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Abstract

In order to investigate the environments for salmon habitat and distributions of salmon in the Bering Sea, the temperature, salinity and abundance of salmon were observed in summer to autumn in 2002. Trawl, hydrobiological, and oceanographic sampling by R/V *Kaiyo maru* was conducted 62 trawl operations at 38 stations from June 29 to July 14 (leg 1) and from August 21 to September 18 (leg 3 and 4) in the Bering Sea. In the Bering Sea, temperatures in the depth of 30 m were about 3 to 8 ºC in leg 1, and they were about 5 to 8 ºC in leg 3 and 4. A total of 11,369 salmon were captured including pink salmon (13), chum salmon (10249), sockeye salmon (738), coho salmon (4) and chinook salmon (445) in the Bering Sea. The total lipid (TL) contained in the white muscle of chum salmon caught in June and July ranged from 0.4% to 15.6% (n=123). The TL contained in chum salmon caught in September ranged from 1.0% to 21.2% (n=334). High lipid content was observed even in small size fish. Haplotypes of mitochondria DNA of chum salmon were mostly analyzed during the cruise of leg 4. The haplotypes characteristics to Asian populations were dominant in the central region of the Bering Sea.

Introduction

Japan continues to monitor summer salmon stocks and environments in the Bering Sea using research gillnets since 1992. However, data of the monitoring research is not sufficient to estimate abundance of salmon and to determine their ecosystem structures, because of the limited survey area in the central Bering Sea. In first year 2002, we conducted intensive surveys in the whole areas of the Bering Sea using trawl nets by R/V *Kaiyo maru* according to the Bering-Aleutian Salmon International Survey (BASIS), which is a North Pacific Anadromous Fish Commission (NPAFC)-coordinated program of cooperative research in the Bering Sea.

Material and methods

In order to investigate the environments for salmon habitat and distributions of salmon in the Bering Sea, the temperature, salinity and abundance of salmon were observed in summer to autumn in 2002. Trawl, hydrobiological, and oceanographic

sampling by R/V *Kaiyo maru* was conducted 62 trawl operations at 38 stations from June 29 to July 14 (leg 1) and from August 21 to September 18 (leg 3 and 4) in the Bering Sea (Fig. 1). Plankton and oceanographic sampling were conducted at every station at the approximate location of the trawl tows. Temperature and salinity data were collected using a SBE9+ CTD from 0 to 3000 meters, or 0 to near sea floor in leg 1, and from 0 to 1000 meters, or 0 to near sea floor in leg 3, 4. CTD Rosette sampling using the 2.5 liter Niskin bottle x 7 depth was made for collection of the water at the depth of 0 (bucket sample), 10, 20, 30, 50, 75, and 100m for the concentration of chlorophyll-a. Zooplankton was collected using a NORPAC net at each station. An ORI net and RMT net were also conducted for zooplankton sampling at nighttime. The ORI net was towed the surface for 10 minutes after sunset 1 hour. RMT net was towed from about 500 meters to surface. Multi layer net was conducted at two stations in the Bering Sea at daytime and nighttime, respectively. The sampling by multi layer net was carried out at each depth layers from 500 m to surface, 0 - 500 m, 500 - 300 m, 300 - 200 m, 200 - 100 m, 100 - 50 m, 50 - 25m, and 25 - 0m, with changing nets. Particle size of plankton, salinity, temperature, fluorescence and dissolved oxygen in the sea surface were measured continuously by EPCS (Electronic Particle Counting and Sizing System). These data were stored every 1 minute. The fluorescence and number of particle corresponds to the phytoplankton and zooplankton biomass, respectively.

DNA samples to be analyzed were obtained from about 1000 chum salmons caught by surface layer trawl operations at 18 stations in the Bering Sea during leg 4. DNA of individual fish was extracted from blood samples collected from caudal vasculatures, and were subjected to PCR amplification with a set of oligonucleotide primers. PCR products were individually applied to a microarray, and were incubated for hybridization and coloration. To determine haplotypes, the patterns of reaction were captured and stored in a personal computer as an image data with an image scanner.

Results

Oceanographic conditions in the Bering Sea during summer 2002

During June to mid of September, hydrographic observations of the total of 84 CTD casts were conducted in the Bering Sea. These hydrographic stations covered in the central Bering Sea. Thus, the hydrographic observations showed the synoptic character of oceanographic conditions in the Bering Sea in summer.

In leg1, temperatures in the depth of 30 m, which was the center of mouth of

trawl net, were about 3 to 8 ºC. In leg 3 and 4, thought these temperatures rose about 2 ºC, temperature did not rise near the Unimak Pass (Fig. 2). Temperature in the eastern Bering Sea shelf was less than 5 ºC. The temperature in the southern area in the Bering Sea basin was lower than that in northeastern area in the Bering Sea basin. There was counterclockwise circulation around the Bering Sea basin in the field of geostrophic currents at the depth of 30 m, using a reference level of 1000 db (Fig. 2). Athough the Bering Slope Current along the eastern Bering Sea shelf break was not clear, there were some anticyclonic eddies. Velocity of inflows through the Amchitka Pass and Amukuta Pass from North Pacific were about 10 cm/s. Figure 3 shows the vertical section of temperature along 180º. The thickness of the surface mixed layer was about 20m. The minimum temperature less than 2 ºC was observed in the depth of 60 m to 200m in the central Bering Sea. Under the minimum temperature layer, the maximum temperature of 3.7 ºC was observed. In the Amchitka Pass, these hydrographic structures were not occurred.

Chlorophyll-a concentration more than 2 mg/m^3 appeared in the eastern Bering Sea shelf break, Unimak Pass and east area of the Bowers Ridge in the depth of 10 m (Fig. 4). Relatively low chlorophyll-a concentration existed around north of the Bowers Ridge, where zooplankton concentration observed by EPCS (Electric Particle Counter System) was the relatively high (Fig. 5). Chlorophyll-a concentration was the highest at 54 ºN, 166 ºW in the Bering Sea. It was suggested that the increasing chlorophyll-a concentration in those areas was caused by the eddy or tidal mixing.

Comparison of fishing gears

To estimate the difference between catch efficiencies of trawl net and of drift gillnet, R/V *Kaiyo maru* and R/V *Wakatake maru* conducted 4 operations at the same stations using a trawl net and research gillnets from July 1 to July 5, 2002, in the central Bering Sea. Many chum salmon were caught using both the trawl net and gillnets. Numbers of chum salmon caught using 30 tans of research gillnets was 4.11-fold of numbers of chum salmon using the trawl net. In one of four operations, fork length of chum salmon caught using research gillnets was larger than fork length using the trawl net.

Distributions of salmon and other fishes

To estimate salmon distribution in the Bering Sea, the R/V *Kaiyo maru* conducted 62 trawl operations at 38 stations from June 29 to July 14 and from August 21 to September 18 in summer of 2002. Main catches included sockeye (*Oncorhynchus*

nerka), chum *(O. keta)*, chinook salmon *(O. tshawytscha)*, Atka mackerel *(Pleurogrammus monopterygius)*, walleye Pollock *(Theragra chalcogramma)*, and other fishes. A total of 11,446 salmon were captured including pink salmon (*O. gorbuscha*) (0.1%), chum salmon (89.5%), sockeye salmon (6.4%), coho salmon (*O.kisutch*) (0.03%) and chinook salmon (3.9%). Horizontal distributions of salmon are shown in Fig. 6. Number weight of samples is listed in Table 1. Total weight of Atka mackerel in the Bering Sea amounts to 15,360kg. CPUEs (number of fish per trawl) of sockeye and chum salmon were larger in the eastern Bering Sea than in the western area. CPUE of Atka mackerel was larger in the western Bering Sea, and CPUEs of chinook salmon and walleye pollock were large in the central Bering Sea. Distributions of sockeye and chinook salmon were affected by sea surface temperature (SST), but distribution of chum salmon was not. Distribution of Atka mackerel differed from distribution of salmon and was not affected by SST.

Total lipid content in the white muscle of chum salmon caught in the Bering Sea in the late summer 2002

Generally, it is assumed that salmon in the ocean consume prey heavily in the summer and, therefore, the lipid level in the muscle of these fish may be higher than in spring and winter. This study is the first to report the total lipid (TL) content in the white muscle of chum salmon caught in the international waters of the Bering Sea in late summer.

The TL was extracted from the white muscle of 457 chum salmon using chloroform and methanol and then measured gravimetrically. The TL contained in chum salmon caught in June and July ranged from 0.4% to 15.6% (n=123). The TL contained in chum salmon caught in September ranged from 1.0% to 21.2% (n=334). High lipid content was observed even in small size fish (Fig. 7). There was significant relationship between total lipid content and moisture content in the muscle (Fig. 8). High lipid content in September may indicate that late summer is the time when lipid is stored at the expense of growth in order to promote survival through a lean winter. We recommend continuing the study of chum salmon TL content by age group with an emphasis on monitoring seasonal changes, particularly those that occur in the fall.

DNA microarray for rapid detection of mitochondrial DNA haplotypes of chum salmon and its application to genetic stock identification

In our previous study, we examined the genetic structure of chum salmon in the Pacific Rim based on the nucleotide sequences of 5' end portion of mitochondrial

DNA (mtDNA) control region, and showed the presence of 20 variable nucleotide sites in 500 bp which define 30 haplotypes of three genealogical groups referred to as A, B, and C in more than 2500 individuals of about 50 populations. The haplotypes composing groups A (yellow in Fig. 9) might be derived from Japanese populations, those composing group C (red) were found mostly in Asian populations, and those characterizing group B (blue) distinguish North American populations.

We considered that these mtDNA data would be useful for constructing the baseline for stock identification of mixed high seas populations of chum salmon, so that developed DNA microarray that can discriminate a particular haplotype among others by use of sequence specific oligonucleotides as captures. Preliminary experiments confirmed rapid and accurate ability of newly developed microaray to detect particular haplotypes from PCR-amplified DNA samples. Hence we tested whether our protocol to analyze haplotypes of chum salmon DNA microarray is effective to rapidly identify genetic stocks on the research vessels.

The haplotypes of chum salmon, which were collected in the leg 4, were mostly analyzed during the cruise. Only very small portion of samples could not be identified, probably because they have unknown sequence variability. According to the preliminary analysis for those one years sample, distribution of haplotypes was not even among the stations. The haplotypes characteristics to Asian populations were dominant in the central region of the Bering Sea, where the water temperature was relatively high compared to neighboring regions (Fig. 2). In contrast, the haplotypes belonging to group B were dominant in rather cooler regions.

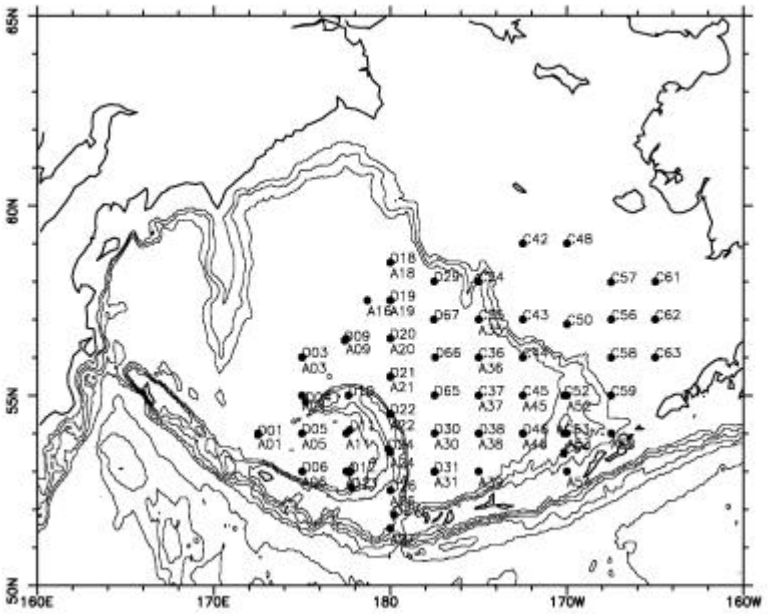

Figure 1. Observation points of R/V *Kaiyo maru* from June to September cruise in 2002. A represents observation points during leg 1, C represents observation points during leg 3, and D represents observation points during leg 4.

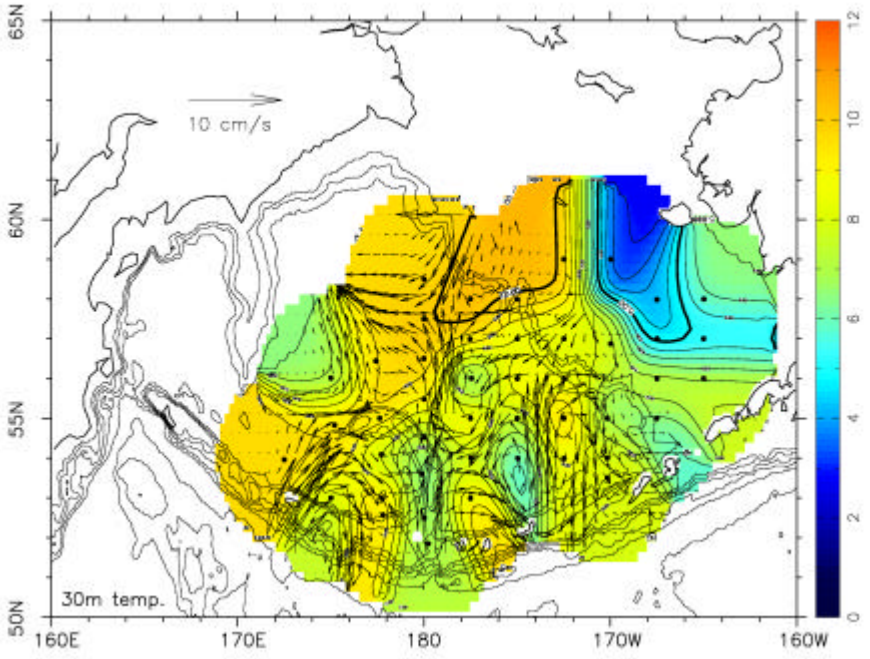

Figure 2. Horizontal distributions of temperature and geostrophic currents during

summer 2002 at depth of 30 m, using a reference level of 1000 db.

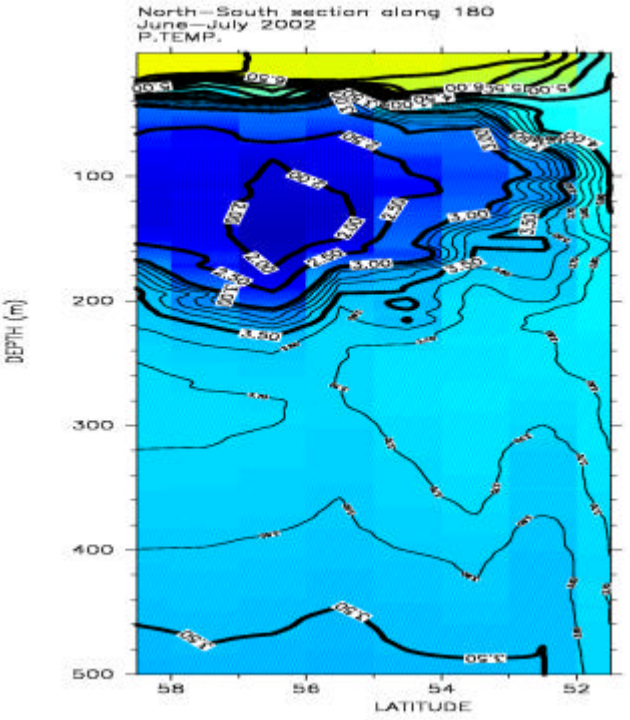

Figure 3. Vertical distributions of temperature along 180°.

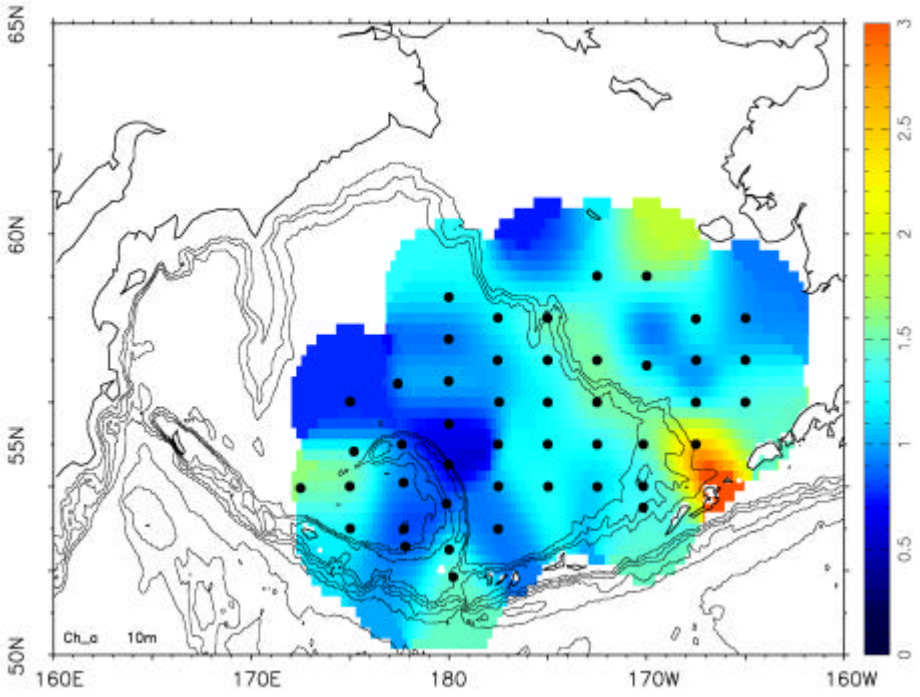

Figure 4. Horizontal distributions of chlorophyll-a concentrations (mg/m3) at the

depth of 10 m during leg 3 and leg 4, 2002.

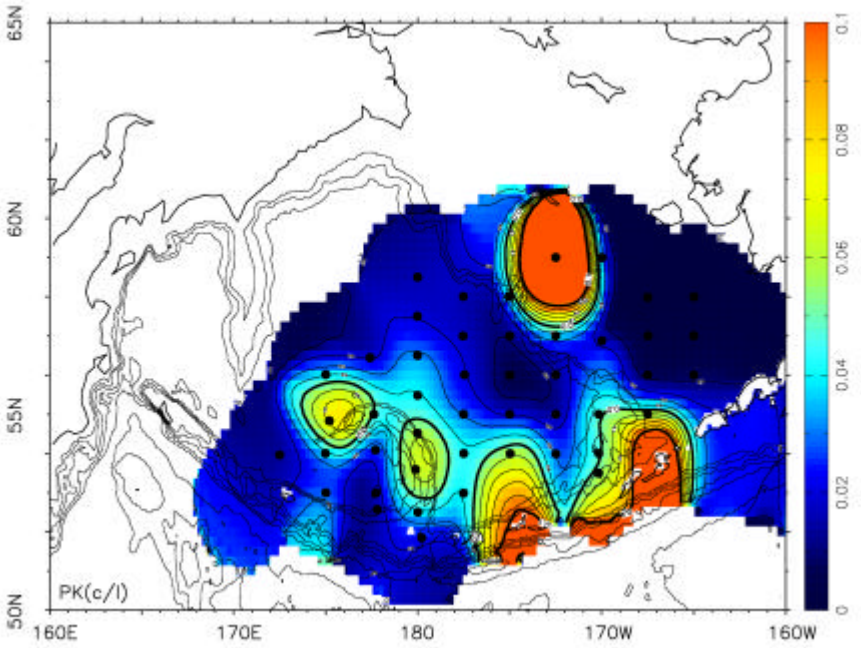

Figure 5. Horizontal distributions of zooplankton at sea surface by the EPCS during leg 3 and 4 in 2002.

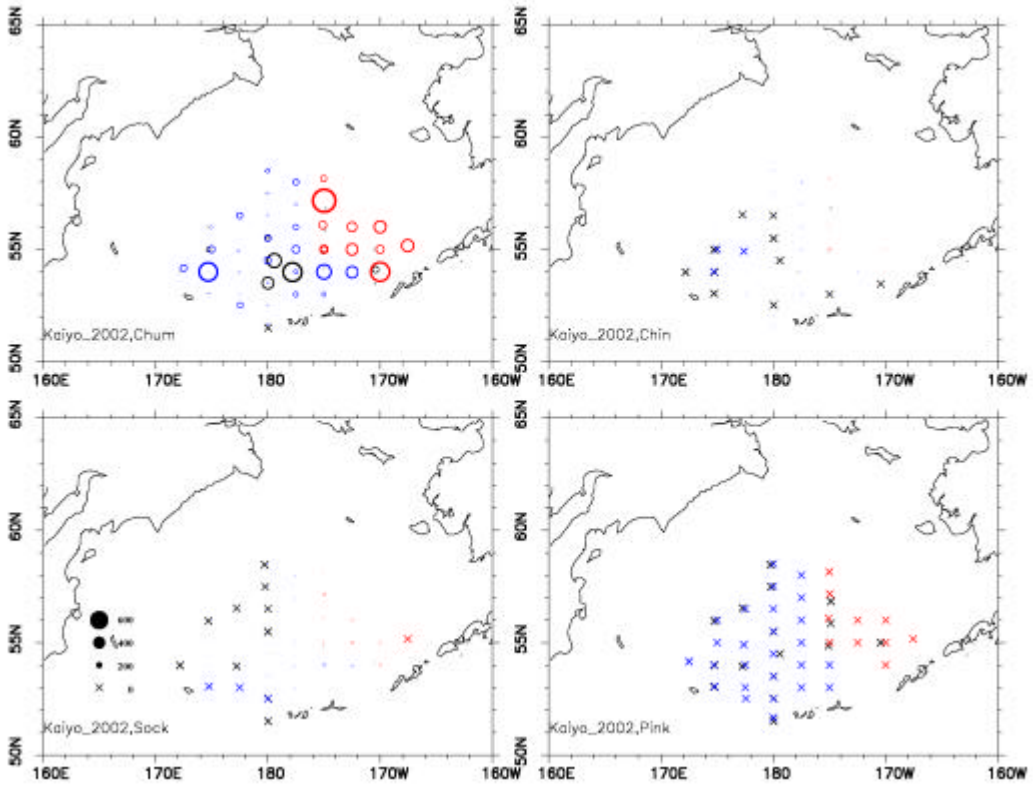

Figure 6. Horizontal distributions of salmon in the Bering Sea. Black circles indicate

leg 1, red leg 3, and blue leg 4.

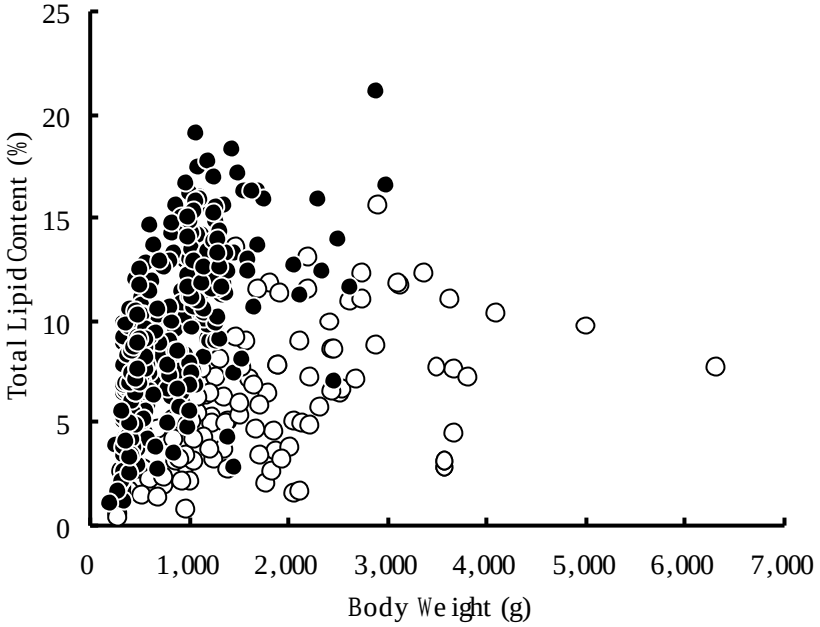

Figure 7. Relationship between total lipid content in white muscle and body weight in chum salmon caught in Bering Sea in June and July (open circle) and September (solid circle).

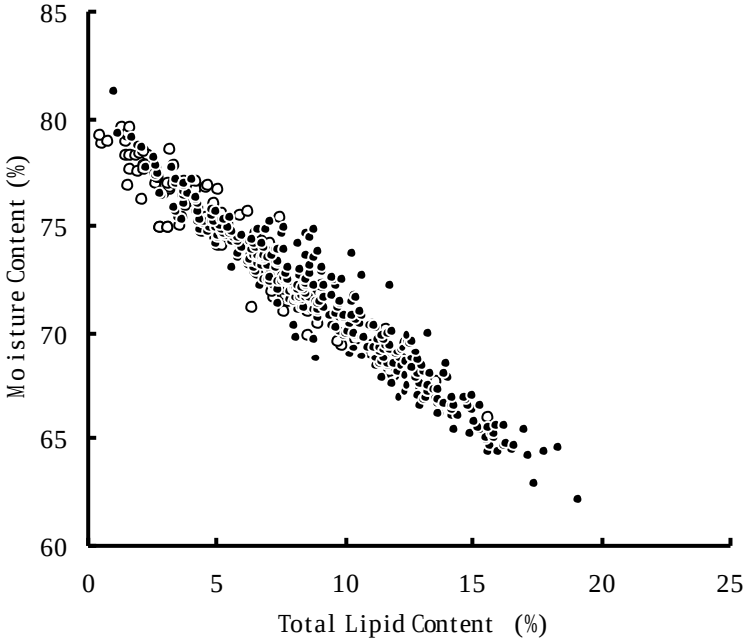

Figure 8. Relationship between total lipid content and moisture content in white muscle in chum salmon caught in Bering Sea in June and July (open circle) and

September (solid circle).

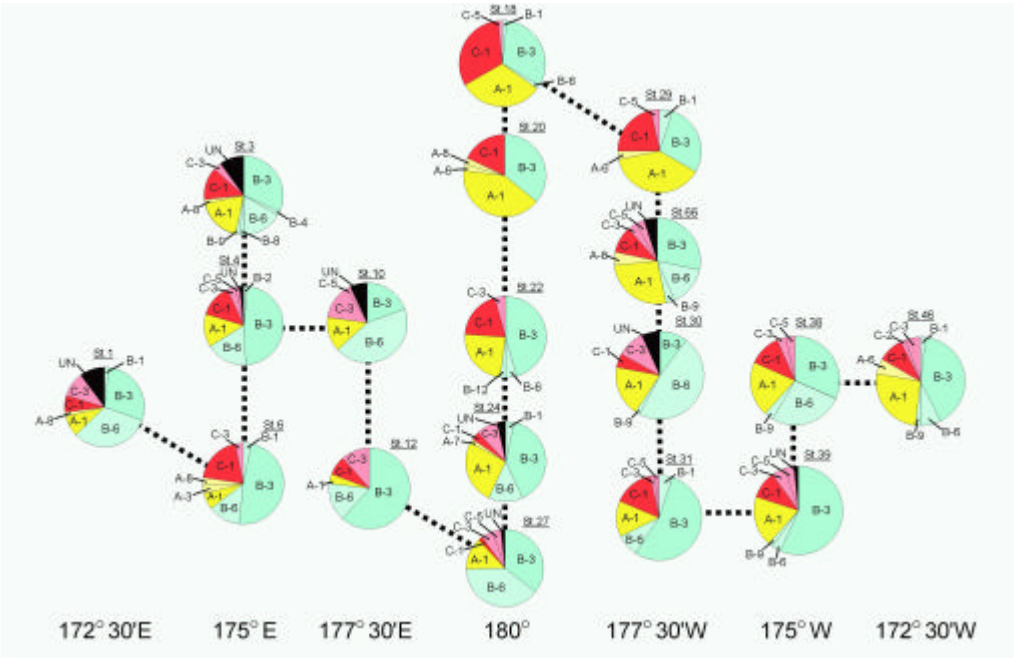

Figure 9. Haplotype distribution of mix stocks of chum salmon in the Bering Sea.

Table 1. Numbers and weight of salmonids and other organisms cought by *Kaiyo maru* in 2002.

	Date		Sock	Chum	Pink	Coho	Chinook	Steelhead	Atka mackerel	Walleye pollock
Leg 1	June 29 -July 14	Numbers	92	2382	11	0	107	0	47391	15
		Weight (kg)	95.15	2457.565	13.65	0	165.95	0	8786.375	19.72
Leg 3	Aug.21-Aug.26	Numbers	365	3704	1	4	101	0	17049	14550
		Weight (kg)	314.16	3760.72	1.4	13.61	158.28	0	594.61	35.95
Leg 4	Sept. 3-Sept. 18	Numbers	281	4163	1	0	237	0	174366	3145
		Weight (kg)	265.33	4375.3	2.27	0	435.27	0	5979.33	9.04
Total		Numbers	738	10249	13	4	445	0	238806	17710
		Weight (kg)	1320.64	18460.585	19.32	17.61	1097.5	0	206775.315	17759.71