

a recent review article,³³ health benefits attributed to astaxanthin included reduced risk of cataracts, diabetes, heart disease, neural deterioration, and certain cancers; however, most of these studies were conducted using in vitro or animal models. In a human trial, young adult male volunteers provided astaxanthin in a daily beverage for 14 d showed protection against premature LDL oxidation in the absence of any other dietary changes.³⁴ Despite the promising results for astaxanthin, research is needed regarding the health benefits of astaxanthin derived from krill.

One of the most important antioxidants is vitamin E, which functions specifically to protect against lipid peroxidation in biological membranes. Due to methodological difficulties, vitamin E has not been determined extensively in foods. According to Table 2, krill meets the RDA for vitamin E and contains higher amounts of vitamin E than either shrimp or fish. Thus, the stability of krill may be attributed to its high levels of vitamin E. However, other components may also protect krill from oxidation. Dunlap et al.³⁵ identified a marine-derived tocopherol with enhanced antioxidant effects on cellular lipids. Venkatraman et al.³⁶ reported that mice fed a diet containing 10% krill oil had higher liver expression of endogenous antioxidant enzymes (i.e., catalase, glutathione peroxidase, and superoxide dismutase) and lower peroxide and thiobarbituric acid values compared with mice fed a 10% corn oil diet. The high content of vitamin E and the presence of other antioxidant components suggest that krill may have beneficial effects against oxidative damage.

The water-soluble vitamins play an important role in maintaining health by acting as coenzymes in energy metabolism. Table 2 shows the levels of selected water-soluble vitamins in krill, shrimp, and fish, along with the RDA for these vitamins in adults. The levels of thiamin, riboflavin, and niacin are higher in fish compared with shellfish (i.e., krill and shrimp), but are below the RDA for adults, indicating that in general seafood is a poor source of these vitamins. Table 2 also shows the levels of the B-complex vitamins B₁₂, B₆, and folate in krill, shrimp, and fish. These vitamins play a role in the metabolism of homocysteine, so deficiencies can lead to high blood homocysteine. This has important implications because a high blood level of homocysteine is an independent predictor of heart disease and stroke.³⁷ The vitamin B₆ content of most seafood is below the RDA, and levels in krill are below that of either shrimp or fish. Although krill is higher in folate than shrimp and fish, levels are still below the RDA. On the other hand, vitamin B₁₂ in krill (16 µg/100 g) is substantially higher than in shrimp (1.16 µg/100g) or fish (4.17 to 4.45 µg/100g) and exceeds the RDA of 2.4 µg/d for adults.

The assessment of the nutritive value of krill based on vitamin content indicates that it has considerable appeal for human consumption because it provides a good source of vitamin B₁₂, vitamin E, and astaxanthin, as well as other potential antioxidant compounds.

KRILL PROTEIN QUALITY

Whole Krill

Krill is a high-protein food, having a protein content estimated in the range of 60% to 65% dry weight.⁸ Similar to other animal foods, the protein derived from krill is a complete protein, as indicated by the presence of all nine of the indispensable amino acids required by adults. As shown in Table 3,³⁸ the level of all of the indispensable amino acids in whole krill met the FAO/WHO/UNU amino acid requirement for adults³⁹ (with the exception of histidine). In contrast, several indispensable amino acids in fish were below the amino acid requirement for adults, most notably methionine and cysteine. However, it should be noted that the indispensable amino acid levels in whole krill were below the reference protein, egg. Tamura⁴⁰ reported an amino acid score in the range of 0.85 to 1.00, indicating that the amount of indispensable amino acid provided by krill protein met the protein requirement of adults. Although considered a high-quality protein, krill is of lower quality than whole egg, which has an amino acid score of 1.21.

In practice, protein quality is determined not only by the amino acid composition but also by its digestibility. Iwantani et al.⁴¹ fed rats a diet containing either egg protein or defatted, freeze-dried whole krill for 4 weeks, and then determined the weight gain of the animals to calculate the protein efficiency ratio. Biological value and net protein utilization were also measured to assess protein availability and digestibility. Rats fed whole krill gained less weight, had a reduced protein efficiency ratio, biological value, and net protein utilization compared with animals fed egg protein. The decreased digestibility of krill protein may have been due to the presence of the exoskeleton. Obatake²² determined the nutritive value of the protein in krill meat after removal of the exoskeleton, and found that it was still inferior to that of egg protein. Findings in the animal studies were not always indicative of the findings in humans. In a human trial, Tamura⁴² fed boiled krill or whole egg to adult men for 21 days and reported a net protein utilization of 55% for krill and 61% for whole egg, with no differences in their digestibility. Based on the evidence, krill appears to be a good source of high-quality protein. The large biomass and high-quality protein offered by krill provides an economical replacement for commer-

Table 3. Amino Acid Composition of Krill and Lean (Trout) and Fatty (Salmon) Fish* Compared With Egg, Along With the Amino Acid Requirement for Adults†					
Amino Acid	Krill	Trout	Salmon	Egg	Requirement
	<i>mg/g</i>				<i>mg/g protein</i>
Isoleucine	25	9.4	10.0	63	13
Leucine	40	16.6	17.6	88	19
Lysine	44	18.8	19.9	70	16
Methionine + cysteine	24	8.3	8.7	56	17
Phenylalanine + tyrosine	50	14.9	15.7	98	19
Threonine	22	9	9.5	49	9
Tryptophan	7	2.3	2.4	16	5
Valine	26	10.5	11.1	72	13
Histidine	11	6	6.4	24	16
Arginine	38	12.3	12.9	28.4	ND
Alanine	29	12.4	13.1	26.4	ND
Aspartic acid	53	21	22.1	47.6	ND
Glutamine	67	30.6	32.3	61.9	ND
Glycine	34	9.8	10.4	15.9	ND
Proline	23	7.2	7.6	18.9	ND
Serine	19	8.4	8.8	35.2	ND
* Whole krill (<i>Euphausia superba</i>), wild rainbow trout (<i>Salmo gairdneri</i> Richardson), and wild Coho salmon (<i>Oncorhynchus kisutch</i>). Values for krill are based on Torres et al. ³ ; values for trout, salmon, and egg are based on the USDA ¹⁰ ; values for egg are also based on Young and Pellett ³⁸ ; requirement for adults based on FAO/WHO/UNU. ³⁹ ND, Not determined.					

cially available protein sources. In addition, commercial protein sources available for human consumption such as casein, whey, soy protein, lactalbumin, and wheat gluten have various limitations. For example, casein is low in sulfur amino acids, particularly cysteine, which must be added to the diet. Products in which krill protein may substitute for other proteins are nutritional supplements, sports drinks, infant formulas, and milk replacers. For these products, protein concentrates rather than whole krill or krill meat are used.

Krill Protein Concentrate

Protein concentrates are produced by technologies that concentrate the proteins in food so that levels are higher than those in the original food, making them an inexpensive source of available protein. Preparation of protein concentrate from fish is accomplished by extracting the lipids, removing the bones, and drying, so that the resultant product is 85% to 95% higher in protein than the original fish. Lack of proper technology for protein recovery from krill has hindered progress in the commercial development of krill protein concentrates. Some research has been carried out that made use of krill's high content of proteolytic enzymes to produce a product with protein recovery as high as 80%.⁴³ There is technology available for isolating muscle protein in a continuous

mode from whole krill to produce krill protein concentrate. The continuous protein recovery technology is based on a principle of isoelectric solubilization followed by isoelectric precipitation of the protein. This basic biochemical principle has long been used in the dairy industry to manufacture cottage cheese, and also in other food processing industries such as those for soy protein isolates and concentrates. More recently, the principle of isoelectric point has been applied to fish muscle⁴⁴ and other animal muscle.⁴⁵ The protein recovery (dry basis) yield from krill using isoelectric solubilization/precipitation is approximately 90% or greater in a continuous mode. A more in-depth discussion of the isoelectrical solubilization recovery of krill is available elsewhere.³

Table 4⁴⁶ shows the amino acid composition of krill protein concentrate compared with that of other commercially available protein concentrates such as casein, whey, and soy protein. The indispensable amino acid profiles of the protein concentrates were compared with the indispensable amino acid requirements for adults and infants because protein concentrates are frequently used in infant formulas. As shown in Table 4, krill protein concentrate contains all indispensable amino acids in amounts that exceed the requirements for healthy adults. Krill protein concentrate also contains all of the indispensable amino acids in amounts that meet the requirements for infants (although leucine, tryptophan, and histidine levels were slightly below). Milk-

Table 4. Amino Acid Composition of Krill (*Euphausia superba*) Protein Concentrate, Whey Protein Concentrate, Sodium Caseinate, and Soy Protein Concentrate Compared with the Amino Acid Requirements for Infants and Adults*

Amino Acid	Krill	Whey	Sodium Caseinate	Soy	Infant Requirement	Adult Requirement
	<i>mg/g</i>				<i>mg/g protein</i>	
Indispensible Amino Acids						
Isoleucine	56	49.7	45.9	29.4	46	13
Leucine	88	106.6	88.9	49.2	93	19
Lysine	84	88.1	77.5	39.3	66	16
Methionine + cysteine	44	79.7	32	17	42	17
Phenylalanine + tyrosine	96	58.2	101.4	55.8	72	19
Threonine	46	68.7	40.5	23	43	9
Tryptophan	16	17.3	10.4	8.3	17	5
Valine	56	18.4	56.4	30.5	55	13
Histidine	25	7.8	25	15.8	26	16
Dispensible Amino Acids						
Arginine	54	27.1	33.5	46.4	ND	ND
Alanine	62	55.5	27.5	26.8	ND	ND
Aspartic acid	53	91.8	75.7	72.5	ND	ND
Glutamine	67	158.4	218.4	120.1	ND	ND
Glycine	34	53.2	17.3	26.9	ND	ND
Proline	23	66.6	93.3	33	ND	ND
Serine	19	53	55.4	33.7	ND	ND

* Values for krill are based on Torres et al.³; values for whey and soy protein concentrate are based on USDA¹⁰; values for sodium caseinate are based on Sindayikengera and Xia⁴⁶; requirements for infants and adults are based on FAO/WHO/UNU³⁹.
ND, Not determined.

derived whey protein concentrate, sodium caseinate, and soy protein concentrates are more deficient in the indispensable amino acids for infants than are krill protein concentrates.

The amount of indispensable amino acids in krill protein concentrate is higher than that in soy protein concentrate and similar to that of the milk protein casein. Sidhu et al.²⁹ reported that rats fed krill protein concentrate had similar weight gain and protein efficiency ratio as rats fed casein. Although higher than casein, krill protein concentrate is low in threonine, leucine, lysine, and the sulfur amino acids (methionine and cysteine) compared with the whey protein concentrate (Table 4). This has important health implications because sulfur amino acids are involved in DNA transcription and RNA translation and may play a role in reducing the risk of CVD, dementia, cirrhosis, and immunomodulation.⁴⁷ The health benefits and safety issues regarding the use of alternative protein sources need to be investigated in more detail.

The Health Benefits and Safety of Krill Protein

Adequate protein intakes are necessary for synthesis of structural components of the muscle and of enzymes,

hormones, hemoglobin, and other body tissues. Sidhu et al.²⁹ reported that rats fed krill protein concentrate showed no difference in organ weights and hemoglobin counts than rats fed casein, indicating that krill protein is capable of supporting protein synthesis. Too much rather than too little protein is typical of the Western diet. Consuming protein beyond recommended amounts is common among athletes interested in increasing their muscle mass. The general population has also recently developed an interest in increasing their protein intakes due to suggestions that high-protein diets support weight loss, and there is some evidence that short-term consumption of high-protein diets increases satiety and thermogenesis and reduces energy intake.⁴⁸ However, despite growing interest in proteins, the long-term effects of high-protein diets on weight loss are unknown and there may be harmful effects associated with such diets. For example, diets high in protein have been suggested to increase the risk for cardiac, renal, bone, and liver disease.⁴⁹

High-protein diets have also been suggested to increase the risk of CVD by inducing negative effects on blood lipid profiles. However, the negative effects on blood lipids are more likely due to the high SFA intakes associated with most high-protein diets. Krill, unlike

other animal sources of proteins, is low in SFAs. Obatake²² reported no difference in serum cholesterol levels for rats fed boiled whole krill or krill meat compared with animals fed casein.

Another area of concern is the use of alternate protein sources in infant formulas. Soy protein has been reported to accelerate puberty in female rats compared with whey and casein.⁵⁰ Other concerns regarding alternative protein sources are their ability to support growth and their long-term health effects.⁵⁰ In addition, all food proteins have the potential to be allergenic to some people. Approximately 1% to 2% of adults and up to 5% to 7% of children experience food allergies with symptoms ranging from a mild rash to life-threatening anaphylaxis.⁵¹ The Food and Agricultural Organization of the United Nations includes crustaceans on its list of the eight most significant food allergens.⁵² According to Wild and Lehrer,⁵³ shellfish is the number-one cause of food allergies in adult Americans. To our knowledge, no studies have examined whether krill protein or krill protein concentrate causes food allergies. Determining the protein allergens present in krill and the relative allergenic activity of krill compared with other major protein substitutes is important. For example, soy protein used to manage cow's milk allergies in infants has been reported to have less allergenic reactivity compared with milk proteins; however, concerns have been raised regarding the isoflavone content in soy infant formulas affecting neurobehavioral development, immune, endocrine, and thyroid function.⁵⁴ Krill protein concentrate, which has an indispensable amino acid profile superior to that of soy protein concentrate, may be a better protein substitute in infant formulas if its allergenic reactivity is found to be lower than that of other protein sources.

Finally, the high protein levels achievable with protein concentrates and isolates have been suggested to increase the risk of kidney damage, particularly in the immature kidneys of infants. Rats fed semi-purified diets containing soy protein or casein have been reported to have an increased incidence of nephrocalcinosis.⁵⁵ Nephrocalcinosis may be caused by a number of conditions, including excretion of excess calcium by the kidneys. Increased calcium excretion (calciuria) by the kidneys due to high-protein diets has been attributed to the sulfur amino acids increasing the acid load in the body, followed by the release of calcium from the tissues to restore acid-base balance.⁵⁶ Calcium released from the bone to restore acid-base balance has led to suggestions that high-protein diets may contribute to bone loss. In addition, rats fed semi-purified diets containing soy protein have been reported to have reduced mineral absorption.⁵⁵ The calciuria and reduced mineral absorption associated with high-protein diets may contribute to

negative mineral balance, leading to bone loss and increasing the future risk of osteoporosis. Furthermore, minerals may be lost during the processing of whole foods into protein concentrates. The next section assesses the mineral content of whole krill and krill following processing into edible products.

KRILL MINERAL CONTENT

The mineral content of shellfish is to a large extent located in the exoskeleton, which is removed to produce krill meat and krill protein concentrate. Figure 4A compares the level of selected minerals in whole krill, krill meat, and krill protein concentrate, along with the RDA of these minerals for adults. In whole krill, the content of major minerals involved in bone health (calcium, phosphorus, and magnesium) met the RDA. Following removal of the exoskeleton to produce krill meat, calcium was reduced by 84.4%, phosphorus by 47.3%, and there were minute effects on magnesium. As a result, krill meat was below the RDA for calcium and phosphorus (Figure 4A). Processing of whole krill into krill protein concentrate reduced calcium by 78.6%, phosphorus by 14.4%, and magnesium by 83.5%. The phosphorus content in krill protein concentrate met the RDA, but calcium and magnesium were both below the RDA (Figure 4A).

Another important dietary mineral is iron. Identifying good food sources of iron is important because iron deficiency is the most common nutritional disorder worldwide.⁵⁷ Anemia due to iron deficiency has been associated with significantly lower scores in cognitive and educational achievement tests in school-age children⁵⁷ and lower work productivity in adults.⁵⁸ Removal of the exoskeleton to obtain krill meat and processing to krill protein concentrate have minute effects on the level of iron in krill (Figure 4B). Still, the iron content of krill does not meet the RDA for adult women (18 mg/d) or the lower RDA for adult men (8 mg/d). King et al.⁵⁹ determined the mineral content of shellfish commonly marketed in the northwestern United States, and found that shellfish in general is low in iron, with the exceptions of oysters, mussels, and clams, which have levels comparable to red meat.

Most foods are low in fluoride, but krill is an exception. Fluoride is important for the mineralization of bone and teeth and in the prevention of dental caries. Furthermore, the use of high-dose fluoride is being investigated for the prevention of osteoporosis.⁶⁰ The fluoride content of krill is concentrated in the exoskeleton, where it may reach concentrations of 350 mg/100g dry weight.⁶¹ At death, the fluoride in krill is capable of migrating from the exoskeleton into the soft tissues.

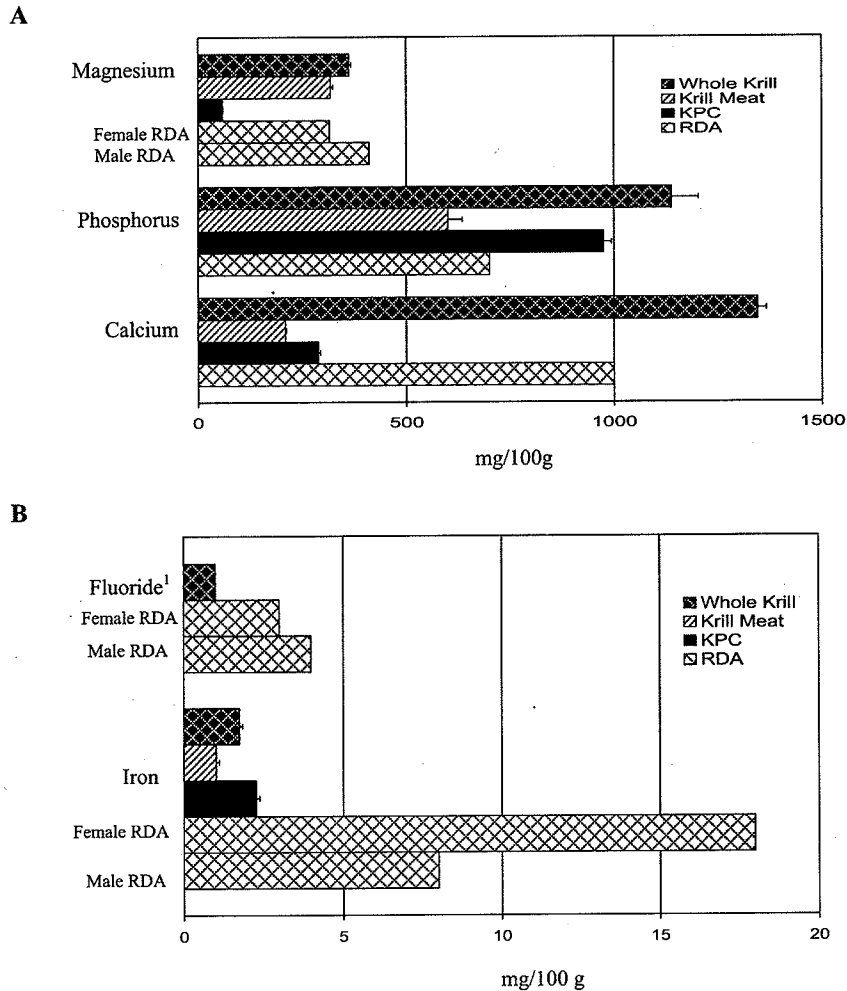


Figure 4. Mineral content of whole krill and krill following processing into meat and protein concentrate along with the recommended dietary allowance of these minerals for adults. (Fluoride value in krill meat is based on Virtue et al.⁶¹)

Budzinski et al.⁶² reported fluoride levels as high as 9 mg/100 g wet weight in krill meat. The fluoride derived from krill is also highly bioavailable. Tenuta-Filho and Alvarenga⁶³ reported that weanling male rats fed krill exoskeleton had 80% absorption of fluoride. The high fluoride content of krill, together with the high bioavailability of this fluoride, has led to concerns about the potential for toxicity. Careful removal of the exoskeleton before consuming krill and immediate removal of the shell upon harvest to prevent migration of fluoride into the muscle can minimize potential toxicity since over 99% of the fluoride content of krill is associated with the exoskeleton.⁶⁴

Overall, the evaluation of the mineral content of krill indicated that it is a poor source of iron. Whole krill meets the RDA for minerals important for bone health; however, depending on the technology used to process krill into edible products, mineral losses may occur that

result in mineral levels below the RDA. On the other hand, processing typically removes potentially toxic levels of fluoride. In the processing of shellfish, by-products represent approximately 50% to 70% of the weight of the raw material, much of which is the exoskeleton.⁶⁵ Whether the by-products generated by processing of krill into food may be developed into value-added products is another important consideration. In the next section, potential value-added products generated from krill are assessed.

VALUE-ADDED PRODUCTS

Chitin

Chitin is a polysaccharide comprised of units of N-acetyl-2 amino-2-deoxy-D-glucopyraose ($C_8H_{15}O_6N$)

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