Further studies of the reactivity of chlorocarbene and the different behavior of methylene bromide toward butyllithium will be the subject of a detailed publication.

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CHROMATOGRAPHY OF MYOSIN

Sir:

The general method of Peterson and Sober[1] has been applied to the muscle protein, myosin or "myosin A." Myosin A[2,3,4] freed of myosin B by dialysis against 0.2 *M* KCl, 0.01 *M* tris *p*H 7.4 in the presence of adenosine triphosphate and by 1 hour of centrifugation at 55,000 × *g* was passed through a diethylaminoethyl cellulose column equilibrated with a solvent 0.2 *M* KCl, 0.01 *M* tris *p*H 7.4. An ascending gradient to 1.0 *M* KCl was applied (Fig. 1), and protein concentration was measured[5] in the effluent. Protein recovery was better than 80%.

TABLE I

Prepn.		α	β
19	$\bar{M}_w \times 10^{-5}$	4.52	6.10
	$\bar{r}_g$	437	474
	V_m (2 d.)	4.7	9.5
22	$\bar{M}_w \times 10^{-5}$	4.55	5.00
	$\bar{r}_g$	434	560
	V_m (12 d.)	0.4	3.8
28	$\bar{M}_w \times 10^{-5}$	4.02	5.60
	$\bar{r}_g$	475	634
	V_m (3 d.)	5.0	17
33	$\bar{M}_w \times 10^{-5}$	4.21	6.36
	$\bar{r}_g$	430	500
	V_m (0 d.)	8.0	8.7
	V_m (11 d.)	1.0	8.0
21	$\bar{M}_w \times 10^{-5}$	4.00	..
	$\bar{r}_g$	434	..

Myosin is resolved into at least two components, α and β (Fig. 1). Neither component shows a turbidity drop on adenosine triphosphate addition, confirming the elimination of myosin B. The α-component probably is highly purified myosin. The data[6] of Table I yield an average $\bar{M}_w$ of 4.3 × 10^5 g. and an average $\bar{r}_g$ of 442 Å. $\bar{M}_w$ from ultracentrifuge work[7] is 4.2 × 10^5 g. This shows that the two methods can agree; moreover the straightness of the Zimm light-scattering plot (Fig. 1) does not encourage speculation about myosin nonuniform substructure.[8] In this work the "full" Zimm plot (*i.e.*, intensities at various concentrations as well as at various angles) was not attempted because it has been shown[8] that in 0.6 *M* KCl the second virial coefficient is essentially zero. The β-component is heavier (average $\bar{M}_w$, 5.77 × 10^5 g.) and more extended (average $\bar{r}_g$ 542 Å.); also its specific ATPase activity,[9] V_m, (Table I) is greater and more thermostable than that of the α-component. Scattered observations suggest that β may be transformable into α, either by warming briefly from 4 to 25°, or by aging.

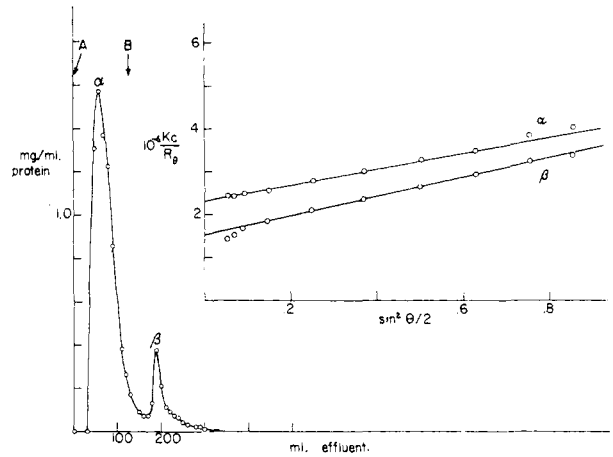

Fig. 1.—Chromatography on a 13 × 2.5 cm. column of diethylaminoethyl cellulose (1 meq./g.); eluting solutions: A—0.2 *M* KCl, 0.01 *M* tris *p*H 7.4; B—gradient elution to 1.0 *M* KCl; flow rate 60 ml./hr.; 10 ml. fractions were collected. The gradient used was composed of two cone-cylindrical vessels filled with 250 ml. of 1.0 *M* KCl, 0.01 *M* tris *p*H 7.4 and 125 ml. of 0.2 *M* KCl, 0.01 *M* tris *p*H 7.4. Insert shows: Zimm plot of α and β fractions in 0.5 *M* KCl, 0.01 *M* tris *p*H 7.4.

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(9) μ mole P·sec.$^{-1}$ g. protein^{-1} in 0.5 *M* KCl, 0.1 *M* tris, 10^{-3} $CaCl_2$, *p*H 8.0, 25°. The age of myosin preparation (in days) is indicated in parentheses.

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OPTICAL ROTATORY DISPERSION STUDIES. XXX.[1] DEMONSTRATION OF BOAT FORM IN A 3-KETO STEROID[2]

Sir:

Kinetically controlled bromination of 2α-methylcholestan-3-one[3] (or of its enol acetate) leads to 2-bromo-2-methylcholestan-3-one (m.p. 136–138°), whose spectral properties ($\lambda_{max}^{CHCl_3}$ 5.84 μ; $\lambda_{max}^{cyclohex}$ 313 mμ) require[4] an axial bromine atom. By

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