Physiology. — *Further experimental investigations of the internal secretion of the pancreas.* (By N. WATERMAN M. D.). (Communicated by Prof. Dr. C. A. PEKELHARING).

(Communicated in the meeting of June 28, 1913).

In a previous publication [1]) we discussed some properties of the blood from the pancreatic vein (containing what may presumably be considered as the internal secretum), and concluded from our experimentation that the secretum of the pancreas subserves the glycogenesis of the liver. No effort was made to detect whether this influence is due to a diminished splitting of the glycogen in the liver or perhaps to an increased formation of glycogen. As known, J. DE MEIJER [2]) has pointed out that various pancreatic extracts, which are made to pass artificially through the liver, cause a relative increase of glycogen. On the basis of these investigations DE MEIJER accepted the second alternative. I have also endeavoured to make up my mind about this question, in a different way from DE MEIJER's though, partly because circumstances compelled me to adopt a different method, in default of the expensive apparatus for artificial circulation, partly because the results of DE MEIJER's experiments did not appear to me altogether conclusive on the following grounds:

1. frequent errors are incidental to artificial circulation, so that out of 17 of DE MEIJER's experiments 6 (= 1/3) were discarded outright;

2. after all, we are disposed to think that critics must find fault with the values DE MEIJER obtains in his glycogen-determinations and especially with his way of calculating large percentages from insignificant absolute differences. Some examples to illustrate this; l.c. page 48 we are told that a double circulation evolves an increase of 0.00158 % of glycogen. In the 4th experiment on page 49 the examination of about 70 grs of liver yielded an absolute difference of 0.00428 grs. of glycogen. Still the percentage differences for these experiments were given even as high as 15 and 20 perc.

I, therefore, followed out another plan, which of course will also be at fault in one way or other, but which, in my opinion, resulted in very serviceable evidence. I proceeded as follows:

A number of dogs were put under a profound narcosis and during one hour and a half they were given in most cases per jugular vein, rarely per pancreatic duodenal vein, a certain dose of LOCKE's

[1]) These Proc. Vol. XVI, p. 2.

[2]) J. DE MEYER, Archives Internat. de Physiologie. Vol. IX, 1910.

olution or of venous blood from a normal dog or of blood from ıe pancreatic duodenal vein. The amount of glycogen was estimated efore and after the irrigation.

To this conduct of the experiment the following objections may ıe raised:

1. One dog is more susceptible to the narcotic than another, and ıoreover reacts in a different way (to mention an essential point, here is a difference in the duration and in the intensity of the stage f excitation).

2. A laparotomy is performed and part of the liver is extirpated lurante vita, to which stimulant acts various dogs are apt to respond n different ways.

3. Some loss of blood is inevitable.

We should bear in mind, however, that every experiment upon , living organism must involve errors, but also that if the experiıent is performed upon a large number of animals these errors ıay probably neutralise each other in part or altogether.

Method: Dogs of from 4 to 22 KGs. were narcotized with chloroorm; the abdomen was opened and a portion of the liver was emoved. The following precautions were taken; the operation was lways performed in a very warm room (21°—26° C.) to minimize he fall of temperature. The narcosis was as profound and as restful s possible. The portions of liver removed varied between 20 and 0 grs.; they were cut off by means of scissors that were rather lunt and had been warmed beforehand; there was no fear of arenchymatous bleeding and only some large veins had to be igated. The abdomen was left open only for a very short space except of course in the two cases of injection per pancreatic vein) nd even then the laparotomy-wound was covered throughout the vhole experiment with a warm tampon, soaked with physiological altsolution. Subsequently fluids were injected per jugular vein for ne hour and a half in most of the experiments. Irrigation was erformed from a burette on stand, so that the velocity of the inlowing liquid could be regulated by means of a tap.

After one hour and a half another piece of liver of approximately qual weight was cut off as fast as possible.

Finally a determination was made of the amount of glycogen of oth pieces of liver. In order to ensure a proper amount of glycogen he animals had been given about eight hours before the operation , diet consisting of 100 grs. of cake. Owing to outward circum-

stances this was not done in one case, and in another the meal was given only 5 hours prior to the operation.

The amount of glycogen was estimated after PFLÜGER's method; immediately after the extirpation the portions of liver were put in a 60 % solution of KOH and were weighed.

The detailed reports of the experiments contain particulars relative to the fluids injected intravenously.

FOURTH SERIES (D).

1. On the 24th of December 1912 I extirpated 38 grs. of the liver of a dog, weighing $5^1/_2$ KG, which had been fed with amylacea eight hours before. The piece of liver was forthwith subjected to the process described. Hereafter I injected per pancreatic duodenal vein 50 c c. of a mixture of 100 cc. of serum + 50 cc.. of defibrinated blood + 50 cc. of LOCKE's solution.

The remaining 150 cc. of this mixture was injected slowly per jugular vein. After an hour's irrigation another portion of liver of 24 grs. was removed and subjected to the same process.

The blood and the serum had been obtained the evening before from the pancreatic vein of a dog under the influence of a subcutaneous injection of 50 cc. of secretin. The blood had been kept cool, not in the refrigerator.

The glycogen of the 1st portion of liver amounted to 3.825 %.
" " " " 2d " " " " " 3.905 %.
So there was a slight increase of glycogen of 3 %.

2. On the 28th of December 1912 26 grs. of liver were taken from a dog of 12 KG. Directly after this 180 cc. of LOCKE's solution was given per jugular vein from a burette in the space of two hours. The laparotomy-wound was covered with warm tampons. At the end of the experiment the rectal temperature was 36.7°. The solution had been heated to 30° C., the room-temperature was 20—22° C After the irrigation a second portion of the liver (29 grs.) was excised

The amount of glycogen of the 1st portion was 1.71 %.
" " " " " " 2d " " 0.895 %.
This reveals a considerable loss of glycogen of 50 %.

3. On the 30th of December 1912 a dog weighing 15 KG. was irrigated in a similar manner with 180 cc. of LOCKE's solution during one hour and a half. Beforehand 41 grs. of liver had been extirpated and again 37 grs. after one hour and a half.

The amount of glycogen of the 1st portion was 3.995 %.
" " " " " " 2d " " 1.712 %.
Consequently a loss of 56 %.

4. A dog of 8 KG., which had not had the ordinary diet, was irrigated intravenously with the following mixture: 60 cc. of defibrinated blood + 40 cc. of serum, obtained from jugular blood collected the day before from a healthy dog. To this mixture was added 50 cc. of LOCKE's solution. The irrigation lasted one hour and a half. Beforehand a portion of the liver (31 grs.) had been removed. After the irrigation a piece of 34 grs.

The amount of glycogen of the 1st piece was 0.171 %.
" " " " " " 2d " " 0.11 %.
So the loss was 36 %.

5 On the 6th of January 1913 the following mixture was thrown into a dog of 9 KG. per jugular vein during one hour and a half: 88 cc. of blood + serum (obtained from the pancreatic duodenal vein under the influence of secretin) + 70 cc. of LOCKE's solution. Room-temperature somewhat lower than usual (20° C.). The abdomen was covered with warm cloths soaked with a physiological salt-solution.

39 grs. of liver had been removed before the process, 30.8 grs. after it.

The amount of glycogen of the first portion was 4.23 %.
" " " " " " second " " 4.17 %.

In neutralizing portion 1 after the inversion a slight excess of KOH was added somewhat too quickly, which originated a light brown decoloration.

6. On the 19th of January 1913 a dog of 11 KG was given per jugular vein during one hour and a half a mixture of 100 cc. of serum + blood + 40 cc. of LOCKE's solution. The blood had been obtained the previous evening from the pancreatic vein under the influence of secretin. The dog from which it had been drawn was very old The pancreas had greyish white spots, rather smaller than a threepence; there was besides suppuration of the cysts in the postate; the urine, however, contained no trace of sugar whatever.

After the first extirpation of liver venous hemorrhage ensued, which could be staunched only by ligation. The room-temperature was 19° C.

Previously 26.6 grs of liver had been excised. After one hour and a half 29 grs.

The amount of glycogen before the irrigation was 3.61 %.
" " " " after " " " 2.87 %.
This points to a loss of 21 %.

7. On the 25th of January 1913 the following mixture was used per jugular vein upon a dog of 5.5 KG.: 60 cc. of defibrinated blood + 30 cc. of serum from the jugular vein of a normal dog + 50 cc. of LOCKE's solution. Room-temperature 26°—22° C. Duration of the test one hour and a half.

32 grs. of liver had been removed previous to the irrigation, 29.7 grs. after it.

Amount of glycogen before the irrigation 2.638 %.
" " " after " " 1.677 %.
Loss 33 %.

8. On the 28th of January 1913 24.8 grs. of the liver of a dog (6 KG.) was excised. Injection per jugular vein of: 25 cc. of defibrinated blood + 40 cc. of serum from the pancreatic vein (obtained the evening before) taken up with 20 cc. of LOCKE's solution are freed from protein by three times their quantity of 96 % alcohol. After filtration the mixture is evaporated down to 80 cc. at 70° C. and again LOCKE's solution is added to 150 cc. A filtration error causes a loss of 50 cc. so that an equivalent volume of LOCKE's solution has to be added again.

24.8 grs. of liver had previously been removed and 23.8 grs. after one hour and a half.

Amount of glycogen before irrigation 2.024 %.
" " " after " 1.03 %.
Loss 50 %.

During the irrigation the dog had two syncopes so that the respiration had to be maintained artificially.

9. On the 31st of January 1913 a dog of 7 KG. was given 200 cc. of a mixture of 50 cc. of blood + 100 cc. of serum obtained the previous evening under the influence of secretin from the pancreatic vein of a dog. LOCKE's solution was added to 200 cc. Besides a secretin injection this dog had also had a light meal of carbohydrates.

The irrigation lasted one hour and a half; only once breathing came to a standstill.

Beforehand 25.1 grs. of the liver had been excised, afterwards 28.4 grs.

Amount of glycogen of the 1st portion was 2.36 %.

" " " " " 2nd " " 1.77 %.

Loss 20 %.

10. The same large dog from which pancreatic blood had been drawn for injection of the dog in N°. 9, had the pancreas removed. Part of this (3/5) was rubbed up with about 40 cc. of LOCKE's solution and mixed with three times this quantity of 96 % alcohol. After filtration this liquid was evaporated down to 1/3 of its volume in a porcelain dish at 70° C. and subsequently LOCKE's solution was added to 180 cc.

Three days after the preparation (on the 3rd of February) the liquid was thrown into a dog of 4 KG.

Previously 13.2 grs. of liver had been extirpated. 12.4 grs. one and a half hour later.

The amount of glycogen of the first portion was 2.41 %.

" " " " " " second " " 1.48 %.

The loss, therefore, appeared to be 37 %.

11. On the 11th of February 1913 a dog of 12 KG. was given during one hour and a half per jugular vein normal blood (100 cc. made up to 140 cc. with LOCKE). First I tried to inject part of it per pancreatic vein. However, when 10 cc. had been injected a venous hemorrhage had to be staunched. We therefore performed further injections per jugularis.

Four hours before the dog had taken a copious meal of amylacea so that the digestive process was at its height. The narcosis proceeded regularly.

Of the liver 14.6 grs had been removed before the injection and 40 grs an hour and a half after it.

Amount of glycogen of the first portion 7 823 %.

" " " " " second " 6.433 %.

Here the loss was 18 %.

12. An extract of the pancreas of the dog whose normal blood had been used in N°. 11 was used upon a dog of 5 KG. The extract had been prepared as follows: The pancreas, weighing 23 grs, was rubbed up with LOCKE, then heated for one hour and a half to 60° C. to destroy the ferments; after this it was subjected to a low temperature (about 5° C.) and used next day after filtration through cotton wool and addition to 190 cc. After precisely an hour's irrigation a shock occurred; in less than no time a portion of the liver was cut off.

Extirpation of 14.6 grs of liver before injection; after an hour 13.3 grs.

Amount of glycogen of the 1st portion 10.88 %.

" " " " " 2nd " 6.76 %.

Loss after an hour 38 %.

13. On the 25th of February a mixture of secretin and LOCKE's solution was thrown into the jugular vein of a dog weighing 7 KG The secretin had been prepared as follows: In the space of 2 × 3 hours an extract was made of the mucous membrane of the duodenum with absolute alcohol in SOXHLET's apparatus; only then it was macerated with HCl and after boiling up neutralized with sodium. There was a secretin solution of 50 cc. to which 130 cc of LOCKE's solution had been added. Irrigation for one hour and a half.

Of the liver 24 grs had been excised before the injection and 33 grs one hour and a half after it.

Amount of glycogen of the 1st portion 4.356 %.
" " " " " 2nd " 2.697 %.
Loss of glycogen 37 %.

14. On the 3rd of March a dog of 6 KG. was given per jugular vein 50 cc. of pancreas blood + 60 cc. of serum + 50 cc. of LOCKE's solution, obtained the preceding evening from a dog under the influence of secretin.

The dog was very restless under the narcosis; vomiting occurred twice.

Before the injection 37 grs of liver had been removed; 21.9 grs one and a half hour after it.

Amount of glycogen of the 1st portion 9.035 %.
" " " " " 2nd " 6.683 %.
Loss 26 %.

TABLE I.

Liquid injected	Gain or Loss of glycogen	Time of irrigation	Particulars
Secretin-Pancreas venous blood	+ 3 %	1 hour	
" " " "	— 20 %	1 1/2 "	Bloodserum from old dog
" " " "	— 1 %	1 1/2 "	
" " " "	— 25 %	1 1/2 "	
" " " "	— 26 %	1 1/2 "	Excitation under narcosis
Normal blood	— 36 %	1 1/2 "	No diet
" "	— 33 %	1 1/2 "	
" "	— 18 %	1 1/2 "	Copious diet
Aqueous extract of the pancreas	— 50 %	1 1/2 "	
Alcoholic extract	— 37 %	1 1/2 "	
" "	+ 5 %	1	Restful narcosis
Secretin	— 37 %	1 1/2 "	
LOCKE's solution	— 47 %	2 "	
" "	— 56 %	1 1/2 "	
Aqueous extract of the pancreas	— 38 %	1	

15. An alcoholic extract of the pancreas, the blood of which had been used in the foregoing test was given to a large dog of 30 KG. The extract had been prepared in the following manner: 20 grs of pancreas had been rubbed up and macerated with 100 cc. of 65 % alcohol. During $^3/_4$ hour this macerated substance was evaporated down to $^1/_3$ of its volume at 65° C., made up to 180 cc with LOCKE's solution and filtered through wadding.

The dog received this fluid during one hour through the jugular.

The first portion removed from the liver was 33 grs.

The second (an hour later) $35^1/_2$ grs.

Amount of glycogen of the 1st portion 4.124 %.

„ „ „ „ „ 2d „ 4.356 %.

The narcosis was remarkably restful, without any disturbance

In this experiment there was a gain of 5%.

For an easy survey we summarize the results of this series of experiments in the preceding table.

CONCLUSIONS.

Above all we observe that narcosis, laparotomy and liver-extirpation, in spite of continual irrigation by injection of a fluid deemed favourable, result in a loss of glycogen. This is after all not surprising, if we bear in mind that a number of glycolytic factors act upon the liver. Among others: the narcosis per se, often associated with considerable agitation; the animal's emotions; sometimes spasmodic contractions caused by vomiting; loss of blood; fall of body-temperature. Nevertheless the fact deserves consideration in view of the surgical process. The loss of glycogen averaged about 50 %. Furthermore it follows from our experiments, that if defibrinated blood and serum is given instead of LOCKE's solution, the decrease of glycogen is considerably less. An injection of venous blood from a normal dog lowers the amount of glycogen to about 29 % i.e. about half of the average. The average fall is still greater if irrigation is accomplished with blood collected from an active pancreas. Then the average decrease is only about 15 %, i.e. $^1/_3$ of the splitting caused by irrigation with LOCKE's solution.

Irrigation with normal blood, more particularly with pancreatic blood, therefore, inhibits the loss of glycogen in the liver, nay in one very successful experiment even a slight increment of glycogen was noticeable.

We have now to consider the problem as to how this result was effected. Three hypotheses offer themselves for discussion.

a. The blood favours glycogenesis.

b. The blood impedes glycogenolysis.

c. The blood neutralizes the inimical influence of the narcosis.

As for the third theory it seems to me that the volume of the injected blood and serum (about $^1/_{10}$ of the blood of the body) is too small to act vigorously. With regard to the other two, I do not think it quite possible as yet to decide in favour of either of them. Still, I incline towards the former, because occasionally an augmentation of glycogen was to be noted, which on no account can be originated by prevention of splitting alone. It is obvious that most often the splitting prevailed over the formation of glycogen.

Additional conclusions from this series of experiments are the following:

In two cases an aqueous extract of the pancreas yielded negative results; positive results, however, were obtained twice from an *alcoholic* extract. Aqueous extracts and LOCKE's solution resulted in an equivalent fall of the glycogen, alcoholic extracts in one instance in $^2/_3$ of the average, in another a rise of 5 % was observed. It seems then that *the exciting agent of the pancreas is soluble in alcohol*, not in *water*. It is probable therefore, that this agent is present in the blood rather in physical adsorption than in chemical solution.

Again the thermostability was proved in both experiments. Lastly we notify that a secretin deprived by alcohol of its vaso-dilating substances acted favourably upon the amount of glycogen in the liver.

Results compared with those of DE MEYER.

Broadly speaking also this series of experiments lends support to J. DE MEYER's evidence. Our methods of investigation differ essentially, which renders the ultimate accordance in our results all the more satisfactory. My plan differs from DE MEYER's 1. in experimenting in vivo with all its drawbacks and advantages; 2. in the lack of double circulations in the liver.

The accordance in our results consists in:

1. aiding the glycogenesis even through normal blood more than through a physiological salt solution.

2. a greater activity in this respect of blood that has passed through the pancreas.

It may also be remarked that the volume of the injected fluid is considerably smaller than DE MEYER uses in his experiments. It should be borne in mind, however, that I worked in vivo, an advantage of which is that glycogenetic factors are allowed to exert an action beforehand, since a normal fluid containing sugar passes through the liver.

Recapitulation:

1. Normal blood, in a higher degree still blood from the pancreatic vein, aids the glycogenesis of the liver.
2. Alcoholic extracts of the pancreas seem to have the same effect.
3. The material which incites this action is thermostable.

FIFTH SERIES (E).

We considered the following problem. If it is a fact that the pancreas furnishes the venous blood with a substance, that promotes the glycogenesis, it follows that in the case of experimental pancreatic diabetes, when especially also the liver-glycogenesis is disturbed, a favourable influence of this blood upon the progress of the disease must be noticeable. We have set ourselves the task to ascertain this.

Method. Of some dogs the pancreas was extirpated in the following way: With the utmost attention to asepsis the abdomen was opened by an incision parallel to the right arcus costalis. While the bleeding was being stopped with due care, the pancreas was searched for and extirpated after HÉDON's method. An essential characteristic of this process is that no ligatures are applied along the duodenum, but that the pancreas is separated from the duodenum by scratching with a sharp nail and the bleeding is staunched only by plugging vigorously with a tampon, another characteristic is that particular caution is given to ligation of a small vein, which joins the tail of the pancreas to the spleen. Now in reality HEDON's method applies to extirpation of the entire pancreas in two stages (part of the proximal extremity of the pancreas, which is provided with a separate vascular bundle, is transplanted under the skin of the abdomen). However, extirpation of the entire pancreas did not answer our purpose, because it quickens and aggravates the pathological progress to such an extent that it becomes difficult to watch the effect of some therapeutic procedures. We did not see the necessity for transplanting part of the pancreas, and only left a piece of it intact (at a rough estimate 2 to 3 grs) in the neighbourhood of the place where the ductus choledochus enters into the duodenum. An additional advantage of this method is that icterus, which often ensues from an extirpation of the pancreas, can be avoided. The abdomen was irrigated with a warm physiological solution, after which the different layers were closed successively. Most times the animals recovered pretty soon. We subjoin the experiments which resulted in diabetes after a partial excision of the pancreas. Parenthetically I wish to recall to mind an extirpation,

TABLE II

Date	Weight	Diet	Urine	*D* percentage	*D* total	*N* total	*D*/*N*	Acetone	GERHARD	Particulars
ne 14	4.4 KG.	none	80 cc	0.4 %	0.32	—	—	—	—	
„ 15	4.4 „	vomiting	175 „	0.2 „	0.35	—	—	—	—	
„ 16	„	100 grs of horse flesh + 350 grs of milk	230 „	6.48„	15.55	2.92	5.36	—	—	
„ 17	„	„	320 „	4.6 „	15.04	3.83	3.82	—	—	very hungry and thirsty as on all following days
„ 18	„	„	120 „ 145 „	4 1 „	10.86	2.445	4.44	—	—	
„ 19	„	„	121 „ **70** „	6.7 „ **10.8** „	8.107 7.56	1.50 1.00	5.30 5.8	—	—	At 8 p. m. i. e. after the first 12 hours subcutaneous injection of 35 c.c of blood from the panc. vein collected the previous evening under the influence of secretin.
					15.66	2.80				
„ 20	4.35 „	„	162 „ 142 „	5.8 „ 4.2 „	9.39 5.96	1.92 1.68	4 88 3.58	—	—	
					15.35	3.60				
„ 21	„	„	144 „ 142 „	4.1 „ 3.8 „	5.96 5.39	1.90 1.60	3.20 3.36	—	—	
					11.35	3.50				
„ 22	„	„	112 „ 140 „	2.43„ 6.35„	12	1.80 3.—	2.6	—	—	
„ 23	4.2 „	„	180 „ 110 „	7.03„ 3.15„	16	5	3.2	—	—	
„ 24	„	„	160 „ 160 „	6.2 „	19.8	4 33	4.6	—	—	
„ 25	4.1 „	„	205 „ 113 „	5.2 „ **10 4** „	10.60 11 3	1.91 2.17	5.56 5.40	—	—	After these 12 hours' another subc. inj. of 20 c c of blood from the pancr. vein. One hour after the inj. the values of bloodsugar are compared. Before the injection **0.227%**; after it **0.237%**. Blood was drawn under a light narcosis by chloroform.
					21 9					
„ 26	4.1 „	„	232 „ 160 „	6.9 „ 6.7 „	26 7	6.2	3.66	—	—	
„ 27	„	„	324 „ 120 „	7.2 „	31.5	7.75	4	—	—	

which did not lead to diabetes even after 6 weeks, although the venous blood of this dog (see Exp. 34. Table V of the previous publication) was competent to occasion a fall of the N-metabolism in another dog in N-equilibrium.

Experiment 55. A little dog ($4^1/_2$ KG.) had on the 13th of June 1912 its pancreas (14 grs.) removed in the manner just described. The animal passed well through the operation. The table on page 257 gives the history of the disease together with the particulars worth mentioning.

For want of further particulars I leave off tabulating. Suffice it to say here, that glucosuria was increasing all along to as much as 40 grs. and more, that the N-metabolism was maintained constant, that, however, on the 1st of July acetone appeared for the first time and the animal was killed on July 4. The section brought to light a small portion of the pancreas of the size of a die, which looked unimpaired.

All organs had exchanged their natural colour for a yellowish white. This was due to strong lipemia, which made the bloodserum creamlike.

The effect of our injections is of course a matter of the highest interest to us. From the values recorded above we may be justified in deducing the following:

The first injection of 35 cc. of pancreatic blood lessens the diuresis and raises the glucose percentage to such an extent (from 6 to 10%) that it cannot be a matter of mere chance. However, the N-percentage does not rise so much, so that D/N increases. For all this there is no augmentation of the quantity of sugar secreted on the day itself nor on the two subsequent days. On the contrary it is slightly lowered (15—11 grs.), which can neither be a casual fluctuation, as for instance on the 18th of June, since the decrease continued for more than twice 24 hours. This decrease coincides with a process in the composition of the urine which is the reverse of the one just mentioned, viz. an increase of the N-amount and of the diuresis and a fall of the glycose-percentage. The effect of the second injection, on the 25th of June, is far less obvious, it is almost negative. It is true, only 20 cc. is injected and the dog is subjected to a slight narcosis with chloroform prior to the puncture of the vein, but here a temporary rise of the glycogen-percentage is followed by a fall; the quantity of urine, however, first remains constant and increases later on. Absolute decrease of the sugar secreted is out of the question, rather the reverse takes place. Neither do we note any favourable influence of the injection on the hyperglykemia.

Experiment 57. On the 19th of September 1912 a rather large dog (11 KG.) has $21^1/_2$ grs. of the pancreas extirpated after the ordinary method: a small duodenal portion is left in the abdomen. The operation was attended with some troubles: frequent escape of blood and repeated vomiting occurred. Recovery ensued without complication and on the 22d of September the dog was restored so far, that he took meat and drank milk. The urine contained bile-pigments but no sugar.

TABLE III.

Date	Diet	Weight	*D* percentage	*D* total	*N* total	D/N	Acetone	Urine	Particulars
Oct. 2	300 grs of horse flesh, 150 grs of cake. 200 grs of water. Afterwards unlimit.	10.7 KG.	3.5 %	7 Gr.	—	—	—	200	Greedy for food and drink.
» 3	»	—	4.25 »	8.5	4.96	1.7	—	200	
» 4	»	—	3.6 »	8.2	8 05	1	—	230	
» 5	»	—	—	—	—	—	—	465	Severe diarrhea interrupting the experiment for some days.
» 10	»	—	9.6 »	46	—	—	—	480	Towards the close of these 24 hours the animal is given subcutaneously serum of the blood from the pancr. vein, obtained the previous evening under the infl. of secretin (100 c c.)
» 11	»	—	9 »	57 6	10.7	5.4	—	640	
» 12	»	10.3	8.2 »	77.9	10.45	7.6	—	950	
» 13	»	—	11.33 »	56.6	not determined	—	—	500	
» 14	»	—	10.2 »	66.3	7.8	8.5	—	650.	
» 15	»	—	8.7 »	73.9	9.35	7.9	—	850	
» 16	»	—	10.5 »	89.2	8.4	8.8	—	850	
» 17	»	—	10.4 »	100.8	10.3	10.1	—	1050	Great thirst. Appetite lessens.
» 18	»	—	10.3 »	103	11.6	9.1	—	1000	
» 19	»	—	10.8 »	92	7.9	11.6	—	850	The animal stops eating and declines rapidly.
» 20	»	?						800	The animal is killed.

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On the 2d of October i.e. only a fortnight after the operation the first traces of sugar appear. The substances just mentioned have disappeared.

The following table gives particulars about the experiment with this animal.

Retrospect. Here we have a case of a typical incomplete pancreatic diabetes (SANDMEYER). Not until a fortnight after the extirpation diabetes is gradually evolved and reaches its acme only after three weeks.

With regard to the injection of blood from the pancreatic vein, obtained the previous evening under the influence of secretin, we notice an effect similar to that of the foregoing experiment. In the first place a smaller quantity of urine associated with a rise of the D-percentage. As for the N-percentage the values are not very demonstrative, because the urine had been standing for more than 24 hours. We could not make a reliable Kjeldahl-determination on the most important day (Oct. 13). Still also in this case a relative decrease seems to have occurred, succeeded by an insignificant increase.

Lastly it appeared again that after injection the total D-secretion was for some days considerably lower than before.

On the 19th of October the animal was sinking so fast, that sub finem vitae D-secretion as well as N-elimination was retroceding. On this account the animal was despatched.

At the section no considerable modifications (lipemia) were detected; owing to several adhesions nothing was to be seen of residues of the pancreas.

Experiment 59. In my judgment the following test yields weighty evidence. The time of observation was longer. Four injections of pancreas secretum were given.

In the ordinary way $20^1/_2$ grs. of the pancreas of a dog is extirpated on the 22d of September 1912. At a rough guess 2 grs. is left in the abdomen. Operation comes off quickly and without a hitch. As early as the 23d of Sept. the dog takes water and milk. A trace of protein and a small quantity of sugar is present in the urine. On the 24th of September the dog is tolerably well, only somewhat languid, vomits once more, but eats some horseflesh and cake. A considerable amount of sugar is present in the urine. The following table illustrates our subsequent experience.

Retrospect. This is a serious case of diabetes, clearly demonstrating the influence of different acts. The first two injections were attended by an irregular secretion of urine on the preceding days, as evidenced respectively by 8 and 17 grs. of sugar on the 27th and 28th of September, which, when expressed graphically, yields

TABLE IV.

	Diet	Urine	D %	D total	N total	D/N	Acetone	Particulars
25	150 grs flesh 10 » cake	292	3.6%	10.6	3.89	2.7	—	Dog languid, not greedy, not very thirsty.
26	»	214	5.1 „	10.9	3.616	3.3	—	
27	200 grs flesh 10 » cake	150	5.3 „	8	2.41	3.—	—	
28	»	234	7.3 „	17	6.22	2.7	—	
29	»	38 144	2.9 „ 7.4 „	1.1 10.6	1.9 3.14	3.3	—	50 c.c of pancreas serum obtained the previous evening and maintained at 0—6° C. is injected intravenously after first 12 hours.
30	»	186	6.1 „	11.3	4 27	2.6	—	
1	»	100	8.6 „	8.6	1.99	4.3	—	Again 50 c c. of the same serum 72 hours old is injected at the beginning ot the following 24 hours.
2	»	228	10.2 „	23.2	7.43	3.1	—	
3	»	150	7.5 „	11.25	3.—	3.75	—	
4	»	240	8.1 „	19.4	6.9	3.2	—	
5	»	180	10.6 „	19	5.99	3.1	—	
6	»	100 78	8.7 „ 9.1 „	8.7 7.1	3 5 3.25	2 5 2.2	— —	After the first 12 hours subcut. inj of 120 c.c of defibrinated blood collected the evening before under the influence of secretin from the pancr. vein and kept cool. Greater appetite.
7	»	183	9.1 „	16.67	5.83	2.9	—	
8	»	182	9.2 „	16.6	4.51	3.3	—	
9	»	170	9 „	15.3	4.42	3.5	—	
10	»	278	6.9 „	18.9	6.83	2.7	±	
11	»	255	8 „	20.4	6.63	3.1	—	
12	»	170 120	9 38	27 2	6.11	4.4	+	After the first half of the 24 hours 95 c.c. of pancreas blood-serum of the previous evening and kept cool, is injected subcutaneously.
13		200	11.1 „	22.2	5.84	3 8	++	Even now the dog refuses any food. No appetite at all.
14		66	7 „	4 6	3.—	1.5	+++	
15		68	10 „	6.8	2.56	3.6	++	
16		224	10.6 „	23.7	5.42	4.—	+++	
17		118 205	8.3 „ 7.5 „	9.79 15.3	4.41 3.15	2.2 4.9	+- ±	After the 17th the dog declines rapidly, does not eat and cannot be made to move.
18								
19		58 78 136	5 „	2.9	1.62	1.7	±	
21								Dog despatched forthwith after complete paralysis had set in also of the muscles of the maxilla.

false apices. To all appearance the average value approximates the truth. It was moreover a mistake to make the second injection after too short an interval from the first; this was done lest the activity of the substance should get lost, it would have been better to inject both portions simultaneously Yet also after these injections a relatively slight fall of the D-secretion appears to take place, which is still perceptible after a few days. Furthermore a short increase of the D-percentage is to be noted especially after the second injection by virtue of which the D/N coefficient rises considerably.

The results of the third and the fourth injections are still more demonstrative. At the third the *D*-secretion during three days falls short of what could be expected from the rise of the previous days. Later on the excretion of sugar, likewise that of urine increases rapidly. The changes in *D*- and *N*-concentration are not conspicuous. The fourth injection results in still more striking alterations. The total *D*-secretion is lowered from 27 grs to 22 grs and 6 grs. The quantity of urine diminishes from about 300 to 200 and 60 cc. The *D*-percentage is suddenly exalted to rather more than 11 % and the *D*/*N*-coefficient to 4,4, and 3,8 to fall again later on to 1,5. Here then we have an accumulation of all the alterations announced beforehand. Some particulars however must not be forgotten, viz. after the 13th of October the dog declines nearly all food. However, this fact does not in itself elucidate matters satisfactorily, since on the 16th of October the sugar-secretion is normal again.

Secondly the table shows that on the 12th of October acetone is found in the urine for the first time, which no doubt will be attended with other changes in the metabolism. Even this, however, does not lessen the value of our results, seeing that acetonuria was on the decrease and ultimately disappeared entirely, when the animal was sinking rapidly and was sub finem vitae on the 17th of October. This proves that spontaneous lowering of the *D*-secretion sub finem vitae (a well-known phenomenon) must be carefully distinguished from the *D*-secretion after injection which contrariwise evolves a separation of large quantities of acetone.

We have to add in this connection, that we suspect the 4th injection to have been noxious to the animal's condition, which was not surprising after all. At the section a considerable loss of flesh was demonstrated; there was no lipemia; no trace of pancreatic residue was found.

We record one noteworthy experience. In examining the mouth cavity of the animal bulbous tumours at the mucous membrane of

the lips drew our attention; at the section it appeared that as well at the lips as at the sublingual and buccal membrane these greyish white protuberances were visible and arranged symmetrically. Especially in the sublingual membrane, symmetry was most pronounced at the excretory ducts of the salivary glands. Thinking of the relation between the pancreas and the salivary glands we supposed this to be a vicarious hypertrophy of the salivary glands. A microscopic examination however did not lead to detection of any glandular tissue but only to a rankling growth of the epithelium.

CONCLUSIONS.

A The blood obtained under the influence of secretin from the pancreatic vein is capable of lowering the total secretion of sugar for some days, but also of raising the sugar-percentage temporarily.

B. The blood commonly evolves a relative fall of *N*-elimination close upon the injection, later on a relative rise

These experiences fall in with those recorded in our earlier publications. For instance the rise of the *D*-percentage after injection of blood from the pancreatic vein is in perfect concordance with the fact that the same blood, thrown into normal dogs, also incites glucosuria. On the other hand the influence on *N*-elimination in dogs suffering from pancreatic diabetes, is just the reverse of the influence in normal dogs.

Critical summary. A comparison of our results with those of other researchers shows some notable differences. It is self-evident that soon after the theory of the internal secretion of the pancreas had been propounded, efforts should be made to render pancreatic diabetes less virulent by throwing blood of healthy dogs into dogs suffering from the disease. MINKOWSKI and HÉDON were the first to make the effort. Their failure strengthened PFLUGER's contrary views. Afterwards, indeed, some positive evidence was brought forward by FORSCHBACH [1]), who, by application of the parabiosis between healthy and diabetic dogs elaborated a decrease of sugar secretion. EHRMANN, who also applied transfusion, failed again. Latterly similar experiments have been made by DRENMAN [2]) and HÉDON [3]). DRENMAN obtained a very marked decrease of sugar secretion, after intravenous injection of large quantities of normal blood. This blood, however, had lost

[1]) FORSCHBACH, Arch. f. exp. Path u. Pharmat Bd. IX, 1908.

[2]) DRENMAN, American Journal of Physiol. Vol. XXVIII, 1111.

[3]) HÉDON, Refer. Dr. v. HERWERDEN. Nederl. Tijdsch. v. Geneesk. 1913.

its activity about 12 hrs after having been collected, which proved, according to DRENMAN that the exciting factor is highly labile and that the dilution of the diabetic blood cannot be responsible for the result. HÉDON, on the contrary, inquiring into the effect of transfusion of blood by vascular connection, from normal into diabetic dogs, ascribes the decrease of sugar secretion in the diabetic animal only to the dilution of the hyperglycemian blood, while he attributes a strong inhibitory influence on the renal secretion to transfusion.

My experience differs from DRENMAN's in that I did not detect anything at all of a marked lability of the active factor in the pancreatic blood; anyhow, after more than 20 hours subsequent to the removal of the blood, activity was still noted. This may be only a quantitative difference, because in theory there are more active materials in the pancreatic blood than in the general circulation. It also seems to me a sheer impossibility, to attribute the results, reported here, to dilution of blood; first and foremost because the quantity injected was too small in most cases; secondly the injection was subcutaneous, so that resorption was slow; thirdly the action was continued too long (on an average 2 days). In concordance with HÉDON's experiments I detected an influence upon the renal secretion, not in such a marked degree, however, that it could bear up my results. Lastly a permanent influence on the N-elimination was demonstrated.

Physics. *"On the interpretation of photospheric phenomena"*. By Prof. W. H. JULIUS.

(Communicated in the meeting of May 31, 1913).

§ 1. It is a common belief that a body always presenting the appearance of a circular disk, from whichever side it is looked at, must be bounded by a spherical surface. The general conviction that the bulk of the sun is an incandescent sphere rests on that belief, and was a natural starting-point for solar theories.

After the effective solar temperature had been found so high as to exceed the critical temperatures of perhaps all known substances, the earlier idea that the main body of the sun was in the liquid or the solid state had to be replaced by the hypothesis that it is substantially gaseous. This new idea involved the necessity of explaining the phenomenon of the apparent "solar surface". One had to choose between YOUNG's view, that the photosphere was a