



Fig. 1. Changes in the amounts of adenine and hypoxanthine compounds in squid muscle during storage at -5°C . \square , Hypoxanthine; \blacktriangle , inosine; \circ , adenosine monophosphate; \triangle , adenosine diphosphate; \bullet , adenosine triphosphate; \circ , total purine compounds

without any conversion to inosine monophosphate, was converted to inosine and then to hypoxanthine. It was observed that these changes, which occurred in squid muscle, are almost quantitative. In squid muscle, as compared with carp muscle, two specific phenomena can be observed. The first is the accumulation of adenosine monophosphate. A possible pathway for the deamination of this substance, other than direct deamination, is by a combination of a 5'-nucleotide phosphatase with adenosine deaminase. This is supported by the fact that adenosine was converted to inosine very rapidly in crude extracts of squid muscle (Saito, T., and Arai, K., unpublished). The second is dependent upon the degree of splitting of adenosine triphosphate. In carp muscle there is a very rapid splitting as soon as freezing is complete, and at the same time inosine monophosphate accumulates; in squid muscle these specific phenomena are not observed.

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Amino-Acid Composition of Hæmoglobin from *Thunnus thynnus*

WE have recently crystallized¹ hæmoglobin and myoglobin of the saltwater fish, *Palamys sarda* and *Thunnus thynnus*. Some physico-chemical and functional properties of these pigments have been described²⁻⁴. In this communication we report the amino-acid composition of *Thunnus thynnus* hæmoglobin, as determined by chromatography on an ion-exchange resin: a slightly modified version⁵ of the method of Moore and Stein.

The hæmoglobin crystals, obtained from ammonium sulphate, were dissolved in water, dialysed against water and hydrolysed with 6*N* hydrochloric acid for 24 hr. The quantity of protein present in each

Table 1. AMINO-ACID COMPOSITION OF CRYSTALLINE *Thunnus thynnus* HÆMOGLOBIN (GM. AMINO-ACID/100 GM. PROTEIN)

Amino-acid	Hydrolysate 1				Hydrolysate 2		Mean
Aspartic acid	11.77	11.00	9.97	11.69	11.4	11.28	11.18 ± 0.31
Threonine	4.67	4.5	4.47	4.48	4.12	4.04	4.38 ± 0.09
Serine	4.71	4.85	4.51	4.51	4.10	4.24	4.48 ± 0.16
Glutamic acid	6.90	6.89	7.14	6.67	6.54	5.88	6.67 ± 0.18
Proline	2.45	1.66	2.58	2.93			2.40 ± 0.28
Glycine	4.45	4.49	5.08	4.38	3.87	3.82	4.34 ± 0.22
Alanine	9.04	8.35	7.68	8.92	8.58	8.39	8.49 ± 0.22
Valine	7.01		7.92	8.12	6.21	6.89	7.28 ± 0.35
Methionine	3.22		2.37	2.37	2.51	3.59	2.81 ± 0.26
Leucine	5.90		5.63	6.10	5.32	5.48	5.68 ± 0.19
Leucine	12.74		11.80	11.73	11.11	11.28	11.78 ± 0.30
Tyrosine	4.81	4.99	4.72	4.73	4.37	4.16	4.63 ± 0.12
Phenylalanine	6.69	6.65	6.82	6.44	5.93	5.70	6.37 ± 0.19
Histidine	4.04	3.98	4.20		4.42	4.02	4.13 ± 0.10
Lysine	8.95	8.40	8.15		8.97	8.75	8.64 ± 0.21
Arginine	5.67	5.73			5.46	5.49	5.58 ± 0.18
Total							98.74

sample used for chromatography was calculated on the basis of the nitrogen content (determined by the micro-Kjeldahl method in quadruplicate), assuming the value 16 for the percentage of nitrogen. The analyses were performed on two different hydrolysates of the same sample of crystals.

The results are given in Table 1. The values for proline were calculated using the formula previously reported⁵, since we did not obtain a good separation of this amino-acid from glutamic acid. No results for cystine are available, since the method used is not appropriate for this amino-acid, but it has been identified as cysteic acid by paper chromatography².

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Absolute Configuration of Enantiomorphous Carbanions Involved in the Aldolase and Triose Phosphate Isomerase Reactions

DIHYDROXYACETONE phosphate is one of many biological compounds which contain a carbon atom attached to two identical and two dissimilar groups, that is, a carbon attached to groups *a*, *a*, *b* and *d*. When one of the identical groups of such an 'Ogston atom'¹ or 'mesocarbon atom'² participates in a reaction it is of interest to establish the identity of that group³⁻⁶. Aldolase and triose phosphate isomerase have been shown to labilize only one of the hydrogens bound to the carbinol-carbon of dihydroxyacetone phosphate, and it has further been demonstrated that a different hydrogen is labilized by each of these enzymes^{7,8}. It is the purpose of this communication to establish the identity of the specific hydrogen