with the rubber stopper is somewhat higher than the exit, and an appropriate glass double bend makes connection with the flask. This tube must be large enough (7 mm o.d.) to prevent the formation of a siphon. The solvent must be introduced first to prevent water from entering the return tube. Long extractions with carbon tetrachloride have been carried out in this way without difficulty.

While this type of extractor had no greater efficiency than the type usually used[2] in removing materials easy to extract, such as succinic acid, an appreciable difference was found where the distribution between the solvents was less favorable. Citric acid provided an example of this type.

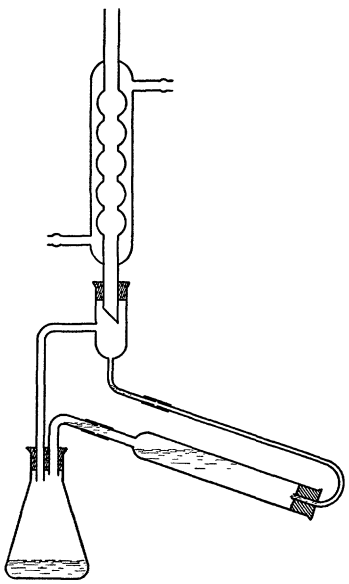

FIG. 1.

Experimental observations are shown in the accompanying tables.

TABLE 1

SUCCINIC ACID

Extraction time (hrs.)	1.5	3.5	6.5
Type A Extractor[2]	0.33g	0.48g	0.50g
Type B Extractor	0.32	0.47	0.50

Table 1 gives the total amount of material extracted in the time given from a solution of 0.50g of succinic acid in 15 ml of water.

TABLE 2

CITRIC ACID

Extraction time (hrs.)	5	10	20	28	35
Type A Extractor ...	0.05g	0.12g	0.30g	0.41g	0.54g
Type B Extractor ...	0.10	0.19	0.38	0.46	0.52

The total amount extracted in the time given from a solution of 0.52g of citric acid in 15 ml of water is shown in Table 2. The rate at which the ether condensed was in all cases about 15 drops in 10 seconds.

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SPECIAL ARTICLES

ST. LOUIS ENCEPHALITIS

SEROLOGICAL RELATION TO JAPANESE ENCEPHALITIS AND EXPERIMENTAL STUDIES ON IMMUNITY

THE severe outbreak of encephalitis in St. Louis[1-4] and Kansas City, Mo., during the late summer of 1933, preceded by a similar epidemic in Paris, Ill., in 1932,[5, 6] has been differentiated clinically and epidemiologically from Economo's disease (lethargic or epidemic encephalitis) and likened to the summer epidemic of encephalitis in Japan, designated as Type B.[7] A further comparison of the Japanese and St. Louis disease has now become possible, following the discovery of a virus as the causative agent of the St. Louis encephalitis[8, 9] and a specific immune reaction between this virus and the blood serum of convalescents.[9, 10] The basis for the immunity test rests on the fact that sera from a large majority of tested convalescents of the St. Louis, Kansas City and Paris disease exhibit a specific protective effect when mixed with living virus obtained from the brains of fatal cases and injected intracerebrally into Swiss mice. Animals receiving these mixtures usually remained well, while those given mixtures of virus plus sera from non-contact individuals invariably died.

[2] Quick, *Ind. Eng. Chem. An. Ed.*, 5: 76, 1933; McNair, *Ind. Eng. Chem. An. Ed.*, 5: 76, 1933.

[1] J. P. Leake, *Jour. Am. Medical Assn.*, 101: 928, 1933.

[2] J. F. Bredeck, *Am. Jour. Pub. Health*, 23: 1135, 1933.

[3] J. P. Leake, *Am. Jour. Pub. Health*, 23: 1140, 1933.

[4] T. C. Hempelmann, *Am. Jour. Pub. Health*, 23: 1149, 1933.

[5] W. E. Conklin, personal communication.

[6] H. S. Houston, *Ill. Health Quart.*, 6: 174, 1932 (No. 4).

[7] R. Kaneko and Y. Aoki, *Erg. d. inn. Med. u. Kinderheilk.*, 34: 342, 1928.

[8] R. S. Muckenfuss, C. Armstrong and H. A. McCordock, *Pub. Health Rep.*, 48: 1341, 1933.

[9] L. T. Webster and G. L. Fite, SCIENCE, 78: 463, 1933.

[10] L. T. Webster and G. L. Fite, *Proc. Soc. Exp. Biol. and Med.*, 31: 344, 1933.

Sera from cases of encephalitis in Japan have now been tested for a similar effect. Professor Inada, of Tokyo, kindly sent us sera from 3 persons with a history of encephalitis in August, 1924, aged at that time 60, 50 and 51 years, respectively, and from 9 persons with a similar history in August and September, 1933, aged 17, 17, 20, 26, 33, 46, 53, 62 and 65 years, respectively. In each case fever was noted for 6 to 9 days. The sera were drawn January 10 to 12, 1934. Professor Takaki, of Tokyo, likewise sent us sera from 3 cases from the August, 1933, outbreak. None of these 15 sera showed any protective action against the virus of the St. Louis disease.

Experiments have also been made on the ability of the encephalitis virus to incite a specific immunity in the mouse. Active brain virus given to mice intranasally in doses as small as 10^{-5} gms or intracerebrally in 10^{-8} gms causes death, while the same virus-containing material injected intraperitoneally or subcutaneously in 10^{-2} gm amounts in $\frac{1}{2}$ cc of diluent (which is a million intracerebral and a thousand intranasal lethal doses) rarely proves fatal. Still smaller amounts, 0.001, 0.0001, 0.00001 and 0.000001 gm, when inoculated subcutaneously, induce no symptoms but render them immune to a million intracerebral and a thousand intranasal doses. This induced active immunity has persisted unchanged for 3 weeks and doubtless endures much longer.

This report, therefore, indicates first, that the Japanese B. type and the St. Louis form of epidemic encephalitis are serologically distinguishable; and second, that animals as highly susceptible to infection as are mice, by certain portals of entry of the virus, may be immunized actively by the introduction of minimal amounts of virus—into parts of the body more refractory to its pathogenic action.

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AN ATTEMPT TO ISOLATE VITAMIN A

UP to this time the most powerful concentrate of Vitamin A ever known was a pale yellow oil prepared from a liver oil by P. Karrer and his associates at Zurich. As he measured its potency with the antimony trichloride color reagent it was approximately 10,500 times as rich in Vitamin A as the standard cod-liver oil.

Karrer and Morf,[1] in a recent paper on this subject, refer to a rich concentrate prepared by Carr and Jewell[2] with a very high cod-liver oil value ("C. L. O. value") of 7,800 and to another preparation by Heilbron, Heslop, Morton, Webster, Rea and Drummond[3] with a C. L. O. value of 6,500. These English workers used high-vacuum distillation methods.

In the Severance Laboratory at Oberlin College we have recently obtained a much richer concentrate of Vitamin A than those listed above. Repeated experiments have yielded products with C. L. O. values ranging from 13,000 to approximately 14,000. A convincing number of products ranked above the 10,500 value previously supposed by many to represent pure Vitamin A. Until we can crystallize our richest product, however, we would not be justified in calling it the isolated vitamin, yet it seems probable that it is extremely close to 100 per cent. pure.

The Parke Davis Company and the Abbott Company generously supplied us with the non-saponifiable portion of halibut liver oil as a starting material. This was chilled in methyl alcohol solution, to freeze out cholesterol, etc., filtered cold under nitrogen, transferred to pentane by addition of water, dried over anhydrous sodium sulfate, and then in pentane solution cooled to about $-70°$ C. with the aid of carbon dioxide snow mixed with alcohol and again filtered with careful exclusion of oxygen to remove impurities frozen out. This procedure was, in general, that of Karrer, but from this point on variations were introduced.

The cold pentane solution was next filtered through a Tswett column of very specially prepared carbon (Karrer used alumina and lime), and washed completely through with pure pentane.

The method of Karrer involved washing the strongest color band into the middle section of his column,

TABLE I

VITAMIN A CONCENTRATES RANGING FROM 14,400 TO 10,500 TIMES THE POTENCY OF STANDARD COD LIVER OIL

No.	C.L.O. value	Worker	No.	C.L.O. value	Worker
1.	14,400	Manly	14.	11,400	Hartzler
2.	13,500	"	15.	11,300	"
3.	13,500	Hartzler	16.	11,300	"
4.	12,900	"	17.	11,300	Manly
5.	12,600	"	18.	10,800	Hartzler
6.	12,500	"	19.	10,700	"
7.	12,000	Manly	20.	10,600	"
8.	12,000	"	21.	10,600	"
9.	12,000	"	22.	10,600	Manly
10.	12,000	Hartzler	23.	10,600	"
11.	11,800	"	24.	10,500	Hartzler
12.	11,800	Manly	25.	10,500	"
13.	11,600	"			

NOTE:
Tswett Column I. includes filtrate fractions 1, 2, 7, 13.
" " II. " " " 3, 4, 5, 6, 14, 15, 21, 24, 25.
" " III. " " " 8, 9, 12, 17, 22, 23.
" " IV. " " " 10, 11, 16, 18, 19, 20.

[1] *Helv. Chim. Acta*, 16: 625, 1933.
[2] *Nature*, 131: 92, 1933.
[3] *Biochem. Jour.*, 26: 1178, 1932.