

Proximate and Lipid Analyses of Krill (*Euphausia* species) and Red Crab (*Pleuroncodes planipes*)

Richard W. Pierce, John Van der Veen, and H. S. Olcott

Proximate analyses were obtained of several samples of krill (*Euphausia* species) and one of "red crab" (*Pleuroncodes planipes*) as examples of abundant zooplankton which might be used as food or feed. The krill samples contained, on a dry weight basis, approximately 53% protein,

17% ash, and lipid varying from 18 to 26%. The decapod contained 32% protein, 36% ash, 8% lipid, and 11% chitin. The lipids were hydrolyzed, methylated, and subjected to gas chromatographic analysis. Highly unsaturated fatty acids accounted for 36 to 48% of the total.

A number of authorities have suggested that man will turn to the ocean for the protein which will be required by the foreseeable increase in population. The possibility of utilizing stages lower than fish in the food chain is appealing because total yields manyfold greater than those now available can be envisioned (Burkholder *et al.*, 1966, 1967; Reay, 1954; Weiss, 1953). However, because most of the primary photosynthetic organisms are small, scattered, and difficult to harvest, projects involving phytoplankton will be costly (Emery and Iselin, 1967; Geiger, 1958; Walford, 1955). On the other hand, some of the larger zooplankton occasionally aggregate in large numbers and possibly then could be harvested economically. One example is the small shrimplike food of the whale called krill. Russian reports indicate that 5 to 10 metric tons of *Euphausia superba* can be caught with industrial pelagic trawls in 30- to 60-minute sweeps of the kilometer-wide blooms which occur in the Antarctic during its summer (Solianek, 1960). In December 1966, a Russian research vessel equipped with special gear studied the feasibility of processing krill into products, the flavor of some of which was said to resemble that of crab (Burukovskiy, 1967).

With the expectation that interest in this resource will continue, the authors report here the proximate analyses and lipid composition of three samples of zooplankton: *Euphausia pacifica*, the Pacific Euphausiid with distribution from latitude 30°N to 50°N (Brinton, 1962); *Euphausia superba*, confined to the Antarctic (Marr, 1962); and the galatheid decapod, *Pleuroncodes planipes*, a pelagic species called red crab, which periodically appears in large numbers off the Pacific coast of North America (Longhurst, 1966). Longhurst (1967) has estimated that 10,000 metric tons of red crab might be available for exploitation annually in Pacific waters adjacent to Mexico. A potential harvest of 50,000,000 tons of krill annually was postulated by Alverson (1967).

EXPERIMENTAL

One sample of *E. pacifica* was collected with a commercial shrimp trawl off the coast of Eureka, Calif.,

September 9, 1966. A second, supplied by the Bureau of Commercial Fisheries, was caught with an Isaacs-Kidd trawl off the coast of Washington, August 3, 1967. The sample of *E. superba* was taken with a triple-O net during the first week in March 1966, near the South Shetland Islands, by the Argentine research vessel *A.R.A. Zapiola*. *P. planipes*, supplied by the Scripps Institution of Oceanography, was collected with a hand dip net off the coast of Baja, Calif., April 28, 1967. All samples were maintained at -18° C. and ground prior to chemical analysis.

Ash was determined in a muffle furnace at 525° C. for 24 hours. Lipids were determined in duplicate by solvent extraction according to the method of Bligh and Dyer (1959), but performed under N₂. The lipid extracts were deep red in color, but the color faded when the extracts were exposed to air or light. They were therefore stored in the dark at -18° and under nitrogen until needed. Fatty acid content was determined by gas-liquid chromatography (GLC) after saponification and methylation (Medwadowski *et al.*, 1967). Thin-layer chromatography (TLC) was used to estimate the lipid classes present (Medwadowski *et al.*, 1967).

Total nitrogen was assayed by Kjeldahl (Association of Official Agricultural Chemists, 1965). Protein nitrogen was determined by biuret (Layne, 1957) after treatment according to the procedure of Torten and Whitaker (1964), with serum albumin as the standard.

Chitin was determined according to the method of Brown (1959) which involves: extraction with 70% acetone, digestion with 90% formic acid to decalcify the material and extract colored impurities and some of the protein, digestion with 5% sodium hydroxide to extract the remainder of the protein, and determination of nitrogen by Kjeldahl on the insoluble residue. Chitin contains 6.89% N. Attempts to separate diffusible nitrogen by dialysis of suspensions of the ground products in cellophane bags were not successful because the bags disintegrated overnight at 4°, presumably evidence of cellulase activity.

RESULTS AND DISCUSSION

The proximate analyses for *E. pacifica* and *E. superba* (Table I) are consistent with those of other authors (Brown, 1959; Suyama *et al.*, 1965; Vinogradova, 1960; Yamada, 1964), but at variance with the data of Burk-

Institute of Marine Resources, Department of Nutritional Sciences, University of California, Berkeley, Calif. 94720

Table I. Proximate Composition of *E. pacifica*, *E. superba*, and *P. planipes*^a

	<i>E. pacifica</i> , %		<i>E. superba</i> , %	<i>P. planipes</i> , %
	Eureka	Washington		
Ash	17.1	20.5	18.9	35.9
Extractable lipids	23.8	18.9	18.7	7.6
Protein (total N × 6.25)	62.4	68.4	61.8	48.7
Protein (biuret)	58.6	53.8	44.8	32.1
Chitin	2.2	2.8	2.3	10.9

^a Wet-ground material contained approximately 90% moisture. Analytical data corrected to dry matter basis.

holder *et al.* (1967) for ash and lipid. Their reported values for ash (1.30%) and lipid (1.77%) were in error and should have been 13.1 and 17.8%, respectively (Burkholder *et al.*, 1968). One sample of *E. pacifica* was washed with distilled water. The ash content was found to be less than half that found with the unwashed sample, probably reflecting a loss of residual salt.

Analyses of *P. planipes* have not been previously reported. According to Lafon (1943) the average chitin content in the integument or shell of several decapods was 13.9%; the remainder is presumed to be inorganic. Since the ground sample of the red crab contained 10.9% chitin (Table I), approximately 70 to 75% of the dry weight of the animal was integument. This agrees with data obtained by physical separations of shell and meat (data not shown).

According to Camien *et al.* (1951) and Suyama *et al.* (1965) the soluble nonprotein nitrogen components of crustacea are composed of amino acids, particularly alanine, glycine, and proline, in addition to taurine, betaine, trimethylamine oxide, and others. In the data of Suyama *et al.*, these amount to 17% of the total nitrogen. The differences in the values for total nitrogen and protein nitrogen for *E. pacifica* and *E. superba* shown in Table I suggest that these products similarly have about 17% of the nitrogen in nonprotein form.

The difference in lipid content in the two samples of *E. pacifica* is consistent with the known seasonal fluctuations which take place in related species of Euphausiids (Giese, 1966; Il'ichev, 1966; Littlepage, 1962). Assuming that the shell of the red crab contains little or no lipid, the residual tissue can be calculated to contain about 30% lipid.

Fatty acid analyses of the total lipid extract are shown in Table II. A considerable proportion of the fatty acids were highly unsaturated; C 20:5 and C 22:6 comprise 39 to 45% of the total. Saturated fatty acids account for about 30%. Red crab was appreciably richer in C 22:6 than in C 20:5. Yellowfin tuna are known to feed on red crab (Longhurst *et al.*, 1967) and their fatty acid composition is similar (Shuster *et al.*, 1964). The krill samples contained more C 20:5 than C 22:6. The reason for the low value of 22:6 (6.4%) in krill caught in Eureka is not known but location of catch, or sea-

Table II. Fatty Acid Methyl Ester Distribution in Various Zooplankton^a

Fatty Acid Chain Length: Double Bond	<i>P. planipes</i> , %	<i>E. pacifica</i> , %		<i>E. superba</i> , %
		Eureka	Washington	
14:0	3.4	4.7	3.8	4.1
15:0	0.8	Tr	0.5	Tr
16:0	18.2	28.5	22.8	24.4
16:1	2.8	6.4	8.1	9.0
17:0	0.6	Tr	Tr	Tr
18:0	3.9	2.5	1.4	1.7
18:1	16.4	15.1	13.9	15.2
18:2	2.3	2.9	1.9	1.7
18:3	1.6	0.6	1.2	1.3
20:0	Tr	0.5	1.0	1.0
20:5	17.4	30.9	27.9	25.3
22:1	3.2	1.3	1.2	1.2
22:6	28.4	6.4	16.1	14.6

^a Average of at least 3 analyses; peaks with less than 0.5% of total area not shown. Tr = trace, less than 0.5%.

sonal changes of krill diet (Littlepage, 1962) may be involved.

Qualitative results by thin-layer chromatography showed that free fatty acids, triglycerides, and cholesterol were the main lipids. The high amount of free fatty acids possibly reflected lipid degradation prior to analysis. Phosphatidylcholine and phosphatidylethanolamine were the major phospholipids present. Other normal phospholipids were present in red crab but did not appear in thin-layer plates of euphausiid extracts. Quantitative studies of the lipid components by column chromatographic techniques are in progress.

Needed for more complete evaluation of the potential of these products for food or feed are actual feeding tests with experimental animals; and detailed analyses of the amino acid composition such as those already reported by Burkholder *et al.* (1966, 1967) for various zooplankton samples.

ACKNOWLEDGMENT

We express our appreciation for samples and for help in collecting the material to the following: Luis G. Fernandez, Serviceo de Hidrografia Navel, Buenos Aires, Argentina, for arranging for the Antarctic collections and to Paul Burkholder for forwarding the material to us; to the Bureau of Commercial Fisheries, Exploratory Fishing and Gear Research Base, Seattle, Wash., for collecting and furnishing samples of *E. pacifica*; to Dan Gotschall and the crew of the *N. B. Schofield*, California Department of Fish and Game, for their assistance in collecting samples of *E. pacifica* off the coast of California, and to Carl Hubbs, Scripps Institution of Oceanography, for collecting the sample of *P. planipes*. Andrew Chang and Barbara Medwadowski contributed analytical data.

LITERATURE CITED

Alverson, D. L., *Comm. Fisheries Rev.* **29**, (6), 24 (1967). Association of Official Agricultural Chemists, "Official Methods of Analysis," 10th ed., 1965.

- Bligh, E. G., Dyer, W. J., *Can. J. Biochem. Physiol.* **37**, 911 (1959).
- Brinton, E., *Bull. Scripps Inst. Oceanog., Univ. Calif.* **8**, 51 (1962).
- Brown, R. L., *Comm. Fisheries Rev.* **21**, 6 (1959).
- Burkholder, P. R., Burkholder, L. M., Centeno, P., *Nature* **211**, 860 (1966).
- Burkholder, P. R., Mandelli, E. F., Centeno, P., *J. AGR. FOOD CHEM.* **15**, 718 (1967).
- Burkholder, P. R., Mandelli, E. F., Centeno, P., *J. AGR. FOOD CHEM.* **16**, 881 (1968).
- Burukovskiy, R. N., ed., "Antarctic Krill," Atlantic Scientific-Research Institute for Fish and Oceanography, p. 9, Kaliningrad, 1967.
- Camien, M. N., Sarlet, H., Duchateau, G., Florkin, M., *J. Biol. Chem.* **193**, 881 (1951).
- Emery, K. O., Iselin, C. O. D., *Science* **157**, 1279 (1967).
- Geiger, E., *Am. J. Clin. Nutr.* **6**, 394 (1958).
- Giese, A. C., *Physiol. Rev.* **46**, 244 (1966).
- Il'ichev, E. F., *Rejbnoe Khozyaistro* **1**, 61 (1966).
- Lafon, M., *Ann. Sci. Nat. Zool. Biol. Animale*, 11° ser. **5**, 134 (1943).
- Layne, E., *Methods Enzymol.* **3**, 450 (1957).
- Littlepage, J. L., "Biologie Antarctique Comptes-rendes," p. 463, Hermann, Paris, 1962.
- Longhurst, A. R., California Cooperative Ocean. Fisheries Investigation, Progr. Rept., 1 July 1963-30 June. Vol. **11**, 142, 1966.
- Longhurst, A. R., FAO World Scientific Conferences on Biology and Culture of Shrimps and Prawns, p. 15, June 1967.
- Longhurst, A. R., Lorengen, C. J., Thomas, W. H., *Ecology* **48**, 190 (1967).
- Marr, J. W. S., *Discovery Reports* **32**, 33 (1962).
- Medwadowski, B. F., Van der Veen, J., Olcott, H. S., *J. Food Sci.* **32**, 361 (1967).
- Reay, G. A., *Food Technol.* **8**, 65 (1954).
- Shuster, C. Y., Froines, J. R., Olcott, H. S., *J. Am. Oil Chemists' Soc.* **41**, 36 (1964).
- Solianek, G. A., *Sovet. Antarktiches Kaia Expeditiia* **14**, 29 (1960).
- Suyama, M., Nakajimi, K., Nonaka, J., *Bull. Japan. Soc. Sci. Fisheries* **31**, 302 (1965).
- Torten, J., Whitaker, J. R., *J. Food Sci.* **29**, 168 (1964).
- Vinogradova, Z. A., *Doklady Akad. Nauk SSSR* **133**, 680 (1960).
- Walford, E. A., "Living Resources of the Sea." Chap. 8, Ronald Press, New York, 1955.
- Weiss, F. J., *J. AGR. FOOD CHEM.* **1**, 822 (1953).
- Yamada, M., *Bull. Japan Soc. Sci. Fisheries* **30**, 673 (1964).

Received for review September 3, 1968. Accepted December 9, 1968. Supported in part by the Bureau of Commercial Fisheries and by Graduate Educational Grant #14-17-0077-399.