TABLE I. Summary of Radioactivity Data of Lipid Fractions of Liver and Mesentery.

Rat No.	Fatty acids fed (mg)	% absorbed	Specific activity* (× 10²) Liver 1	Liver 2	Liver 3	Liver 4	Liver 4/3	Mesentery 1	Mesentery 2	Mesentery 3	Mesentery 4
2	107	83	2.53	1.21	1.08	3.02	2.79	1.15	1.90	0.79	0.68
3	83	85	1.63	1.11	1.15	2.07	1.80	—	0.92	0.06	—
4	275	73	2.81	2.42	1.90	4.30	2.26	0.17	0.80	0.43	0.17
5	258	90	0.91	0.67	0.65	1.35	2.08	0.23	0.67	0.29	0.24
6	127	81	4.73	2.48	2.34	5.34	2.28	1.34	1.54	0.77	1.04
Mean			2.52	1.58	1.42	3.22	2.24	0.72	1.17	0.47	0.53
Stand. error of mean			±0.65	±0.37	±0.30	±0.72	±0.16	±0.31	±0.24	±0.14	±0.20

* Specific activity $= \frac{\%\text{ of administered counts}}{\text{mg C}}$.

1. Total fatty acids; 2. Free fatty acids; 3. Phospholipid fatty acids; 4. Neutral fat fatty acids.

TABLE II. Specific Activity of Liver Fatty Acids Isolated from Triglycerides Alone and from a Mixture of Triglycerides and Cholesterol Esters.

Rat No.	Specific activity (× 10²) Triglyceride fatty acids	Triglyceride + cholesterol ester fatty acids
2	3.02	3.86
3	2.07	2.07
5	1.35	1.54
6	5.34	3.97

tion of the phospholipids of the excised liver is in keeping with Fairbairn's observation(22) of the rapid hydrolysis of phospholipids of extirpated livers of cats and rats. The specific activity of the free fatty acids in the mesentery was always greater than the activity of any other fraction. They must, therefore, be regarded as preformed material, perhaps being deposited as soaps from the chyle.

In Table II a comparison is made of the concentration of C^{14} in the fatty acids of the triglycerides and of the fraction containing both triglycerides and cholesterol esters. Generally speaking, the activity of cholesterol esters was equal to or greater than the activity of the glycerol esters, an indication of the importance of cholesterol in the metabolism of ingested fatty acids.

Summary. Biosynthesized lipids labeled with C^{14} was fed to rats and the lipids from various organs examined for radioactivity. The importance of the observations in studies of lipid metabolism was pointed out.

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Effect of Leucocytes and Bacteria on Glucose Content of the Cerebrospinal Fluid in Meningitis. (18300)

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The subnormal level of glucose in the cerebrospinal fluid of patients with bacterial meningitis is frequently of diagnostic value in differentiating bacterial from aseptic meningitis(1). Reasons for the reduced concentration of glucose in bacterial meningitis are not understood although it is stated that the main factor is utilization of glucose by bacteria growing in the fluid(1). On the other hand, the concentration of glucose in aseptic

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meningitis is said to be normal because the leucocytes do not metabolize it(1). The object of the present investigation was to assess the importance of bacterial growth and leucocytic metabolism as factors tending to lower levels of glucose in the cerebrospinal fluid. It has been found that leucocytes and pneumococci utilize glucose in the cerebrospinal fluids of dogs but the data suggest that these factors are not sufficient in themselves to account for the low levels of glucose in cerebrospinal fluid of bacterial meningitis.

Methods. Tests were carried out with the cerebrospinal fluids of dogs rather than human beings because the production of aseptic meningitis by intracisternal injection of irritating substances made it possible to exclude more surely the participation of bacteria. The dogs were anaesthetized by intravenous nembutal and subjected to cisternal puncture with a 20 gauge needle. After withdrawal of 3 to 5 ml of cerebrospinal fluid, equal amounts of sterile normal rabbit serum or distilled water containing 5000 units of penicillin were administered intracisternally. Bacterial meningitis was produced similarly by intracisternal injection of 0.5 to 1 ml of a 12 to 18 hour culture of virulent Type III pneumococci. Further cisternal punctures were done after varying periods for examination and tests of fluid. The number of polymorphonuclear leucocytes per cubic millimeter of cerebrospinal fluid was determined in the usual manner(1). Absence of bacteria from fluids of dogs with aseptic meningitis was confirmed by aerobic and anaerobic cultures on rabbit blood agar plates(2). Pneumococci to be placed in normal cerebrospinal fluid were grown for 4 hours in beef infusion broth containing 10% normal sheep serum and 0.2% glucose. Small amounts of young cultures prepared in this manner were transferred by pipette to the cerebrospinal fluids, and numbers of pneumococci in the fluids were determined at hourly intervals. Bacterial content was determined by removing 0.5 ml of the fluid, making serial ten-fold dilutions in broth, followed by colony counts of poured blood agar plates. Concentrations of glucose were determined by standard methods(3,4). For measurement of the rate of consumption of glucose *in vitro,* fluids were shaken constantly at 37°C in a Warburg water bath as recommended by Barron and Harrop(5).

Results. Concentrations of glucose in the cerebrospinal fluids of dogs with aseptic meningitis are shown in Table I and were found to be essentially normal as judged by standards in human beings(1). Similar determina-

TABLE I. Glucose Levels in Cerebrospinal Fluids of Dogs with Aseptic and Pneumococcal Meningitis.

Exp. No.*	Type of meningitis	Leucocytes per cu mm	Conc. of glucose (mg per 100 ml)
1	Aseptic	2227	80
8	"	4284	61
19	"	30000	50
23	"	13000	53
24	Pneumococcal	25000	28
28	"	8000	17
29	"	30000	22
31	"	11520	23†

* To conserve space, only representative experiments are presented.

† Bacterial count of this specimen revealed 250000 pneumococci per ml of fluid.

TABLE II. Consumption of Glucose *In Vitro* by Cerebrospinal Fluids of Dogs with Aseptic Meningitis.

Exp. No.	Leucocytes per cu mm	Conc. of glucose: Before incubation	Conc. of glucose: After incubation	Consumption of glucose per hr (mg per 100 ml)
1	1305	72	61	11
2	3000	78	70	8
6	6400	45	47	0
5	13000	48	32	16
7	15000	74	63	11
9	20700	53	42	11
12	25920	66	49	17
3	30000	50	26	24
11	37000	49	36	13
10	43000	60	33	27
4	57600	128	98	30

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TABLE III. Consumption of Glucose *In Vitro* by Pneumococci Growing in Normal Cerebrospinal Fluid.

Exp. No.	Time (hr)	No. of pneumococci	Conc. of glucose (mg per 100 ml)	Consumption of glucose per hr (mg per 100 ml)
1	0	6×10^7	80	
				3
	1	15×10^7	77	
				26
	2	95×10^7	51	
				48
	3	15×10^8	3	
	4	15×10^8	0	
2*	1	6.7×10^6	85	
				3
	2	79×10^6	82	
				8
	3	34×10^7	74	
				20
	4	9×10^8	54	
				23
	5	2×10^8	31	

* For this experiment, glassware was cleaned by boiling in equal parts of concentrated nitric acid and distilled water.

tions in pneumococcal meningitis are also recorded in Table I and are lower than normal by the same standards. In Table II, it may be seen that definite amounts of glucose were consumed by fluids of dogs with aseptic meningitis and that the rate of utilization appears to be more rapid in fluids with higher cell counts. Table III shows the consumption of glucose *in vitro* by pneumococci growing in normal cerebrospinal fluid and it may be noted that only with extremely large numbers of pneumococci growing rapidly did the rate of consumption of glucose approximate that utilized by the leucocytes in fluid of aseptic meningitis.* In control experiments not recorded in the tables, prolonged incubation of normal cerebrospinal fluids did not result in diminished concentrations of glucose.

Discussion. Measurements of glucose before and after incubation of cerebrospinal fluids of dogs with aseptic meningitis have demonstrated that glucose is utilized by such fluids. Although two previous studies(1,6) report failure to detect consumption of glucose under somewhat similar conditions, several others(7-11) have presented qualitative experiments indicating that cerebrospinal fluid containing leucocytes but no bacteria is capable of using glucose. Consumption of glucose by these fluids must be due to the leucocytes and is not surprising in view of the considerable published evidence(5,12-17) showing that leucocytes from other sources metabolize glucose *in vitro*.

Normal levels of glucose are known to occur in aseptic meningitis of human beings(1) and this fact has been confirmed for dogs in the present experiments. Normal concentrations of glucose under these circumstances might not be expected in view of the fact that fluids from such dogs were shown to utilize glucose. However, an adequate explanation for this discrepancy would appear to be that glucose of the cerebrospinal fluid is replenished more rapidly by the body than it is utilized by the leucocytes.

It is important to note that *in vitro* tremendous numbers of rapidly growing pneumo-

* In the single instance where utilization of glucose by the bacteria was greater than that of leucocytes, the number of pneumococci was particularly large and the rate of growth especially rapid.

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cocci were necessary in order to consume glucose in cerebrospinal fluid as rapidly as the leucocytes in the fluids from dogs with aseptic meningitis. Since the utilization of glucose by less than 150 million pneumococci per ml was so low that it could hardly be measured it is probable that the consumption of glucose by the much smaller numbers of pneumococci usually present in meningitis would be so small as to be insignificant.† Previous workers(7,8,10,11) have added bacteria to cerebrospinal fluid and have shown consumption of glucose but have not attempted to measure rates of utilization of glucose under these conditions. On the other hand, studies of the consumption of glucose by bacteria in culture media have shown even lower rates than were found in the present experiments (18-20).

Inasmuch as normal mechanisms of replenishment appear to suffice to maintain glucose levels in the cerebrospinal fluid when leucocytes are present without bacteria, and since the numbers of bacteria in the cerebrospinal fluid in meningitis are usually much smaller than were used in the *in vitro* tests, it seems likely that additional factors must be operating to cause the lowered levels of glucose in the cerebrospinal fluid of bacterial meningitis. Especially, does it seem probable that other factors than utilization of glucose by leucocytes and bacteria are present in tuberculous meningitis. The numbers of leucocytes and bacteria are not great enough in this disease to overcome normal mechanisms of replenishment, particularly since there is evidence that lymphocytes consume less glucose than granulocytes(5,16) and that tubercle bacilli grow slowly with a low rate of utilization of glucose(21).

Other possibilities have been suggested as mechanisms for low levels of glucose in the cerebrospinal fluid in bacterial meningitis. For example, there may be a change in the permeability of the blood-cerebrospinal fluid barrier(9,11,22-24); or damage to meninges and choroid plexus may increase the glycolytic rate of these tissues(7,9). To support these theories there is as yet no direct experimental evidence.

Summary. 1. Levels of glucose in cerebrospinal fluids of dogs with aseptic meningitis were normal, while incubation *in vitro* of such fluids resulted in consumption of glucose. Evidently, glucose is transported into the cerebrospinal fluid *in vivo* more rapidly than it is utilized by the leucocytes under these conditions. 2. Very large numbers of pneumococci growing rapidly in normal cerebrospinal fluid were needed in order to consume glucose at rates approximating those at which glucose is utilized *in vitro* by fluids of dogs with aseptic meningitis. 3. Consumption of glucose by leucocytes and bacteria does not appear to account for lowered levels of glucose in bacterial meningitis.

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† The number of pneumococci in the cerebrospinal fluid of a dog with acute meningitis was found to be 250,000 per ml (Table I, Experiment 31).

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