Myosin was prepared in the standard way[2,3] and complete light scattering, viscosity and ultracentrifuge measurements were made within sixty hours after the rabbit's demise. Within this period the ultracentrifuge showed a single peak. Longer storage (at 4°) caused the appearance of a second, faster peak indicating some dimer formation. All the data reported here, therefore, refer to a monodisperse protein.

Results for three samples are given in the table.

TABLE I

Sample	$[S_{20,w}] \times 10^{13}$	$[\eta]$ (dl/g)	$M \times 10^{-5}$	$(\overline{\rho^2})^{1/2}$(Å.)
VII	6.38	2.3	570	487
VIII	6.43	2.2	500	473
IX	....	2.4	526	470

A typical angular scattering envelope, extrapolated to zero concentration, is shown in Fig. 1. The molecular weight, M, and r.m.s. radius of gyration, $(\overline{\rho^2})^{1/2}$, were determined from the curve in the usual way.[4] The intrinsic viscosity, $[\eta]$, and sedimentation constant, $[S_{20,w}]$, were obtained by extrapolation to infinite dilution. The solvent was 0.6 M KCl and the temperature of measurement 25°.

The values for $[\eta]$ and $[S_{20,w}]$ found here are in good agreement with some of those in the literature.[5,6,7] The observed molecular weight, though lower than earlier values,[6,7] agrees well with the more recent estimate of Laki and Carroll.[8]

These data may be used to elucidate the molecular configuration. In particular, we can test the consistency of the most likely models, the rod and random coil.

To test the rod model we use the theoretical relations for intrinsic viscosity and sedimentation constant.[9,10] The equations relate these properties to the length, L, diameter, d, and mass of a rigid string of spherical beads. Since the diameter is the only parameter not directly measured, the consistency of this model depends upon whether a single value of this quantity fits the viscosity and sedimentation behavior.

The viscosity equation can be written[10]: $[\eta] = 24\bar{\nu}J^2/9000 \ln J$, where J is the axial ratio and $\bar{\nu}$ the partial specific volume.[11] From this we get $J = 71$. Since the light-scattering radius corresponds to a rod-length of 1650 Å., we obtain 23.3 Å. for the diameter.

If Stokes' law is assumed for the spherical rod-segments, the sedimentation equation becomes[9]: $[S] = (1 - \bar{\nu}\rho_0)d^2 \ln (6M\bar{\nu}/N\pi d^3)/18\ \bar{\nu}\eta_0$ with η_0 the solvent viscosity, and N Avogadro's number. This equation yields 27.4 Å. for the diameter.

(2) A. Szent-Györgyi, "The Chemistry of Muscular Contraction," Academic Press, New York, N. Y., 1951, 2nd edition, p. 146.

(3) W. F. H. M. Mommaerts and R. G. Parrish, *J. Biol. Chem.*, **188**, 545 (1951).

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(7) R. G. Parrish and W. F. H. M. Mommaerts, *J. Biol. Chem.*, **209**, 901 (1954).

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(10) J. G. Kirkwood and P. Auer, *ibid.*, **19**, 281 (1951).

(11) We use the value 0.728 as determined by W. F. H. M. Mommaerts and R. G. Parrish, ref. 7.

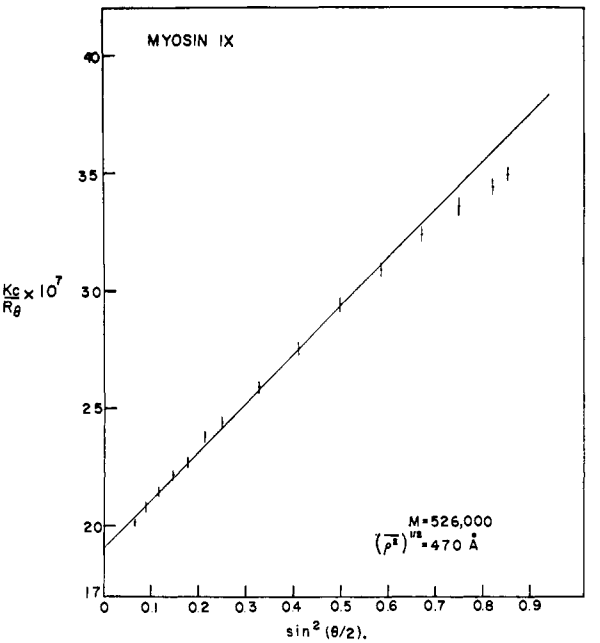

Fig. 1.—Extrapolated angular scattering curve for pure myosin.

A third estimate can be made from the definition of density: $1/\bar{\nu} = 6M/N\pi d^2L$, which gives 27.9 Å. for d.

The agreement, within error, of these three values of d demonstrates the consistency of the rod model.

The inappropriateness of the random coil model may be shown by using the Flory–Fox relation for $[\eta]$ and the Flory–Mandelkern relation for [S].[12] These equations incorrectly predict values of 335 and 300 Å., respectively, for $(\overline{\rho^2})^{1/2}$.

We conclude that the appropriate model is a rod 1650 Å. long, 26 Å. thick, and of molecular weight 530,000.

(12) P. J. Flory, "Principles of Polymer Chemistry," Cornell University Press, Ithaca, N. Y., 1953, pp. 616 and 609.

CONTRIBUTION 1402 FROM THE
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SYNTHESIS OF POTASSIUM HEXACHLORORHENATE AND POTASSIUM HEXABROMORHENATE

Sir:

It has been shown that small amounts of perrhenate can be converted quantitatively to hexachlororhenate(IV) ion by chromium(II) chloride reduction in strong hydrochloric acid solution.[1] The same reaction may be used for the synthesis of macro amounts of potassium hexachlororhenate-(IV).

The synthesis described here may be accomplished in considerably less time than with the other preparations of this salt,[2,3] yet the purity remains at

(1) V. W. Meloche and R. L. Martin, *Anal. Chem.*, **28**, 1671 (1956).

(2) L. C. Hurd and V. A. Reinders, "Inorganic Synthesis," Vol. I, McGraw-Hill Book Co., New York, N. Y., 1939, pp. 178–180.

(3) C. L. Rulfs and R. J. Meyer, THIS JOURNAL, **77**, 4505 (1955).

least as high. Hydriodic acid[2] and hypophosphorous acid[3] have been used previously to reduce the perrhenate in the preparation of potassium hexachlororhenate(IV).

Potassium hexabromorhenate(IV) is formed in the same manner when chromium(II) bromide and hydrobromic acid are substituted for the chloride reagents. This rhenium salt may be prepared in higher yields, in better purity, and in considerably less time than with the existing method of preparation.[3]

Procedure.—Add approximately 3 g. of potassium perrhenate, a stoichiometric amount of potassium chloride (0.77 g.), 100 ml. of concentrated hydrochloric acid (12.4 *N*), and 20 ml. of water to a 250-ml. Erlenmeyer flask. Heat the contents of the flask for several minutes until the potassium perrhenate goes into solution. While the solution is still warm, add about 25 ml. of 1.7 *M* chromium(II) chloride, preferably under an atmosphere of nitrogen. Prepare the chromium(II) chloride by running a 1.7 *M* chromium(III) chloride solution, which is about 0.2 *M* in hydrochloric acid, through a Jones reductor. After cooling the flask in an ice-bath, suction filter the green precipitate into a sintered glass filter crucible. Wash the salt with two 5-ml. volumes of ice-cold 10% hydrochloric acid and then, twice each, with ethyl alcohol and diethyl ether. Air dry the salt by drawing air through the filter by means of suction, and finally dry in an oven at 110° for one hour.

The yield of the potassium hexachlororhenate is about 55%, with a purity of about 98.5%. The yield can be increased to about 85%, with no appreciable sacrifice in purity, by boiling the solution down to a volume of about 30 ml. before filtering.

The same procedure is used for the preparation of potassium hexabromorhenate except that the chloride reagents are replaced with bromide. The 3 g. of potassium perrhenate are dissolved in 120 ml. of 48% hydrobromic acid. The chromium(III) bromide solution may be prepared conveniently by addition of hydrobromic acid to chromium(III) carbonate.

The yield of potassium hexabromorhenate is about 60%, with a purity of 99%. Here also the yield can be increased to about 85% with no sacrifice in purity by boiling the solution down to a volume of about 30 ml. before filtering.

Either of the preparations can be completed in less than an hour, not including the final drying.

The extension of this procedure for the preparation of potassium hexaiodorhenate(IV) and potassium hexafluororhenate(IV) was not successful.

Analyses.—The rhenium in the hexahalorhenates was oxidized to perrhenate with hydrogen peroxide after hydrolysis of the tetravalent salt. After boiling to destroy the excess hydrogen peroxide, aliquots of the solutions were analyzed for rhenium spectrophotometrically with alpha furil dioxime.[4] Other aliquots, after acidification with nitric acid, were assayed for halide by conventional gravimetric techniques.

(4) V. W. Meloche, R. L. Martin and W. H. Webb, submitted for publication.

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FORMATION OF α,γ-DIAMINOBUTYRIC ACID FROM ASPARAGINE-CONTAINING PEPTIDES[1]

Sir:

During the recent synthesis of the cyclic disulfide of L-cysteinyl-L-tyrosyl-L-isoleucyl-L-glutaminyl-L-asparaginyl-L-cysteinamide (I),[2] of interest because of its relationship to the pentapeptide ring moiety of oxytocin,[3] an unusual side reaction of some interest was observed. The route to I utilized several standard methods of peptide synthesis currently in wide use: namely, (1) couplings using the tetraethyl pyrophosphite reagent,[4] for the preparation of tosyl-L-isoleucyl-L-glutaminyl-L-asparaginyl-S-benzyl-L-cysteinamide (II) from tosyl-L-isoleucyl-L-glutaminyl-L-asparagine and S-benzyl-L-cysteinamide, and of carbobenzoxy-S-benzyl-L-cysteinyl-L-tyrosyl-L-isoleucyl-L-glutaminyl-L-asparaginyl-S-benzyl-L-cysteinamide (III) from L-isoleucyl-L-glutaminyl-L-asparaginyl-S-benzyl-L-cysteinamide and carbobenzoxy-S-benzyl-L-cysteinyl-L-tyrosine; and (2) removal of tosyl, benzyl, and carbobenzoxy protecting groups with sodium in liquid NH_3,[5,6] from II and III. Another product (IV) (K = 0.14), found to be present in 23% yield during the final isolation of I (K = 0.19) by countercurrent distribution,[7] has now been further purified by distribution in *sec*-butyl alcohol–0.01 *M* NH_3 (K = 0.77). Unlike I, it possessed no significant oxytocic activity.

Analysis of an acid hydrolysate of IV by the starch column procedure[8] in the solvent system (A) 1:2:1 *n*-BuOH–*n*-PrOH–0.1 *N* HCl followed by 2:1 *n*-PrOH–0.5 *N* HCl indicated cystine, tyrosine, isoleucine, and glutamic acid in molar amounts, 2 moles of ammonia, and an unidentified ninhydrin-positive substance (V), which was eluted at fraction 352 soon after cystine. These results were confirmed qualitatively by paper chromatography. V showed chromatographic behavior characteristic of the basic amino acids. Treatment of IV with 2,4-dinitrofluorobenzene (DNFB) followed by hydrolysis, resulted in the replacement of V by a yellow, acid-soluble, ninhydrin-sensitine DNP derivative (VI), R_f 0.52, 5:1:5 *n*-BuOH-HAc–H_2O; R_f 0.79, phenol, as well as affecting the cystine and tyrosine.

(1) This work was supported in part by a grant (H-1675) from the National Heart Institute, Public Health Service.

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