Is the Hyperglycaemia-Provoking Capacity of Asphyxial Blood Due to Epinephrine ?

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That asphyxial blood when introduced into another animal can give rise to glycosuria without the co-existence of hyperglycaemia has been reported by former investigators.¹⁾ The asphyxial blood of a dog or an extract of it was applied to another dog or guinea pig.

In connection with the question of the augmented epinephrine discharge by asphyxia, this matter, which had fallen into oblivion for a long time, was later studied more systematically by Yamakami,²⁾ who was able to see, in the first place, that the asphyxial blood of a rabbit can call forth hyperglycaemia in another rabbit when intravenously introduced, and, in the second place, that the surplus of sugar content in the asphyxial blood is too much small to be taken as responsible for this increase of the blood sugar concentration in the recipient, and neutralization of the asphyxial blood does not abolish this glycaemic capacity of asphyxial blood. And that epinephrine is responsible for this glycaemic property of asphyxial blood was tentatively suggested by him.

Nowadays where it has been well substantiated that asphyxia is capable of evoking an acceleration in the rate of epinephrine output from the suprarenal gland, for an account of which we may refer to a paper of Kodama,³⁾ this question which has been attacked by Yamakami, but not yet wholly settled, is seemingly more significant and interesting.

1) Lépine and Bouloud, C. R. Acad. Sc., 1902, **134**, 582; R. Lépine, Le sucre du sang, Paris 1921, 339 f.

2) K. Yamakami, Am. J. Physiol. 1919 (-20), **50**, 177.

3) Sakuji Kodama, Tohoku J. Exp. Med., 1924, **5**, 47.

The general procedures and methods applied in the present investigations are the same as those described in our previous papers. Rabbits were exclusively employed.

The cervical sympathetic and great auricular nerves of the recipient rabbit were previously cut on both sides, for collecting blood specimens for sugar estimation on one left side and introducing the blood from the donor rabbit into the marginal ear vein on the other right side, with ease and without causing pain. Bang (1920) was used for the blood sugar estimation, Bertrand for that of the urine sugar.

I.

As the control 10 to 15 c.c. of normal rabbit blood were intravenously injected into the recipient rabbit. The blood shed from a carotid artery of a normal rabbit was defibrinated by whipping. The results yielded were negative, in accordance with Yamakami. Namely the blood sugar concentration of the recipient after receiving some cubic centimeters of blood from another rabbit remained either wholly unchanged or showed only a trace of in-

TABLE I.
Control experiments.

No.	Date	Body weight (kilos)	Defibr. normal blood injected				Blood sugar				Urine sugar			Body temperature (°C)	Frequency of respiration (per min.)	Frequency of heart beat (per min.)	Room temperature (°C)
			Amount injected (c.c.)	Blood sugar (%)	Duration of fastening (hrs.)	Region of collection	Before injection (%)	Max. after injection (%)	Time to Maximum (hrs.)	Before injection (%)	Max. after injection (%)	Time to Maximum (hrs.)	Maximum				
1	17. VII. '25	2.0	13	0.17		Carotid	0.10	0.12	1	0.020	0.049	1½	39.3-41.0	84-168	192-254	21.5-25.0	
2	17. VII. '25	1.94	10	0.17		"	0.10	0.12					39.2-39.8	72-120	204-228	22.0-25.5	
3	18. IX. '25	2.16	15	0.17	1	"	0.10	0.13					38.6-39.2	48-96	204-228	18.0-20.5	
4	18. IX. '25	1.84	14	0.17	1	"	0.09	0.11		0.029	0.029		37.2-39.0	96-180	180-240	18.0-20.5	
5	22. IX. '25	2.54	15	0.12	1	Heart	0.10	0.12	1½	0.039	0.059	2½	39.1-40.3	48-132	228-312	19.5-24.0	
6	22. IX. '25	2.32	15	0.12	1	"	0.11	0.12	3½				38.2-39.8	54-84	264-276	20.0-24.5	

creasing, as shown in Table I. Also no trace of glycosuria was detected.

In passing it may be noticed that a recent investigator has reported an unmistakable glycaemic effect of transfusion of citrate blood in a dose of 16 to 7 c.c. per kilo of body weight. Rabbits were used as the recipient and the donor.⁴⁾

II.

In 11 normal rabbits, summerized in Table II the asphyxial blood from other rabbits was introduced. The donors were asphyxiated by means of applying a clamp on the trachea, operatively exposed, and really, either suddenly, so that it proved fatal within a few minutes, or gradually, so that complete suffocation occurred first at the end of one hour. Then the blood was obtained from a carotid artery or from the heart by puncture, and forthwith whipped. The manipulations were carried out aseptically. About half an hour later the defibrinated blood was injected into the marginal ear vein of the recipient. Yamakami made use of hiru-

EXAMPLE 4. (Table II.)

21. VII. 1925. Rabbit ♂. 1.84 kilos.

Time	Room temperature (°C.)	Body temperature (°C.)	Frequency of respiration (per min.)	Frequency of heart beat (per min.)	Blood sugar (%)	Urine		
						Quantity (c.c.)	Reaction	Sugar (%)
10:38 a.m.					0.11			
10:42	26.5	38.9	216	228				
10:47							neutral	0.029
11:22					0.13			
11:32	Intravenous infusion of 13 c.c. of defibrinated asphyxial blood (0.18% sugar (obtained from right heart of another rabbit.							
11:42	$\frac{1}{6}$ hour after infusion.							
12:02 p.m.	$\frac{1}{2}$	"	"	"	0.14			
12:32	1	"	"	"	0.15			
1:02					0.12			
1:17	28.5	41.2	300	228	0.14			
1:22						10	neutral	0.059
1:32					0.12			
2:32					0.10			
3:32					0.11			
5:00					0.09			
5:10	28.0	40.5	228	240				
5:12						8	acid	0.049

4) II. Takemasa, Tokyo Igakkwai Zasshi, 1926, 40, 398 f.

TABLE II.

Experiments on normal rabbits.

No.	Date	Body weight (kilos)	Defibr. asphyxial blood inj.				Blood sugar			Urine sugar			Body temperature (°C)	Frequency of respiration (per min.)	Frequency of heart beat (per min.)	Room temperature (°C)
			Amount injected (c.c.)	Blood sugar (%)	Duration of asphy- xiation (hrs.)	Region of collection	Before injection (%)	Max. after injection (%)	Time to Maximum (hrs.)	Before injection (%)	Max. after injection (%)	Time to Maximum (hrs.)				
1	9. VII. '25	1.98	12			Carotid	0.12	0.17	1	0.020	0.034	5½	39.3-40.2	48-180	216-276	20.0-21.5
2	10. VII. '25	1.72	12			"	0.10	0.16	½	0.015	0.118	6	39.0-39.9	72-216	216-276	19.0-21.0
3	11. VII. '25	1.70	15			"	0.10	0.15	1				38.9-39.3	72-180	216-228	20.0-22.0
4	21. VII. '25	1.84	13	0.18	½	Heart	0.11	0.15	½	0.029	0.059	15	38.9-41.2	216-300	228-240	26.5-28.5
5	24. VII. '25	1.78	12	0.10	½	"	0.12	0.14	2	0.020	0.029	6½	38.6-40.3	108-216	156-228	25.5-28.0
6	24. VII. '25	1.86	14	0.10	½	"	0.10	0.14	3	0.020	0.029	6½	39.5-40.5	168-204	168-204	25.5-28.0
7	24. IX. '25	2.32	15	0.23	½	Carotid	0.10	0.13	1½	0.020	0.049	3½	38.4-40.0	72-156	204-252	21.0-25.5
8	24. IX. '25	1.55	10	0.23	½	"	0.10	0.14	½				38.6-40.0	60-132	228-240	21.0-25.5
9	29. X. '25	2.00	14	0.14	1	Heart	0.09	0.11	½				39.5-40.5	48-48	228-238	13.0-17.0
10	9. VI. '26	2.52	12	0.12	½	"	0.11	0.11	1½				38.7-40.7	72-144	240-252	20.0-20.0
11	9. VI. '26	1.84	13	0.12	½	"	0.12	0.13	1½				38.6-39.6	72-120	204-228	20.0-20.0

din in order to prevent clotting.

On receiving 10 to 15 c.c. of defibrinated asphyxial blood, the sugar concentration of which was estimated as 0.10 to 0.23 % (of the bloods of Nos. 1, 2 & 3 estimation was not carried out), the blood sugar concentration of 8 recipient rabbits increased from 0.10-0.12% to 0.13-0.17% in the lapse of ten minutes to three hours. In the remaining three cases the variation was very small or nil. A surplus of 0.05-0.06% was detected in 3 instances out of our 11 rabbits, while an excess of 0.05-0.08% in 5 rabbits out of 11 was noted in the paper of Yamakami. Entirely negative results were yielded in two of his cases.

III.

The defibrinated asphyxiated blood was intravenously applied into the previously

doubly splanchnectomized rabbit in the same manner as above described. This time the donors were suffocated gradually. The results were similar to the cases of the normal recipients. An excess of blood over 0.04% was perceived in 5 cases out of 9, in the other 3 it was 0.03%, and in the rest practically no alteration in the blood sugar concentration occurred.

EXAMPLE 8. (Table III.)

13. X. 1926. Rabbit ♂. 1.78 kilos.

(25. IX. 1926. 1.79 kilos. Double splanchnectomy.)

Time	Room temperature (°C.)	Body temperature (°C.)	Frequency of respiration (per min.)	Frequency of heart beat (per min.)	Blood sugar (%)	Urine		
						Quantity (c.c.)	Reaction	Sugar (%)
9:40 a.m.	14	38.3	72	180	0.12			
9:56							neutral	0.020
10:04								
10:30	Intravenous infusion of 12 c.c. of asphyxial blood (0.22% sugar) obtained from right heart of another rabbit.							
10:35					0.15			
10:45					0.12			
10:53	15.5	37.8	96	168				
11:00					0.14			
11:20					0.13			
11:45					0.12			
11:50						10	neutral	0.059
11:55	17	38.6	72	204				
12:15 p.m.					0.12			
1:15					0.13			
1:17	17	39.1	72	180				
1:29						18	neutral	0.049

Whether or not the splanchnic nerves on the both sides have been divided apparently has no bearing upon the glycaemic effect of the asphyxiated blood.

IV.

Thus to the finding of Yamakami, that asphyxial blood is capable of inducing an increase of the blood sugar content in the recipient, we have now been able to afford corroboratory evidence, and further agree with

TABLE III.
Experiments on bilaterally splanchnectomized rabbits.

No.	Date	Body weight (kilos)	Defibr. asphyxial blood inj.				Blood sugar			Urine sugar			Body temperature (°C)	Frequency of respiration (per min.)	Frequency of heart beat (per min.)	Room temperature (°C)
			Amount injected (c.c.)	Blood sugar (%)	Duration of asphy- xiation (hrs.)	Region of collection	Before injection (%)	Max. after injection (%)	Time to Maximum (hrs.)	Before injection (%)	Max. after injection (%)	Time to Maximum (hrs.)				
1	8. V. '25	1.46	5	0.13	1	Carotid	0.09	0.13	$\frac{1}{2}$	0.059	0.078	2	38.7-39.1	42-120	238-348	17.5-20.0
2	14. V. '25	2.08	9	0.12	1	"	0.09	0.10	$\frac{1}{2}$	0.020	0.039	$4\frac{1}{2}$	39.9-41.0	66-252	298-360	19.0-21.0
3	21. VII. '25	1.9	15	0.21	$\frac{1}{2}$	"	0.10	0.16	$\frac{1}{2}$				39.8-41.0	288-336	204-240	26.0-28.5
4	3. VIII. '25	2.16	15	0.30	1	Heart	0.11	0.15	$\frac{1}{2}$				40.2-40.5	120-264	204-240	25.0-27.0
5	3. VIII. '25	2.21	15	0.30	1	"	0.10	0.16	$\frac{1}{2}$				39.9-40.1	156-168	298-264	25.5-27.0
6	2. X. '25	1.99	13	0.15	1	Carotid	0.09	0.13	$\frac{1}{2}$	0.029	0.069	1 $\frac{1}{2}$	38.8-40.0	48-108	216-240	19.5-21.5
7	2. X. '25	1.47	10	0.15	1	"	0.10	0.12	$\frac{1}{2}$				38.6-39.6	60-96	228-264	19.5-21.5
8	13. X. '26	1.78	12	0.22	5	Heart	0.12	0.15	$\frac{1}{2}$	0.020	0.059	1 $\frac{1}{2}$	37.8-39.1	72-96	168-204	14.0-17.0
9	13. X. '26	1.8	12	0.22	5	"	0.11	0.14	$\frac{1}{2}$				38.2-39.5	120-168	240-276	14.0-17.5

him in disregarding the excess of sugar in the transfused asphyxial blood as the unique source of increased sugar in the recipient after the transfusion. In Experiment No. 6 in Table II, for example, 14 c.c. of asphyxiated blood gave rise to an increase of blood sugar in the recipient, the preformative blood sugar content of which was estimated as exactly the same as the transfused asphyxial blood. Further, if in the cases, e. g. No. 7 in Table II and Nos. 4 & 5 in Table III, in which a large amount of excess sugar was transfused into the recipient, the increase of the blood sugar concentration in the recipient due to the transfusion of sugar itself be calculated by taking the total blood volume as one eighteenth of the body weight, it should be 0.114% in No. 7, 0.131% in No. 4 and 0.122 in No. 5, but in fact it was 0.13%, 0.15% 0.16% respectively. Although the excess of sugar in the transfused blood over the preformative one in

the recipient, if it exists, must be taken, as a matter of course, into account as a source of the increase of blood sugar in the recipient, it can not be assumed as the sole origin of the latter fact.

In the next place the effect of the augmented epinephrine discharge by asphyxia upon this occurrence of hyperglycaemia, suggested by Yamakami, has been submitted to the test of experiment. Blood was taken from a carotid artery of the heart of rabbits, asphyxiated in the above mentioned manner, and its inhibiting ability on the rabbit intestine segment was estimated by one of us (Saito) in the manner usual in our laboratory.⁵⁾ The indifferent blood used for the assay was taken from a carotid before the start of asphyxiation.

In all cases as shown in Table IV, epinephrine was not detected with certainty by means of the rabbit intestine strip worked with, the bloods acted upon the strip nearly as the indifferent bloods, or at most the epinephrine content was estimated as undoubtedly less than 0.00005 mgrm. in 1 c.c.

TABLE IV.
*Epinephrine content of asphyxial blood estimated by the
rabbit intestine strip method.*

No.	Date	Body weight (kilos)	Duration of asphyxiation (hrs.)	Region of blood collection	Sugar content of asphyxial blood (%)	Epinephrine content
						In 1 c.c. of asphyxial blood (mgrm.)
1	1. VI. '26	2.2	$\frac{1}{10}$	Carotid	0.16	Below than 0.00005, not different from the in- different blood
2	1. VI. '26	1.9	1	"	0.25	
3	3. VI. '26	1.62	$\frac{1}{10}$	"	0.15	
4	3. VI. '26	1.5	$\frac{1}{10}$	"	0.17	
5	8. VI. '26	1.92	$\frac{1}{10}$	Heart	0.11	
6	8. VI. '26	1.86	1	"	0.16	
7	22. VI. '26	1.87	$\frac{1}{10}$	"	0.11	
8	22. VI. '26	1.5	1	"	0.27	
9	7. VII. '26	1.45	$\frac{1}{3}$	"	0.08	
10	28. IX. '26	1.66	1	"		
11	28. IX. '26	1.6	1	"		
12	28. IX. '26	1.6	1	Carotid		
13	7. X. '27	1.72	$\frac{1}{2}$	"		
14	7. X. '27	1.72	$\frac{1}{2}$	Heart		
15	7. X. '27	1.65	$\frac{1}{2}$	"		

5) Tadashi Sugawara, Masa, Watanabé and Shidzuka Saito, Tohoku J. Exp. Med., 1926, 7, 6-19.

A tracing belonging to Exp. No. 3 is reproduced as an example of the epinephrine assay.

No. 3 (Table IV)

At $\times$ atropin-Tyrode's solution was displaced by indifferent blood solution, at 4 and 6 by the asphyxiated blood solution, at 5 by the indifferent blood solution, and at 7 by the adrenaline blood solution containing 0.000025 mgrm. in 0.5 c.c. The inhibition produced by the asphyxiated blood sample is far less than that by the adrenaline blood solution, and not so different from that by the indifferent blood solution. It was therefore taken as far less than 0.00005 grm. in 1 c.c., not so different from the indifferent blood.

At all events, we must assume that the asphyxiated blood may contain epinephrine of less than 0.0005 mgrm. in 1 c.c. Then it may be now further questioned whether or not some cubic centimeters of blood containing epinephrine in such a quantity are capable of causing an increase of blood sugar concentration, portrayed in the foregoing paragraphs with subtraction of the increase due to the transfused excess sugar.

There are some previous investigations regarding the threshold of adrenaline amount which is just sufficient to evoke hyperglycaemia in rabbits, but continuous intravenous infusion of a diluted adrenaline solution was only tried therein, so that they can not be made use of for the present purpose. Consequently we have carried out another set of experiment on

TABLE V.

Infusion of diluted adrenaline saline solution corresponding to the epinephrine content of asphyxial blood into doubly splanchnectomized rabbits.

No.	Date	Body weight (kilos)	Epinephrine content in 15 c.c. of physiologic saline solution (mgrm.)	Blood sugar					
				Before infusion	$\frac{1}{2}$ hour after infusion	$\frac{1}{3}$ hour after infusion	$\frac{1}{2}$ hour after infusion	1 hour after infusion	2 hour after infusion
1	24. XI. '26	2.2	0.00075	0.11	0.11	0.11	0.10	0.10	0.11
2	24. XI. '26	1.75	0.00075	0.11	0.11	0.12	0.12	0.10	0.11
3	24. XI. '26	1.55	0.00075	0.11		0.12	0.11	0.11	0.11
4	24. XI. '26	2.0	0.00075	0.10		0.11	0.10	0.10	0.11

some rabbits. Physiological saline solution containing 0.00005 mgrm. in 1 c.c. was intravenously administered to rabbits in a dose of 15 c.c. per animal, this being the maximum amount of asphyxiated bloods introduced. No increase of the blood sugar content was established.

V.

To sum up :

(1) The asphyxiated blood of rabbits from the general circulation is capable, though not invariably, of causing hyperglycaemia of a slight degree excepting the increase of sugar concentration due to the transfused excess sugar in the asphyxiated blood, when intravenously introduced in a dose of ten to fifteen cubic centimeters for a rabbit weighing about two kilos.

(2) Practically no epinephrine was discovered in the defibrinated asphyxial rabbit blood from the general circulation by means of the rabbit intestine segment.

Towards the expenses of this research a grant was received from the Saito Hōonkai in Sendai.

Prof. Y. Sataké.