

Nomograph for Estimating Histamine Formation in Skipjack Tuna at Elevated Temperatures

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Introduction

Fresh tuna contains almost no histamine (Geiger, 1944; Frank et al., 1981; Yoshinaga and Frank, 1982), but substantial histamine is formed in the muscle tissue during spoilage of skipjack tuna and other scombroid fish (Tomiyasu and Zenitani, 1957; Kimata, 1961; Ferencik, 1970; Omura et al., 1978), sometimes in excess of 0.5 percent of the fresh tissue weight (Hillig, 1956; Ienistea, 1973; Arnold and Brown, 1978; Lerke et al., 1978; Frank et al., 1981; Yoshinaga and Frank, 1982). Histamine is produced via microbial decarboxylation of free histidine which is abundant in the loins of scombroid fish (Shewan, 1955; Lukton and Olcott, 1958; Shifrine et al., 1959; Ferencik, 1970). A large number of organisms have been reported to be responsible for histamine formation in several kinds of scombroid fish (Shifrine et al., 1959; Mossel, 1968; Ienistea, 1973; Omura

et al., 1978; Arnold and Brown, 1978; Taylor et al., 1978; Arnold et al., 1980; Niven et al., 1981). Recently we found histamine-forming isolates of *Clostridium perfringens*, *Enterobacter aerogenes*, *Klebsiella pneumoniae*, *Proteus mirabilis*, and *Vibrio alginolyticus* in skipjack tuna that had decomposed under experimental conditions in seawater (Yoshinaga and Frank, 1982).

Because it is present in spoiled scombroid fish, histamine has been considered as a suitable index of microbial decomposition in tuna (Geiger, 1944; Hillig, 1954, 1956; Williams, 1954; Ferencik et al., 1961; Arnold and Brown, 1978; Lieber and Taylor, 1978). The purpose of this investigation was to construct a scale to estimate the histamine level in skipjack tuna, providing an indication of the extent of spoilage after exposure to warm conditions.

Previously we reported that the optimum temperature for histamine formation in skipjack tuna was 100°F (Frank et al., 1981). In the present study, fresh skipjack tuna were placed in seawater for various periods at five temperatures. After incubation, the histamine contents of the decomposed fish were estimated, and a nomographic scale was made from the his-

tamine levels observed at the time-temperature combinations employed. It is recommended that such a scale be used as a guide on fishing vessels to estimate histamine formation in skipjack tuna prior to freezing.

Materials and Methods

Skipjack tuna, weighing about 4-5 pounds, were held alive for 12-18 hours in circulating seawater until shortly before experimental incubation, as described previously (Frank et al., 1981; Yoshinaga and Frank, 1982). Individual fish were placed in separate polyethylene bags containing 4-5 l of filtered fresh seawater and incubated in a water bath for the desired times at 70°, 77.5°, 85°, 90°, or 100°F. Two fish were employed for each time-temperature combination in most experiments.

After incubation, the tuna were eviscerated and decapitated, and the two sides were separated and steam-heated for 15 minutes at 220°F in a home-style pressure cooker. Each side was debrided, comminuted twice with a household meat grinder and analyzed for histamine by the fluorometric AOAC (1975) method employed previously (Frank et al., 1981; Yoshinaga and Frank, 1982). Data reported below for each time-temperature combination represent the mean values of eight estimations, being taken from duplicate analyses by two technicians for both sides of the fish. Histamine content (mg histamine per 100 g of the entire loin tissue) is equivalent to the composite histamine value described earlier (Frank et al., 1981).

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ABSTRACT—Histamine formation in skipjack tuna was measured in fresh, whole fish incubated at five temperatures between 70° and 100°F. Employing the data collected, a nomograph was constructed to show the relationship between the histamine content and incubation times over the temperature range studied. The equation for this nomograph is:

$$H = 2.8 \cdot 10^{-32} \cdot T^{4.86} \cdot F^{13.85}$$

where H = histamine (mg/100 g tuna), T = time (hours), and F = temperature (°F). Since histamine is generally considered to be an index of microbial decomposition in tuna, the nomograph can be used to estimate spoilage in scombroid fish that have been held too long in warm environments.

Results and Discussion

Derivation of Equation Describing Histamine Formation

Table 1 shows the effect of incu-

bation time on histamine formation in skipjack tuna over the range of 70°-100°F. When these data are plotted on a log-log scale, a set of straight lines can be drawn to show the rela-

tionship between incubation time and histamine formation at the temperatures tested (Fig. 1). This relationship can be expressed mathematically by a power function equation,

Table 1.—Histamine formation in skipjack tuna at elevated temperatures¹.

Temperature and time (hours)	Histamine (mg/100 g tuna)			Temperature and time (hours)	Histamine (mg/100 g tuna)			Temperature and time (hours)	Histamine (mg/100 g tuna)			
	Trial 1	Trial 2	Mean		Trial 1	Trial 2	Mean		Trial 1	Trial 2	Mean	
70°F				18	1.40	6.25	3.83	90°F				
0	—	— ¹	0.11 ²	24	45.4	66.7	56.1	0	—	—	0.11 ²	
6	0.31	0.59	0.45	30	86.9	28.2	57.6	7.5	0.34	0.52	0.43	
12	0.26	0.30	0.28	85°F				15	5.98	24.7	15.3	
18	0.41	0.32	0.36		0	—	—	0.11 ²	22.5	189	102	146
24	0.44	1.32	0.88		3	0.07	0.22	0.15	30	449	330	390
30	4.59	2.16	3.38		6	0.11	0.23	0.17	37.5	275	— ³	275
36	69	6.09	37.5		9	0.35	0.34	0.35	100°F			
42	48.4	114	81.2	12	0.53	3.31	1.92	0	—	—	0.11 ²	
77.5°F				15	1.53	2.38	1.96	6	0.50	0.49	0.50	
0	—	—	0.11 ²	18	32.4	— ³	32.4	12	2.57	27.7	15.1	
6	0.32	0.26	0.29	21	49.9	— ³	49.9	18	80.9	53.6	67.3	
12	0.41	0.41	0.41	24	137	— ³	137	24	219	467	343	

¹Fresh, whole fish were placed in separate plastic bags containing seawater, incubated for the desired time at the temperature shown, and analyzed for histamine.

²Six fish.

³No data.

⁴From Frank et al., 1981 (Table 4).

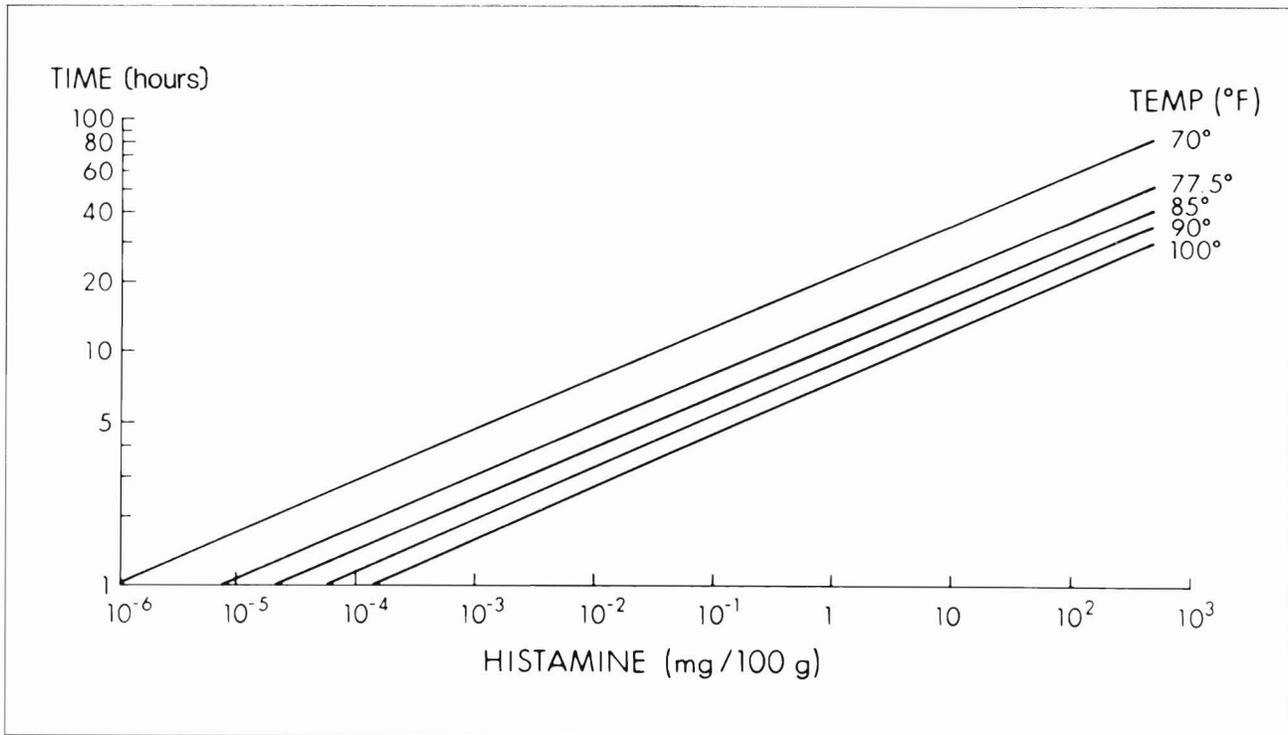


Figure 1.—Straight-line curves for log-log plot of histamine formation in skipjack tuna at elevated temperatures.

$$H = a \cdot T^x \cdot F^y \quad (1) \quad 1 = 2.8 \cdot 10^{-32} \cdot 7.1^x \cdot 100^{13.85} \quad (1c)$$

where

H = histamine (mg/100 g tuna),
 T = time (hours),
 F = temperature ($^{\circ}$ F), and
 a, χ, γ = constants.

To determine the constants in Equation (1), we selected two points in Figure 1 and observed that when

$$F = 100 \text{ and } T = 1, \\ H = 1.4 \cdot 10^{-4}$$

and Equation (1) becomes

$$1.4 \cdot 10^{-4} = a \cdot 1^x \cdot 100^y = a \cdot 100^y \quad (1a)$$

Also, when

$$F = 70 \text{ and } T = 1, \\ H = 1.4 \cdot 10^{-6}$$

and Equation (1) becomes

$$10^{-6} = a \cdot 1^x \cdot 70^y = a \cdot 70^y \quad (1b)$$

Dividing Equation (1a) by Equation (1b) yields

$$\left(\frac{100}{70}\right)^y = \frac{1.4 \cdot 10^{-4}}{10^{-6}} = 140 \\ (1.4286)^y = 140 \\ \gamma \log 1.4286 = \log 140 \\ \gamma = \frac{\log 140}{\log 1.4286} \\ = \frac{2.146}{0.155} = 13.85$$

and Equation (1a) now becomes

$$1.4 \cdot 10^{-4} = a \cdot 100^{13.85} = a \cdot 10^{27.7}$$

and

$$a = \frac{1.40 \cdot 10^{-4}}{10^{27.7}} = \frac{1.40 \cdot 10^{-4}}{10^{27} \cdot 10^{0.7}} \\ = \frac{1.40 \cdot 10^{-4} \cdot 10^{-27}}{5.01} \\ a = 0.28 \cdot 10^{-31} = 2.8 \cdot 10^{-32}$$

Figure 1 also shows that when

$$H = 1 \text{ and } F = 100, T = 7.1$$

and Equation (1) becomes

Also, when

$$H = 10 \text{ and } F = 100, T = 11.4$$

and Equation (1) becomes

$$10 = 2.8 \cdot 10^{-32} \cdot 11.4^x \cdot 100^{13.85} \quad (1d)$$

Dividing Equation (1c) by Equation (1d) yields

$$\frac{1}{10} = \left(\frac{7.1}{11.4}\right)^x \\ 0.1 = (0.6228)^x$$

and

$$\chi = \frac{\log 0.1}{\log 0.6228} = \frac{-1}{-0.20565} = 4.86$$

Finally, then,

$$H = 2.8 \cdot 10^{-32} \cdot T^{4.86} \cdot F^{13.85} \quad (2)$$

Employing Equation (2) and procedures described by Johnson (1966), we constructed a nomograph for histamine formation over the range of 70 $^{\circ}$ -100 $^{\circ}$ F (Fig. 2).

The histamine content for a given set of conditions can be found on the middle scale of the nomograph (Fig. 2) after drawing a straight line between the particular time and temperature conditions prevailing. For example, a level of 10 mg histamine per 100 g would be present after 29 hours at 70 $^{\circ}$ F, 14 hours at 90 $^{\circ}$ F, or 11 hours at 100 $^{\circ}$ F.

Use of Nomograph

Because it provides a means for estimating histamine content, the nomograph can be used to evaluate the effect of exposing skipjack tuna to warm temperatures during handling. Table 2, which was assembled from the nomograph, lists the times at 70 $^{\circ}$, 90 $^{\circ}$, and 100 $^{\circ}$ F required for formation of three levels of histamine. Figure 2 and Table 2 show that a substantial delay preceded the formation of detectable amounts of histamine, especially at low temperatures. For example, up to 18 hours at 70 $^{\circ}$ F were required for production of only 1 mg of histamine per 100 g tuna. How-

ever, the rate of histamine formation accelerated as incubation continued, and the time needed to produce higher levels of histamine decreased accordingly.

Extensive studies in our laboratory have shown that a reduction in several quality attributes (appearance, texture, odor, flavor) is associated with decomposition in skipjack tuna (Frank and Yoshinaga, 1980¹). Fish containing more than 10 mg of histamine per 100 g invariably were found to be unacceptable by an experienced panel of sensory evaluation judges.

It has been suggested that 10 mg histamine per 100 g is the maximum allowable in canned tuna, and that tuna containing more than this amount should be considered decomposed (Geiger, 1944; Arnold and Brown, 1978). Assuming that 10 mg histamine per 100 g tuna is an acceptable limit, the time interval during which histamine increases from 1 to 10 mg becomes crucial in controlling decomposition. Table 2 shows that histamine formation could be reduced by keeping exposure times under 20 hours at 70 $^{\circ}$ F, 10 hours at 90 $^{\circ}$ F, 8 hours at 100 $^{\circ}$ F, or below comparable times at other temperatures in the 70 $^{\circ}$ -100 $^{\circ}$ F range (Fig. 2).

Recently the U.S. Food and Drug Administration declared that 20 mg/100 g and 50 mg/100 g tuna should be considered as defect and hazard levels for histamine in tuna products (Federal Register, September 14, 1982).

Table 2.—Histamine formation in skipjack tuna¹.

Histamine (mg/100 g)	70 $^{\circ}$ F	90 $^{\circ}$ F	100 $^{\circ}$ F
	Time (hours)		
1	18	9	7
10	29	14	11
100	46	23	17

¹Taken from nomographic scale (Fig. 2).

¹Frank, H. A., and D. H. Yoshinaga. 1980. Unpubl. data. Univ. Hawaii, Honolulu.

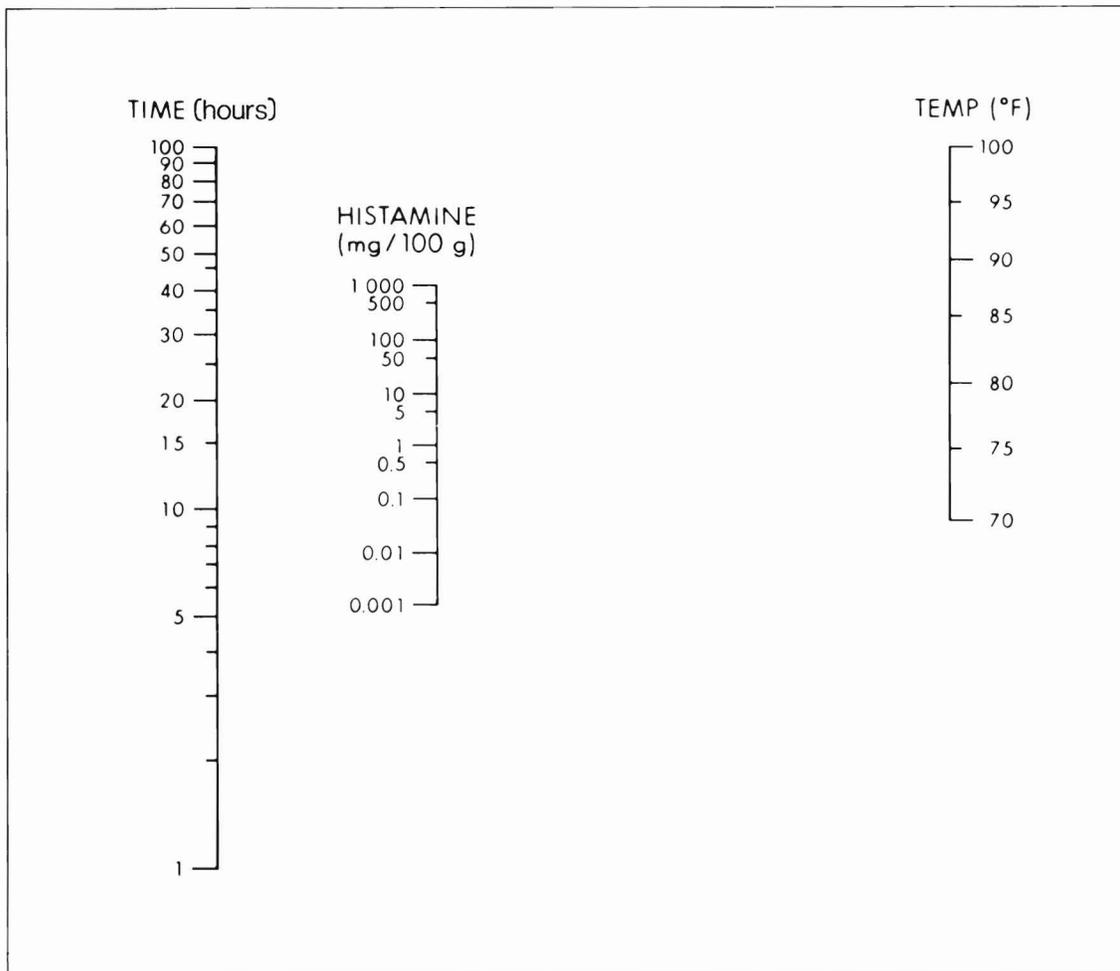


Figure 2.—Nomograph for $H = 2.8 \cdot 10^{-32} \cdot T^{4.86} \cdot F^{13.85}$, where H = histamine (mg/100 g tuna), T = time (hours), and F = temperature (°F).

Other Considerations

The nomograph was prepared from data collected with fresh, relatively small skipjack tuna incubated under controlled (laboratory) conditions at constant incubation temperatures. Applicability of the nomograph to larger fish, to varying environmental temperatures, and to other scombroid species must await further testing, preferably aboard a commercial fishing vessel. It is unlikely that the nomograph will be generally suitable for estimating histamine levels in all types of scombroid fish particularly because of differences in histidine content among various species (Shew-

an, 1955; Lukton and Olcott, 1958; Kimata, 1961; Ienistea, 1973). On one occasion, however, the nomograph was used successfully to find an appropriate time-temperature combination to produce a desired level of about 30 mg histamine per 100 g in a 7-pound, commercially fresh mahi mahi (dolphin fish), *Coryphaena hippurus* (footnote 1).

Histamine is not distributed uniformly throughout a decomposed fish (Hillig, 1956; Ienistea, 1973; Lerke et al., 1978). Instead, histamine is present as a gradient that is higher in the anterior loin tissue and diminishes gradually toward the tail (Frank et al.,

1981; Yoshinaga and Frank, 1982). Consequently, samples taken from different locations in a fish do not have the same level of histamine. Since it is based on composite measurements from the entire loin, the nomograph does not provide information about the histamine content at specific locations.

We have found that the relationship between composite histamine content (Figure 2 and Table 2) and the histamine level in the anterior loin section depended upon the extent of spoilage. Based on analyses of seven slightly decomposed fish (where the anterior section had <3 mg histamine

per 100 g tuna), the histamine content of the anterior section was 1.14 times that of the composite histamine. With 21 fish in advanced stages of decomposition (histamine in the anterior section was between 10 and 650 mg per 100 g of tuna), the histamine content of the anterior section was 1.93 times that of the composite histamine. Thus, for mildly spoiled tuna, the composite histamine content is generally representative of any location in the fish loins. However, in tuna that have undergone substantial decomposition, the nomographic estimate will be about one-half the histamine content in the anterior section of the fish.

Temperature Integrators

Miniaturized electronic devices have been developed recently to measure continuously the thermal history of perishable foods, including fish (Olley and Ratkowsky, 1973a, b; Olley, 1978; Olley et al., 1978). These devices can integrate the time-temperature exposures into a single value for the spoilage that has accumulated during storage. Thus, the residual shelf life of a perishable food at a given temperature can be determined at any time during storage.

Temperature function integrators could be used to estimate deterioration in tuna held aboard fishing vessels for long periods, especially at cooler temperatures. However, decomposition data at several storage temperatures (such as are presented in this paper) would be needed before the circuitry of the devices could be adjusted correctly. These integrators also would be useful for determining the accuracy of the nomographic histamine estimations developed in this publication.

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