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#### MORPHOLOGICAL FEATURES USED TO IDENTIFY CHINOOK SALMON SEX DURING FISH PASSAGE

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ABSTRACT—We compared several external morphological features for determining sex of adult fall-run chinook salmon (*Oncorhynchus tschawytscha*) migrating to spawning grounds in the Central Valley of California. Adult fish carcasses of known sex were measured at fish hatcheries or during angler surveys. These data were used to develop predictive morphometric discriminant function models for potential incorporation in an automated monitoring system. The best predictor for determining sex of handled fish was snout length to fork length ratio, which correctly classified 96% of individuals tested. In contrast, adipose fin length to fork length ratio was the best predictor of sex when measurements were obtained from video images at a fish passage facility. Of these fish, 86% were correctly identified. Combining both ratios with a third (head length) increased model accuracy to 92% for video images.

RESUMEN—Comparamos el uso de varias características morfológicas externas para determinar el sexo de adultos de salmones (*Oncorhynchus tschawytscha*) durante su migración otoñal rumbo a su sitio de desove en el Valle Central de California. Cadáveres de peces adultos de sexo conocido se midieron en criaderos de pez o durante entrevistas con pescadores. Estos datos se usaron para hacer modelos morfométricos de predicción de función discriminante. El mejor pronosticador para determinar el sexo fue la proporción de la longitud del hocico a la longitud corporal (hasta la bifurcación del ala caudal), que clasificó correctamente 96% de los individuos probados. En contraste, la proporción de la longitud de la aleta adiposa a la longitud corporal (hasta la bifurcación del ala caudal) fue el mejor pronosticador del sexo de adultos salmónidos cuando las medidas se obtuvieron de imágenes de video en una facilidad del pasaje de pez. De éstos peces, 86% fueron identificados correctamente. Al combinar ambas proporciones con una tercera (usando la longitud de la cabeza) la precisión del modelo aumentó hasta 92% para imágenes de video.

Salmonid passage counts at dams are important to fisheries managers for setting fishing seasons, estimating run size, determining inriver survival, estimating escapement to spawning grounds, and establishing and monitoring various compensation and enhancement programs (Hatch et al., 1998; Dauble and Mueller, 2000). Determining the proportion of male and female adult salmonids returning to natal streams for spawning is important in evaluating production goals and estimating stock reproductive potential (Crim and Glebe, 1990). In many cases, physically counting and identifying sex of salmonids at passage facilities is time consuming and expensive (Hatch et al., 1998), and there is a strong interest in developing rapid and practical methods of sexing live fish (Crim and Glebe, 1990).

Sexual dimorphism is common in many salmonid species (Morton, 1965; Grunchy and Vladykov, 1968). Although external determination of the sex of immature Pacific salmon (Oncorhynchus) is difficult (Morton, 1965; Beacham and Murray, 1985), dimorphic secondary sexual characteristics, such as coloration, body and fin shape, and jaw morphology, develop during the spawning run (Beacham and Murray, 1983; Beacham and Murray, 1985; Quinn and Foote, 1994). Dimorphic extremes range from pronounced dorsal humps and distinct kypes in male pink salmon (O. gorbuscha) and sockeye salmon (O. nerka) to less pronounced sexual differences in chinook salmon (O. tschawytscha) (Foerster, 1968; Groot and Margolis, 1991).

Although several computerized systems em-



FIG. 1—Morphometric measurements made on adult chinook salmon collected from the Mokelumne and American rivers, California: A, fork length (FL); B, head length (HL); C, snout length (SNL); D, adipose fin length (AFL); E, adipose fin height (AFH).

ploy automated photography or video recording, the determination of fish sex must still be made by trained personnel (Hatch et al., 1994; Pippy et al., 1997; Hatch et al., 1998). High turbidity, poor lighting, or multiple fish passing the system at one time can impair the ability of the viewer to make an objective assessment, reducing accuracy.

Using predictive morphometric discriminant function models to classify sex of animals in field studies is well documented (Holmgren, 1993; Martin et al., 2000; Love 2002). In this study, we examined the use of various body measurements of fall-run chinook salmon measured directly from hatchery and angler survey specimens and images recorded by underwater video camera at a fish passage facility in the Central Valley of California to determine sex of migrating adults and assess the possibility of incorporating such predictive models into automated monitoring systems.

METHODS—We examined 216 (110 female and 106 male) fall-run chinook salmon (fork length range: 468 to 1,071 mm) in the fall and winter of 1998 through 2000 from angler surveys and from the Mokelumne River Fish Hatchery on the lower Mokelumne River and the Nimbus Fish Hatchery on the American River. Measurements were taken from freshly killed (<12 h) fish. For each individual, sex was recorded after internal inspection. Fork length (FL), head length (HL), snout length (SNL), adipose fin length (AFL), and adipose fin height (AFH)

were measured to the nearest 1 mm with a measuring tape (Fig. 1).

We used ratios of HL to FL, SNL to FL, AFL to FL, and AFH to FL to standardize morphological data (Reist, 1985; Beacham and Murray, 1985) and to minimize the effects of measurement error attributable to video lens distortion and fish distance from video lens. We assessed the relationships between these morphometrics and sex by calculating Pearson product-moment correlation coefficients between measures for the total sample and for each sex. Simple linear regression was used to assess the relationship between FL and each of the other measurements recorded. We used one-way analysis of variance to compare the ratios between the sexes (Motulsky, 1995). Linear discriminate function analysis (Straus and Bond, 1990) was used to classify individuals by sex based on the ratios HL:FL, SNL:FL, AFL:FL, and AFH:FL. Analyses were performed with Statistical Programs for the Social Sciences, version 10, software (SPSS, Chicago, Illinois).

A total of 83 (27 females and 56 males) upmigrating chinook salmon were recorded at the Woodbridge Dam fish passage facility on the lower Mokelumne River with a closed-circuit color video camera with a 3.6-mm, wide-angle lens installed in an underwater housing. Fish were recorded as they passed between a clear Plexiglas panel and a white plastic sheet with black vertical lines spaced at 5-cm intervals inside one of the fish ladder bays. At the end of the monitoring season (15 August through 15 March), 10-minute to 15-minute sections were randomly selected from 8-h videotapes and viewed using a 53.3-cm color monitor. As fish came into view on the monitor, the video was paused and FL, HL,

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TABLE 1—Means and	standard deviations for	4 ratios employ	ed with angler and	d hatchery data	for chinook
salmon in California. V	ariables illustrated on F	Fig. 1.			

Sex	Variable	Minimum	Maximum	Mean	SD	Skewness	Kurtosis
Female	HL:FL	0.1926	0.2503	0.2268	0.0106	-0.2213	-0.0520
n = 110	AFL:FL	0.0385	0.0814	0.0596	0.0078	0.1219	-0.0520
	AFH:FL	0.0128	0.0371	0.0250	0.0037	-0.1241	1.8936
	SNL:FL	0.0742	0.1181	0.0870	0.0072	1.1721	3.4247
Male	HL:FL	0.1835	0.2971	0.2581	0.0154	-1.1937	5.0617
n = 106	AFL:FL	0.0576	0.1084	0.0405	0.0102	-0.0517	-0.0046
	AFH:FL	0.0215	0.0711	0.0405	0.0063	1.1013	4.9370
	SNL:FL	0.0734	0.1570	0.1156	0.0118	-0.0572	1.8802

AFL, and AFH were measured to the nearest 1 mm on the monitor, and sex was determined from the images by trained personnel. Snout measurements obtained with the videotaping technique were unreadable because water turbidity and lens distortion made observation of fish eyes difficult. We assumed that ratios between specific measurements would remain constant for individual fish, swimming parallel to the lens, no matter what distance the fish was from the camera. Twenty-two of these videotaped fish were trapped upstream of the video chamber to assess sex identification accuracy by personnel viewing the tapes.

RESULTS-Neither skewness nor kurtosis for any of the ratios in the hand-measured group exceeded a magnitude of 0.88 (Table 1). However, for each sex, the distribution of SNL:FL was markedly leptokurtic, although skewness for neither sex exceeded a magnitude of 1.19 for this ratio. In each instance, the difference between means of ratios for females was smaller than that of males, while variances of the ratios were no more than 1.7 times smaller. Two ratios did not meet the assumption of homogeneity of variance, HL:FL and AFL:FL. However, if sample sizes are approximately equal, violations of that assumption minimally affect the significance of statistical tests of difference between means, especially if skewness in each group is not extreme and is in the same direction (Samuels and Witmer, 1999). Both conditions are true in these data, suggesting that differences in ratios might be used to differentiate females from males. Relationships of HL, SNL, AFL, and AFH to FL were typically stronger for males than females, but were significant for both sexes for all 4 relationships (Table 2). Strongest and weakest relationships were observed for HL:FL and AFH: FL, respectively (Fig. 2). Means of ratios were significantly different between sexes for all recorded measurements, with AFH: FL (F = 484.2; df = 214; P < 0.001) and HL: FL (F = 302.8; df = 214; P < 0.001) having greatest and smallest differences in mean ratios (Fig. 3).

Linear discriminant function models identified 91 to 96% of all fish correctly with SNL: FL the best predictor. A multiple regression model using all 4 ratios correctly identified the greatest percent of fish (96%).

During video-recorded surveys, relationships of HL, AFL, and AFH with FL were significant for both sexes for all 3 relationships. However, differences in means were less obvious for measurements recorded by video than those made by hand. Utilizing the linear discriminant function procedure, morphometric ratios, recorded from video monitoring, correctly classified 71 to 86% of fish. Using all 3 ratios together provided the greatest accuracy (92%) in chinook salmon sex identification.

DISCUSSION—In general, male chinook salmon had significantly larger HL, SNL, and AFL to FL ratios than females. Such differences are consistent with other studies on sexual dimorphism (Keenleyside and Dupuis, 1988; Cooper and Vitt, 1989). Overlap in size ratios was most apparent with females and precocious males that were smaller than 60 cm FL. However, even within the Mokelumne River, a system that consistently has high jack to adult ratios (up to 40%), we were able to correctly classify 97% of all chinook salmon measured by hand (15% with FL < 60 cm). We encountered several problems in the video-monitoring assessment, especially turbidity, underwater lighting,

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TABLE 2—Body morphometric ratios used for determining fish sex, associated statistical results, and percent of fish correctly identified for chinook salmon handled and video-recorded in California. Variables illustrated in Fig. 1.

	]	Female		Male						Both sexes
						%				%
			%			cor-				cor-
Variables	Mode	$R^2$	correct	Mode	$R^2$	rect	F	df	P	rect
Handled										
HL:FL	0.23	0.79	93	0.26	0.92	87	303	214	< 0.01	91
SNL:FL	0.09	0.58	97	0.12	0.89	94	468	214	< 0.01	96
AFL:FL	0.06	0.32	95	0.09	0.69	88	408	214	< 0.01	92
AFH : FL	0.03	0.27	97	0.04	0.57	93	484	214	< 0.01	95
All added	0.41	0.81	99	0.52	0.92	95	176	211	< 0.01	97
All multiplied	0.0004	0.56	100	0.0007	0.74	91	34	214	< 0.01	96
Video-recorded										
HL:FL	0.23	0.87	67	0.27	0.85	73	20	81	< 0.01	71
AFL:FL	0.06	0.64	85	0.08	0.68	86	49	81	< 0.01	86
AFH : FL	0.02	0.62	90	0.03	0.6	77	42	81	< 0.01	81
All added	0.30	0.92	93	0.36	0.89	91	34	79	< 0.01	92
All multiplied	0.0003	0.88	100	0.0005	0.77	79	64	81	< 0.01	86

and distorted wide-angle images (Herbert, 1987). Even so, by combining the 3 recorded measurements, we were able to correctly classify recorded fish 92% of the time. These data suggest that morphological measurements taken in automated systems (Gatlin et al., 1993; Pippy et al., 1997; Norman, 2001) provide reliable estimates of sex ratio for chinook salmon returning to spawn through fish passage facilities.

Although morphometric relationships for hand-measured chinook salmon from the American and Mokelumne rivers were virtually identical, it is important to note that measurements for adult chinook salmon migrating past fish passage facilities on other systems might be different. This is especially true where monitoring occurs closer to the estuary and secondary sexual characteristics might be less developed. Morphometric differences have been observed for salmonids of the same species, specifically chinook salmon and chum salmon (*O. keta*), found in separate drainages (Beacham et al., 1988; Kinnison et al., 1998). Therefore, specific morphometric ratios might need to be developed for specific drainages to accurately determine sex of individual stocks moving through these passage facilities.



FIG. 2—Relationships of adipose fin height and head length to fork length of female and male chinook salmon from California.

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FIG. 3—Frequency of California female and male chinook salmon adipose fin height and head length measurements divided by fork length measurements.

Great strides are being made in the area of image recording to identify fish species (Scalabrin, 1996; Cadrin and Friedland, 1999; Le-Feuvre et al., 2000). Furthermore, image processing and computer analysis programs allow precise measurement, reorientation, scaling, and marker transfer for images of specific organisms recorded by numerous media, including video (Vogt, 1995; Seibert et al., 1996; Zhang et al., 2000). Coupling of these improving technologies with drainage-specific morphometric data as described here might improve the ability to identify fish sex for Pacific salmonid species within fully automated systems, reducing both labor hours and subjective determination of fish sex. Further research is warranted in this area.

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