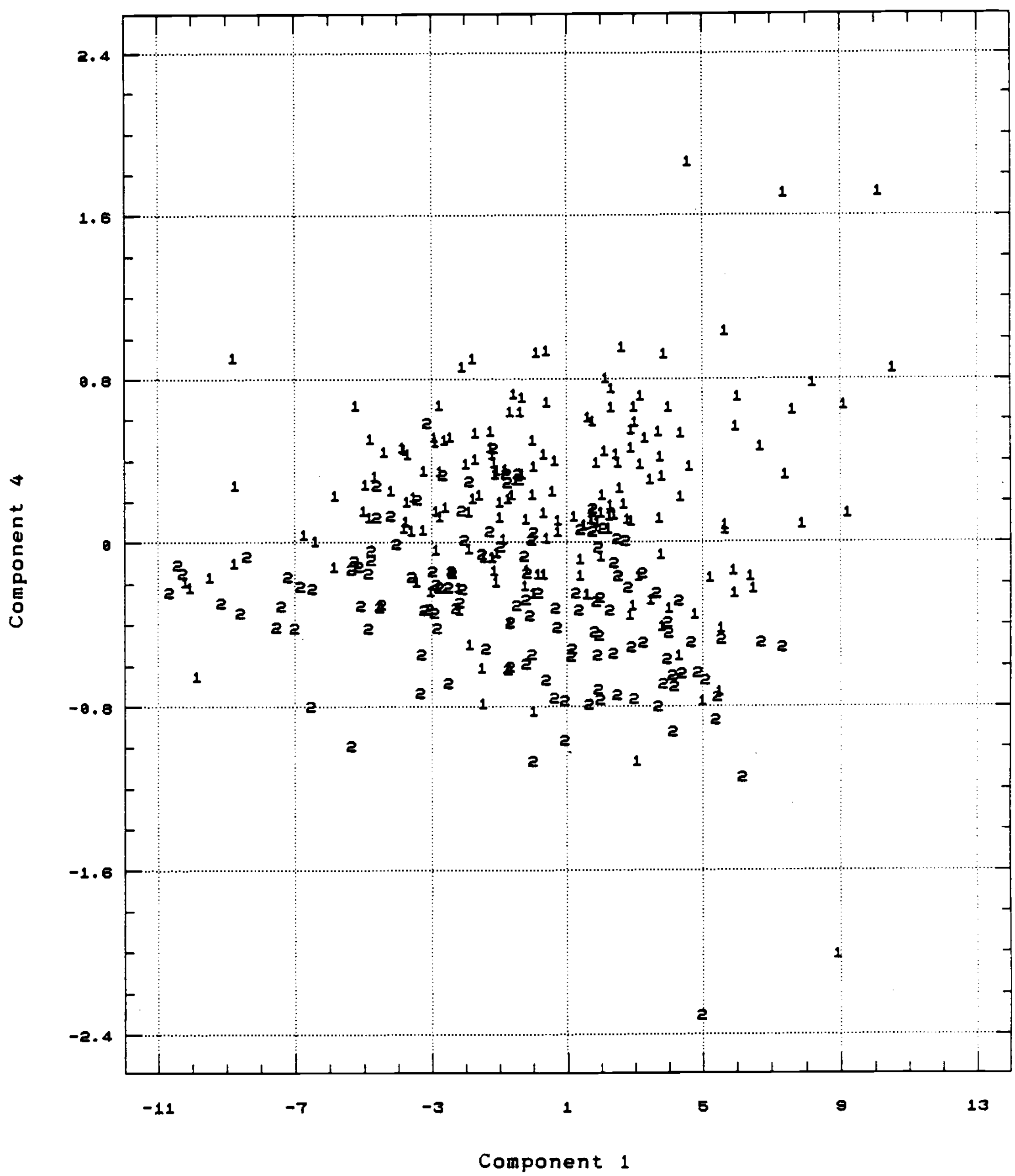


Figure 5. Scatterplot of PC4 and PC1 of morphometric variables for the Willamette and the Umpqua Oregon chub (1 = Willamette, N=181; 2 = Umpqua, N=131).

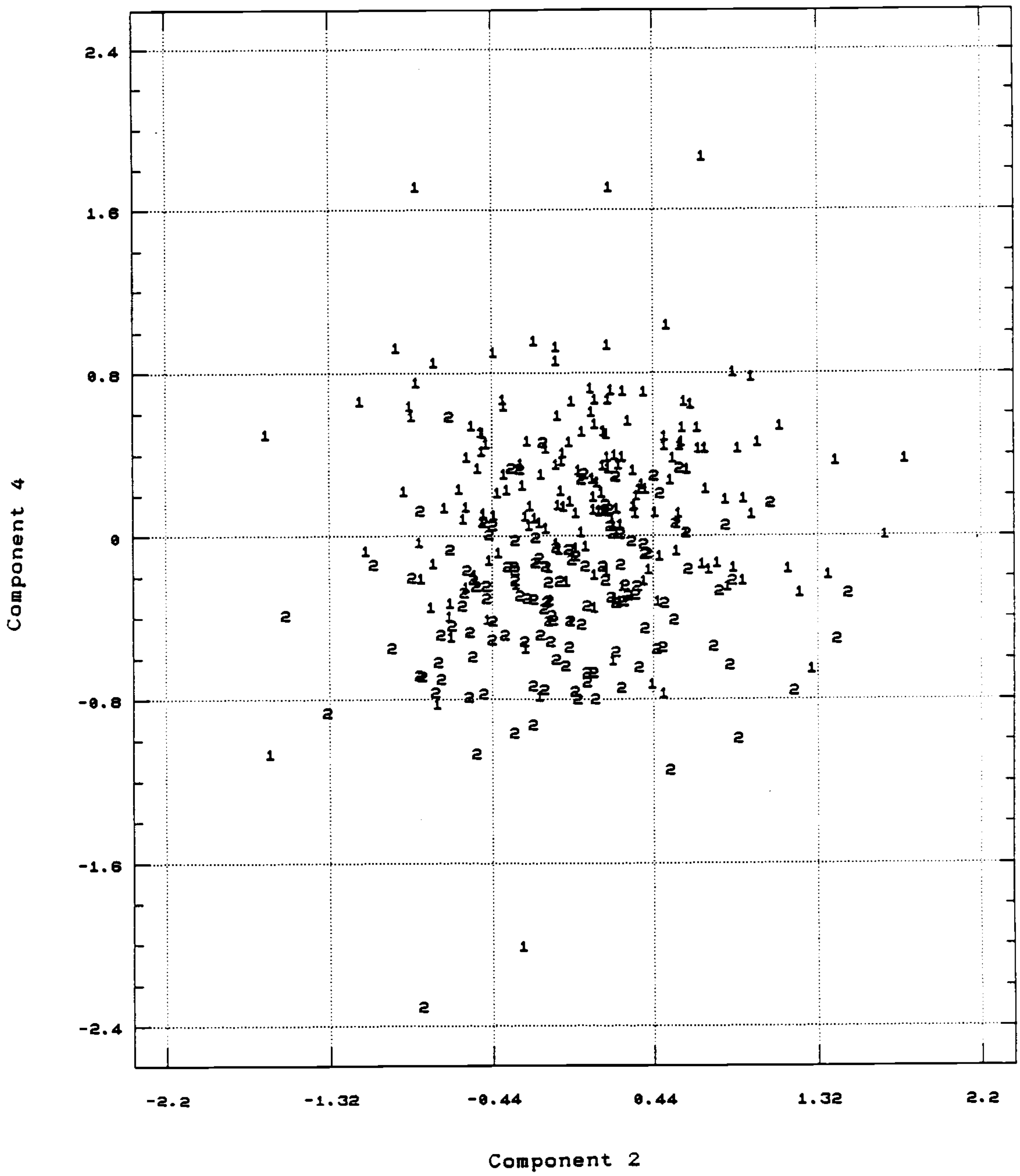


Figure 6. Scatterplot of PC4 and PC2 of morphometric variables for the Willamette and the Umpqua Oregon chub (1 = Willamette, N=181; 2 = Umpqua, N=131).

Figure 7. Illustration of lactate dehydrogenase in muscle tissue in the Willamette and Umpqua Oregon chub.

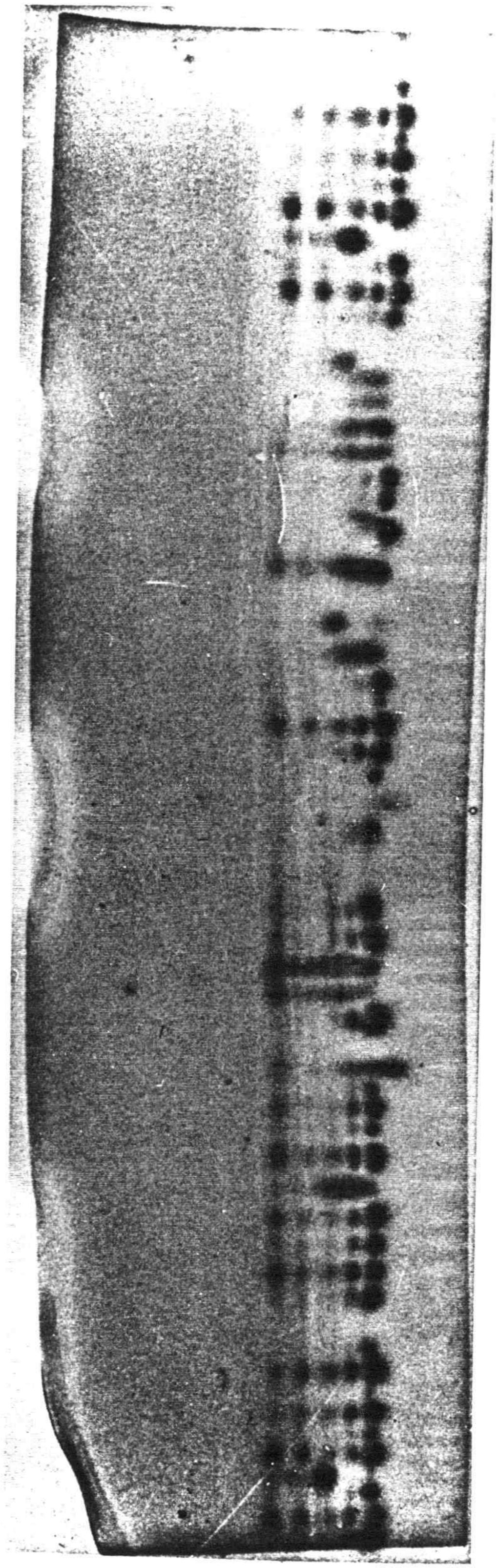


Figure 7.

Figure 8. Left lateral view of Willamette Oregon chub, OS 9620, 50.6 mm SL.

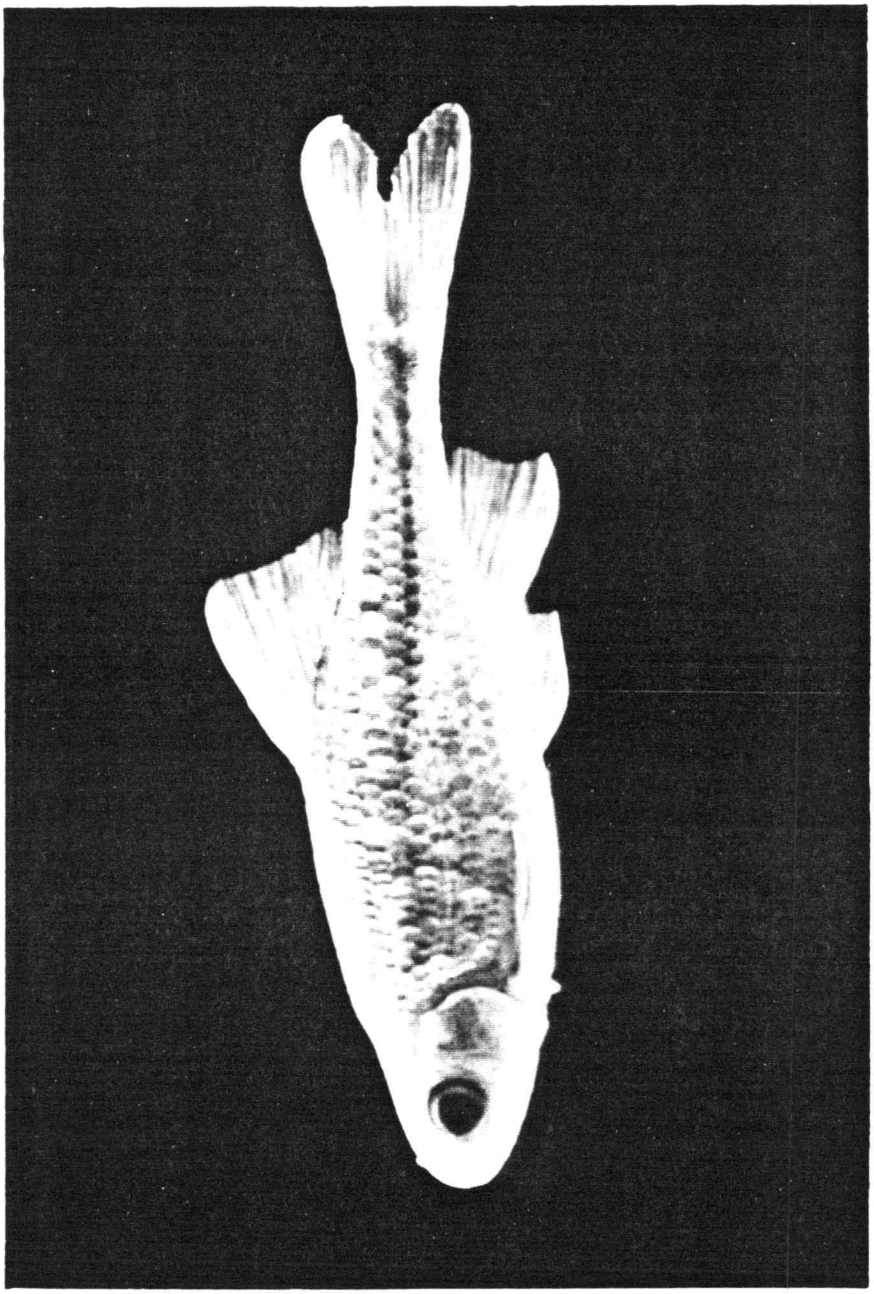


Figure 8.

Figure 9. Left lateral view of Umpqua Oregon chub, OS 10903, 46.3 mm SL.

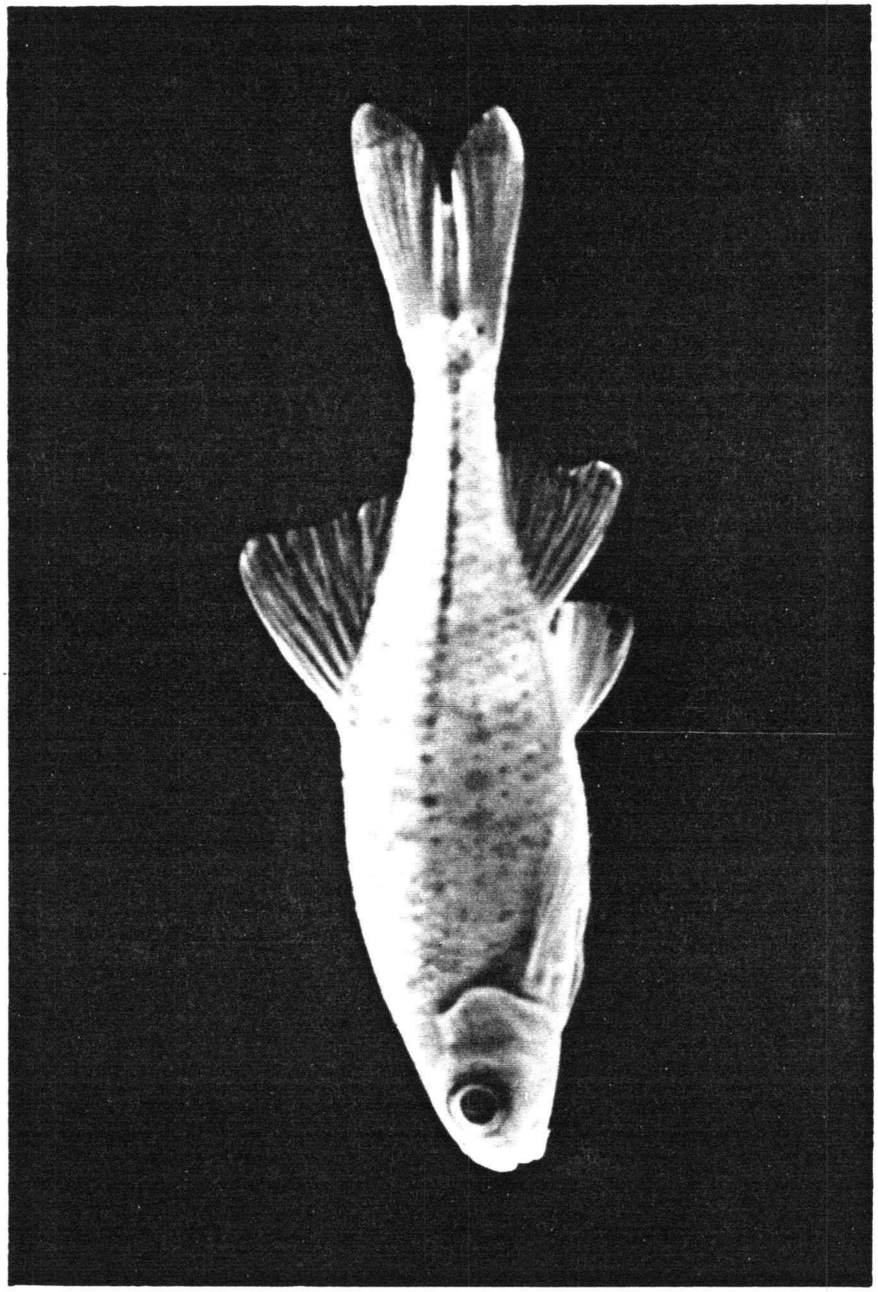


Figure 9.

Table 1. Location and numbers of Oregon chub included in all meristic and morphometric analysis, and those included only in the lateralis system analysis.

Location	all	lateralis
Willamette		
Northern mainstem		
Mouth of Clackamas	9	
Oregon City paratypes, Clear Creek	1	
Willamette-Oregon City	2	
Middle area		
McFaddens Pond	26	
N. Santiam-Aumsville, Jefferson	12	11
S. Santiam-Crabtree, Lebanon	3	
Willamette at Corvallis	24	
Southern area		
(Middle Fork)		
Lookout Point Res.	22	11
Shady Dell Pond	29	11
Dexter Reservoir	20	
Buckhead Cr./Lookout Point Res.	20	
Willamette at Eugene	9	
(Coast Fork)		
Gravel Pit Ponds	4	
Umpqua		
Northern area		
Mainstem		
Elk Creek at Drain	18	11
Elk Creek near mouth	4	
Elk Creek at Schad Road	21	
Scotts Creek Park	12	
Middle area		
Calapooya	31	11
Olalla Creek	5	
Tenmile	11	
Southern area		
Canyonville	6	
Cow Creek near Polan Cr.	16	11
3C Rock near Tiller	5	
South Umpqua	1	
Fords Camp	1	

Table 2. Location and numbers of specimens used in electrophoresis, and number of pharyngeal teeth arches.

Location	Electrophoresis	Pharyngeal teeth
Willamette		
Middle Fork		
a. Shady Dell	17	14
b. Dexter Reservoir	16	16
	33	30
Umpqua		
Mainstem		
a. Elk Creek	14	18
b. Smith River	3	
North Umpqua		
b. Calapooya	23	5
d. Scotts Creek Park	8	11
South Umpqua		
e. Bunch Bar	3	5
f. Cow Creek	4	7
	55	46

Table 3. List of the enzymes, their abbreviations, and enzyme commission numbers (E.C.N.), evaluated in this study.

Enzyme	Abbreviation	E.C.N.
phosphoglucIose isomerase	PGI	5.3.1.9
isocitrate dehydrogenase	IDH	1.1.1.42
lactate dehydrogenase	LDH	1.1.1.27
malate dehydrogenase	MDH	1.1.1.37
malic enzyme	ME	1.1.1.40
superoxide dismutase	SOD	1.15.1.1
tripeptide aminopeptidase	TAPEP	3.4.11.4
dipeptide	DPEP	3.4.13.11
asparate aminotransferase	AAT	2.6.1.1
phosphoglucomutase	PGM	2.7.5.1
acid phosphatase	ACP	3.1.3.2
aconitate hydratase	AH	4.2.1.3
alcohol dehydrogenase	ADH	1.1.1.1
creatine kinase	CK	2.7.3.2
monnosephosphate isomerase	MPI	5.3.1.8
xanthine oxidase	XO	1.2.3.2
L-iditol dehydrogenase	SDH	1.1.1.14
phosphogluconate dehydrogenase	6PG	1.1.1.44
gluclose-6-phosphate dehydrogenase	G6PDH	1.1.1.49

Table 4. Percentage counts and means of precaudal vertebrae for each area and region of the Oregon chub.

Location	N	Number of precaudal vertebrae 17	18	19	20	Mean
Willamette	181					18.36
Northern	12		58.3	41.7		18.42
Middle	65	3.1	63.1	33.8		18.31
Southern	104	1.0	58.7	40.4		18.39
% of Total*		1.7	60.2	38.1		

Location	N	Number of precaudal vertebrae 17	18	19	20	Mean
Umpqua	131					18.86
Northern	55		20.0	78.2	1.8	18.82
Middle	47		14.9	83.0	2.1	18.87
Southern	29		10.3	86.2	3.4	18.93
% of Total*			16.0	81.7	2.3	

*% of Total represents the percentage calculated from the total frequency count.

Table 5. Percentage counts and means of caudal vertebrae for each area and region of the Oregon chub.

Location	N	Number of caudal vertebrae 15	16	17	18	19	Mean
Willamette	181						17.42
Northern	12			41.7	58.3		17.58
Middle	65		4.6	52.3	40.0	3.1	17.42
Southern	104		5.8	51.0	41.3	1.9	17.39
% of Total*			5.0	50.8	42.0	2.2	

Location	N	Number of caudal vertebrae 15	16	17	18	19	Mean
Umpqua	131						16.83
Northern	55		21.8	70.9	5.5	1.8	16.87
Middle	47	2.1	31.9	53.2	12.8		16.77
Southern	29		17.2	79.8	3.4		16.86
% of Total*		0.8	24.4	66.4	7.6	0.8	

*% of Total represents the percentage calculated from the total frequency count.

Table 6. Percentage counts and means of the difference between precaudal vertebrae and caudal vertebrae for each area and region of the Oregon chub.

Location	N	Precaudal minus caudal vertebrae -1	0	1	2	3	4	Mean
<u>Willamette</u>	181							0.95
Northern	12		50.0	16.7	33.3			0.83
Middle	65	4.6	33.8	33.8	23.1	4.6		0.89
Southern	104	2.9	32.7	30.8	28.8	4.8		1.00
% of Total*		3.3	34.3	30.9	27.1	4.4		

Location	N	Precaudal minus caudal vertebrae -1	0	1	2	3	4	Mean
<u>Umpqua</u>	131							2.03
Northern	55	1.8	1.8	20.0	54.5	20.0	1.8	1.95
Middle	47		10.6	6.4	48.9	29.8	4.3	2.11
Southern	29			13.8	69.0	13.8	3.4	2.07
% of Total*		0.8	4.6	13.7	55.7	22.1	3.1	

*% of Total represents the percentage calculated from the total frequency count.

Table 7. Percentage counts and means of pelvic fin rays for each area and region of the Oregon chub.

Location	N	Number of pelvic fin rays 7	8	9	Mean
Willamette	180				7.99
Northern	11	27.3	72.7		7.73
Middle	65	4.6	87.7	7.7	8.03
Southern	104	1.9	97.1	1.0	7.99
% of Total*		4.4	92.2	3.3	

Location	N	Number of pelvic fin rays 7	8	9	Mean
Umpqua	131				7.92
Northern	55	5.5	94.5		7.95
Middle	47	17.0	78.7	4.3	7.87
Southern	29	6.9	93.1		7.93
% of Total*		9.9	88.5	1.5	

*% of Total represents the percentage calculated from the total frequency count.

Table 8. Percentage counts and means of upper procurrent caudal rays for each area and region of the Oregon chub.

Location	N	Number of upper procurrent caudal rays 6	7	8	Mean
Willamette	175				6.99
Northern	10	10.0	70.0	20.0	7.10
Middle	64	15.6	76.6	7.8	6.92
Southern	101	12.9	71.3	15.8	7.03
% of Total*		13.7	73.1	13.1	

Location	N	Number of upper procurrent caudal rays 6	7	8	Mean
Umpqua	114				6.65
Northern	40	37.8	59.5	2.7	6.65
Middle	47	44.4	55.6		6.56
Southern	27	18.5	81.5		6.81
% of Total*		35.8	63.3	0.9	

*% of Total represents the percentage calculated from the total frequency count.

Table 9. Percentage counts and means of total caudal rays for each area and region of the Oregon chub.

Location	N	29	30	31	32	33	34	35	Mean
		Number of total caudal rays							
Willamette	173								32.35
Northern	10			10.0	50.0	20.0	20.0		32.50
Middle	64		6.2	6.2	42.2	39.1	4.7	1.6	32.34
Southern	99	2.0	2.0	12.1	42.4	28.3	11.1	2.0	32.34
% of Total*		1.2	3.5	9.2	42.8	31.8	9.2	1.7	

Location	N	29	30	31	32	33	34	35	Mean
		Number of total caudal rays							
Umpqua	113								32.01
Northern	39			31.4	40.0	22.9	5.7		32.03
Middle	47		6.7	33.3	26.7	33.3			31.87
Southern	27			16.7	41.7	41.7			32.25
% of Total*			2.9	28.8	34.6	31.7	1.9		

*% of Total represents the percentage calculated from the total frequency count.

Table 10. Percentage counts and means of rudimentary dorsal rays for each area and region of the Oregon chub.

Location	N	Number of rudimentary dorsal rays 1	2	Mean
<u>Willamette</u>	176			1.06
Northern	11	100.0	0.0	1.00
Middle	63	96.8	3.2	1.03
Southern	102	92.2	7.8	1.08
% of Total*		94.3	5.7	

Location	N	Number of rudimentary dorsal rays 1	2	Mean
<u>Umpqua</u>	128			1.28
Northern	53	69.2	30.8	1.31
Middle	47	78.7	21.3	1.21
Southern	28	67.9	32.1	1.32
% of Total*		72.4	27.6	

*% of Total represents the percentage calculated from the total frequency count.

Table 11. Percentage counts and means of total dorsal rays for each area and region of the Oregon chub.

Location	N	Number of total dorsal rays 10	11	12	Mean
Willamette	176				10.07
Northern	11	100.0			10.00
Middle	63	96.8	3.2		10.03
Southern	102	90.2	9.8		10.10
% of Total*		93.1	6.9		

Location	N	Number of total dorsal rays 10	11	12	Mean
Umpqua	128				10.28
Northern	53	69.2	30.8		10.31
Middle	47	78.7	21.3		10.21
Southern	28	67.9	28.6	3.6	10.36
% of Total*		72.4	26.8	0.8	

*% of Total represents the percentage calculated from the total frequency count.

Table 12. Percentage counts and means of maxillary barbels for each area and region of the Oregon chub.

Location	N	Number of maxillary barbels 0	1	2	Mean
Willamette	179				1.10
Northern	10	60.0	20.0	20.0	0.60
Middle	65	43.1	20.0	36.9	0.94
Southern	104	25.0	25.0	50.0	1.25
% of Total		33.5	22.9	43.6	

Location	N	Number of maxillary barbels 0	1	2	
Umpqua	129				1.45
Northern	55	34.5	12.7	52.7	1.18
Middle	47	10.6	14.9	74.5	1.64
Southern	27	7.8	18.5	74.1	1.67
% of Total		20.2	14.7	65.1	

*% of Total represents the percentage calculated from the total frequency count.

Table 13. Univariate summary of meristic variables, means and ranges (in parenthesis). Includes variables significantly different between the Willamette and the Umpqua Oregon chub ($P < 0.05$) and those not significantly different ($P > 0.05$) as determined by Tukey's Studentized Range Test.

Variables with significant differences

Variable	Willamette	Umpqua
Precaudal vertebrae	18.36 (17-19)	18.86 (18-20)
Caudal vertebrae	17.41 (16-19)	16.83 (15-19)
Precaudal-caudal	0.95 (-1-4)	2.03 (-1-4)
Rudimentary dorsal rays	1.05 (1-2)	1.28 (1-2)
Total dorsal rays	10.07 (10-11)	10.28 (10-12)
Pelvic rays	7.99 (7-9)	7.92 (7-9)
Upper procurrent caudal rays	6.99 (6-8)	6.65 (6-8)
Total caudal rays	32.35 (29-35)	32.01 (30-34)
Barbels	1.10 (0-2)	1.45 (0-2)

Variables without significant differences

Variable	Willamette	Umpqua
Total vertebrae	35.80 (34-37)	35.69 (34-37)
Principal dorsal rays	9.01 (9-10)	9.01 (9-10)
Rudimentary anal rays	1.18 (1-2)	1.25 (1-2)
Principal anal rays	8.02 (8-9)	8.00 (7-9)
Total anal rays	9.22 (9-11)	9.27 (8-10)
Pectoral rays	12.83 (12-14)	12.85 (12-14)
Upper primary caudal	9.97 (8-10)	9.97 (9-10)
Lower primary caudal	9.01 (8-10)	9.00 (9-9)
Lower procurrent caudal	6.41 (5-8)	6.39 (5-8)

Table 14. Summary of supraorbital, infraorbital, preoperculomandibular, and supratemporal cephalic lateral line canals in the Willamette Oregon chub.

Aumsville N=11

	minimum	maximum	average	% broken
supraorbital	8	11	8.9	70
infraorbital	11	15	13.1	50
preoperculomandibular	10	14	12.5	90
supratemporal	3	4	3.7	10

Shady Dell N=11

	minimum	maximum	average	% broken
supraorbital	9	11	9.9	100
infraorbital	11	15	12.6	18
preoperculomandibular	10	15	12.0	64
supratemporal	3	5	4.1	36

Lookout Point N=11

	minimum	maximum	average	% broken
supraorbital	9	10	9.4	100
infraorbital	12	14	12.5	0
preoperculomandibular	12	14	13.1	100
supratemporal	3	5	3.8	27

Summary N=33

	minimum	maximum	average
supraorbital	8	11	9.4
infraorbital	11	15	12.7
preoperculomandibular	10	15	12.5
supratemporal	3	5	3.9

Table 15. Summary of supraorbital, infraorbital, preoperculomandibular, and supratemporal cephalic lateral line canals in the Umpqua Oregon chub.

<u>Elk Creek</u> N=11

	minimum	maximum	average	% broken
supraorbital	7	10	8.9	73
infraorbital	9	13	11.2	9
preoperculomandibular	10	13	12.0	64
supratemporal	3	4	3.6	0

<u>Cow Creek</u> N=11

	minimum	maximum	average	% broken
supraorbital	7	10	8.7	45
infraorbital	10	13	10.5	27
preoperculomandibular	12	13	12.5	91
supratemporal	4	5	4.1	0

<u>Calapooya</u> N=11

	minimum	maximum	average	% broken
supraorbital	8	11	8.9	64
infraorbital	10	12	11.1	9
preoperculomandibular	10	14	12.3	100
supratemporal	3	5	3.3	0

<u>Summary</u> N=33

	minimum	maximum	average
supraorbital	7	11	8.8
infraorbital	9	13	10.9
preoperculomandibular	10	14	12.3
supratemporal	3	5	3.7

Table 16. Discriminant analysis - Resubstitution summary - for all meristic variables for the Willamette and the Umpqua Oregon chub.

Number of Observations and Percent Classified in Region:

From Region	Willamette	Umpqua	Total
Willamette	152	16	168
	90.48	9.52	100.00
Umpqua	35	65	100
	35.00	65.00	100.00
Total	187	81	268
Percent	62.78	30.22	100.00

Table 17. Discriminant analysis - Resubstitution summary - for meristic variables with significant univariate results between the Willamette and Umpqua Oregon chub (includes precaudal, caudal, and precaudal minus caudal vertebrae, upper procurrent caudal rays, total caudal rays, rudimentary dorsal rays, total dorsal rays, pelvic rays, and maxillary barbels).

Number of Observations and Percent Classified in Region:

From Region	Willamette	Umpqua	Total
Willamette	154	14	168
	91.67	8.33	100.00
Umpqua	39	62	101
	38.61	61.39	100.00
Total	193	76	269
Percent	71.75	28.25	100.00

Table 18. Eigenvalues of the prinicipal component analysis on meristic variables for Willamette and the Umpqua Oregon chub.

Component Number	Percent of Variance	Cumulative Percentage
1	18.26	18.26
2	16.09	34.35
3	8.95	43.30
4	8.18	51.48
5	6.98	58.46
6	6.79	65.25
7	6.10	71.35
8	5.86	77.21
9	5.47	82.68
10	5.20	87.88
11	4.74	92.62
12	4.36	96.98
13	3.02	100.00

Table 19. Eigenvectors and eigenvalue proportions of the correlation matrix for twelve principal components from the meristic variables (precaudal vertebrae, caudal vertebrae, total vertebrae, precaudal-caudal vertebrae, rudimentary dorsal rays, principal dorsal rays, total dorsal rays, rudimentary anal rays, principal anal rays, total anal rays, pectoral rays, pelvic rays, upper procurrent caudal rays, upper primary caudal rays, lower primary caudal rays, lower procurrent caudal rays, total caudal rays, and maxillary barbels) of the Willamette Oregon chub.

	Eigenvectors					
	PC1	PC2	PC3	PC4	PC5	PC6
PRECAUDAL	0.109	-.410	-.004	0.381	-.422	-.049
CAUD/VERT	-.084	0.582	0.094	0.146	0.176	0.003
TOTVERT	0.006	0.248	0.092	0.463	0.527	-.037
PRE-CAU	0.113	-.598	-.063	0.109	0.111	-.028
RUDI/DOR	0.339	0.016	0.375	-.204	0.261	-.058
PRIN/DOR	0.038	-.029	0.045	-.320	0.123	0.630
DORSALS	0.327	0.002	0.364	-.322	0.291	0.212
RUDI/ANA	0.335	0.042	0.274	0.294	-.296	-.045
PRIN/ANA	0.058	0.017	-.038	0.238	-.198	0.489
ANALS	0.341	0.047	0.247	0.370	-.358	0.143
PECTORAL	0.167	-.017	0.243	-.076	-.200	-.242
PELVIC	0.089	0.009	0.137	-.241	0.030	-.372
UP/PROC	0.353	0.153	-.336	-.010	0.059	0.037
UP/PRIM	0.186	0.036	-.194	-.027	-.056	-.088
LO/PRIM	0.136	0.026	0.024	-.010	-.083	-.271
LO/PRO	0.328	0.074	-.394	-.051	0.075	-.008
CAUDALS	0.421	0.310	-.425	-.055	0.046	-.049
BARBELS	0.119	-.141	-.000	0.002	-.125	0.083
Eigenvalue Proportion	.17	.14	.12	.10	.08	.06
Cumulative Proportion	.17	.32	.43	.53	.61	.67

Table 19 (con't). Eigenvectors and eigenvalue proportions of the correlation matrix for twelve principal components from the meristic variables (precaudal vertebrae, caudal vertebrae, total vertebrae, precaudal-caudal vertebrae, rudimentary dorsal rays, principal dorsal rays, total dorsal rays, rudimentary anal rays, principal anal rays, total anal rays, pectoral rays, pelvic rays, upper procurrent caudal rays, upper primary caudal rays, lower primary caudal rays, lower procurrent caudal rays, total caudal rays, and maxillary barbels) of the Willamette Oregon chub.

	PC7	PC8	PC9	PC10	PC11	PC12
PRECAUDAL	0.054	-.026	0.014	0.064	0.058	0.126
CAUD/VERT	0.021	0.052	0.089	-.011	0.038	0.086
TOTVERT	0.065	0.031	0.102	0.042	0.087	0.191
PRE-CAU	0.015	-.047	-.051	0.041	0.006	0.012
RUDI/DOR	0.058	0.181	-.077	-.070	-.196	-.356
PRIN/DOR	-.010	-.347	0.131	0.063	0.217	0.452
DORSALS	0.049	0.020	-.016	-.038	-.089	-.137
RUDI/ANA	-.238	-.342	0.069	-.115	0.024	0.014
PRIN/ANA	0.305	0.438	-.162	0.504	0.032	-.154
ANALS	-.111	-.160	0.004	0.081	0.036	-.046
PECTORAL	0.243	0.311	-.304	-.091	-.169	-.722
PELVIC	-.378	0.100	0.184	0.703	0.276	0.101
UP/PROC	-.086	0.129	-.095	-.173	0.185	-.023
UP/PRIM	0.355	-.175	0.587	0.224	-.570	0.068
LO/PRIM	0.657	-.234	0.020	-.006	0.589	-.129
LO/PRO	-.211	-.002	-.228	0.063	-.068	0.078
CAUDALS	-.021	-.011	-.050	-.008	0.036	0.024
BARBELS	-.110	0.545	0.624	-.350	0.273	0.060
Eigenvalue Proportion	.06	.06	.05	.04	.04	.04
Cumulative Proportion	.74	.80	.85	.89	.93	.97

Table 20. Eigenvectors and eigenvalue proportions of the correlation matrix for twelve principal components from the meristic variables (precaudal vertebrae, caudal vertebrae, total vertebrae, precaudal-caudal vertebrae, rudimentary dorsal rays, principal dorsal rays, total dorsal rays, rudimentary anal rays, principal anal rays, total anal rays, pectoral rays, pelvic rays, upper procurrent caudal rays, upper primary caudal rays, lower primary caudal rays, lower procurrent caudal rays, total caudal rays, and maxillary barbels) of the Umpqua Oregon chub.

	Eigenvectors					
	PC1	PC2	PC3	PC4	PC5	PC6
PRECAUDAL	0.297	-.213	0.094	-.264	0.439	0.245
CAUD/VERT	-.366	0.448	0.049	0.087	0.188	0.040
TOTVERT	-.174	0.327	0.125	-.104	0.536	0.229
PRE-CAU	0.393	-.409	0.011	-.185	0.080	0.090
RUDI/DOR	0.229	0.042	0.499	0.302	0.046	-.165
PRIN/DOR	0.064	0.105	0.295	0.088	-.086	0.387
DORSALS	0.230	0.061	0.534	0.305	0.026	-.078
RUDI/ANA	0.259	0.077	-.337	0.469	0.198	-.044
PRIN/ANA	0.014	-.029	-.231	0.132	-.044	0.525
ANALS	0.252	0.068	-.373	0.480	0.181	0.068
PECTORAL	0.151	0.071	-.023	-.003	0.195	-.443
PELVIC	0.035	0.104	0.001	-.125	0.406	0.003
UP/PROC	0.251	0.389	-.074	-.211	-.214	-.114
UP/PRIM	0.055	0.162	-.071	0.102	-.181	-.030
LO/PRIM	-.000	-.000	-.000	0.000	-.000	-.000
LO/PRO	0.348	0.280	0.005	-.190	-.105	0.234
CAUDALS	0.358	0.416	-.052	-.212	-.217	0.069
BARBELS	0.119	0.087	-.176	-.256	0.233	-.378
Eigenvalue Proportion	.19	.14	.13	.11	.09	.07
Cumulative Proportion	.19	.33	.46	.56	.65	.72

Table 20 (con't). Eigenvectors and eigenvalue proportions of the correlation matrix for twelve principal components from the meristic variables (precaudal vertebrae, caudal vertebrae, total vertebrae, precaudal-caudal vertebrae, rudimentary dorsal rays, principal dorsal rays, total dorsal rays, rudimentary anal rays, principal anal rays, total anal rays, pectoral rays, pelvic rays, upper procurrent caudal rays, upper primary caudal rays, lower primary caudal rays, lower procurrent caudal rays, total caudal rays, and maxillary barbels) of the Umpqua Oregon chub.

	PC7	PC8	PC9	PC10	PC11	PC12
PRECAUDAL	-.027	-.272	0.063	0.078	-.175	0.078
CAUD/VERT	-.084	-.026	0.050	0.017	-.057	-.041
TOTVERT	-.112	-.234	0.102	0.077	-.194	0.015
PRE-CAU	0.044	-.112	-.004	0.025	-.045	0.065
RUDI/DOR	-.163	0.032	0.141	-.233	0.001	0.096
PRIM/DOR	0.310	0.160	-.155	0.688	0.304	0.032
DORSALS	-.092	0.063	0.103	-.082	0.063	0.098
RUDI/ANA	-.072	-.092	-.236	0.104	-.010	-.036
PRIN/ANA	0.031	0.392	0.615	-.190	-.008	0.243
ANALS	-.063	-.006	-.098	0.061	-.011	0.017
PECTORAL	0.503	0.264	0.375	0.230	-.358	-.304
PELVIC	0.484	0.282	-.347	-.479	0.355	0.139
UP/PROC	0.020	0.064	-.147	0.072	-.323	0.596
UP/PRIM	-.679	0.462	0.127	0.338	-.163	0.146
LO/PRIM	-.000	-.000	-.000	-.000	-.000	0.000
LO/PRO	-.173	0.097	-.008	-.165	0.114	-.642
CAUDALS	-.010	-.040	-.022	-.079	-.053	-.035
BARBELS	-.370	0.126	0.296	0.260	0.594	0.155
Eigenvalue Proportion	.07	.06	.05	.05	.03	.03
Cumulative Proportion	.78	.84	.89	.94	.97	1.00

Table 21. Univariate summary of morphometric characters for specimens between 25 - 45 mm in the Willamette (N=138) and Umpqua (N=108) Oregon chub, means and ranges (in parentheses).

Variable	Willamette		Umpqua	
Standard length	37.71	(27.75-44.65)	36.67	(25.20-44.85)
Predorsal length	20.23	(10.70-28.90)	19.83	(10.30-25.00)
Body depth	9.84	(6.60-13.40)	9.52	(6.35-13.20)
Body width*	5.24	(3.45-7.80)	5.54	(3.55-8.80)
Head length	9.17	(6.75-11.20)	9.15	(6.45-11.65)
Head depth*	5.02	(3.65-6.95)	4.80	(3.50-6.55)
Head width	5.35	(3.55-7.30)	5.33	(3.70-7.10)
Caud. ped. length*	10.53	(7.45-15.70)	9.74	(6.60-12.35)
Caud. ped. depth	3.25	(2.35-4.50)	3.25	(2.15-4.10)
Eye length*	2.76	(2.00-3.60)	2.65	(2.00-3.40)
Snout length	2.27	(1.40-3.50)	2.36	(0.90-3.20)
Dorsal length	8.66	(6.30-11.10)	8.66	(5.60-11.25)
Dorsal base length	5.67	(3.80-7.50)	5.85	(3.90-7.90)
Pectoral length	6.77	(4.60-8.50)	6.72	(4.45-8.95)
Pect. base length	1.63	(0.90-2.50)	1.65	(1.00-2.50)
Pelv length	6.01	(4.20-8.10)	5.90	(4.00-8.10)
Pelv base length	1.47	(0.90-2.20)	1.46	(0.85-2.10)
Anal length	7.60	(5.45-9.70)	7.79	(5.50-10.00)
Anal base length*	4.31	(2.95-5.80)	4.50	(3.30-5.75)

Variables marked with an asterisk (*) are significantly different ($P < 0.05$) from each other in Tukey's Studentized Range Test

Table 22. Eigenvalues of the principal component analysis on morphometric variables for the Willamette and Umpqua Oregon chub (N=312).

Component Number	Percent of Variance	Cumulative Percentage
1	87.42	87.42
2	2.99	90.41
3	2.10	92.51
4	1.55	94.06
5	1.19	95.25
6	1.10	96.35
7	0.71	97.06
8	0.59	97.65
9	0.48	98.13
10	0.46	98.59
11	0.45	99.04
12	0.36	99.40
13	0.24	99.64
14	0.20	99.84
15	0.16	100.00

Table 23. Eigenvectors and eigenvalue proportions of the correlation matrix for six principal components of morphometric characters (predorsal length, standard length, body depth, body width, head length, head depth, head width, caudal peduncle length, caudal peduncle depth, eye length, snout length, dorsal fin length, dorsal fin base length, pectoral fin length, pectoral fin base length, pelvic fin length, pelvic fin base length, anal fin length, anal fin base length) for the Willamette Oregon chub, specimens 25 - 45 mm.

	PC1	PC2	PC3	PC4	PC5	PC6
PREDORL	0.237	-.242	-.237	0.048	0.102	0.027
SL	0.252	-.093	-.066	-.070	-.027	-.063
BODYD	0.223	-.335	-.020	-.124	-.173	-.210
BODYW	0.221	-.520	0.016	-.072	-.165	0.070
HEADL	0.254	-.008	-.082	0.043	0.111	-.053
HEADD	0.237	-.105	0.186	0.113	0.151	-.018
HEADW	0.235	-.244	0.084	-.023	0.220	0.075
CAUDPEDL	0.233	0.017	0.095	-.040	0.029	-.322
CAUDPEDD	0.234	-.125	0.044	-.291	-.170	-.038
EYEL	0.206	0.121	-.505	0.442	0.047	0.386
SNOUTL	0.213	0.007	-.096	-.004	0.752	-.014
DORSAL	0.243	0.082	-.091	-.096	-.172	0.159
DBASE	0.237	0.049	-.004	0.018	-.178	-.002
PECTL	0.238	0.280	-.030	-.203	-.104	0.024
PBASE	0.206	0.353	0.441	-.336	0.261	0.155
PELVL	0.242	0.254	-.005	-.111	-.204	0.108
PELVBL	0.192	-.043	0.609	0.581	-.153	0.284
ANALL	0.230	0.293	-.198	-.112	-.200	0.185
ANALBL	0.213	0.290	-.037	0.387	-.069	-.712
Eigenvalue proportion	.76	.04	.03	.03	.02	.02
Cumulative Proportion	.76	.80	.83	.86	.88	.90

Table 24. Eigenvectors and eigenvalue proportions of the correlation matrix for six principal components of morphometric characters (predorsal length, standard length, body depth, body width, head length, head depth, head width, caudal peduncle length, caudal peduncle depth, eye length, snout length, dorsal fin length, dorsal fin base length, pectoral fin length, pectoral fin base length, pelvic fin length, pelvic fin base length, anal fin length, and anal fin base length) for the Umpqua Oregon chub, specimens 25 - 45 mm.

	PC1	PC2	PC3	PC4	PC5	PC6
PREDORL	0.242	-.009	-.088	0.079	-.178	-.070
SL	0.243	-.032	-.078	0.023	-.156	-.100
BODYD	0.237	0.010	-.168	0.188	-.176	-.113
BODYW	0.222	-.165	-.326	0.379	0.070	-.220
HEADL	0.242	-.187	0.029	0.055	0.054	0.028
HEADD	0.233	-.050	0.002	0.058	0.401	-.055
HEADW	0.241	-.008	-.088	0.136	0.074	-.194
CAUDPEDL	0.231	0.153	-.160	0.136	-.129	-.348
CAUDPEDD	0.241	0.099	-.030	-.047	-.056	-.056
EYEL	0.215	-.388	-.169	-.020	0.589	0.299
SNOUTL	0.192	-.138	0.711	0.549	-.135	0.204
DORSAL	0.237	-.048	-.020	-.255	-.182	0.219
DBASE	0.235	-.069	-.108	-.228	-.046	0.142
PECTL	0.234	0.172	0.103	-.083	-.135	-.089
PBASE	0.205	0.448	0.397	-.251	0.468	-.378
PELVL	0.240	-.045	0.101	-.185	-.127	0.033
PELVBL	0.199	0.664	-.232	0.173	0.030	0.596
ANALL	0.236	-.132	0.181	-.195	-.018	0.226
ANALBL	0.233	-.184	0.062	-.421	-.252	-.028
Eigenvalue Proportion	.85	.03	.03	.02	.01	.01
Cumulative Proportion	.85	.88	.91	.93	.94	.95

Table 25. Discriminant analysis - Resubstitution summary - for morphometric variables from specimens 25-45 mm of the Willamette and Umpqua Oregon chub.

Number of Observations and Percent Classified into Region:

From Region	Willamette	Umpqua	Total
Willamette	124 90.51	13 9.49	137 100.00
Umpqua	13 12.04	95 87.96	108 100.00
Total	137	108	245
Percent	55.92	44.08	100.00

Table 26. Discriminant analysis - Resubstitution Summary - for morphometric data for all Willamette and Umpqua Oregon chub specimens (N=310).

Number of Observations and Percent Classified into Region:

From Region	Willamette	Umpqua	Total
Willamette	160	19	179
	89.39	10.61	100.00
Umpqua	20	111	131
	15.27	84.73	100.00
Total	180	130	310
Percent	58.06	41.94	100.00

Table 27. Lists of the enzyme systems, their abbreviations, and resolution with muscle tissue and Ridgeway buffer system.

Enzyme	Abbreviation	Results*
lactate dehydrogenase	LDH	1
phosphogluclose isomerase	PGI	2
malate dehydrogenase	MDH	2
phosphogluconate dehydrogenase	6PG	2
isocitrate dehydrogenase	IDH	3
malic enzyme	ME	3
superoxide dismutase	SOD	3
tripeptide aminopeptidase	TAPEP	3
dipeptide	DPEP	3
creatine kinase	CK	3
asparate aminotransferase	AAT	4
phosphoglucomutase	PGM	4
acid phosphatase	ACP	4
monnosephosphate isomerase	MPI	4
xanthine oxidase	XO	4
gluclose-6-phosphate dehydrogenase	G6PDH	4
aconitate hydratase	AH	4
alcohol dehydrogenase	ADH	4
L-iditol dehydrogenase	SDH	4

*1. good resolution, repeatable variation
2. good resolution, possible rare variation
3. fair resolution, no apparent variation
4. no resolution, no apparent variation

Table 28. Summary of lactate dehydrogenase in the Willamette and Umpqua Oregon chubs: frequency of genotypes in lactate dehydrogenase, allele counts, and allele frequencies.

Location	Genotypes			
Willamette	AA	AC	CC	N
Shady Dell	17	0	0	
Dexter	14	2	0	
	31	2	0	33
Umpqua	AA	AB	BB	N
Elk Cr.	11	2	1	
Calapooya	14	6	3	
Smith River	0	2	1	
Scotts Creek	4	2	2	
Bunch Bar	3	0	0	
Cow Creek	3	1	0	
	35	13	7	55

Allele counts	A	B	C
Willamette	64	0	2
Umpqua	83	27	0

Allele counts equal two times the specified homozygote plus the number of heterozygotes

Allele frequencies	A	B	C
Willamette	.97	.00	.03
Umpqua	.75	.25	.00

Allele frequencies equal the number of counts for a specified allele divided by two times the number of individuals.

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APPENDIX

Appendix 1

Materials Examined

Catalog Number, Location, County, Number of specimens, Size range, Collectors, and Date

<u>Willamette River</u>

Northern area
USNM 58088, paratype, Oregon City, Clear Creek, n=1, 47.40; Cramer and Otaki, 1907. USNM 61575, paratypes, Willamette at Oregon City, n=2, 38.60-40.10; Cramer and Otaki. OS 9760, Mouth of Clackamas, Clackamas Co., n=9, 19.00-29.00 mm; Kerns, McHugh, and Davison, 1953.

Middle area
UW 12758, McFaddens Swamp, 12 miles south of Corvallis, Benton Co., n=4, 40.7-43.50; G. Deschamps, 1951. UW 12769, McFaddens Swamp, 12 miles south of Corvallis, Benton Co., n=3, 32.70-36.00; C. Bond and class, 1952. OS 682, McFaddens Pond, 12 miles south of Corvallis, n=19, 34.20-48.45 mm; G. Deschamps, 1951. OS 11440, N. Santiam-Aumsville, Marion Co., n=12, 40.65-60.20; J. Long, 1984. OS 9612, S. Santiam-Crabtree, Linn Co., n=3, 33.05-36.60; C. Ziebeil and R. McHugh, 1953. OS 5404, Willamette at Corvallis, Benton Co., n=3, 45.00-49.60; 1945. USNM 58138, Willamette at Corvallis, n=10, 30.45-43.60. USNM 58088, paratype, Willamette at Corvallis, n=11, 28.60-51.00.

Southern area
OS 9605, Lookout Point Res., Lane Co., n=5, 42.55-47.70; Hasselman and Garrison, 1957. OS 10551, Lookout Point Res., Lane Co., n=17, 21.45-48.55; R. Hasselman and R. Garrison, 1957. OS 9632, Shady Dell Pond, Lane Co., n=30, 30.60-49.40; J. Long and K. Howe, 1977. OS 9632, Shady Dell Pond, approximately 7 miles west of Oakridge, Lane Co., n=29, 30.60-49.40; J. Long and K. Howe, 1977. OS 9604, Dexter Reservoir, Lane Co., n=7, 35.10-51.50; Garrison and Hasselman, 1957. OS 2733, Dexter Reservoir, Lane Co., n=13, 31.80-46.50; Hasselman and Garrison, 1957. OS 9620, Buckhead Creek, Lane Co., n=20, 27.90-51.85; J. Long, 1981. USNM 58138, paratypes, Willamette at Eugene, n=9, 32.30-44.00; Cramer and Otaki, 1894. OS 9603, Gravel Pit Ponds, Lane Co., n=4, 41.40-47.90; Reimers and Wallace, 1967.

<u>Umpqua River</u>

Northern area
OS 9611, Elk Creek at Drain, Douglas Co., n=12, 28.80-35.85; Reimers and Forsberg, 1967. OS 9601, Elk Creek 1 mile above mouth, Douglas Co., n=4, 27.90-33.75; Reimers and Forsberg, 1967. OS 10907, Elk Creek at Drain, Douglas Co., n=7, 21.40-32.85; J. Long and B. Bolding, 1983. OS 11439, Elk

Creek and Calapooya Creek, Douglas Co., n=31, 28.50-49.50; T. Pearsons, 1988. OS 11503, Elk Creek at Schad Road, n=21, 28.50-49.50; Markle *et al.*, 1987. OS 11507, Scotts Creek Park, n=12, 25.20-39.25. OS 11518, Smith River at Vincent Creek Recreation site, n=3, 26-38; Pearsons and Bills, 1987.

Middle area
OS 11506, Olalla Creek, n=5, 18.50-31.60; Markle *et al.*, 1987. OS 9608, Tenmile Creek, Douglas Co., n=11, 33.45-40.50; P. Reimers, 1971.

Southern area
OS 11519, Umpqua at Bunch Bar Public Access, n=4, 37.00-41.00 mm; Pearsons and Bills, 1987. OS 10903, South Umpqua at Canyonville at bridge, Douglas Co., n=6, 24.10-45.90; J. Long and B. Bolding, 1983. OS 5494, Cow Creek near Polan Creek, Douglas Co., n=16, 39.55-45.00; Bond and Juntunen, 1970. OS 11504, 3C Rock near Tiller, n=5, 23.10-44.85; Markle *et al.*, 1987. OS 3683, Fords Camp, n=1, 24.10; Ballantyne and Borland, 1945. USNM 58116, South Umpqua River, n=1, 41.60; 1907.