3,4-Dihydro-2H-1,4-benzoxazine was prepared from 2H-1,4-benzoxazin-3(4H)-one by reduction with lithium aluminium hydride [2]. UV-Spectrum (in ethanol) max. 274 mμ, $\varepsilon = 5\,350$, max. 296, $\varepsilon = 3\,150$.

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Amino Acid Composition of Seal Myoglobin I

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The isolation and purification of seal myoglobin I has been described recently [1]. The quantitative composition of amino acids has been determined by the method of Moore and Stein [3] using Amberlite IR-120 columns. For each analysis, 20 mg of salt-free myoglobin I was dissolved in 6 N HCl, the solution was cooled in an ice bath and the tubes were evacuated with a water pump and sealed. The hydrolyses were conducted in pairs in an oven at 110°C for 20 and 70 h. The cooled tubes were opened and the contents centrifuged and evaporated to dryness over NaOH pellets in a vacuum desiccator at room temperature. The dry material was dissolved in a small amount of water and dried. Each, nearly colorless, residue was dissolved in 0.2 N sodium citrate buffer, pH 3.25 just before putting it on the column. A sample containing 0.0949 μmole of amino acids, calculated on the basis of protein, was used in each run.

The calculations are based on Moore and Stein's [3] values for color yields, and residues/mole are based on a molecular weight of 18 600 deduced from the iron content [1]. It has been possible to detect small peaks for methionine sulfoxide in 20 and 70 h hydrolysates. Table 1 shows the values obtained.

The amide ammonia values are not included in the summation of amino acid residues.

In order to provide a check on the amide NH_3 values calculated from the chromatographic results, the amide nitrogen was determined by two different methods, micro Kjeldahl and Nessler. For the Nessler nitrogen a solution of 3 mg of protein in 2 ml N H_2SO_4 was heated for 4, 6 and 8 h in a sealed tube at 105°C[4]. An ammonium sulfate solution was used as a standard. In the micro-Kjeldahl technique 22 mg of protein was heated with 0.9 ml of 6 N HCl for 20 h in a sealed tube at 110°C. The resulting ammonia was titrated with 0.5 N H_2SO_4 using an "Agla" micrometer

Table 1. Amino acid composition of hydrolysates of seal myoglobin I.

Amino acid	Time of hydrolysis 20 h	70 h	Number of residues to nearest integer
	Residues/mole		
Aspartic acid	10.5	10.4	11
Threonine	4.7	4.7	5
Serine	6.8	5.8	7
Glutamic acid	15.8	15.7	16
Proline	4.2	3.9	4
Glycine	11.8	11.5	12
Alanine	13.9	13.8	14
Valine	5.3	5.9	6
Methionine	1.6	1.3	2
*Iso*leucine	6.3	7.2	7
Leucine	17.9	18.0	18
Tyrosine	1.8	1.6	2
Phenylalanine	6.7	6.6	7
Lysine	17.9	17.9	18
Histidine	12.3	12.0	12
Arginine	4.6	4.9	5
Amide NH_2	6.7	8.1	7
Total			146

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syringe. The Nessler results after 8 h hydrolyses were the same as the 20 h micro-Kjeldahl.

As Kendrew [5] has mentioned in *Structure and function in myoglobins and other proteins*, "myoglobins from different species have amino acid compositions which are broadly similar, but differ in detail". We may only confirm his statement by comparing the compositions of the purified myoglobins which are presently available, namely sperm whale [6], seal and horse [7] myoglobin. The results show that these myoglobins differ only in details.

The amino acid analysis of Mb II, Mb III, Mb IV and Mb V are in progress using the procedure of Moore and Stein.

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Structure of Rhodan Hydrate

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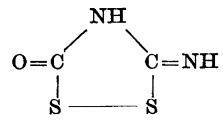
So far X-ray structure determinations of two unsaturated five-membered cyclic disulphides have been reported [1,2]. Observed bond lengths in the thiuret ion [1] and in 4 - methyl - 1,2 - dithia - 4 - *cyclo*pentene - 3 - thione [2] indicate that the five-membered ring has some aromatic character. This seems to explain the relatively high stability of unsaturated five-membered cyclic disulphides as contrasted with saturated ones. It is hoped that a determination of the molecular structure of rhodan hydrate will give further information that can be used to test this idea. The rhodan hydrate molecule has, according to Söderbäck [3], the following structure:



The preparation of rhodan hydrate has been described by Söderbäck [3,4], and the unit cell and space group have been reported by Foss [5]. The crystals are monoclinic, $a = 12.50$ Å, $b = 5.24$ Å, $c = 14.67$ Å,

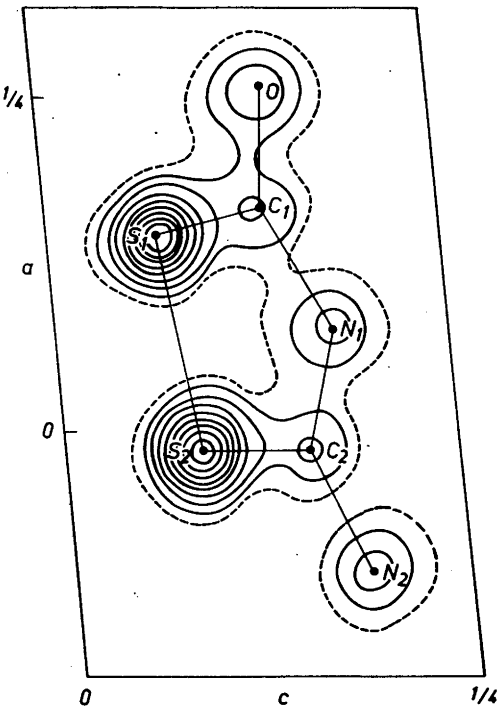


Fig. 1. Electron density projection of rhodan hydrate along the *b*-axis showing one asymmetric unit. Plane group *p*2 and origin in center of symmetry in the projection. Contours at arbitrary but equal intervals.