lating to central nervous action have been described.

Experimental. Table I shows the dose of the various compounds administered to mice required to paralyze half the animals (ED_{50}, loss of righting reflex). Also shown are the doses required to protect half the mice against the tonic phase of maximal electrical seizure pattern induced by a shock of 8 milliamperes of current applied for 0.5 second through ear electrodes. The ratios of these doses to the median lethal dose are given also for each compound. In most instances, several groups of 5 or 10 mice each were used to determine each value shown. The various dosages shown in Table I were obtained together with their standard errors where shown, by the plotting procedure of Miller and Tainter(7).

These compounds showed no significant protection against strychnine or metrazol convulsions in experiments in which 2 lethal doses of the convulsant were injected into mice intraperitoneally 5 minutes after the intraperitoneal administration of the compound to be tested. Survivals at the end of one hour were counted and a mean protecting dose, if any, was calculated.

EJA II, phenyl γ-(2-pyridyl) propyl ketone, was the most potent compound studied.

Discussion. It is noteworthy that many of these compounds exhibit potent action both as paralyzing agents and as anti-convulsants against electro-shock. However, the great majority of these compounds were characterized by a short duration of action, *viz.*, 5 to 20 minutes. Since our screening procedures measure protective activity against strychnine or metrazol for one hour, few of the compounds appeared to be effective anti-convulsants against these agents although in many cases there was a significant prolongation of survival time. Protection technics utilizing intravenous titration-to-convulsion or death(8) measure the instantaneous potency of the anticonvulsant without the immediate consideration of the probable duration of action. Such studies should provide more useful information concerning the site of action of anti-convulsant drugs having short action. These compounds have been found to be primarily central nervous depressants, although in two instances (M-85 and 170-2) stimulant activity is observed as indicated by excitement and convulsions (Table I). In mice the depressant action is characterized in most cases by an initial arching of the neck and stiffening of the limbs, with paralysis ensuing shortly thereafter. A similar action has been observed in preliminary experiments on rats, rabbits, and cats.

Because of the short duration of action of these compounds, they show little promise of clinical application at present. However, in view of their interesting central action, further studies on these and related compounds are being undertaken.

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8. Orloff, M. J., Williams, H. L., and Pfeiffer, C. C., PROC. SOC. EXP. BIOL. AND MED., 1949, v70, 254.

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Distribution of Radioactivity in the Egg after Feeding Sodium Acetate-1-C^{14}* (18615)

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Earlier work on the distribution of radioactivity in eggs following the administration of radioactive isotopes of phosphorus, strontium and iron has been discussed in the com-

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prehensive treatise of Romanoff and Romanoff (1). More recently, further work has been carried out using isotopic phosphorus(2) and calcium(3,4).

The present work was undertaken with the two-fold purpose of securing data to indicate the uptake of radioactive carbon in the egg and of obtaining from these eggs products of high enough specific activities to warrant their use in subsequent work. The isotopic carbon used was in the form of $CH_3C^{14}OONa$ and was mixed with the feed over a period of several days with a view to obtaining high activities in the eggs produced. The radioactive carbon was incorporated into the shell, albumen and yolk of every egg laid during the experimental period.

Experimental procedures. The sodium acetate-1-C^{14} (9.55 millicuries, 0.787 g, sp. act. 12.1 μc/mg) was prepared by carbonation of methyl magnesium iodide with $C^{14}O_2$ (5). The sodium acetate-1-C^{14} was dissolved in a small amount of water and the resulting solution was sprayed over the feed (a practical laying ration) which was being stirred by hand. After all the solution was added, the feed was thoroughly mixed. The labeled acetate was administered in a total of 3 feedings. The first feeding comprised 4.78 mc of acetate in 350 g of feed. Then 2.39 mc of acetate in 175 g of feed and 2.38 mc in 175 g of feed were fed on the sixth and eighth days, respectively, after the initial feeding. As soon as one portion of feed was exhausted, a new one was prepared and fed. Thus, the hen was fed labeled acetate for ten days, at which time she was sacrificed. The hen used was a Single Comb White Leghorn, maintained in a wire cage with a raised floor. The shells were washed free of adhering matter and the yolk and albumen of each egg was lyophilized separately. Each shell was freed of its shell membrane, which was lyophilized with the albumen, and then air dried and pulverized. Aliquots of the shell, yolk and albumen were converted by dry combustion to CO_2, which was collected and counted as barium carbonate. Seven eggs were collected over the 10 day period of the experiment. The eggs were collected on the second, third, fifth, sixth, eighth, ninth and tenth days after feeding was begun. The hen was sacrificed on the tenth day and the contents of the ovary were separated into 4 fractions comprising 2 large ova, a number of easily separated small ova and the ovary with the remaining ova. Each of these fractions was homogenized, lyophilized and the activity of an aliquot was determined.

Results and discussion. Romanoff and Romanoff(6) present data to show that the egg, in passing through the oviduct, spends 3 hours in the albumen secreting region and 19 hours in the uterus, where the shell is laid down. The fact that in egg 1 (which was laid within 24 hours after feeding was begun) radioactivity was present in the yolk, albumen and shell, reflects the very rapid metabolic utilization of the acetate.

In the first egg, most of the activity was found in the shell, which is understandable, since radioactive carbonate was being produced at the same time that carbonate was being utilized for shell formation. The specific activities of shells 2, 3, 5, 6 and 7 were roughly the same, the carbonate for these shells probably having been supplied by the metabolic pool. The activity of shell 4, formed soon after fresh feed was offered, was considerably higher.

The yolks exhibited a specific activity variation that parallels that of the shell, with one exception which will be noted shortly. The specific activities began to fall off as the feed was exhausted and had the hen been allowed to lay the 2 eggs represented by yolks 8 and 9, the yolk specific activities would have been 75×10^{-4} μc/mg and 36×10^{-4} μc/mg, respec-

1. Romanoff, A. L., and Romanoff, A. J., *The Avian Egg*, John Wiley and Sons, Inc., New York, 1949, p. 363.

2. O'Neil, J. B., Jowsey, R., Lee, C. C., Reade, M. A., and Spinks, J. W. T., *Science*, 1948, v107, 295.

3. Comar, C. L. and Driggers, J. C., *Ibid.*, 1949, v109, 282.

4. Spinks, J. W. T., Berlie, M. R., and O'Neil, J. B., *Ibid.*, 1949, v110, 332.

5. Calvin, M., *et al.*, *Isotopic Carbon*, John Wiley and Sons, Inc., New York, 1949, p. 178.

6. loc. cit., p. 223 ff.

TABLE I. Weights and Activities of Collected Eggs and Ova.*

Day	Acetate fed	Egg	Total activity of egg	Shell		Albumen		Yolk	
				Wt	Act.	Wt	Act.	Wt	Act.
1	4780								
2		1	6.4	6.50	4.8	3.35	0.2	8.56	1.4
3		2	53.0	6.22	16.9	3.27	3.8	8.61	32.3
5		3	57.3	6.95	16.1	4.40	8.3	8.81	32.9
6	2390	4	115.5	6.13	33.4	4.08	6.8	8.48	75.3
8	2380	5	56.4	6.35	20.6	4.30	11.3	8.30	24.5
9		6	132.7	6.11	13.7	3.54	8.1	8.37	110.9
10		7	129.6	5.00	10.7	4.21	5.3	9.33	113.6
—		Ovum 1	64.8	—	—	—	—	5.85	64.8
—		" 2	30.4	—	—	—	—	3.25	30.4
—		Ova	9.4	—	—	—	—	1.43	9.4
—		Ovary and ova	10.5	—	—	—	—	2.44	10.5

* All weights of lyophilized solid, given in g. All activities as μc.

tively, based upon average weight of yolks obtained.

Possible explanations for the low activity of yolk 5 are: (1) error in marking and perhaps in combustion of the yolk, or (2) failure of the hen to ovulate this yolk at the normal time, that is, to have produced an egg on the fourth day between eggs 2 and 3. No instances of such "held" yolks were found in the studies of Warren and Conrad(7). Further investigation into the specific activities of various yolk fractions will indicate which of these possibilities is the more valid.

The specific activity of all the albumen fractions was lower, which might be expected considering that for inclusion into albumen, the acetate fragment or its carboxyl moiety would be required to traverse longer metabolic pathways than for inclusion into shell or yolk. In general, a gradual increase of specific activity of the albumen fractions was observed, which would seem to indicate that the radioactive portion of this fraction was being supplied by the metabolic pool.

In all, 666 μc (6.96%) of the total fed activity was recovered in the eggs; of this, the yolks contained 495.5 μc (5.18%), the shells 116.2 μc (1.21%), the albumen 43.8 μc (0.46%) and the ovary and ova 10.5 μc (0.11%).

7. Warren, D. C. and Conrad, R. M., *J. Agr. Research*, 1939, v58, 875.

Summary. The data indicate that upon feeding of carboxyl-labeled sodium acetate, the activity is rapidly incorporated into all portions of the egg. The acetate is utilized immediately and a comparatively small percentage of radioactivity is diverted to the metabolic pool. Steady feeding of labeled material is required in order to maintain a high specific activity in all portions of the egg.

It is a pleasure to acknowledge the interest shown by Professors Melvin Calvin and Samuel Lephovsky during the course of this work.

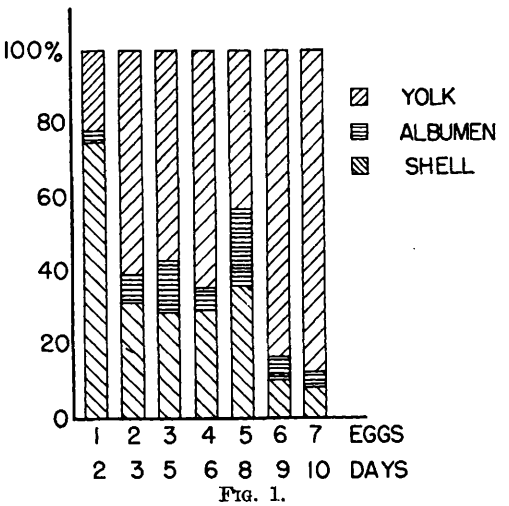

FIG. 1.
Distribution of total activity in laid eggs.

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