cessation of sino-auricular activity. This ventricles- auricles-sinus order of stoppage was characteristic. In a number of tests after complete stoppage had been brought about in the heart, washing with Ringer's brought back normal beat, and as was expected, the resumption of sino-auricular activity preceded that of auriculo-ventricular activity.

Electrocardiograms of hearts treated with concentrations inadequate to produce stoppage showed a decrease in rate and a lengthening of the P-R interval. If concentrations great enough to produce stoppage were used, there occurred first a lengthening of the P-R interval and later disappearance of the QRST complex with transient persistence of the P wave. Finally the P wave disappeared. The disappearance of the QRST complex coincided exactly with the observed cessation of ventricular activity. Recovery in Ringer's showed the same reversal of activity as did the mechanocardiograms. It was noted in both mechano- and electrocardiograms that low concentrations brought about a decrease in rate, while with high concentrations no perceptible decrease in rate occurred before cessation of activity.

To conclude, it may be said that *Triturus* embryonic toxin primarily affects the conducting mechanisms in the frog heart and secondarily contractility. Auriculo-ventricular conduction is first impaired, then sino-auricular activity, and lastly the contractility of the heart muscle itself.

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Estimation of Tryptophane Content of Various Proteins.

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An extensive study of the tryptophane content of various proteins was made by Jones, Gersdorff and Moeller.[1] They employed the Holm and Greenbank[2] modification of the May and Rose[3] method, in which the protein, together with a solution of paradimethylaminobenzaldehyde in 17.5% hydrochloric acid is incubated at 37° until maximum intensity of color develops. This usually requires from 6 to 8 days. Comparison is made with a standard casein similarly treated.

The desirability of shortening the time required for maximum color development led Bates[4] and Sullivan, Milone and Everitt[5] to propose modifications based on the suggestions of Komm[6] and of Boyd,[7] involving the use of mild oxidizing agents as accelerators. Bates[4] adds sodium nitrate to the mixture of casein and reagent in hydrochloric acid, and obtains maximum intensity of color very speedily at room temperature. Sullivan, Milone, and Everitt[5] digest the mixture of protein and reagent in 17.5% hydrochloric acid at 85° for 15 min, add a 0.3% solution of hydrogen peroxide, and read after cooling to room temperature.

The short procedure of Sullivan *et al.*[5] would permit the rapid estimation of the tryptophane content of a large number of proteins. Accordingly, 20 proteins, obtained, with the exception of egg albumin, from Jones,[1] were analyzed by this method. This permitted the comparison of the results so obtained with those reported by Jones, Gersdorff and Moeller.[1]

1 Jones, D. B., Gersdorff, C. E. F., and Moeller, O., *J. Biol. Chem.*, 1924, **62**, 183.

2 Holm, G. E., and Greenbank, G. R., *J. Am. Chem. Soc.*, 1923, **45**, 1788.

3 May, C. E., and Rose, E. R., *J. Biol. Chem.*, 1922, **54**, 213.

4 Bates, R. W., *Proc. Am. Soc. Biol. Chem.*, *J. Biol. Chem.*, 1937, **119**, vii.

5 Sullivan, M. X., Milone, H. S., and Everitt, E. L., *J. Biol. Chem.*, 1938, **125**, 471.

6 Komm, E., *Z. physiol. Chem.*, 1926, **156**, 35.

7 Boyd, W. J., *Biochem. J.*, 1929, **23**, 78.

Experimental. The colored complex formed as a result of the reaction of para-dimethylaminobenzaldehyde with casein in the rapid procedure, and that formed by casein and by tryptophane in the long procedure were analyzed, using a Coleman Regional Spectrophotometer, Model 10 S. The results, when plotted, yielded 3 curves exhibiting the same general absorption characteristics, with maximum absorption at a wave length of 590 mμ in each case.

The tryptophane content of 20 proteins was determined by the rapid procedure,[5] using casein as a standard. The casein, obtained from the Will Corporation, was analyzed by the Holm and Greenbank[2] method. The first standardization of this sample was carried out in June 1940 and showed 2.44% tryptophane, after correcting for moisture and ash. Subsequent standardizations over a period of 2 years showed no material variation. Hence there is no need to restandardize a given sample of casein, once its tryptophane value has been determined.

TABLE I.
Percentage of Tryptophane in Various Proteins.

Natural source and name of protein	Found by authors	Reported by Jones *et al.*
Cow's Milk		
Casein	2.44	2.2
Lactalbumin	2.69	2.69
Hen's Egg		
Ovalbumin	1.77	2.25
Hemp Seed		
Edestin	2.56	2.48
Wheat		
Gliadin	1.10	1.09
Soy Bean		
Glycinin	1.78	1.66
Mung Bean		
Alpha-globulin	1.97	2.03
Beta-globulin	1.12	1.18
Navy Bean		
Phaseolin	1.76	0.94
Conphaseolin	2.14	2.79
Lima Bean		
Alpha-globulin	1.96	1.92
Beta-globulin	1.58	2.16
Locust Bark		
Albumin	3.29	4.18
Globulin	3.19	3.17
Peanut		
Arachin	1.80	0.88
Conarachin	1.78	2.13
Chinese Velvet Bean		
Stizolobin	1.22	1.36
Tomato Seed		
Beta-globulin	1.23	1.45
Coconut		
Globulin	1.39	1.25
Almond		
Globulin	1.45	1.37
Flax Seed		
Globulin	3.95	3.98

The results presented in Table I represent, for each protein, an average of at least 4 separate determinations on solutions of the same concentration. All values are corrected for moisture and ash.

Discussion. The short procedure herein employed permits the rapid estimation of the tryptophane content of a large number of proteins. It requires the use of casein or some other protein as a reference standard. Sullivan, Milone and Everitt[5] found that although tryptophane itself could not be used, because of its instability in hot acid, casein standardized against tryptophane by the method of Holm and Greenbank[2] could be used, since equivalent amounts of casein tested by both the long and short procedures yielded the same maximum intensity of color. The color developed by casein in the rapid procedure and by both casein and tryptophane in the long procedure, when analyzed spectrophotometrically exhibited the same general absorption characteristics with maximum absorption in each case at 590 mμ.

The results obtained by the short procedure are in fair agreement with those found by Jones, Gersdorff and Moeller.[1] The lower value for egg albumin may be due to the fact that our sample was a commercial preparation of unknown purity. Of the remaining 19 proteins, 13 agree within 10% of the values of Jones *et al.*,[1] obtained by the long procedure of Holm and Greenbank.[2] The remaining 6 proteins for the most part gave lower values than found by these workers, a phenomenon we are at present unable to explain.

Summary. 1. The colored complexes formed as a result of the reaction of pure tryptophane[2] and of casein[2,5] with para-dimethylaminobenzaldehyde appear to be similar. 2. The rapid procedure[5] when employed in the determination of the tryptophane content of various proteins gives results which are in fair agreement with those reported by other investigators[1] using a long procedure.[2]