LXX. STUDIES IN CARBOHYDRATE METABOLISM.

VI. THE ANTAGONISTIC ACTION OF PITUITRIN AND ADRENALINE UPON CARBOHYDRATE METABOLISM WITH SPECIAL REFERENCE TO THE GASEOUS EXCHANGE, THE INORGANIC BLOOD-PHOSPHATE AND THE BLOOD-SUGAR.

BY CHARLES GEORGE LAMBIE AND FRANCES AGNES REDHEAD.

From the Clinical Research Laboratory, Royal Infirmary, and the Research Laboratory of the Royal College of Physicians, Edinburgh.

(Received June 17th, 1929.)

In recent years a considerable amount of attention has been directed to the question of the physiological antagonism between pituitrin and insulin, particularly in relationship to carbohydrate metabolism.

With the discovery of insulin opportunities were afforded for studying more directly the interaction of these hormones, and interest in the subject was stimulated afresh by the observations of Burn [1923], who demonstrated that pituitrin could inhibit the fall in blood-sugar following administration of insulin and bring about recovery from insulin hypoglycaemia in rabbits. These results were confirmed by numerous observers [Olmstead and Logan, 1923; Moehlig and Ainslee, 1925; Lawrence and Hewlett, 1925; Voegtlin, Thompson and Dunn, 1925; Geiling and Britton, 1927]. The suggestion put forward by Burn that these findings might be explained by a direct and specific antagonism between pituitrin and insulin has resulted in attempts being made by other workers to look for evidence of this antagonism in other directions. Thus Joachimoglu and Metz [1924] have employed the isolated uterus, but the evidence of antagonism was not conclusive. Serebrijski and Vollmer [1925] and Koref and Mautner [1926] found that when insulin was injected simultaneously with pituitrin it did not inhibit the antidiuretic effect of the latter, but during insulin hypoglycaemia the antidiuretic action was abolished. Given separately both insulin and pituitrin were antidiuretic, insulin causing water retention in the tissues, and pituitrin acting directly upon the kidneys. The peculiar and variable effect of insulin upon blood concentration, producing hydraemia or anhydraemia according to the conditions, is a factor which will have to be taken into consideration in

interpreting these results [Drabkin and Edwards, 1924; Levine and Kolars, 1924-1925; Villa, 1924; Olmstead and Logan, 1923; Haldane, Kay and Smith, 1924; Olmstead and Taylor, 1924].

Coope and Chamberlain have shown that pituitrin and insulin are antagonistic as regards certain changes in fat metabolism [Coope, 1925; Coope and Chamberlain, 1925].

The facts so far accumulated show that certain metabolic processes are influenced in opposite directions by insulin and pituitrin, and to this extent they may be said to be mutually antagonistic, but conclusive evidence of a direct inactivation of one hormone by the other has not been obtained. The changes which occur in fat metabolism and water metabolism under insulin may merely be secondary results of the action of insulin upon the combustion and storage of carbohydrate. With regard to carbohydrate metabolism, it may be noted that investigations regarding the modification of insulin action by pituitrin have been almost entirely confined to observations upon the blood-sugar or upon symptoms dependent upon changes in blood-sugar concentration, and the same question arises as to whether the fact that the blood-sugar concentration is altered in the opposite direction by insulin and pituitrin is to be explained by direct inactivation or by the result of some secondary effects of the hormone. In this connection it may be mentioned that Lambie [1926] and Clark [1928] have found that in the decerebrate cat, with the liver excluded from the circulation and constant transfusion of dextrose, pituitrin is unable to prevent the fall in blood-sugar caused by insulin, and this would suggest that, in the intact animal, either the liver is necessary to effect the mutual inactivation of insulin and pituitrin, or it is merely the source of dextrose yielded up under the influence of pituitrin to maintain the blood-sugar at or above the normal level in experiments in which the organ is intact. In so far, however, as changes in the concentration of the blood-sugar are not necessarily a measure of utilisation of sugar in the tissues, it cannot be inferred with certainty that even with the liver excluded from the circulation pituitrin is without influence upon carbohydrate metabolism.

In order, if possible, to throw light upon the effect of pituitrin on the metabolism of carbohydrate, it was thought desirable to study the respiratory metabolism and the inorganic blood-phosphate after administration of carbohydrate or insulin together with pituitrin, and the experiments described below were undertaken with that object.

If pituitrin inactivates insulin, changes of a similar character to those met with in diabetes might be looked for.

Hines, Leese and Boyd [1927], in experiments upon dogs with continuous transfusion of dextrose and pituitrin, came to the conclusion that while pituitrin caused less sugar to be retained than in controls, it was without effect upon the respiratory quotient and heat production: but the conditions of these experiments may not have been favourable for obtaining a positive result.

In view of the alleged capacity of pituitrin to annul the action of insulin upon carbohydrate metabolism, it was thought that an opportunity might also be afforded to study the metabolism of dihydroxyacetone under these conditions and thus gain further information upon the question as to whether this triose can be utilised even when the metabolism of dextrose is inhibited.

Since adrenaline is another hormone which, like pituitrin, has an action opposite to that of insulin, in so far as it can raise the blood-sugar and cause recovery from insulin hypoglycaemia, it was deemed of interest to carry out some experiments in order to determine how the changes in the respiratory metabolism compared with those obtained with pituitrin.

EXPERIMENTAL.

A. RESPIRATORY EXCHANGE, BLOOD SUGAR, AND INORGANIC BLOOD-PHOSPHATE IN THE HUMAN SUBJECT.

As in previous experiments [Lambie and Redhead, 1927] the subject was in the post-absorptive condition. The expired air was collected in 6-minute samples, using the Douglas bag method, and was analysed by means of the Haldane apparatus. The basal metabolic rate and fasting blood-sugar were first determined, after which the substance to be investigated was administered, and subsequently samples of expired air and blood were taken at intervals. The blood-sugar was estimated by the Hagedorn-Jensen method. When intravenous administration was employed, the sugar (20 g.) was dissolved in 100 cc. of saline and sterilised by filtering through a Seitz germicide (EK) filter No. 6 using pads impermeable to organisms. The injection was made at a uniform rate and occupied a period of exactly 7 minutes.

Exp. 1 (a). Respiratory metabolism and blood-sugar curve in normal student after intravenous injection of 20 g. of dextrose.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.
0	+ 7	1.62	0.99	57.25	0.086
1-7	Dextrose intravenously				
1-3	+28	2.00	0.87	52.0	—
3-6	+ 6	1.72	0.73	45.0	—
10	+ 4	1.63	0.85	41.25	0.133
20	+11	1.73	0.89	44.0	0.108
30	+17	1.86	0.81	45.0	0.100
40	+ 8	1.72	0.82	44.75	0.087

Exp. 1 (b). Same as preceding + 0.75 cc. of pituitrin* administered subcutaneously while dextrose transfusion was proceeding. (Dextrose dissolved in 200 cc. saline.)

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.
0	+ 3	1.56	0.88	76	0.079
1-7	Dextrose intravenously + pituitrin subcutaneously				
10	+17	1.81	0.79	71	0.166
25	+ 4	1.56	0.91	71	0.146
40	+ 7	1.62	0.90	70.71	0.139
50	+18	1.80	0.87	77.65	0.070
80	+15	1.72	0.95	77.5	0.097
120	+17	1.87	0.67	71.5	0.106

* The pituitrin used was that manufactured by Parke Davis and Co.

Exp. 2 (a). Respiratory metabolism in diabetic patient on low diet, after intravenous injection of 20 g. of dextrose.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 9	1.27	0.72	36.66
1-7	Dextrose intravenously			
10	- 6	1.30	0.84	42.0
20	- 4	1.27	1.00	52.5
30	- 7	1.14	0.82	42.5
40	-24	1.09	0.72	37.0
50	-20	1.11	0.72	42.0

Exp. 2 (b). Same as preceding + 1 cc. pituitrin administered subcutaneously at end of dextrose injection.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	+ 4	1.49	0.67	38.5
1-7	Dextrose intravenously + pituitrin subcutaneously			
10	+ 6	1.53	0.64	38.0
20	+ 7	1.56	0.65	38.5
30	+ 7	1.6	0.65	39.0
40	-16	1.18	0.75	37.0
50	-11	1.25	0.74	38.0

Exp. 3 (a). Respiratory metabolism in normal male after intravenous injection of 20 g. of dihydroxyacetone.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 7	1.59	0.83	29.35
1-7	Dihydroxyacetone intravenously			
1-3	+15	1.94	0.87	42.0
3-6	+50	2.51	0.92	61.5
10	+49	2.54	0.86	57.0
20	+36	2.35	0.81	51.0
30	+12	1.91	0.84	47.0
40	+ 7	1.85	0.80	46.5
60	+ 3	1.80	0.76	40.25

Exp. 3 (b). Same as preceding + 1 cc. pituitrin.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	+ 5	1.84	0.77	40.2
1-7	Dihydroxyacetone intravenously + pituitrin subcutaneously			
1-3	+10	1.91	0.78	40.5
3-6	+32	2.25	0.86	50.4
10	+17	1.97	0.89	52.25
20	+15	1.99	0.81	46.5
30	+35	2.40	0.71	48.0
40	+16	1.98	0.84	54.5
70	+19	2.01	0.89	52.0
100	+ 8	1.88	0.76	38.75

Exp. 4 (a). Respiratory metabolism, blood-sugar and inorganic blood-phosphate in normal male after injection of 15 units of insulin subcutaneously.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	-10	1.34	0.86	38.5	0.100	3.428
Insulin 15 units subcutaneously						
15	-18	1.20	0.92	37.0	0.095	3.387
30	- 2	1.45	0.87	43.0	0.085	3.170
45	-11	1.40	0.85	39.25	0.079	2.928
60	- 9	1.35	0.88	38.5	0.079	2.991

Exp. 4 (b). Respiratory metabolism in normal male after subcutaneous injection of 1 cc. of pituitrin.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	- 20	1.20	0.72	36	0.099	3.377
Pituitrin subcutaneously						
10	- 12	1.29	0.94	44	0.107	3.172
20	- 10	1.34	0.85	42	0.112	2.906
40	- 13	1.32	0.78	39.5	0.113	2.777
50	- 15	1.31	0.79	37	0.107	2.777

Exp. 5 (a). Respiratory metabolism in normal male after subcutaneous injection of 10 units of insulin.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 3	1.24	0.95	37.2
Insulin subcutaneously				
10	+ 31	1.76	0.73	38.2
20	+ 20	1.62	0.72	38.2
30	+ 19	1.58	0.80	36.0
40	+ 17	1.58	0.72	34.7
50	+ 7	1.36	0.78	30.5
80	+ 124	2.41	0.71	56.9*

* Symptoms of hypoglycaemia.

Exp. 5 (b). Same subject as preceding (5 a). Respiratory metabolism after subcutaneous injection of 1 cc. of pituitrin.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 10	1.21	0.71	25.1
Pituitrin subcutaneously (injection painful)				
10	- 6	1.27	0.72	26.1
20	- 19	1.06	0.81	25.0
30	- 19	1.09	0.74	25.1
40	- 12	1.17	0.77	25.2
70	- 13	1.16	0.76	26.7

Exp. 5 (c). Same subject as in two preceding experiments (5 a and 5 b). Respiratory metabolism after simultaneous injection of insulin (10 units) and pituitrin (1 cc.) subcutaneously.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 2	1.33	0.72	38.5
Insulin and pituitrin subcutaneously				
10	- 5	1.28	0.74	38.5
20	- 2	1.35	0.70	39.5
30	+ 17	1.58	0.71	40.0
40	+ 1	1.31	0.89	42.5
70	- 9	1.21	0.77	37.8

Exp. 6 (a). Respiratory metabolism in normal male after subcutaneous injection of 15 units of insulin.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	- 3	1.41	0.73	34.75	0.104	2.906
Insulin 15 units subcutaneously						
10	- 6	1.36	0.72	31.72	0.104	2.777
20	+ 5	1.51	0.74	36.5	0.074	2.765
30	- 2	1.41	0.77	37.6	0.066	2.687
40	- 1	1.42	0.73	38.5	—	2.705

Exp. 6 (b). Same subject as preceding (6 a). Respiratory metabolism in normal male after 1 cc. pituitrin subcutaneously.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	-3	1.32	0.81	34.5
Pituitrin subcutaneously				
15	-5	1.27	0.77	35.0
25	+5	1.386	0.86	36.0
35	+1	1.380	0.73	34.5

Exp. 6 (c). Same subject as preceding experiments (6 a and 6 b). Respiratory metabolism, blood-sugar and inorganic blood-phosphate in normal male after subcutaneous injection of insulin, 15 units, followed 7 mins. later by pituitrin 1 cc.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	+6	1.61	0.74	40.75	0.111	2.928
Insulin subcutaneously						
6	+11	1.7	0.79	42.25	0.111	2.875
7	Pituitrin subcutaneously					
22	+4	1.5	0.96	46.25	0.100	2.823
30	+11	1.66	0.85	45.5	0.104	2.703
50	-14	1.25	1.1	43.5	0.111	—
65	-18	1.42	1.0	40.75	0.111	2.652

Exp. 6 (d). Same as preceding but pituitrin given 25 mins. after insulin. Respiratory metabolism.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	-16	1.19	0.81	29.5
Insulin subcutaneously				
10	-14	1.26	0.78	28.5
Pituitrin subcutaneously				
35	-13	1.21	0.86	33
45	-5	1.32	0.86	39.5
55	-9	1.27	0.85	34
65	-11	1.25	0.84	32

Exp. 6 (e). Respiratory metabolism, blood-sugar and inorganic blood-phosphate in normal male (different subject from preceding) after insulin, 15 units, followed 62 mins. later by pituitrin 1 cc.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	+11	1.76	0.76	31	0.100	3.515
Insulin subcutaneously						
30	+30	2.06	0.78	37.5	0.082	2.928
60	+24	1.17	0.76	36	0.076	2.510
Pituitrin subcutaneously						
64	+30	2.01	0.86	42.35	0.076	2.130
74	+32	2.07	0.80	43.25	0.079	2.162
84	+18	1.85	0.81	36.2	0.082	2.381
94	+26	2.01	0.75	37.52	0.086	2.343
104	+11	1.75	0.77	33.75	0.086	2.421

Exp. 7 (a). Respiratory metabolism, blood-sugar and inorganic blood-phosphate in normal subject after subcutaneous injection of 0.75 cc. of adrenaline (1 in 1000).

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	+ 8	1.99	0.81	40.5	0.091	3.652
Adrenaline subcutaneously						
10	+35	2.41	0.94	57.0	0.117	3.255
20	+40	2.59	0.80	54.25	0.133	3.181
30	+45	2.69	0.78	53.0	0.133	3.232
40	- 7	1.69	0.93	41.25	0.25	3.308

Exp. 7 (b). Respiratory metabolism in normal male after 0.75 cc. of adrenaline (1 in 1000) subcutaneously.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 13	1.39	0.76	42
Adrenaline subcutaneously				
10	+ 8	1.62	0.84	53.7
20	+ 8	1.6	0.82	50.25
30	- 5	1.5	0.76	47
40	- 8	1.34	0.86	49
50	- 9	1.39	0.75	43

Exp. 7 (c). Same as 7 (b). Dextrose 20 g. injected intravenously and adrenaline 0.75 cc. subcutaneously.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.
0	- 27	1.3	0.72	38
Dextrose intravenously; adrenaline subcutaneously				
10	+42	2.15	0.82	72
20	+ 7	1.63	0.78	54
30	- 4	1.46	0.80	48
40	-18	1.25	0.76	38
50	-16	1.28	0.78	40

Exp. 8. Respiratory metabolism, blood-sugar and inorganic blood-phosphate in a patient suffering from exophthalmic goitre, after oral administration of 50 g. of dextrose.

Time mins.	Metabolic rate % of normal B.M.R.	Oxygen consumption l. per 6 mins.	R.Q.	Expired air l. per 6 mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	+ 52	1.58	0.85	46.75	0.075	3.947
Dextrose orally						
30	+102	2.17	0.81	51.25	0.145	3.448
60	+ 48	1.88	0.71	58.0	0.205	3.355
90	+ 52	1.86	0.85	54.5	0.162	3.488
120	+ 37	1.71	0.78	57.5	—	—

B. ANIMAL EXPERIMENTS.

Exp. 1. Rabbit 2340 g. Effect of pituitrin alone on blood-sugar and inorganic blood-phosphate.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.124	3.472
Pituitrin 0.5 cc. intravenously		
15	0.159	3.874
35	0.124	3.204

Exp. 2 (a). Rabbit 2950 g. Effect of dihydroxyacetone alone.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.145	4.166
Dihydroxyacetone 2.5 g. intravenously		
15	0.288	3.424
30	0.247	3.100
45	0.236	2.976

Exp. 2 (b). Same animal as *Exp. 2 (a)*. Effect of pituitrin and dihydroxyacetone together.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.111	2.702
Pituitrin 0.5 cc. + dihydroxyacetone 2.5 g. intravenously.		
15	0.255	3.120
30	0.238	3.030
45	0.217	2.630

Exp. 3 (a). Rabbit 2500 g. Effect of dextrose alone.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.097	3.900
Dextrose 2.5 g. intravenously		
15	0.186	3.676
30	0.115	3.472
45	0.084	3.378

Exp. 3 (b). Same animal. Effect of dextrose and pituitrin together.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.132	3.570
Pituitrin 0.5 cc. + dextrose 2.5 g. intravenously		
15	0.302	3.56
30	0.164	3.444

Exp. 4 (a). Rabbit 1390 g. Effect of dihydroxyacetone and pituitrin together, using smaller dose of pituitrin than in preceding experiments

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.113	3.387
Pituitrin 0.1 cc. + dihydroxyacetone 1.39 g. intravenously		
20	0.294	2.876
45	0.168	2.763

Exp. 4 (b). Same animal as *Exp. 3 (a)*. Effect of dextrose and pituitrin together in similar doses.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.120	3.402
Pituitrin 0.1 cc. + dextrose 1.39 g. intravenously		
20	0.175	3.735
45	0.121	3.206

Exp. 5 (a). Rabbit 3000 g. Effect of dihydroxyacetone and pituitrin together.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.125	3.786
Pituitrin 0.1 cc. + dihydroxyacetone 2.5 g. intravenously		
15	0.264	3.204
30	0.187	3.110

Exp. 5 (b). Same animal. Effect of dextrose and pituitrin together in similar doses.

Time mins.	Blood-sugar g. per 100 cc.	Inorganic blood-phosphate mg. per 100 cc.
0	0.115	3.2110
Pituitrin 0.1 cc. + dextrose 2.5 g. intravenously		
15	0.285	3.7668
30	0.135	3.1865

DISCUSSION.

The above experiments appear to show that pituitrin has a marked inhibitory action upon the metabolism of carbohydrates. Thus Exp. 1 (*a*) shows the typical changes which occur in the respiratory metabolism and blood-sugar in the healthy individual following the administration of 20 g. of dextrose intravenously. There is first a gradual rise in the heat production and oxygen consumption, the R.Q. and the respiratory volume, maximal values being attained in approximately 20–30 minutes. (The high initial R.Q. in this experiment may have been due to overbreathing.) The blood-sugar following the injection falls rapidly, reaching the base line in 30–40 minutes. When the same amount of dextrose was administered to this individual along with pituitrin, injected subcutaneously, there was little if any rise in the heat production, oxygen consumption, R.Q. or respiratory volume, by the time these would have been at their height in the control. In fact, save for an initial increase in the metabolic rate and oxygen consumption, for which changes in blood flow resulting from the injection of fluid may have been responsible, there was remarkably little alteration in the metabolism for the first 30 or 40 minutes. After this, however, coinciding with the passing off of the vaso-constriction, as evidenced by diminution in the pallor of the skin, there was a rise in all these values with the exception of the R.Q. which fell a little when the heat production and oxygen consumption were at a maximum. The R.Q. rose to its highest at a later period. It is interesting to note that the blood-sugar fell very slowly during the period when the respiratory metabolism showed little change, being still well above the base line at the end of 40 minutes. After this, however, it fell while the metabolism rose.

In Exp. 2, which was performed upon a diabetic patient on a low diet, the changes are less striking but are of a similar character. The R.Q., which was remarkably low, here attained its maximum after all the other values had fallen.

The effect of pituitrin upon the changes in respiratory metabolism which follow the intravenous administration of dihydroxyacetone is equally striking. In spite of the administration of a dose of pituitrin sufficient almost completely to annul the rise in metabolism due to dextrose during the first 20 minutes of the experiment, there was still a considerable rise in the metabolism during this period; in fact the actual rise was even greater than that obtained with dextrose alone. Nevertheless, the rise in metabolism was not nearly as great as with dihydroxyacetone alone, the percentage inhibition due to pituitrin being just as great as in the case of dextrose. In addition to the inhibition of the metabolism, however, the form of the curves for heat production and oxygen consumption were entirely altered, resembling in certain respects those obtained with dextrose alone. Thus, whereas dihydroxyacetone alone caused an immediate rise in metabolism with maximal values

soon after the completion of the injection, followed by a rapid fall to almost normal at the end of 30–40 minutes, pituitrin given along with dihydroxy-acetone inhibited the initial acceleration in metabolism (see 10 minutes' sample), there being instead a gradual rise reaching a maximum in 30 minutes. The R.Q., however, varied inversely with the oxygen consumption and heat production during this period. A slight secondary rise in the metabolic rate, oxygen consumption, R.Q. and respiratory volume was observed at the end of 70 minutes.

In the experiments in which insulin and pituitrin were injected together, the results were not nearly so clear cut as when pituitrin was given along with carbohydrate, owing to the changes in respiratory metabolism produced by either hormone during the first 30–40 minutes after their administration being so slight as to be of doubtful significance. McKinlay [1921], Bowman and Grabfield [1926] and Castex and Schteingart [1926] report a rise in metabolism of about 10 % occurring 20–30 minutes after the administration of pituitrin alone; but our results are not so uniform. Thus, in Exp. 4 (*b*) there was a slight rise in the heat production, oxygen consumption and R.Q.; in Exp. 6 (*b*) there was a preliminary fall during the first 15 minutes, followed by a rise, while in Exp. 5 (*b*) there was a more prolonged fall in the heat production and oxygen consumption, but not in the R.Q. The slight rise in metabolism obtained in the 10 minutes' sample in this experiment may have been occasioned by the pain of the injection.

These irregular results may be due to pituitrin on the one hand stimulating intestinal peristalsis, which would tend to increase the oxygen consumption, and on the other hand depressing the metabolism in other tissues. The effect upon the respiratory metabolism would depend upon the time relationship and relative potency of these two factors. In both Exps. 5 (*c*) and 6, (*c*) in which insulin and pituitrin were injected either simultaneously or within a comparatively short interval of one another, pituitrin seemed to inhibit the rise in metabolism or cause a fall at the time of the corresponding observations in the control with insulin alone. Since, however, pituitrin by itself caused a fall in metabolism at these times in this particular series of experiments, the effect of the administration of the two hormones together was apparently such as might have resulted from a combination of their independent actions.

When pituitrin was injected at a longer interval (half an hour to an hour) after insulin (Exps. 6 (*d*) and 6 (*e*)) a slight rise in the heat production and R.Q. was observed. This may possibly be connected with the liberation of glucose from the liver or with the contractions of the stomach and intestine which are more readily excited in hypoglycaemia. (Hunger pains are common during hypoglycaemia.)

Our findings regarding the effect of pituitrin upon the blood-sugar are in agreement with those of other observers, namely, a delay in the disappearance of injected dextrose from the blood [Hines, Leese and Boyd, 1927; Achard, Ribot and Binet, 1919], inhibition of hypoglycaemia or a return of the blood-

sugar towards the normal level in hypoglycaemic animals after insulin [cf. Burn, 1923; Olmstead and Logan, 1923; Moehlig and Ainslee, 1925; Lawrence and Hewlett, 1925; Geiling and Britton, 1927], while injection of pituitrin alone caused a transient but inconstant rise in the blood-sugar during the first 30 minutes after its administration [cf. Borchardt, 1908; Goetsch, Cushing and Jackson, 1911; Parton and Katz-Klein, 1921; Lindlau, 1928; Blotner and Fitz, 1928].

With regard to phosphate metabolism, pituitrin alone in the doses employed in the human subject caused a slight fall in the percentage of inorganic phosphate in the blood, but given in relatively large doses to rabbits, it caused a marked but transient rise. When administered to either human beings or rabbits with hypoglycaemia and a low inorganic blood-phosphate resulting from insulin administration, it caused the percentage of inorganic blood-phosphate to rise. Similar results were obtained with adrenaline as with pituitrin.

The results so far reviewed, taken as a whole, might at first sight seem to bear out the contention that pituitrin inactivates insulin, thereby bringing about, temporarily, a state of affairs analogous to that obtaining in diabetes. Hyperglycaemia, delayed disappearance from the blood of injected carbohydrate, diminished response in respiratory metabolism following administration of carbohydrate and an increased percentage in inorganic blood-phosphate, all of which may follow the injection of suitable doses of pituitrin, are also features characteristic of the diabetic condition. Taken in conjunction with the observation that pituitrin seems to inhibit any slight rise in metabolism following administration of insulin, the evidence in support of a direct antagonism would at first sight appear to be strong and to have the advantage of economy of hypothesis. Nevertheless, in certain respects this explanation does not appear to be wholly satisfactory. Thus, pituitrin, except in the large doses employed in rabbits, did not, when given alone, cause any rise in the inorganic blood-phosphate, but rather a slight fall; moreover, in spite of being given in doses sufficient completely to prevent the fall in blood-sugar following insulin administration in the human subject, it did not prevent the inorganic blood-phosphate from falling (Exp. 6 (c)), and, although the fall may have been temporarily checked, the blood-sugar remained at the normal level while the level of inorganic blood-phosphate continued to decline.

Again, pituitrin caused no definite fall in the respiratory quotient, but more often a rise.

Further, in the experiment with dihydroxyacetone and pituitrin (Exp. 3 (b)), the curves of metabolism fail to reproduce the features of those obtained in diabetes. In the latter [cf. Lambie and Redhead, 1927] the heat production, oxygen consumption and R.Q. attained their maxima together soon after the injection, whereas in the experiment referred to the heat production and oxygen consumption gradually rose, reaching their height at the end of 30 minutes, while the respiratory quotient was at its highest in 10 minutes and declined *pari passu* with the rise in the metabolic rate.

In view of these differences between the effects of deficiency of insulin and the alleged inactivation of insulin by pituitrin, it is necessary to consider other possible explanations of the phenomena. Krogh [1922] has shown that pituitrin not only constricts the arterioles, but markedly increases the contractile tonus of capillaries, the effect of which would be to diminish the capillary bed. This would lead to a decreased rate of diffusion of injected material out of the blood into the tissues in which it was to be metabolised and in this way it may be possible to explain the delay in the rise in metabolism with dihydroxyacetone or dextrose, and the delay in the disappearance of sugar from the blood after intravenous injection. The occasional sudden rise in metabolism during and immediately after intravenous injection may have been the result of a temporary increase of blood flow due to the volume of fluid injected, especially since the fluid was hypertonic. The experiments of Hines, Leese and Boyd [1927] did not show any definite change in the heat production or R.Q. as a result of injecting pituitrin during continuous transfusion of dextrose, but this may have been due partly to the very large quantity of dextrose injected and to the fact that there was a continuous transfusion of fluid into the circulation. Their results are therefore not inconsistent with our own, but are due to the different conditions of the experiments.

The depression in metabolism sometimes observed following injection of pituitrin alone may again be due to diminished blood flow through the muscles, while the subsequent rise may perhaps be related to the contraction of non-striated muscle, particularly that of the intestine (borborygmi being frequently audible) and to increased work of the heart.

The rise in inorganic blood-phosphate noted after injection of large doses of pituitrin might also be explainable as a result of local asphyxia in the muscle cells, while increase in the blood-sugar may perhaps be related to changes in the portal circulation [cf. Clark, 1928].

It appears reasonable to expect that drugs, which, like pituitrin and adrenaline, alter the blood flow through organs, particularly those which, like muscle, are important seats of heat production, should be capable of modifying metabolism, and, if at the same time they cause change in the tonus of muscle or the work of the heart, further complications are to be anticipated. The metabolic changes which result from the administration of adrenaline should therefore be considered in this light also, but further experiment is necessary before definite conclusions can be drawn. Krogh [1922] points out that adrenaline differs from pituitrin in having a more powerful action on the arterioles than upon the capillaries, but all arterioles in the body are not equally affected. Hoskins, Gunning and Berry [1916] and Gunning [1917] have brought forward evidence to show that adrenaline in small doses causes blood to be diverted from the splanchnic area and skin while the blood flow through the voluntary muscles and myocardium is increased. This, together with the increased work of the heart, may be responsible for the

marked and sudden rise in metabolism following the administration of adrenaline alone, but it remains an open question as to how the metabolic processes in and the blood flow through the muscles are causally related.

Our experiments do not support the view that the rise in metabolism is to be ascribed to the hyperglycaemia. In the first place, the rise in metabolism following injection of adrenaline is very great even with a slight hyperglycaemia, whereas with a much greater hyperglycaemia produced by intravenous injection of dextrose there was less increase in the metabolism [cf. Boothby and Sandiford, 1921]. The time relationship between hyperglycaemia and the change in respiratory metabolism is also quite different in the two cases. Moreover, the rise in metabolism which occurs after injecting adrenaline is much more rapid than after intravenous injection of dextrose. Thus, 10 minutes after administration of adrenaline in Exps. 7 (*a*) and 7 (*b*) there was a 20 % rise in the metabolism at a time when the blood-sugar had risen (cf. Exp. 7 (*a*)) only slightly above the fasting level, whereas 10 minutes after injection of 20 g. of dextrose intravenously (Exp. 1 (*a*)) there was practically no increase in respiratory metabolism although there was a definite hyperglycaemia. It may be suggested as a possible explanation of these results that the dextrose liberated by the liver is more immediately available for combustion than ordinary dextrose, but apart from the lack of evidence to support this view, there is the observation that in Exp. 7 (*a*) with adrenaline the blood-sugar continued to rise even when the metabolic rate was falling. There appears, therefore, to be no high degree of correlation between hyperglycaemia and heightened metabolism after administration of adrenaline.

As to the method by which adrenaline causes hyperglycaemia, stimulation of the endings of the splanchnic nerves in the liver, changes in the portal circulation [cf. Clark, 1928], and perhaps the breaking down of muscle-glycogen, may all play a part; but there is no need to assume a direct antagonism to insulin.

The influence of adrenaline upon phosphorus metabolism has been investigated by Allan, Dickson and Markowitz [1924] and the results obtained were similar to those produced by insulin, indicating that, as in the case of pituitrin, there was no antagonism between the two in this respect.

The effect of simultaneous injection of adrenaline and insulin upon the respiratory metabolism is such as would be expected to result from the combined effect of each separately. Thus, in Exp. 7 (*b*), the percentage rise in calorie consumption 20 and 30 minutes after adrenaline alone was 21 % and 8 % respectively. In most experiments with 20 g. of dextrose alone, intravenously, the percentage rise at these periods is 5 % and 10 % respectively. The combined effects of dextrose and adrenaline would therefore give + 26 % and + 18 %. In Exp. 7 (*c*) with both adrenaline and dextrose together the actual figures obtained were + 34 % and + 23 %; there is, therefore, no indication of antagonism here.

As in the case of adrenaline, no evidence could be obtained of any

inhibitory effect of excessive thyroid activity upon the metabolism of dextrose, but rather the reverse. Thus, in Exp. 8 the metabolic rate rose to over 100 % 30 minutes after the administration of 50 g. of dextrose by the mouth to a patient with exophthalmic goitre.

The facts so far brought forward, therefore, afford no definite proof of there being a direct inactivation of one hormone by the other, and, until the changes in the circulation can be excluded as the cause of the apparent antagonism, it appears unnecessary to assume that any more direct and specific action is involved. It is true that in certain forms of pituitary disease (acromegaly) there is often diminished sugar tolerance, or even a condition indistinguishable from true diabetes, together with marked resistance to insulin [Ulrich, 1928], but no obvious disturbance of the circulation is present in such cases. The assumption that the low carbohydrate tolerance is due to excessive activity of the posterior lobe of the pituitary body is, however, unwarranted, since the glycosuria occurs in connection with a definite type of tumour of the anterior lobe, namely, the eosinophil adenoma, and not with other tumours, although the latter are more numerous [cf. Davidoff and Cushing, 1927]. Although nothing is at present known regarding the effect of repeated small doses of pituitrin administered over a long period, the fact that the apparent inhibition of carbohydrate metabolism has so far only been obtained with doses sufficiently large to cause obvious circulatory disturbances (*e.g.* blanching in man), makes the resemblance to the clinical pituitary glycosuria incomplete. Since it has been impossible to settle the question of the nature of the antagonism between pituitrin and insulin the significance to be attached to the peculiar behaviour of dihydroxyacetone in the above experiment must likewise remain an open question. The fact that it caused a greater rise in metabolism than dextrose, and brought about a fall in inorganic blood-phosphate in rabbits when dextrose failed to do so after the administration of pituitrin, is merely in conformity with previous findings [Lambie and Redhead, 1927].

SUMMARY.

1. In the human subject pituitrin frequently causes a transient fall in metabolism followed by a rise. Sometimes only a rise is observed. In small doses it causes a slight fall in the inorganic phosphate of the blood.

2. Pituitrin apparently inhibits the slight rise in metabolism which occurs after the administration of insulin; but the changes produced by either insulin or pituitrin are not sufficiently marked to be of much significance.

3. Pituitrin delays and reduces the rise in metabolism and the fall in blood-sugar following the intravenous administration of dextrose or dihydroxyacetone.

4. Pituitrin in small doses sufficient to prevent the fall in blood-sugar resulting from administration of 15 units of insulin did not prevent the fall in inorganic blood-phosphate.

5. Pituitrin administered in large doses to rabbits causes a transitory rise in the inorganic blood-phosphate and for a short period inhibits the fall in inorganic phosphate resulting from administration of dextrose or dihydroxyacetone. A dose of pituitrin which just suffices to prevent the fall in inorganic phosphate following administration of dextrose may fail to overcome the fall due to dihydroxyacetone.

6. In animals rendered hypoglycaemic as a result of insulin administration pituitrin causes the blood-sugar and inorganic blood-phosphate to return to the normal level or even to exceed it.

7. Adrenaline administered alone causes a marked and rapid rise in metabolism which is not solely dependent upon the rise in blood-sugar.

8. Adrenaline does not inhibit the rise in metabolism following intravenous administration of dextrose.

9. Adrenaline administered in large doses to hypoglycaemic animals causes a rise in inorganic blood-phosphate.

10. The question is discussed as to how far the effects of pituitrin and adrenaline upon metabolism result from the changes in the circulation and distribution of blood in the body or whether some more direct and specific antagonism to insulin is to be assumed. It is concluded that experiments so far recorded have not proved unequivocally that such a direct action exists.

REFERENCES.

- Achard, Ribot and Binet (1919). *Compt. Rend. Soc. Biol.* **82**, 788.
 Allan, Dickson and Markowitz (1924). *Amer. J. Physiol.* **70**, 333.
 Blotner and Fitz (1928). *J. Clin. Invest.* **5**, 51.
 Boothby and Sandiford (1921). *Amer. J. Physiol.* **55**, 293.
 Borchardt (1908). *Deutsch. med. Woch.* **34**, 946.
 Bowman and Grabfield (1926). *Endocrinology*, **10**, 201.
 Burn (1923). *J. Physiol.* **57**, 318.
 Castex and Scheingart (1926). *Compt. Rend. Soc. Biol.* **95**, 1512.
 Clark (1928). *J. Physiol.* **64**, 324; **66**, 274.
 Coope (1925). *J. Physiol.* **60**, 92.
 — and Chamberlain (1925). *J. Physiol.* **60**, 69.
 Davidoff and Cushing (1927). *Arch. Int. Med.* **39**, 751.
 Drabkin and Edwards (1924). *Amer. J. Physiol.* **70**, 273.
 Geiling and Britton (1927). *Amer. J. Physiol.* **81**, 478.
 Goetsch, Cushing and Jackson (1911). *Bull. Johns Hopkins Hosp.* **22**, 165.
 Gunning (1917). *Amer. J. Physiol.* **43**, 395.
 Haldane, Kay and Smith (1924). *J. Physiol.* **59**, 193.
 Hines, Leese and Boyd (1927). *Amer. J. Physiol.* **81**, 27.
 Hoskins, Gunning and Berry (1916). *Amer. J. Physiol.* **41**, 513.
 Joachimoglu and Metz (1924). *Deutsch. med. Woch.* **50**, 1787.
 Koref and Mautner (1926). *Arch. exp. Path. Pharm.* **113**, 127.
 Krogh (1922). *Anatomy and physiology of capillaries*, 158, 159.
 Lambie (1926). *Brit. J. Exp. Path.* **7**, 22.
 — and Redhead (1927). *Biochem. J.* **21**, 549.
 Lawrence and Hewlett (1925). *Brit. Med. J.* **i**, 998.
 Levine and Kolars (1924–25). *Proc. Soc. Exp. Biol. Med.* **22**, 169.
 Lindlau (1928). *Z. ges. exp. Med.* **58**, 507.

- McKinlay (1921). *Arch. Int. Med.* **23**, 703.
Moehlig and Ainslee (1925). *J. Amer. Med. Assoc.* **84**, 1398.
Olmstead and Logan (1923). *Amer. J. Physiol.* **66**, 437.
—— and Taylor (1924). *Amer. J. Physiol.* **69**, 142.
Parton and Katz-Klein (1921). *Z. ges. exp. Med.* **25**, 98.
Serebrijski and Vollmer (1925). *Klin. Woch.* **4**, 2256.
Ulrich (1928). *Arch. Int. Med.* **41**, 875.
Villa (1924). *Boll. Soc. Med. Chir. Pavia*, **35**, 239.
Voegtlin, Thompson and Dunn (1925). *J. Pharm. Exp. Ther.* **25**, 137.