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STUDIES ON POST-MORTEM CHANGES IN THE CHEMICAL CONSTITUTION OF THE MEAT OF SEA CUCUMBER (*STICHOPUS JAPONICUS* SELENKA)

II. Autolysis of the Meat of *Stichopus japonicus*

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There is no published study on the autolysis of the meat of *Stichopus japonicus*. The so-called "meat" (edible part) of sea cucumber is not true muscular tissue histologically, but is mainly connective tissue.¹⁾ Therefore there may perhaps be some differences between the so-called "meat" and fish meat muscle.

The authors undertook to investigate the autolytic phenomena when the bodies of *Stichopus japonicus* are left in the air. They have determined the optimum pH for autolysis, and have estimated the periodical changes of the amount of autolyzed products in the solutions of various pH values, and velocity of the autolysis of the meat of *Stichopus japonicus* at various temperatures.

1. Phenomena of autolysis when the bodies of *Stichopus japonicus* are left in the air

After the death of *Stichopus japonicus*, and while the body is left in the air, the meat is observed to soften gradually with the lapse of the time and to become mucous matter.

When the body of *Stichopus japonicus* is left on straw, the phenomena of autolysis occur rapidly; when the body is bound up with straw rope, the body is divided into two parts from the point of the binding. It is said that those phenomena are due to the action of some enzymes in the straw, but that point remains yet unknown.

The authors have observed the degrees of the phenomena of autolysis in the appearance of the bodies of *Stichopus japonicus* under various conditions of leaving.

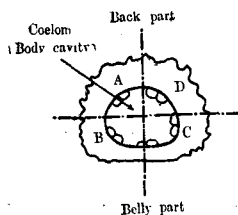


Fig. 1. Eviscerated *Stichopus japonicus* preparation

(1) Samples

As samples, the bodies of *Stichopus japonicus* were left as they were and eviscerated bodies were also employed. The eviscerated body was quartered as shown in Fig. 1.

(2) Experimental method

Each piece of the eviscerated body and non-eviscerated body was left on an enamelled iron square dish or on the straw, in the ice box ($10^{\circ} \pm 4^{\circ} \text{C}$) and in the room ($20^{\circ} \pm 4^{\circ} \text{C}$) respectively.

Those pieces so left were taken up at definite hour intervals, and the mucous matter which had formed on the surface of the sample was removed carefully by pincette. Then the residual solids were weighed. The percentage of the difference in the weight of the

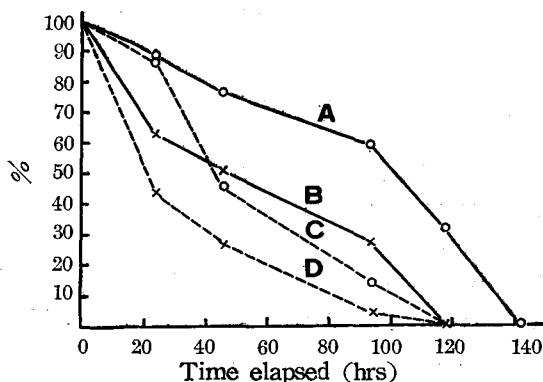


Fig. 2. Auto-viscidation of eviscerated meat

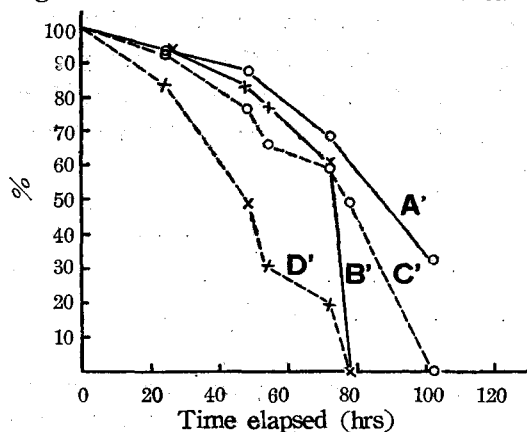


Fig. 3. Auto-viscidation of non-eviscerated meat

Then, whether the pieces of the body of *Stichopus japonicus* which were left on the straw have more rapid autolysis than pieces which were not so left was observed, and results are shown in Table 2.

Table 1. Differences in the degree of auto-viscidation of *Stichopus japonicus* meat stored at various temperatures (%)

Differences	Time (hrs)											
	0	24	46	48	54	72	78	94	102	118	142	
I Sample (C-A)	0	2.15	31.29	—	—	—	—	45.15	—	31.22	—	
II Sample (D-B)	0	19.46	24.10	—	—	—	—	24.28	—	—	—	
III Sample (C'-A')	0	1.00	—	10.84	13.59	7.24	9.36	—	32.63	—	—	
IV Sample (D'-B')	0	8.89	—	34.16	45.69	40.86	—	—	—	—	—	

piece before leaving and the weight of the piece from which the mucous matter was removed was calculated for the weight of the piece before having been left.

(3) Experimental results

The results obtained are shown in Figs. 2 and 3. Table 1 shows the difference (%) of the degree of autolytic phenomena of *Stichopus japonicus* at various temperatures.

As seen in Table 1, samples C, D (eviscerated quarter piece left in the room) and C', D' (non-eviscerated body left in the ice box) show larger degree of autolysis than samples A, B (eviscerated quarter piece left in the room) and A', B' (non-eviscerated body left in the ice box).

Samples left at the room temperature evidence a larger degree of autolysis than samples left at ice box temperature. That is to say, the samples at higher temperature are autolyzed more rapidly than those at lower temperature.

Table 2. Differences in the degree of auto-viscidation of *Stichopus japonicus* meat stored on straw mat or iron square dish (%)

Differences	Time(hrs)										
	0	24	46	48	54	72	78	94	102	118	142
I Sample (A-B)	0	25.74	25.82	—	—	—	—	31.94	—	31.22	—
II Sample (C-D)	0	43.05	18.63	—	—	—	—	10.07	—	—	—
III Sample (A'-B')	0	0.39	—	4.38	2.97	7.45	58.28	—	—	—	—
IV Sample (C'-D')	0	8.28	—	27.70	35.07	30.06	48.92	—	—	—	—

As seen in Table 2, the piece left on the straw shows more rapid autolysis than that on the enamelled iron square dish. The factor of leaving of the piece on the straw exerts greater influence than the factors of temperature and treatment. More explicitly, there is no difference of the degree of autolysis between eviscerated and non-eviscerated bodies, or between the bodies left at room temperature and at ice box temperatures. Comparing the time in which the entire bodies of *Stichopus japonicus* become mucous matter as a result of autolysis, the eviscerated sample requires longer time than the non-eviscerated sample.

2. Relation between the autolysis of the meat of *Stichopus japonicus* and hydrogen ion concentration

According to Ōya²⁾, in the autolysis of fish meat, the addition of acid stimulates the velocity of the autolytic activity, but the addition of alkali inhibits autolysis. The autolytic activity of fish meat shows generally the most active at about pH 4.5.

At this value of pH, the increase in the amounts of soluble nitrogen, polypeptide nitrogen and amino acid nitrogen are the largest.

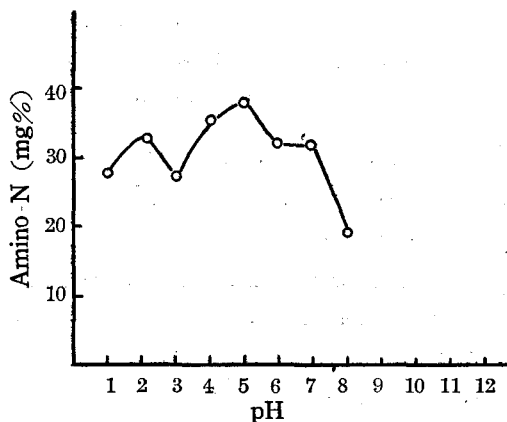


Fig. 4. Changes in the amount of amino acid nitrogen in the solutions adjusted to various pH values

In order to learn the optimum pH of the meat of *Stichopus japonicus*, the following experiments were made.

(1) Sample

Bodies of *Stichopus japonicus* which were caught in October in the sea near Hakodate were eviscerated and skinned as reported previously.⁷⁾ The meat part was crushed and employed as the sample.

As seen in Fig. 4, the amounts of amino acid nitrogen shows two peaks pH 2.2 and pH 4.8. In any case, the

optimum pH of the meat of *Stichopus japonicus* for autolysis is considered to be pH 4.8.

3. Relation between the amounts of amino acid formed in the meat of *Stichopus japonicus* which was autolyzed in various pH solutions and the autolyzing time

Measurements were made of the amounts of volatile base nitrogen, amino acid nitrogen, peptone nitrogen and protein nitrogen which were formed by the autolysis in the

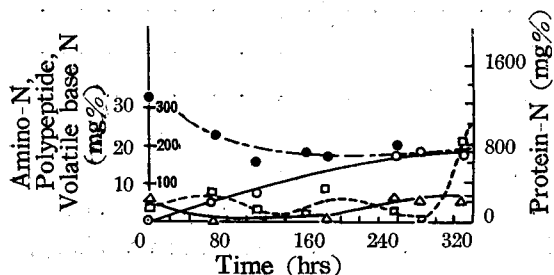


Fig. 5. Changes in the amounts of several fractionated nitrogens in distilled water (pH 6.7)

○—○ Volatile base-N △—△ Amino-N
□—□ Polypeptide-N ●—● Protein-N

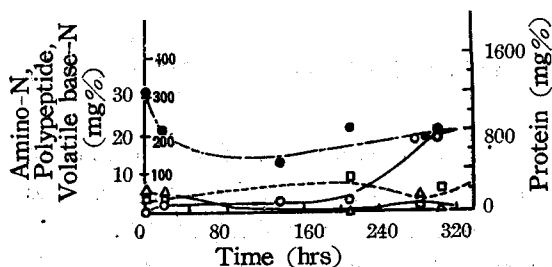


Fig. 6. Changes in the amounts of several fractionated nitrogens in alkali solution (pH 8.9)

○—○ Volatile base-N △—△ Amino-N
□—□ Polypeptide-N ●—● Protein-N

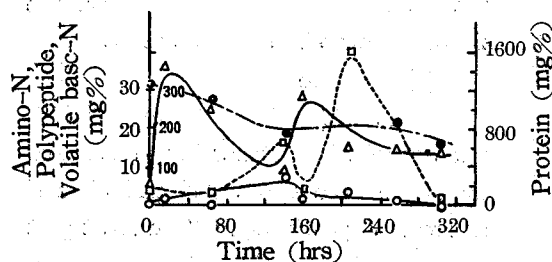


Fig. 7. Changes in the amounts of several fractionated nitrogens in acid solution (pH 4.5)

○—○ Volatile base-N △—△ Amino-N
□—□ Polypeptide-N ●—● Protein-N

meat of *Stichopus japonicus* in solutions of pH 4.5 and pH 8.9 and in distilled water (pH 6.7) at definite time intervals.

(1) Sample and experimental method

Sample was the same as described above in Article 1. Measurements of the amount of volatile base nitrogen were made by Weber and Wilson's method; the pepton nitrogen was measured by the A. O. A. C.³⁾ method; and the protein nitrogen was measured by usual method after removing of protein with trichloroacetic acid solution.

(2) Experimental results

Results obtained are shown in Figs. 5, 6 and 7.

As seen in the Figs., the speed of autolysis in distilled water or alkali solution is slow, but the velocity is rapid in acid solution, because the optimum pH of the autolysis of the meat of *Stichopus japonicus* is on the acid side.

4. The influence of temperature upon the velocity of autolysis of the meat of *Stichopus japonicus*

In order to ascertain the optimum temperature for the autolytic action of the *Stichopus japonicus* meat, the meat was autolyzed at $30^{\circ} \pm 1^{\circ}\text{C}$, $15^{\circ} \pm 1^{\circ}\text{C}$, $3^{\circ} \pm 1^{\circ}\text{C}$ in a solution of the optimum pH value and the amounts of amino acid nitrogen, protein nitrogen and peptone

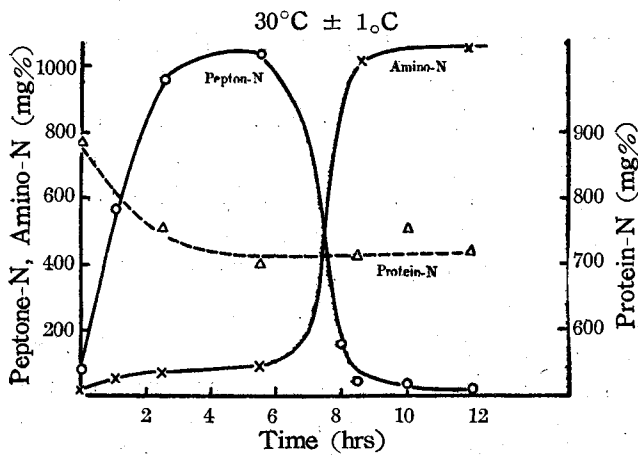


Fig. 8. Autolytic action of *Stichopus japonicus* meat at 30° ± 1°C

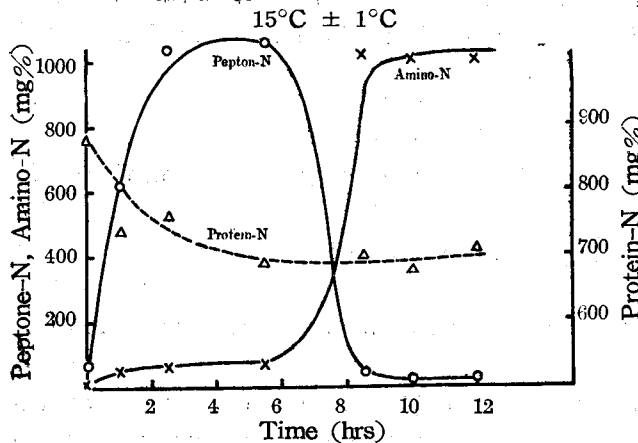


Fig. 9. Autolytic action of *Stichopus japonicus* meat at 15° ± 1°C

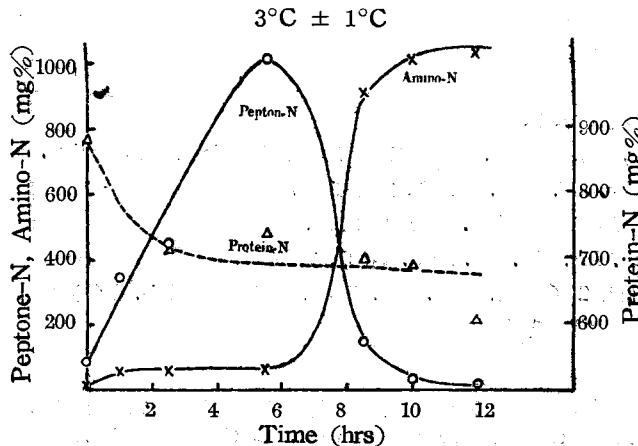


Fig. 10. Autolytic action of *Stichopus japonicus* meat at 3° ± 1°C

nitrogen were measured.

(1) Sample and experimental method

Sample and experimental method were the same respectively as described above in Article 1.

(2) Experimental results

Results obtained are shown in Figs. 8, 9 and 10.

As seen in the Figs, the amount of amino acid nitrogen produced in the course of *Stichopus japonicus* meat autolysis is larger at comparatively higher temperatures within the range of 3°~30°C.

Discussion

As stated above, the purpose of this paper has been to report on the influences of hydrogen ion concentration and temperature upon the autolytic action of the meat of *Stichopus japonicus*. From the results obtained, the amount of the decomposed product was ascertained to attain equilibrium at pH 4.5 after 20 hours. Within the range of 3° and 30°C, the autolytic action was more greater in high temperature than in low temperature. The optimum pH value for the autolytic action of the meat was 4.8 at room temperature (15~18°C). As above stated, Oya has studied

the autolysis of the meat of various kinds of fish²⁾, and he has observed that the optimum pH of fish meat autolysis was generally near 4.5*. Explaining a higher amino acid values obtained in 2.2 of the pH value, according to the present author, the tissue of *Stichopus japonicus* meat are considered to contain various protein components in the network of collagen fiber, differing from the tissue of fish meat¹⁾. There seems to be some relation between the histological characteristic and the chemical behavior in pH 2.2 of the meat of *Stichopus japonicus*. For example, according to Takahashi,⁴⁾ collagen fiber shows the maximum swelling at near pH 2 in the experiment on the skin of fish. The maximum swelling of the meat of *Stichopus japonicus* is considered to occur at and also to be decomposable at near pH 2. There also seems to be some relation between the chemical behavior in pH 2.2 of the decomposition and the isoelectric point of the meat. For example, the isoelectric point of the meat of *Stichopus japonicus* is one-sided being more acidic than that of fish meat—the isoelectric point of the meat of *Stichopus japonicus* is pH 4.2, the Atka mackerel meat is pH 5.2,⁵⁾ and the horse mackerel is pH 5.6⁶⁾.

Summary

(1) After the period of rigor mortis is passed, the autolysis was observed also in the meat of *Stichopus japonicus*.

(2) The optimum pH of the autolysis of the meat of *Stichopus japonicus* was considered to be near 4.8.

(3) The amount of the decomposition product reached equilibrium at pH 4.5 at 20 hours after leaving.

(4) In the range of experimental temperatures (3°~30°C), the higher the temperature is, the more active is the autolysis. This result is generally similar to what is observed in the case of fish meat.

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* Ōya has examined the optimum pH for the autolysis of fish meat by using fish meat suspension. On the contrary, the present authors have employed raw crushed meat. So the results which were obtained by the former and by the latter can not be compared mutually.