

Blood Plasma Levels of Insulin-Like Growth Factor-I in Pacific Salmon in Offshore Waters in Winter

Katherine W. Myers and Nancy D. Davis
University of Washington, Fisheries Research Institute
Box 357980, Seattle, WA 98195

Walton W. Dickhoff
Northwest Fisheries Science Center, National Marine Fisheries Service
Seattle, WA 98112

and

Shigehiko Urawa
National Salmon Resources Center, Fisheries Agency of Japan
Sapporo 062, Japan



Myers, K.W., N.D. Davis, W.W. Dickhoff, and S. Urawa. 1998. Blood plasma levels of insulin-like growth factor-I in Pacific salmon in offshore waters in winter. N. Pac. Anadr. Fish Comm. Bull. No. 1: 129-137

This paper reports on the first study of blood plasma levels of insulin-like growth factor-I (IGF-I) in Pacific salmon (*Oncorhynchus* spp.) in offshore waters of the North Pacific Ocean. Environmental and physiological regulation of salmon growth in offshore waters is poorly understood. Physiological indices of growth are needed to better understand the factors that affect ocean production of salmon. IGF-I is an important regulator of growth and development in teleosts. Previous research has shown that seasonal increases in IGF-I in salmon precede rapid growth, and may be associated with environmental cues (water temperature, photoperiod). We collected blood samples from salmon caught by research trawl in three regions of the North Pacific Ocean (western, central, eastern) during an offshore survey in January 1996. Plasma IGF-I levels were determined by radioimmunoassay. All of the samples were analyzed by the same assay so that inter- and intra-specific comparisons could be made. Mean IGF-I levels were significantly different among the five species sampled: pink (*O. gorbuscha*; mean = 28 ng/ml, n = 14), chum (*O. keta*; mean = 33 ng/ml, n=28), chinook (*O. tshawytscha*; mean = 48 ng/ml, n = 15), sockeye (*O. nerka*; mean = 84 ng/ml, n = 13), and coho (*O. kisutch*; mean = 120 ng/ml, n =26) salmon. High levels of IGF-I in coho salmon correlates with high growth rates in this species found in other studies. There were significant positive correlations between body weight, liver weight, and IGF-I levels in all species except pink salmon. Samples were not collected at enough stations to adequately evaluate correlations between IGF-I and environmental factors. There were significant intra-specific differences in mean IGF-I levels by ocean age and region. Size of age .1 pink salmon was not significantly different by ocean region, but mean IGF-I levels in pink salmon were significantly higher in the eastern North Pacific than in the western region, which may indicate earlier resumption of rapid spring growth in the Gulf of Alaska. Similarly, body size and IGF-I levels in age .1 coho salmon were significantly higher in the eastern North Pacific, than in the central region. We conclude that IGF-I may be a useful measure of ocean growth rates of salmon in the North Pacific Ocean, but additional data on the significance of high or low levels of IGF-I are needed.



INTRODUCTION

Environmental and physiological regulation of growth of Pacific salmon (*Oncorhynchus* spp.) in offshore waters is poorly understood. Physiological indices of growth are needed to better understand the

factors that affect ocean production of salmon. Insulin-like growth factor-I (IGF-I), a 70-amino-acid polypeptide, is an important regulator of growth and development in mammals, birds, and teleosts (e.g., Bern et al. 1991; Kikuchi et al. 1991; Cohick and Clemmons 1993; Duan et al. 1994, 1995). In yearling

coho salmon (*O. kisutch*) held in seawater net pens, seasonal (springtime) increases in IGF-I precede rapid growth, and are associated with environmental (photoperiod, water temperature, food availability) cues (Duan et al. 1995).

Factors affecting changes in the ocean growth of salmon were identified by the North Pacific Anadromous Fish Commission as a critical issue for research (NPAFC 1996). In January 1996, we initiated a cooperative Japan-U.S. study of blood plasma levels of IGF-I in salmon caught during a trans-Pacific survey (Ueno et al. 1996). In this paper, we present the first information on plasma levels of IGF-I in salmon caught in offshore waters. We evaluate inter- and intra-specific differences in IGF-I by sex, age, and ocean region (western, central, and eastern North Pacific) and correlations between IGF-I levels and various biological and environmental factors.

METHODS

Study Area and Biological and Oceanographic Sampling

Samples were collected aboard a Fisheries Agency of Japan (FAJ) research vessel, R/V *Kaiyo maru*, during a cooperative NPAFC-related trans-Pacific salmon survey in January 1996 (Ueno et al. 1996). A large surface rope trawl (208-m long, 63.2-m headrope, and 400-m warp) was used to catch salmon in three offshore regions (western, central, and

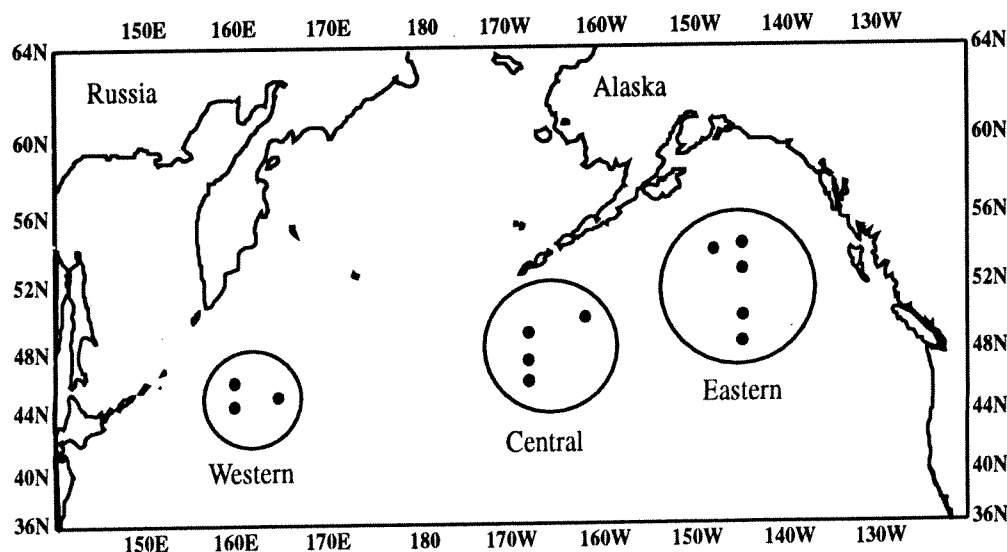
eastern North Pacific Ocean; Fig. 1). A CTD was used to collect data on conductivity, temperature, depth, and dissolved oxygen before or after trawl operations. Seawater was collected by a bucket at the surface and by a CTD rosette sampling system. Seawater nutrients (nitrite-nitrate, silicate, phosphate) and chlorophyll-a were determined from seawater samples within a day of sampling.

Biological Data and Ocean Age Determination

After the net was hauled, salmon in the catch were immediately sorted to species, scales were collected for age determination, the fish were measured (fork length and body weight), blood samples were taken, and the fish were frozen whole (-40°C). After the R/V *Kaiyo maru* returned to Japan, the frozen fish samples were transferred to the National Research Institute of Far Seas Fisheries, FAJ, where additional biological data (sex, gonad weight, liver weight, and stomach content weight) were collected.

Ocean ages of salmon were determined by scientists at the Fisheries Research Institute, University of Washington, from counts of annuli on acetate impressions of scale samples. Ocean age is designated by the European method (a period followed by a number, which is the number of ocean annuli or winters in the ocean). By convention, all salmon caught on the high seas after 1 January are one year older, even if an annulus has not yet formed at the edge of the scale.

Fig. 1 Map showing locations where salmon were sampled for blood during the January 1996 salmon research cruise of R/V *Kaiyo maru* in the western, central, and eastern regions of the North Pacific Ocean.



*Blood Sampling and Measurement of IGF-I***RESULTS**

We attempted to collect blood samples from at least 15 fish per species over the entire survey area (Fig. 1). Blood samples were drawn from the caudal veins of fish (within approximately 1/2 hr after landing) using 10-ml vacuum tubes and holders or syringes fitted with 19-gauge needles. Plasma was separated by centrifugation at 3000 rpm for 15 min. The plasma was auto-pipetted into 1.5-ml cryo-tubes (approximately 1.0 ml of plasma), and stored in a shipboard freezer (-70°C). During the U.S. port call of the R/V *Kaiyo maru*, the frozen plasma samples were transferred to a freezer (-80°C) at the Northwest Fisheries Science Center, National Marine Fisheries Service. Plasma levels of IGF-I in the samples were measured by a homologous radioimmunoassay (RIA; Moriyama et al. 1994). All of the samples were analyzed by the same RIA so that inter- and intra-specific comparisons could be made. Samples with plasma IGF-I levels outside of the area of sensitivity of the RIA were not included in the statistical analyses.

Statistical Analyses

The purposes of our statistical analyses were to evaluate: (1) if the fish in the IGF-I samples were representative of the entire catch, (2) inter-specific differences in plasma IGF-I levels, (3) correlations between IGF-I levels and various biological and environmental variables, and (4) inter- and intra-specific differences in IGF-I levels by sex, age, and ocean region. To evaluate whether fish in the IGF-I samples were representative of the entire catch, body weights of the fish in the IGF-I and non-IGF-I (those fish not sampled for IGF-I) samples were compared by analysis of variance (ANOVA; Super-ANOVA, Abacus Concepts, Berkeley, CA). Inter- and intra-specific differences in plasma IGF-I levels were assessed by ANOVA and Tukey multiple comparisons tests. Plasma IGF-I levels and other biological variables (length, body weight, liver weight, gonad weight, stomach content weight, stomach content index [$100 \times \text{stomach content weight}/\text{body weight}$], and condition factor [$\text{body weight (g)}/\text{fork length (mm)}^3 \times 10^7$]) were tested for correlations (Pearson r statistics, significance tested by Fisher's r to z transformation; StatView, Abacus Concepts, Berkeley, CA). Mean plasma IGF-I levels and environmental data collected at each station (sea surface temperature, surface salinity, surface dissolved oxygen, surface chlorophyll-a, surface nutrients: silicate, phosphate, and nitrate+nitrite) were also tested for correlations. Statistical differences at the $p < 0.05$ level were considered to be significant.

Samples from 98 fish were analyzed by RIA for plasma IGF-I levels: 13 sockeye (*O. nerka*), 29 chum (*O. keta*), 15 pink (*O. gorbuscha*), 26 coho, and 15 chinook (*O. tshawytscha*) salmon. One pink salmon and one chum salmon had IGF-I levels outside of the range of sensitivity of the RIA, and were not included in the statistical analyses. The mean body weights of sockeye and chum salmon in the IGF-I samples were significantly larger than in the non-IGF-I samples (sockeye: $p = 0.03$; chum: $p < 0.01$). There were no significant differences in mean body weights between samples for the other species (pink: $p = 0.08$; coho: $p = 0.20$; and chinook: $p = 0.99$). In the central and eastern regions, the mean body weights of sockeye and chum salmon in the IGF-I samples were larger than those in the entire catch. The mean weights of all species except pink salmon in both the IGF-I samples and in the entire catch increased from west to east across the North Pacific in January (Fig. 2a, b).

For samples pooled over all sex, size, and age groups and ocean regions, mean IGF-I levels were significantly different among the five species sampled: pink (mean = 28 ng/ml, $n = 14$), chum (mean = 33 ng/ml, $n = 28$), chinook (mean = 48 ng/ml, $n = 15$), sockeye (mean = 84 ng/ml, $n = 13$), and coho (mean = 120 ng/ml, $n = 26$) salmon ($p < 0.01$, Table 1). A multiple comparisons test indicated that plasma IGF-I levels in pink, chum, and chinook salmon were not significantly different; levels in pink and chum salmon were significantly different than in sockeye and coho salmon; levels in chinook salmon were significantly different than in coho salmon, but not in sockeye salmon; and levels in sockeye and coho salmon were not significantly different.

Correlations between IGF-I and the biological and environmental variables varied among the species (Table 2). For all species except pink salmon, fork length, body weight, and liver weight were positively correlated with IGF-I levels, and condition factor was negatively correlated with IGF-I levels (Fig. 3, Table 2). Pink salmon IGF-I levels were not correlated with any of the biological variables. Because IGF-I samples from sockeye and pink salmon were collected at only three stations, they were not analyzed for correlations with the environmental data. For chum, chinook, and coho salmon, none of the correlations between mean IGF-I and the environmental data were statistically significant (Table 3).

The species and age composition of fish in the R/V *Kaiyo maru* catches and IGF-I samples varied considerably by ocean region (Ueno et al. 1996; Table 1, Fig. 2). All of the chum, pink, and chinook salmon caught in the western North Pacific were age .1. No sockeye or coho salmon were caught in the western

Fig. 2 Mean body weights and plasma IGF-I levels in five species of salmon by region of the North Pacific Ocean in January 1996. (a) Mean body weight of salmon in IGF-I sample; (b) mean body weight of salmon in the January 1996 *Kaiyo maru* catch; (c) mean levels of plasma IGF-I. Values are mean + SEM. The numbers over the SEM bars in (a) and (b) are sample size.

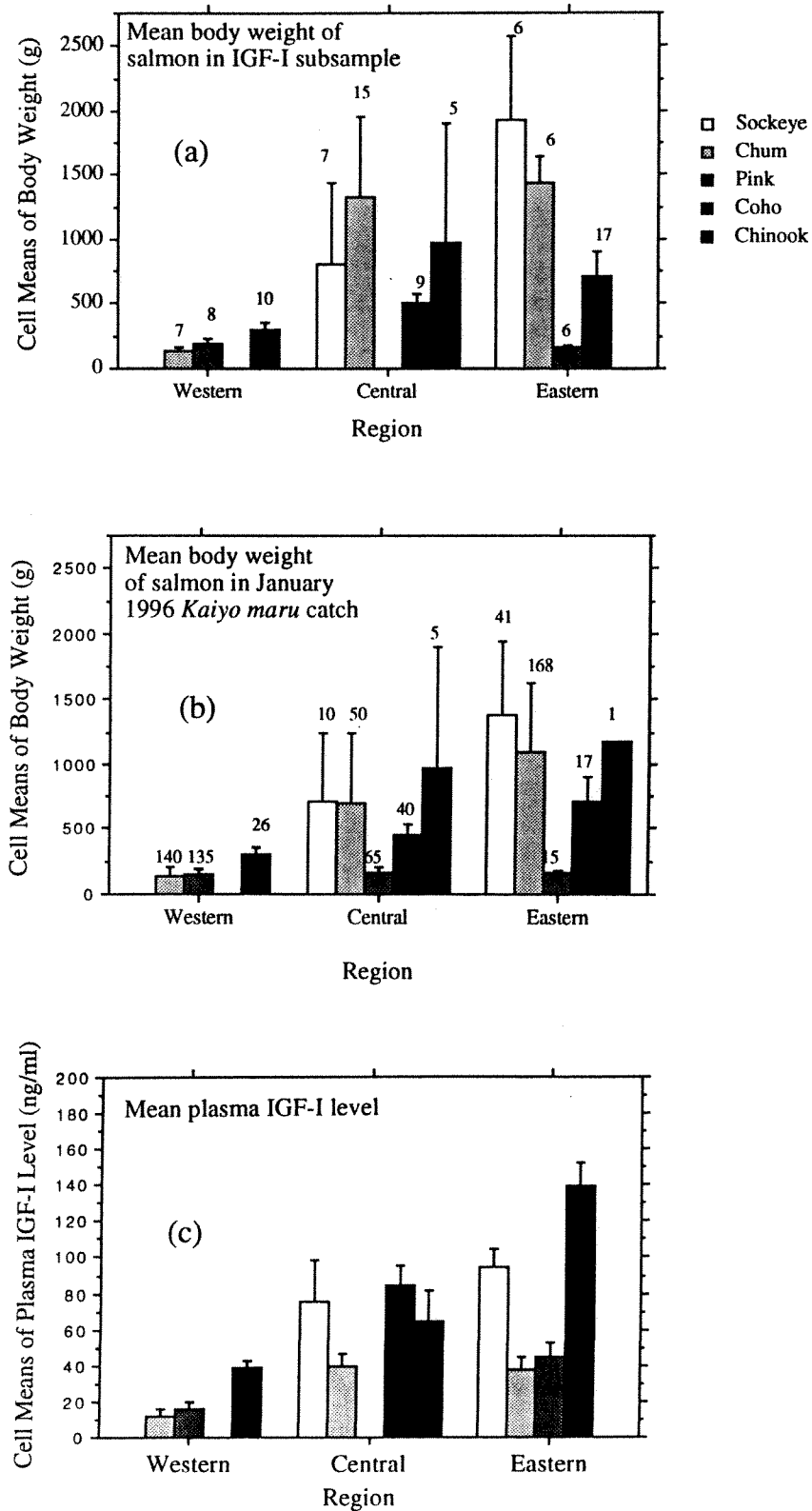


Table 1. Sample size (n) by species, ocean region, and age group, and mean and standard deviation (S.D.) of plasma IGF-I levels.

Species	Region	Age .1			Age .2			Age .3			Age .4			Total		
		n	mean	S.D.	n	mean	S.D.	n	mean	S.D.	n	mean	S.D.	n	mean	S.D.
Sockeye	Western															
	Central	1	44.3		5	54.5	9.8	1	211.4					7	75.5	60.6
	Eastern				1	101.9		5	92.8	27.3				6	94.3	24.7
	Total	1	44.3		6	62.4	21.3	6	112.6	54.2				13	84.2	46.8
Chum	Western	7	12.3	9.9										7	12.3	9.9
	Central				6	16.5	7.3	4	65.2	16.4	5	48.2	26.4	15	40.0	26.8
	Eastern				1	61.9		5	33.0	14.2				6	37.8	17.3
	Total	7	12.3	9.9	7	22.9	18.4	9	47.3	22.1	5	48.2	26.4	28	32.6	24.4
Pink	Western	8	16.1	9.7										8	16.1	9.7
	Central															
	Eastern	6	44.5	20.3										6	44.5	20.3
	Total	14	28.3	20.6										14	28.3	20.6
Coho	Western															
	Central	9	84.5	33.1										9	84.5	33.1
	Eastern	17	138.9	55.3										17	138.9	55.3
	Total	26	120.1	54.8										26	120.1	54.8
Chinook	Western	10	38.8	14.0										10	38.8	14.0
	Central	3	59.6	41.7	2	73.5	39.2							5	65.2	36.2
	Eastern															
	Total	13	43.6	22.8	2	73.5	39.2							15	47.6	25.8
Total n		61			15			15			5			96		
% of total		63.5			15.6			15.6			5.2					

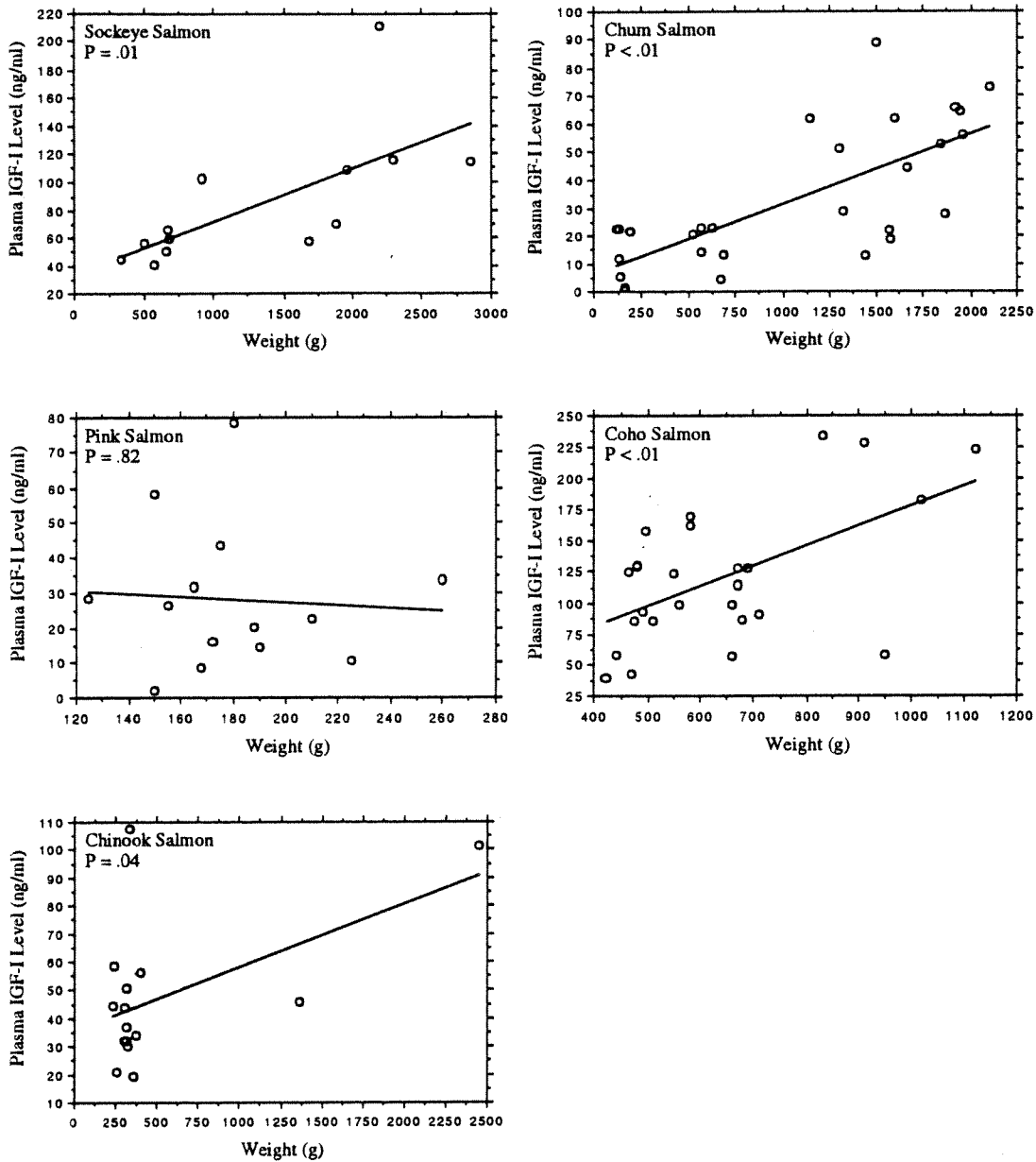
Table 2. Correlations (Pearson r) between plasma IGF-I levels and other biological variables by species. * = statistically significant.

Variables	Sockeye		Chum		Pink		Coho		Chinook	
	r	P-value	r	P-value	r	P-value	r	P-value	r	P-value
Fork length	0.699	0.0062 *	0.654	<0.0001 *	0.008	0.9779	0.538	0.0039 *	0.490	0.0632
Body weight	0.682	0.0084 *	0.723	<0.0001 *	-0.068	0.8225	0.557	0.0026 *	0.524	0.0439 *
Liver weight	0.588	0.0327 *	0.651	0.0001 *	0.376	0.1903	0.519	0.0058 *	0.582	0.0212 *
Gonad weight	0.512	0.0738	0.069	0.7305	0.065	0.8282	-0.216	0.2933	0.580	0.0218 *
Stomach content weight	0.321	0.2919	0.443	0.0172 *	-0.179	0.5495	-0.093	0.6553	-0.100	0.7278
Stomach content index	0.213	0.4939	-0.109	0.5847	-0.241	0.4142	0.254	0.2138	-0.299	0.2853
Condition factor	-0.632	0.0186 *	-0.583	0.0009 *	0.095	0.7520	-0.545	0.0034 *	-0.418	0.1231
Number of observations	13		28		14		26		15	

Table 3. Correlations (Pearson r) between mean plasma IGF-I levels and environmental variables by species. None of the correlations were statistically significant.

Variables	Chum		Coho		Chinook	
	r	P-value	r	P-value	r	P-value
Sea temperature	-0.141	0.7764	0.288	0.6751	0.450	0.4927
Salinity	-0.599	0.1669	-0.386	0.5653	-0.763	0.1555
Dissolved oxygen	0.253	0.6048	-0.023	0.9739	-0.125	0.8594
Chlorophyll-a	-0.577	0.1884	-0.047	0.9467	-0.416	0.5316
Silicate	0.629	0.1392	-0.075	0.9153	0.744	0.1750
Phosphate	-0.404	0.3911	0.385	0.5654	-0.659	0.2630
Nitrite-Nitrate	-0.119	0.8109	0.205	0.7685	-0.582	0.3465
Number of observations	7		5		5	

Fig. 3 Scatter plots showing correlations (Pearsons r) between plasma IGF-I levels and body weight in five species of salmon sampled in the North Pacific Ocean in January 1996. Regression lines are also plotted.



North Pacific. All of the pink and coho salmon in the catches were age .1 (first winter in the ocean). Pink salmon were caught in all regions, but they were not sampled for IGF-I in the central region. The ages of sockeye, chum, and chinook salmon increased from west to east across the North Pacific. Only one chinook salmon was caught in the eastern region, and was not sampled for IGF-I. For samples pooled over all regions, there were no significant differences in mean IGF-I levels by sex for pooled ages .2 and .3 sockeye ($p = 0.75$), age .1 pink ($p = 0.97$), age .1 coho ($p = 0.09$), age .1 chinook ($p = 0.90$), and

ocean ages .1 ($p = 0.47$), .2 ($p = 0.19$), and .3 ($p = 0.28$) chum salmon. Only one female age .4 chum salmon was sampled.

There were significant inter- and intra-specific differences in mean IGF-I levels by age group and by region (Table 1). In general, mean IGF-I levels in all species increased from west to east across the North Pacific Ocean in January (Fig. 2c). The mean IGF-I levels of age .1 chum, pink, and chinook salmon in the western North Pacific were significantly different ($p < 0.01$). A multiple comparisons test indicated that the mean IGF-I in age .1 chinook salmon was

significantly higher than in age .1 pink and chum salmon in the western North Pacific, and levels in pink and chum salmon were not significantly different. The mean IGF-I of age .1 pink salmon was significantly higher in the eastern than in the western region ($p < 0.01$). The mean IGF-I of coho salmon was significantly higher in the eastern than in the central region ($p = 0.01$). For samples pooled over both the central and eastern North Pacific regions, age .3 sockeye salmon had significantly higher IGF-I levels than age .2 fish ($p < 0.01$).

For chum salmon samples pooled over all regions, there were significant differences ($p = 0.01$) in mean IGF-I levels by ocean age group. A multiple comparisons test indicated that mean IGF-I levels were significantly higher for older (ocean ages .3 and .4) than for younger (ocean ages .1 and .2) chum salmon, but differences between ages .1 and .2 fish and between ages .3 and .4 fish were not significant. Samples were generally not sufficient to make comparisons for the differences in IGF-I levels of the four age groups of chum salmon within and between regions, but mean IGF-I levels of age .3 chum salmon were significantly higher ($p = 0.02$) in the central than in the eastern region. In the central region, mean IGF-I levels of ages .2, .3, and .4 chum salmon were significantly different ($p < 0.01$), and a multiple comparisons test indicated that levels in younger (age .2) chum salmon were significantly lower than older (ages .3 and .4) fish. The IGF-I levels in older chum salmon in the central region were not significantly different from each other.

DISCUSSION

In this paper, we used a new physiological approach to investigate the growth of salmon in offshore waters. We selected serum IGF-I levels as a growth index because a previous study showed that IGF-I was significantly correlated with specific growth rates for coho salmon held in seawater net pens (Duan et al. 1995). In salmon, the liver is the major site of IGF-I synthesis, and growth hormone (GH) is the primary positive regulator of IGF-I synthesis (Duan et al. 1994). Insulin and thyroid hormone also stimulate IGF-I expression (e.g., Duan et al. 1992; Duguay et al. 1994; Plisetskaya and Duan 1994). Through a negative feedback loop, IGF-I inhibits synthesis, secretion, and release of GH (Tannenbaum et al. 1983; Yamashita and Melmed 1986; Perez-Sanchez et al. 1993). Duan et al. (1995) hypothesized that elevated GH levels in stunted coho salmon are due to low plasma IGF-I, which results in reduced negative feedback on GH production. Studies on seasonal and developmental (during the parr-smolt transformation and sexual maturation periods) changes of plasma-circulating IGF-I in salmonid fish are in progress

(Moriyama, Duan, Dickhoff, Larsen, Beckman, Swanson, and Plisetskaya, unpublished, cited by Duan et al. 1995). There were no previous data, however, on plasma IGF-I levels for salmon in offshore waters. Our study was limited to a relatively small number of samples collected in the North Pacific Ocean in January 1996. Additional samples from broader times and areas are needed to better understand the role of IGF-I in growth of salmon in offshore waters.

The quality of blood samples from trawl-caught salmon for RIA was excellent (only 2% of the samples were unusable). However, the samples of chum and sockeye salmon from the central and eastern regions were not representative of the entire catch (significantly larger body weights, Fig. 2a, b). Because plasma IGF-I levels are correlated with body size of fish in most species (Fig. 3), future high-seas samples should be stratified by body size of fish if possible. In reality, because many variables and factors are involved (e.g., origin, season, race, sex, maturity, age, gear selectivity, and variations between years, areas, and conditions), random sampling of salmon in offshore waters is difficult, and there is never certainty that samples are unbiased (Takagi et al. 1981). In addition, we encourage the collection of samples from as many different stations as possible. In our study, the number of stations sampled for each species was not sufficient to evaluate correlations between mean IGF-I levels and environmental factors.

The inter-specific differences in IGF-I levels in our samples probably reflect differences in growth rates among species of salmon in the North Pacific Ocean, but we need more information on the significance of high and low levels of IGF-I. According to Ishida et al. (1998), data on monthly changes in average fork length, body weight, and condition factor of salmonids caught in offshore waters of the North Pacific Ocean indicates that pink, coho, and sockeye salmon continue to grow throughout the winter; coho salmon, which had the highest levels of IGF-I in our study, grow faster in winter than any of the other species; sockeye and chinook salmon, which did not have significantly different levels of IGF-I, may have similar winter growth patterns; and chum salmon, which had low levels of IGF-I, apparently do not grow during winter months. IGF-I levels might be expected to be lower in pink salmon than in coho salmon, because pink salmon apparently grow more slowly than coho salmon (Ishida et al. 1998). But if pink salmon continue to grow throughout the winter and chum salmon do not, we do not understand why IGF-I levels in pink and chum salmon caught in the western region were not significantly different.

In all of the species except pink salmon, IGF-I levels were positively correlated with fork length, body weight, and liver weight, and negatively

correlated with condition factor (Table 2, Fig. 3). Perhaps pink salmon IGF-I levels were not correlated with any of the biological variables because there was less variation in the size of pink salmon than there was in the other species. The significant negative correlations between IGF-I and condition factor in sockeye, chum, and coho salmon are interesting. In the North Pacific Ocean, condition factors in all species of salmon are lowest in winter (Ishida et al. 1998). Perhaps the inverse relation between condition factor and IGF-I may be due to IGF-I having a greater effect on growth in length than weight.

That there are significant intra-specific differences in the ocean growth of salmon by sex, age, origin, maturity group, ocean region, and many other factors is well known (e.g., Takagi et al. 1981). Ueno et al. (1996) hypothesized that in the western North Pacific Ocean young (age .1) pink and chum salmon move offshore from coastal waters in January, and immature (including young) sockeye and chum salmon migrate eastward from the northwestern Pacific to the central North Pacific in winter. Because salmon make rapid and extensive migrations in offshore waters, their plasma IGF-I levels may not be directly related to local environmental conditions. Some of the intra-specific differences in IGF-I levels that we observed could be stock-specific, but origins of fish in our samples were not known. Significantly higher mean IGF-I levels in pink salmon in the eastern than in the western North Pacific, may indicate the earlier resumption of rapid growth in pink salmon in the Gulf of Alaska.

North Pacific researchers have used stomach content data as an indicator of how well fish are feeding and growing (e.g., Ishida et al. 1996). In all but one case, IGF-I levels were not significantly correlated with stomach content variables (chum salmon stomach content weight, Table 2). Duan et al. (1995) cautioned that poor growth in salmon may sometimes be linked to GH resistance and reduced IGF-I production rather than to fasting or starvation. In their study, stunted fish fed actively and most had full stomachs, but they had significantly lower IGF-I levels, growth rates, body weights, lengths, and condition factors than normally growing fish. We think that IGF-I levels may provide a useful tool for detecting poorly growing fish in high-seas samples.

CONCLUSIONS

Additional data on the significance of high or low levels of IGF-I in salmon are needed. The most straight-forward interpretation of our results is that high IGF-I levels correlate with high growth rates. However, there may be some exceptions that we do not know about. Cooperative Japan-U.S. investigations of IGF-I in North Pacific salmon are

continuing. Salmon blood samples were collected in June-July 1996 aboard the Japanese vessels R/V *Wakatake maru* and T/S *Oshoro maru* in the central North Pacific, Bering Sea, and Gulf of Alaska (Davis et al. 1996; Myers et al. 1996). These samples will be evaluated with respect to biological and environmental factors and scale-pattern measures of growth (Walker et al. 1998). We also hope to incorporate information on growth rates from IGF-I into species-, region-, and stock-specific bioenergetic models being developed for salmon in offshore waters (Davis et al. 1998).

ACKNOWLEDGEMENTS

We thank Capt. Kakusuke Nakayama and the officers and crew of the R/V *Kaiyo maru*, Fisheries Agency of Japan (FAJ), and Drs. Yukimasa Ishida, Yasuhiro Ueno, and Akihiro Shiimoto, National Research Institute of Far Seas Fisheries (NRIFSF), FAJ, for their assistance in sample and data collection. We especially thank the NRIFSF for giving us permission to use the R/V *Kaiyo maru* data in our analyses. Beeda Lee-Pawlak, U.S. National Marine Fisheries Service (NMFS), Northwest Fisheries Science Center, is gratefully acknowledged for her laboratory analyses of blood samples and for providing a computer spreadsheet of IGF-I values. Robert V. Walker provided useful comments on the manuscript. Funding for Fisheries Research Institute participation was provided by NMFS, Alaska Fisheries Science Center, Auke Bay Laboratory (NOAA contract Nos. 50ABNF400001 and 50ABNF700003).

REFERENCES

- Bern, H.A., S.D. McCormick, K.M. Kelley, E.S. Gray, R.S. Nishioka, S.S. Madsen, and P.I. Tsai. 1991. Insulin-like growth factors "under water": role in growth and function of fish and other poikilothermic vertebrates. p. 85-96 in E.M. Spencer (ed.) *Modern Concepts of Insulin-Like Growth Factors*. Elsevier, New York.
- Cohick, W.S., and D.R. Clemmons. 1993. The insulin-like growth factors. *Annu. Rev. Physiol.* 55:131-153.
- Davis, N.D., M. Takahashi, and Y. Ishida. 1996. The 1996 Japan-U.S. cooperative high-seas salmon research cruise of the *Wakatake maru* and a summary of 1991-1996 results. (NPAFC Doc. 194.) FRI-UW-9617. University of Washington, Fisheries Research Institute, Seattle. 45 p.
- Davis, N.D., K.W. Myers, and Y. Ishida. 1998. Caloric value of high-seas salmon prey organisms and simulated salmon ocean growth and prey consumption. *N. Pac. Anadr. Fish Comm.*

- Bull. No. 1: 146-162
- Duan, C., S.J. Duguay, and E.M. Plisetskaya. 1992. Hormonal regulation of insulin-like growth factor I (IGF-I) mRNA expression in coho salmon. *Am. Zool.* 32:13. (Abstract)
- Duan, C., S.J. Duguay, P. Swanson, W.W. Dickhoff, and E.M. Plisetskaya. 1994. Tissue-specific expression of insulin-like growth factor-I ribonucleic acids in salmonids: developmental, hormonal, and nutritional regulation. Pages 365-372 in K.G. Davey, S.S. Tobe, and D.E. Peter (eds.) *Perspectives in Comparative Endocrinology*. National Research Council of Canada, Toronto.
- Duan, C., E.M. Plisetskaya, and W.W. Dickhoff. 1995. Expression of insulin-like growth factor-I in normally and abnormally developing coho salmon (*Oncorhynchus kisutch*). *Endocrinology* 136(2):446-452.
- Duguay, S.J., P. Swanson, and W.W. Dickhoff. 1994. Differential expression and hormonal regulation of alternatively spliced IGF-I mRNA transcripts in salmon. *J. Mol. Endocrinol.* 12:25-37.
- Ishida, Y., S. Ito, G. Anma, T. Meguro, S. Takagi, Y. Kamei, N.D. Davis, and K.W. Myers. 1996. Salmon stock assessment aboard the Japanese salmon research vessels in the North Pacific Ocean, 1996. (NPAFC Doc. 210.) *Nat. Res. Inst. Far Seas Fish., Japan; Hokkaido Univ., Japan; Fish. Res. Inst., USA.* 28 p.
- Ishida, Y., S. Ito, Y. Ueno, and J. Sakai. 1998. Seasonal growth patterns of Pacific salmon (*Oncorhynchus* spp.) in offshore waters of the North Pacific Ocean. *N. Pac. Anadr. Fish Comm. Bull. No. 1:* 66-80
- Kikuchi, K., F.C. Buonomo, Y. Kajimoto, and P. Rotwein. 1991. Expression of insulin-like growth factor-I during chicken development. *Endocrinology* 128:1323-1328.
- Moriyama, S., P. Swanson, M. Nishii, A. Takahashi, H. Kawachi, W.W. Dickhoff, and E.M. Plisetskaya. 1994. Development of a homologous radioimmunoassay for coho salmon insulin-like growth factor-I. *Gen. Comp. Endocrinol.* 96:149-161.
- Myers, K.W., K. Y. Aydin, and G. Anma. 1996. The 1996 international cooperative salmon research cruise of the *Oshoro maru* and a summary of 1994-1996 results. (NPAFC Doc. 195.) FRI-UW-9613. University of Washington, Fisheries Research Institute, Seattle. 32 p.
- North Pacific Anadromous Fish Commission. 1996. Science plan 1995-96. *N. Pac. Anadr. Fish Comm., Vancouver, B.C.* 10 p.
- Perez-Sanchez, J., C. Weil, and P-Y. Le Bail. 1993. Effects of human insulin-like growth factor-I on the release of growth hormone by rainbow trout (*Oncorhynchus mykiss*) pituitary cells. *J. Exp. Zool.* 262:287-290.
- Plisetskaya, E.M., and C. Duan. 1994. Insulin and insulin-like growth factor I in coho salmon, *Oncorhynchus kisutch*, injected with streptozotocin. *Am. J. Physiol.* 267:R1408-1412.
- Takagi, K., K.V. Aro, A.C. Hartt, and M.B. Dell. 1981. Distribution and origin of pink salmon (*Oncorhynchus gorbuscha*) in offshore waters of the North Pacific Ocean. *Int. N. Pac. Fish. Comm. Bull.* 40. 195 p.
- Tannenbaum, G.S., H.J. Guyda, and B.I. Posner. 1983. Insulin-like growth factors: a role in growth hormone negative feedback and body weight regulation via brain. *Science* 220:77-79.
- Ueno, Y., Y. Ishida, A. Shiomoto, S. Urawa, K.W. Myers, J. Morris, and M.V. Koval. 1996. Summary of wintering salmon research aboard the research vessel *Kaiyo maru* in January 1996. (NPAFC Doc. 213.) National Research Institute of Far Seas Fisheries, Japan. 20 p.
- Walker, R.V., K.W. Myers, and S. Ito. 1998. Growth studies from 1956-95 collections of pink and chum scales in the central North Pacific Ocean. *N. Pac. Anadr. Fish Comm. Bull. No. 1:* 54-65
- Yamashita, S., and S. Melmed. 1986. Insulin-like growth factor I action on rat anterior pituitary cells: suppression of growth hormone secretion and messenger ribonucleic acid levels. *Endocrinology* 118:176-184.