method.[11] 100 - 120 g female albino rats were used and that portion of each internal oblique muscle which could be separated as a thin layer without appreciable trauma was placed intact in the incubating flask. When the muscle was cut into a series of thin strips, little or no glycogen synthesis took place.

The results reported here are evidence that insulin-glycogenesis stimulation *in vitro* is not peculiar to diaphragm muscle. New material is available for further investigation of the action of insulin and other factors controlling glycogen formation and utilization.

Summary. If used in thin sheets with a minimum of tissue injury isolated skeletal muscle in the form of the internal oblique muscle of the rat forms more glycogen under the influence of insulin just as does the specialized muscle comprising the diaphragm. Desoxycorticosterone reduces the amount of glycogen formed by incubation with glucose alone as in the case of diaphragm muscle. Skeletal muscle offers new material for further investigation of the action of insulin.

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17235. Normal and Seizure Levels of Lactate, Pyruvate and Acid-Soluble Phosphates in the Cerebellum and Cerebrum.*

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Seizures induced by various means have been shown to result in an increase in lactic acid, pyruvic acid and inorganic phosphorus and a decrease in high energy phosphate levels in the cerebral cortex[1,2] or whole brain[3] of various experimental animals. There is little data on the several parts of the brain. As reported previously it is possible to show that seizures induced by low doses of nitrogen mustards start first in the cerebellum; further, the convulsive activity of these compounds is potentiated by inhalation of carbon dioxide.[4] Inhalation of carbon dioxide tends to prevent the changes in lactic acid and phosphate attendant upon seizures.[5] As a sequel to the above studies, seizures were induced in a series of cats using representative nitrogen mustards and various other convulsants, the brains were frozen *in situ* and the cerebrum and cerebellum then analyzed separately for lactic acid, pyruvic acid, inorganic phosphorus, and high energy phosphates.

Experimental. Preparation of the animals, methods of EEG and EKG recording, procedure of freezing with liquid air and methods of analysis have been reported in detail in previous papers from this laboratory.[1,5] Seizures were induced by intravenous injections of the compounds indicated in Table I. Cerebral and cerebellar cortex were easily dissected from the frozen brains with a small chisel and mallet. During the dissection frequent applications of liquid air were used to maintain the tissue in a frozen state.

Results. The data are summarized in Table I. In normal brains frozen without seizures the levels of the constituents determined were very similar in both cerebellum and cerebrum and compared well with those previously reported[1,5] except that the phosphocreatine levels were somewhat low. In brains frozen during seizures, however, marked dif-

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TABLE I.
Normal and Seizure Levels of Lactate, Pyruvate and Acid-Soluble Phosphates in the Cerebellum and Cerebrum.

Lactate*		Pyruvate		Inorg. P		Phospho-creatine		Adenosine Tri-PO_4		Adenosine Di-PO_4		
Cb†	Cx	Cb	Cx	Cb	Cx	Cb	Cx	Cb	Cx	Cb	Cx	Remarks
							Controls.					
1.37	1.36	.069	.077	6.72	5.11	1.58	1.49	0.81	0.48	2.56	2.70	
1.50	1.40	.120	.136	4.70	4.93	2.10	1.95	1.30	0.90	1.85	2.06	
1.43	1.38	.095	.106	5.71	5.02	1.84	1.72	1.06	0.69	2.20	2.38	Avg
							Nitrogen Mustards.‡					
1.80	4.30	.177	.213	4.17	4.24	2.10	1.20	0.39	1.69	3.15	1.32	$C_3H_7N(C_2H_4Cl)_2$
3.48	5.86	.193	.268	3.84	3.60	2.28	1.91	1.43	0.85	1.82	2.51	$H_2C=CHN(C_2H_4Cl)_2$
1.66	5.64	.134	.208	4.56	4.20	2.29	1.92	1.13	2.25	2.19	1.00	$C_6H_5CH_2N(C_2H_4Cl)_2$
1.18	2.76	—	—	4.08	4.56	2.77	1.69	1.71	1.49	1.73	1.57	$OC_4H_8NC_2H_4Cl$
3.45	4.08	.277	—	5.19	3.58	1.38	2.30	1.58	1.46	1.97	1.60	$C_4H_9N(C_2H_4Cl)_2$
1.86	2.10	.132	.146	5.94	4.56	3.08	2.16	1.59	0.79	2.00	2.10	$C_3H_7N(C_2H_4Cl)_2$
3.82	7.60	.250	.280	3.72	3.48	3.13	2.03	—	1.36	3.15	1.77	$(C_2H_4Cl)_2NCH_2CH_2N(C_2H_4Cl)_2$
2.46	4.62	.194	.227	4.50	4.03	2.43	1.89	1.30	1.41	2.30	1.70	Avg
							Metrazol.					
1.37	2.42	.087	.100	5.18	4.83	2.52	1.50	0.38	0.52	3.48	2.73	
1.50	2.66	.104	.110	4.97	4.68	1.87	1.20	0.53	0.71	2.84	2.35	
1.43	2.54	.095	.105	5.07	4.53	2.19	1.35	0.45	0.62	3.16	2.54	Avg
							Aminophylline.					
2.76	9.72	.116	.145	5.88	6.54	1.52	0.42	0.42	0.58	3.00	2.96	16′ convulsion
1.56	6.06	.154	.293		(8.21	5.88)		0.75	1.10	3.11	2.18	13′ ″
4.34	4.08	.126	.156	5.90	5.64	0.82	0.42	0.11	0.40	3.02	2.70	8′ ″
3.96	9.72	.112	.106	5.94	6.74	0.60	0.08	0.33	0.56	2.90	2.74	4′ ″
3.16	7.40	.127	.175	6.48	6.20	0.99	0.31	0.40	0.66	3.01	2.64	Avg
							Various.					
2.64	3.51	.240	.130	4.92	3.96	2.05	1.81	1.42	2.28	2.19	0.97	Castorix‖
1.62	3.48	.126	.164		(6.48	5.62)§		1.62	1.52	1.85	1.78	Dibenamine
2.68	4.50	.188	.254		(6.72	5.36)		1.16	1.64	2.24	1.42	Br. Camphor
5.74	6.04	.259	.339		(6.12	5.02)		—	—	—	—	Coramine
2.74	3.60	.164	.206		(7.10	5.18)		1.44	1.16	2.18	1.74	Strychnine
2.20	4.08	.115	.156	3.90	4.14	2.46	2.22	0.92	0.80	1.86	2.30	Electric shock

* All values mM/1000 g (wet wt).
† "Cb" and "Cx" designate cerebellar and cerebral cortex, respectively.
‡ Obtained through the courtesy of the University of Chicago Toxicity Laboratory.
§ Pairs of figures enclosed in parentheses are the sum of inorganic phosphate and phosphocreatine for cerebellum and cerebrum, respectively.
‖ 2-Chloro 4-dimethyl 1,6-methyl pyrimidine.

ferences were found. The lactic acid level rose only about half as much and the phosphocreatine level was appreciably higher in the cerebellum as compared to the cerebrum. There were no systematic differences discernible in the inorganic phosphorus and ATP-ADP levels; however, this may be due to the fact that many of the seizures were potentiated with carbon dioxide[4] a procedure which tends to minimize changes in the assayed constituents during seizures.[5] These results obtained despite the fact that seizure activity was as violent in the cerebellum as in the cerebrum as judged from the EEG recordings.

Discussion. The results reported above point to a quantitative if not qualitative difference in the metabolism of the cerebellum and the cerebrum under the stress of seizures. It is not surprising that this should be so since there are marked histological differences between the two parts. Furthermore, the various parts of the central nervous system

have been shown to differ in their sensitivity to cyanide,[6] their carbonic anhydrase activity,[7] their cholinesterase activity,[8] and many other particulars.

Summary. Lactic acid, pyruvic acid, inorganic phosphorus, phosphocreatine, and ATP-ADP were determined in liquid air fixed brains of normal and convulsed cats. In normal brains the levels in cerebellum and cerebrum were approximately the same. In convulsed brains, however, the lactic acid increases were much smaller and phosphocreatine levels were higher in cerebellum than in cerebrum despite the fact that seizure activity was approximately of equal intensity in both parts.

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17236. Lactate, Pyruvate, and Acid-Soluble Phosphates in Monkey Brains Treated with Carbon Dioxide and Electric Shock.*

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There now exists in the literature a considerable body of data regarding the effect of seizures upon the lactate, pyruvate, and acid-soluble phosphate levels in the brains of dogs,[1] rats,[2] and cats[3,4,5] under various conditions. It seemed, therefore, desirable to extend these analyses to animals higher in the phylogenetic scale, including man. As a step toward this end, a limited series of monkeys has been studied in the normal state, treated with carbon dioxide,[4] electric shock, and electric shock plus carbon dioxide.[6]

Methods and Results. Treatment of the animals, brain wave recordings, and analysis of the liquid air fixed brains were carried out as described in previous reports from this laboratory.[3-6] The results are summarized in Table I. Comparison with the data on other animals shows that the levels of all constituents assayed are of the same magnitude in monkeys as in dogs, cats, and rats and the changes brought about by seizures and carbon dioxide inhalation are in the expected direction.

Discussion. The results reported show that it is probably justifiable to extend conclusions drawn from data on experimental animals to man, as far as the brain constituents here assayed are concerned, since it seems unlikely that there would be a sharp change between man and monkey when monkey does not differ from other mammals.

Another point which is clearly demonstrated by a comparison of animals 3 and 4 is that the rise in lactate, pyruvate, and inorganic phosphate elicited by seizures is the result of the hyperactivity of the brain and not of the stimulus which initiates said hyperactivity. There is an indication in the data that the directly driven response to the stimulus alone, without seizures, may cause a fall in phosphocreatine (animal 4) and possible adenosine triphosphate; but in the case of the latter

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