performed cardiac arrest would ensue. These results are illustrated in Table I.

Since artificial stimulation of the superior laryngeal nerve[1] causes reflex closure of the glottis by contraction of the adductor muscles, it is possible that the symptoms here observed are of parasympathetic origin. In order to test the validity of this assumption experiments were performed to study the modification of this symptom-complex by drugs affecting the sympathetic and parasympathetic divisions of the autonomic nervous system and by section of the vagi. The results may be summarized thus:

Atropin sulphate (3 to 5 mg. per kilo intravenously) always caused immediate relaxation of the vocal cords, a return of normal respiration, disappearance of cyanosis, a shortening of anesthesia time, and an increase in tolerance to the M.L.D.

Ephedrin sulphate, in therapeutic doses (20 mg. per kilo intravenously) caused only partial improvement characterized by relief of dyspnea and slight relief of laryngospasm but with persistence of hyper-active laryngeal reflex. Increasing the dose caused complete relaxation of the vocal cords but was usually followed by ventricular fibrillation.

Adrenalin hydrochloride (0.15 mg. per kilo intravenously) caused immediate improvement but the untoward symptoms reappeared after 10 to 15 minutes.

Finally, bilateral cervical vagotomy invariably relieved the laryngospasm.

From the above data it is concluded that the closure of the glottis following the intravenous administration of these short-acting barbiturates in cats is due to stimulation of the parasympathetic division of the autonomic nervous system.

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Use of Cerebro-Spinal Fluid and Synthetic Salt Solutions in Studies of Tissue Metabolism.

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The desirability of having available a physiologically normal fluid medium in which tissue slices may be immersed for the study of their metabolism is widely recognized. The use of serum as such a

[1] Howell, W. H., *Textbook of Physiology*, ninth edition, p. 697.

TABLE I.
Composition of the Synthetic Medium.

Salt	Concentration of Stock Solution mM/L	Amt. Required per 100 cc. cc.	Final Concentration mM/L
NaCl	154	77.5	119
KCl	154	2.6	4
$CaCl_2$	104	1.15	1.2
$MgCl_2$	104	1.44	1.5
Phosphate Buffer (7.4)	67	0.75	0.5*
Phenol Red	0.1%	0.75	0.00075%
After mixing, and saturating the above solutions with 5.5% CO_2:94.5%O_2, $NaHCO_3$ solution was added as follows:			
$NaHCO_3$	154	15.8	24.3
Glucose	—	—	0.2%
pH			7.4

*The amount of phosphate added varied from 0.49 to 0.63 mM/L depending upon the amount present in the spinal fluid.

medium, though possible, presents technical complexities which preclude its wide application. It was suggested by Davis and Hastings[1] that cerebro-spinal fluid, being a protein-free biological fluid, had the advantages but not the disadvantages of serum. A comparison of the metabolism of liver in cerebro-spinal fluid and their standard synthetic solution indicated that oxygen consumption determinations made in the spinal fluid were greater than those made in their synthetic solution. These authors did not attempt to prepare a synthetic solution whose composition closely paralleled that of the spinal fluid.

The present communication is concerned with the metabolism of tissue slices in cerebro-spinal fluid and in a synthetic fluid medium whose essential inorganic constituents equalled in concentrations those of spinal fluid.

The indirect Warburg[2] technique was used throughout, making it possible to measure the consumption of oxygen, Q_{O_2}, and the production of carbon dioxide, $Q\frac{O_2}{CO_2}$. White rats of homogeneous strain were used in all experiments. The animals were killed by decapitation in a room maintained at 37°, the liver immediately sliced and placed in warmed salt solution. From this, they were transferred to their appropriate fluid medium and their metabolism measured at 38°. Observations were made at 15-minute intervals for a total of 75 minutes. The cerebro-spinal fluid used in the experiments was obtained through the kindness of the neurological services of the Boston City and Massachusetts General Hospitals, to

1 Davis, J. E., and Hastings, A. B., PROC. SOC. EXP. BIOL. AND MED., 1935, **32**, 1449.

2 Warburg, O., *Biochem. Z.*, 1924, **152**, 51.

TABLE II.
Apparent Effect of Presence of Phosphate on Calculated Oxygen Consumption and CO_2 Production.

	PO_4 Absent		PO_4 Present (0.5 mM/L)	
	h	H	h	H
Manometer Reading, mm.	—8	—91	—20	—104
Oxygen Consumption, cmm.	127		125	
Carbon Dioxide Production, cmm.	129		110	

whom the authors are indebted. The fluid was drawn under sterile precautions and kept refrigerated until used. Immediately before use, it was passed through a Berkefeld filter.

In view of the impracticability of carrying out complete inorganic analyses of each sample of spinal fluid before use, average values of the inorganic ions of spinal fluid (except phosphate) were taken as the basis of the preparation of the synthetic medium. The composition of the synthetic medium is given in Table I. In all experiments, the concentration of inorganic phosphate was determined in each sample of spinal fluid, and a similar concentration was added to the synthetic medium with which each was compared. This was done in order to obviate the possibility of having differences in results which were due to differences in the buffer values of the spinal fluid and the synthetic medium.

A critical mathematical analysis of the differential method revealed that although the presence of 0.5 mM of inorganic phosphate per liter had a marked influence on the manometer readings, the calculated oxygen consumption was not appreciably altered. However,

TABLE III.
Oxygen Consumption and CO_2 Production of Liver in Spinal Fluid and the Synthetic Medium.

	Q_{O_2}			$Q_{CO_2}^{O_2}$		
	cmm./mg./hr.			cmm./mg./hr.		
Exp. No.	Spinal Fluid	Synthetic Fluid	Difference	Spinal Fluid	Synthetic Fluid	Difference
1	9.5	9.8	—0.3	11.1	11.6	—0.5
2	11.0	8.6	+2.4	12.8	10.0	+2.8
3	8.6	9.8	—1.2	7.3	9.6	—2.3
4	10.5	9.2	+1.3	11.5	12.0	—0.5
5	8.6	9.7	—1.1	8.5	11.2	—2.7
6	11.8	12.0	—0.2	9.0	8.5	+0.5
7	9.2	7.5	+1.7	10.3	9.5	+0.8
8	8.7	7.1	+1.6	10.0	7.9	+2.1
9	9.1	9.0	+0.1	10.6	10.6	0.0
Aver.	9.7	9.2	+0.5	10.1	10.1	0

the apparent carbon dioxide production is significantly reduced. An example of such a calculation is given in Table II.

Glucose was added to the spinal fluid to bring the concentration to approximately 0.2%. All experiments were run in duplicate. No specimens of spinal fluid were used which contained enough protein to give a visible precipitate with 20% trichloracetic acid.

The results of the comparison of the oxygen consumption, Q_{O_2}, and carbon dioxide production, $Q\,{}^{O_2}_{CO_2}$, of liver slices in 9 experiments are given in Table III.

It may be concluded from these experiments that the metabolism of rat liver in the synthetic medium herein used is not significantly different from that observed in cerebro-spinal fluid, when care is taken to make the composition of the synthetic medium approximately equal to that of the spinal fluid.

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Possible Rôle of Glutathione as a Detoxifying Agent.

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Aside from the probable rôle which glutathione plays in oxidative mechanisms, the suggestion has been advanced that it may be of importance as a detoxifying agent.[1] It is, to say the least, extremely suggestive that this polypeptide consists of 3 amino acids, each one of which is known to act as a detoxifying agent.

It occurred to us that experimental evidence for or against such a hypothesis might be gathered by injecting a substance which is known to form a detoxified product with one of the amino acids of the glutathione molecule, and then determining the glutathione content of the blood. We proceeded on the hypothesis that such an injection would be followed by a mobilization of the glutathione in order to furnish the required amino acid for detoxifying purposes.

Waelsch,[2] using tribromoethanol and phenylacetic acid, claims to have obtained a decrease in the glutathione of the blood.

The substances we selected for subcutaneous injection were benzoic acid and bromobenzene. The former is known to combine

1 Harrow, B., and Sherwin, C. P., *Annual Rev. Biochem.*, 1935, **4**, 263.
2 Waelsch, H., *Arch. Exp. Path. Pharm.*, 1933, **169**, 625.