A New Method for Estimating the Freshness of Fish

Tsuneyuki SAITO Ken-ichi ARAI and Minoru MATSUYOSHI
(Faculty of Fisheries, Hokkaido University)

In spite of the extreme importance of phosphate esters as metabolic intermediates, published reports on studies of their changes in muscle of fish, especially such dealing with acid soluble nucleotides, are few in number[1),2)].

According to the facts related to the role of these esters in living muscle, it may be considered that the freshness of fish must be closely related to the changes in the acid soluble nucleotides. By determining the contents of ribose derived from the nucleotides, J. M. SHEWAN and N. R. JONES[3)] applied the principle to estimating the freshness of fish. On the other hand, in the previous papers[4),5),6)] it has been reported that when carp muscles are frozen at a slow rate there occur rapid splitting of ATP* and instantaneous accumulation of IMP. Furthermore IMP thus formed is slowly converted to inosine and then to hypoxanthine. At room temperature these changes are accelerated and as a result inosine and hypoxanthine, especially the latter, are accumulated predominantly. Using these results as a basis it has been considered that the freshness of raw or frozen fishes may be judged by estimating their contents of inosine and hypoxanthine.

Two grams of fish muscle was ground in 20 ml. of 4 per cent cold perchloric acid solution by hand in a mortar. The extract was filtered and 10 ml. of the filtrate was used for analysis. Hypoxanthine, inosine and nucleotides were separated by the hydrochloric acid gradient elution system on Amberlite IRA-400. The details of the system have been described previously[6)].

The rate of freshness (K) is determined by the following formula:

$$K=\frac{B}{A}\times 100$$

where A is total optical density at 250 mμ of perchloric acid extract and B is optical density at the same wavelength of inosine and hypoxanthine fraction (eluted with 0.01 N ammonium chloride solution). The relation between the freshness of frozen fishes and the values of K are presented in Table 1. It can be seen that these values, from 0–0.1 to 80–90, coincide with the freshness of the fish samples, the former corresponding to perfectly fresh muscle, the latter to the muscle of inferior quality. The relation between this new method and others, which have been universally applied, will be discussed elsewhere.

Received Aug. 22, 1958.

* The following abbreviations are used:
ATP, Adenosinetriphosphate;
IMP, Inosinemonophosphate;

Table 1. Objective index (K) and the freshness of various frozen fishes.

Species	Grade*	Periods of frozen storage (month, at −18∼−20°C.)	Optical density at 250 mμ		K	Time elapsed after catching
			A	B		
Yellow-tail	medium	3.5	5.200	2.202	42	100–120 hrs.
Swordfish	medium	15	7.000	3.279	46	100–120 hrs.
Sardine	high	6	6.250	0.636	10	40 hrs.
Mackerel pike	high	3	6.690	0.828	12	
Mackerel	high	3.5	6.245	1.226	20	
Mackerel	medium	7	6.500	3.019	46	
Mackerel	medium	7	6.600	1.490	23	
Mackerel	medium	7	6.260	1.664	27	
Codling	low	7	4.600	4.083	89	7 days
Rockfish	low	3.5	3.210	2.520	78	10 days
Mackerel (or Salmon) shark	medium	2	5.000	1.246	25	
Great blue shark	low	5	4.100	3.404	83	20–50 days
Long-jaw flounder	medium	4	4.600	1.834	40	
Halibut	high	3	4.480	1.300	29	
Carp	very high	0	4.300	0.000	0	
Squid	very high	0	7.322 (at 260 mμ)	0.008	0.1	freshly caught

* Grade was determined by sensory judgment of raw fish before freezing.

References

1) PARTMANN, W.: Arch. Fischereiwiss., **5**, 159 (1954).
2) JONES, N. R. and MURRAY, J.: Biochem. J., **66**, 5P (1957).
3) SHEWAN, J. M. and JONES, N. R.: J. Sci. Food Agr., **8**, 491 (1957).
4) SAITO, T. and ARAI, K.: Nature, **179**, 820 (1957).
5) ———: Bull. Japan. Soc. Sci. Fisheries, **23**, 265 (1957).
6) ———: Arch. Biochem. Biophys., **73**, 315 (1958).