# METHOD OF DETERMINING CAROTENOID CONTENTS OF ALASKA PINK SHRIMP AND REPRESENTATIVE VALUES FOR SEVERAL SHRIMP PRODUCTS

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#### ABSTRACT

An extraction method is described for estimating the amount of carotenoids in pink shrimp. The carotenoid index is useful as a measure of quality and as an indicator of changes during storage. Values for several shrimp products are reported.

The carotenoid content of Alaska pink shrimp is affected by many conditions and can be used as an index of the general quality of canned shrimp and of the changes in quality of frozen shrimp during storage. It has been also used as a factor in determining optimum peeling characteristics of shrimp (Collins and Kelley, 1969) and in selecting desirable retorting conditions (Kelley, 1971<sup>a</sup>). Color differences in shrimp at different seasons and in different areas may be important in harvesting and marketing practices.

The carotenoid in Alaska pink shrimp is primarily astaxanthin. Both total astaxanthin and astacin, its oxidation product, are measured by the method to be described, which was developed for use with frozen Alaska king crab (Ravesi, 1965') and adapted to Alaska pink shrimp which contain more interfering protein and moisture than crab.

zen storage on the carotenoid pigments of Alaska king crab. Unpublished manuscript filed at NMFS, Kodiak, Alaska.

# METHOD OF DETERMINING CAROTENOID CONTENTS

To 50 g of blended meat add approximately 10 g of silica gel and 100 ml of the proper acetone solution:

- 1. 75% acetone for canned shrimp with liquor.
- 2. 65% acetone for frozen cooked or raw meats

3. 50% acetone for raw shrimp with shells on. The silica gel, which serves as a filter aid, is not essential but makes subsequent extraction and filtration easier. Blend just enough to ensure complete mixing and filter through a medium porosity fritted glass funnel, maintaining suction until dripping ceases. Rinse container and filter as needed with 50% acetone. Discard colorless filtrate and blend residue about 2 min with 15 to 20 g anhydrous sodium sulfate and 100 ml of 1:1 2-propanol:chloroform. Filter and re-extract with 50 ml solvent one or two times as needed to get a colorless meat. Use 2propanol: chloroform as rinse solution during these extractions. Transfer filtrate to 500 ml round bottom flask and strip the solvent, using a rotating vacuum evaporator. Add 5 to 10 ml chloroform and evaporate to dryness. Dissolve residue in enough pure cyclohexane to wash sides of flask and add 10 to 15 g anhydrous sodium sulfate. Let set for a few minutes and filter through sodium sulfate on a fine porosity fritted

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glass funnel, washing sodium sulfate with cyclohexane to remove all traces of color. If filtrate is clear, dilute to 100 ml. If it appears hazy, repeat the filtration, allowing solution to remain in fresh sodium sulfate for a brief time. Read at 474 nm on spectrophotometer, using cyclohexane as a blank.

The precision of the method was determined by analyzing 11 identical samples in quadruplicate on 11 different days. Twenty-two cans of the same code of canned shrimp were blended in a Waring blendor, and the homogeneous mixture was sealed in cans and frozen at  $-60^{\circ}$  F. For each day's sampling, two cans of the frozen mixture were thawed and blended together.

Different lots of solvents were used at intervals to determine the sensitivity to slight variations in solvents. The solvent lot was not critical but the cyclohexane used in the spectrophotometer should be carefully distilled within a few days of use. We used a Gilford modification of a Beckman DU spectrophotometer<sup>5</sup> which gives readings with three place accuracy. The range of absorbance readings was 0.420 to 0.452, the average was 0.436, and the standard deviation was 0.008.

The carotenoid content, expressed as the carotenoid index, is a calculated value based on dry weight. The solids content of the shrimp was determined by the Association of Official Agricultural Chemists method (Horwitz, 1965: 346), using 5 to 10 g of the blended meat sample and heating at  $105^{\circ}$  C for 18 to 24 hr. The carotenoid index represents the absorbance (A) in 100 ml of solvent of the carotenoids from 1 g of dry sample, measured in a 1-cm cell. It is calculated as follows:

 $C_I = \frac{(A \text{ at } 474 \text{ nm in } 100 \text{ ml cyclohexane}) (100)}{(50\text{-g wet sample}) (\% \text{ dry weight})}$ 

The absorbance reading of a shrimp sample with average moisture content is roughly 10 times larger than the carotenoid index; therefore the carotenoid index equivalent of the standard deviation is slightly less than 0.001. The amount of carotenoid can also be expressed as grams of pigment/gram tissue by using the extinction coefficient of 2150, as reported by Kanemitsu and Aoe (1958). The amount of astacin present in fresh shrimp is small and since the extinction coefficients of astacin and astaxanthin for calculation purposes do not introduce significant error for routine analytical work, the calculation would be:

grams pigment/1 g tissue =  $\frac{(A \text{ at } 474 \text{ nm})(100 \text{ ml})}{100 (50 \text{ g}) (d) (2150)}$ 

where d is the cell width in centimeters. This could be converted to dry tissue weight by multiplying by the percent of solids in the sample.

## CAROTENOID CONTENTS OF VARIOUS TYPES OF SHRIMP SAMPLES

Table 1 gives carotenoid values of various types of shrimp samples described. Most of the data were collected as part of some other project so these samples are from several lots of shrimp caught at different times of the year. Only those grouped together in the table can be accurately compared with each other. All data, however, represent an average figure for the given sample and may be used to compare types of sample products or processing methods.

TABLE 1.-Carotenoid values for 11 shrimp samples.

	Sampling conditions	Carotenoid index	Cause of color differences indicated by data
1.	Raw tails, shells on	0.237	
	Raw meats ·	0.086	
2.	Whole cooked, hand-peeled meats, frozen	0.112	
3.	Precooked, machine-peeled meats, frozen	0.086	
	After 6 months' storage	0.070	Storage time
	After 12 months' storage	0.062	-
4.	Precooked, machine-peeled canned	0.073	Precook versus ice held conditioning
	2-day iced, machine-peeled, canned	0.059	
5.	1-day not iced, precooked, machine-peeled, canned	0.080	
	2-day iced, precooked, machine-peeled, canned	0.066	Time of ice holding
	3-day iced, precooked, machine-peeled, canned	0.064	

<sup>&</sup>lt;sup>5</sup> The use of trade names is merely to facilitate description and does not imply endorsement of a product.

#### EXPERIMENTAL PROCEDURE

1. The shrimp were frozen whole as soon as possible after being caught, then were shipped to the laboratory. They were partially thawed, weighed, and separated according to weights. The shrimp used were about 80 count. All were headed and some were hand peeled to obtain meats. The tails with shells on and the peeled meats were refrozen as needed until analyses could be made.

2. Whole cooked, hand-peeled frozen shrimp meats were obtained from a commercial processor. This is the conventional, cocktail style product.

3. Precooked, machine-peeled shrimp were produced under experimental conditions in a commercial plant. Shrimp were landed within 24 hr of catching, held overnight without ice, and precooked at  $165^{\circ}$  F for 10 sec,  $110^{\circ}$  F for 2 min, and machine peeled. The meats were collected at the end of the inspection belt and frozen in cans without vacuum. Analyses were made within a few days, after 6 months, and after 12 months of  $0^{\circ}$  F storage.

4. Precooked, machine-peeled canned shrimp were produced as described above except they were routinely retorted. The 2-day iced, machine-peeled shrimp are a standard commercial pack from the same lot of shrimp.

5. The 1-day not iced; 2-day iced; and 3-day iced, precooked machine-peeled, canned shrimp were also experimentally produced in a commercial plant. The 1-day not iced shrimp were held in the wooden boxes in which they were landed. The 2- and 3-day iced shrimp were held in large tanks and ice added as needed to keep them cool. All of these shrimp were precooked at 165° F for 10 sec, 110° F for 2 min, and then routinely peeled and canned.

All samples were analyzed using the previously described method of determining carotenoid contents. The averages reported in Table 1 represent 3 to 12 determinations under the given sampling conditions.

Some of the factors which cause differences in the carotenoid content of shrimp are shown in Table 1. These include several processing variations which can be controlled by processors and fishermen.

## CONCLUSIONS

The method of determining carotenoid content described is simple and precise and may be used on a variety of shrimp product forms.

The carotenoid index for Alaska pink shrimp varies from 0.267 in raw tails to 0.059 in ice held, machine-peeled canned shrimp. Correlation with subjective color rating is quite good (Collins and Kelley, 1969). At the higher color levels found in raw, hand-picked, or precooked shrimp, small differences are difficult to detect visually and the determination of the carotenoid index becomes even more useful in evaluating samples.

Since the carotenoid content is usually closely correlated with other quality characteristics, the carotenoid determination may be useful in making decisions about the best ways to process or handle shrimp.

## LITERATURE CITED

HORWITZ, W. (chairman and editor).

1965. Official methods of analysis of the Association of Official Agricultural Chemists. 10th ed. Association of Official Agricultural Chemists, Washington, D.C., xx + 957 p.

COLLINS, J., AND C. KELLEY.

1969. Alaska pink shrimp, *Pandalus borealis*: Effects of heat treatment on color and machine peelability. U.S. Fish Wildl. Serv., Fish. Ind. Res. 5: 181-189.

1958. Studies on the carotenoids of salmon. I. Identification of the muscle pigments. Bull. Jap. Soc. Sci. Fish. 24: 209-215.

KANEMITSU, T., AND H. AOE.