The Physiology of the Carbohydrates

An Epicriticism

by

F. W. Pavy M.D., LL.D., F.R.S.

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TO THE READER.

THE circumstances connected with the publication of this EPICRITICISM are peculiar, and I solicit the attention of the reader to the following narrative.

The early history of my connection with the branch of physiology herein dealt with is sufficiently well known to exclude the necessity of any particulars being here given upon the point. That the problem presented for solution is one of great importance alike to physiological science and the art of medicine no one I think will dispute. It involves nothing less broad than the application of the carbohydrate principles of food to the purposes of animal life, and nothing less deserving serious attention than the consideration of their mal-application in Diabetes.

The views I have been led to enunciate have been pressed upon me not only by the weight of evidence derived from experimental research, but also by the teachings afforded by a long and close study of diabetes. The latter source of information has been of great aid in the matter; and without it I do not know whether my present position would have been attained. In diabetes a mal-application of the carbo-hydrates has to be dealt with; and, it is not difficult to understand that, through the phenomena observable in connection therewith, light would be likely to be shed for affording help to discover their mode of application in health.

No subject of physiological research is, perhaps, beset with sources of fallacy in so many directions, or entangled in greater intricacy than the one under consideration. The closest watchfulness over every experimental detail is needed to escape from error, and it will scarcely be denied that the wider the field of personal observation the more advantageous the position for discerning the truth. Experimental physiology freed from results that have been fallaciously interpreted gives, I contend, no facts in support of the glycogenic doctrine. When we associate with physiology the knowledge derivable from the study of diabetes, and thus look at the matter from a comprehensive point of view, it becomes, I think it may be said, self evident that the liver cannot exercise a glycogenic function; for, to escape from diabetes sugar must be kept out of, instead of being poured into, the general circulation. I refer to what is said in this work (pp. 46—64, 134—136) for the grounds upon which this assertion is made.

Although I have for many years held the conviction, grounded upon the evidence of my personal experience, that the glycogenic doctrine must sooner or later fall, I have not until lately been able to advance anything to take its place. In what way does carbohydrate matter become disposed of in the animal system? This I have felt to be the problem to work at, and I have steadily directed my researches towards its solution, instead of sacrificing time in controversy upon the question of glycogenesis. With the discovery of the glucoside constitution of proteid matter a new system of knowledge opened itself out before me which carries us in exactly the reverse direction of that in which investigators have hitherto been looking. This system has been fully set forth in my work on the *Physiology of the Carbohydrates* published last year. It brings the operations of animal and vegetable life into harmony with each other and reduces to the greatest simplicity the explanation of the phenomena belonging to diabetes.

About the time the work to which I have just referred was issued, a paper by Dr. Noël Paton, bearing the title "On Hepatic Glycogenesis," was published in the 'Transactions of the Royal Society.' The view advanced in this paper stands at variance with the results of my work; and, to make way for it the course is attempted to be cleared by the foot-note on the first page running as follows:—"The theory of Pavy, repeated in nearly every text-book, that the liver is 'a sugar destroying, and not a sugar forming organ,' rests on so unsubstantial a basis and has been so completely refuted by the work of Seegen and other investigators that it need not be considered."

Whilst fully conceding to the author of the paper the right to form and express any opinion he likes with regard to my work, I demur to the 'Philosophical Transactions' being applied as the medium for the publication of a defamatory statement of so gratuitous and unsubstantiated a nature as that inserted.

If looked at, what does it amount to? A mere assertion that the outcome of my work "rests on so unsubstantial a basis, and has been so completely refuted by the work of Seegen and other investigators that it need not be considered." Incongruous as it may seem, the text immediately above the foot-note contains the most condemnatory expressions in relation to Seegen's work. For text service Seegen's work is spoken of in a highly condemnatory manner, whilst for foot-note service it is made to play the *rôle* that has been represented. And, moreover, the condemnatory remarks are applied to the actual portion of Seegen's work—that bearing on the question of sugar production—in which the difference between Seegen and myself is especially to be found. As regards the question of sugar destruction, it happens that we stand in accord in saying that the evidence of the disappearance of sugar in the transit of the blood through the systemic capillaries that is implied under the glycogenic doctrine does not exist.

In reply to a letter asking for the names of the other investigators upon whose work the statement is based, I was informed by Dr. Paton that they are to be found in his article in the 'Edinburgh Medical Journal' which is dealt with in the succeeding pages. To these pages I refer the reader for evidence showing that nowhere in the article is there in reality to be discovered any valid support to his assertion.

Looking at the experimental material upon which the paper, for the sake of which my work is so peremptorily cleared out of the way, is based, what do we find? I have experimentally investigated the matter and my results are embodied in the criticism to follow at pp. 71–102. From these results it is seen that the existence of a faulty experimental foundation is disclosed.

Apart from the question of experimental validity which can only be tested by an actual repetition of the experiments, I submit that the paper, upon the strength of what is adduced in the following pages with regard to it, bears on the face of it sufficient to force the inquiry on the mind—How has it come to pass, in view of the traditions of the Royal Society, that it has found a place in the 'Philosophical Transactions'?

Let me at once state that it has no statutory right to be there; and, in order to show the position in which the matter stands I will give an extract from a letter I addressed to the President and Council of the Society.

"I now come to the procedure adopted in the case of the paper in question and have to state that I have sought competent advice and I am advised that under the statutes of the Society the paper has no *locus standi* in the 'Transactions.' If reference be made to the statutes, it will be found that even where the ordinary procedure by Committee is adopted no paper is in a position to be dealt with in relation to publication till after it has been 'communicated to the Society at their weekly meetings' (*vide* chap. xiii, sec. i, and other statutes bearing on the communication, reading and publication of papers). *The*

Journal Books of the Society contain an entry for July 5th, 1893, showing that the Committee of Papers gave authority to the officers to refer papers during the recess, and empowered the President and officers should they think fit to order for publication papers on which the reports were favorable. The last weekly meeting of the Society took place in June. Dr. Paton's paper according to the announcement upon it was received July 24th. The officers proceeded at once to deal with it and ordered it to be published in the 'Transactions.' The action of the officers was confirmed at a meeting of the Committee of Papers held Oct. 26th. In this action, as I am advised, the officers exercised a power for which they had no authority. It would not have been competent for the Committee of Papers itself to take such action, and it could not delegate to the officers a power which it did not possess itself. The action of the officers was an action outside the statutes and therefore devoid of authority. Being invalid at its inception it could not be rendered valid by the confirming action of the Committee of Papers at its meeting on Oct. 26th.

I consequently respectfully submit that the paper stands without right to do so in the 'Philosophical Transactions.'"

I may here mention that the President and Council have not disputed the point raised.

The final words of my communication ran as follows:—

"The Society guards itself in its Statutes against defamation by its Fellows. Have not the Fellows a right to expect that the publications of the Society should not be allowed to constitute a medium for defaming them? My work stands unwarrantably branded with the stigma to be handed henceforth down in the 'Philosophical Transactions' of having been so completely refuted that it need not be considered; and, I consider I have a right to look for such action being taken by the Council as will remove the stigma resting upon me."

To this communication I received a reply containing an expression of regret on the part of the President and Council that the statement should have been published; but, as I failed to see that this in any way nullified the effect of the publication, I wrote another letter, which, as it serves to explain the step I am now taking, I will here transcribe in full :

"When your letter, in reply to mine of June 11th, 1895, reached me the vacation had commenced, and I deemed it advisable to allow it to pass over before communicating with you further. The time for doing so has now arrived.

"The complaint that I laid before you has not been met in a manner that I can accept as a settlement of the matter.

"I stated at the end of my letter that I considered I had a right to look for such action being taken by the Council as would remove the stigma thrown upon my work by the statement that had been allowed to appear in the 'Philosophical Transactions.'

"I respectfully submit that the expression of regret by the President and Council that the statement should have been published, conveyed in a private letter, does not annul the effect of publication.

"I am loth to publish anything reflecting upon the Royal Society, but in justice to myself I am driven to give publicity to the circumstances of the case.

"It is impossible for me to allow the matter to remain in its present position. An expression of regret in a private communication does not nullify what has been done, and I feel that it is incumbent upon me to insert in the publication that is on the eve of being issued the particulars I conceive to be necessary in the interests of my own vindication.

"I cannot permit the remark at the end of the letter received to pass unnoticed: 'nor do they think it right to take any notice of that part of your letter which seems to impute unbecoming motives to the Senior Secretary.'

"I repudiate warmly the charge that my words convey any imputation of *motive* and am surprised that any such conception could have been formed. I have no ground for doing otherwise than give credit for the existence of pure and proper motives. I thought it would be understood when I spoke of the Society and its *unbiassed* relation to Science, and stated what followed, that what was meant was that the action of the Senior Secretary in the position in which he stood was 'contrary to what is considered ethically becoming.' I am sorry I failed to express myself in a sufficiently intelligible manner; and, to explain still further, I will say that a variety of illustrations are constantly presenting themselves from every-day life showing that persons, as an established line of conduct, refrain from adjudicating upon a matter in connection with which the opportunity has occurred for the mind to become biassed. Had this course been pursued in the present instance, I feel assured the Royal Society would have been saved from the introduction into the 'Transactions' of what I am persuaded cannot escape being realised as a serious blot; for, I consider it justifiable to conclude that a mind in an unbiassed state—a mind, in other words, uninfluenced by an inclination to favour the notion advocated—would have regarded the evidence put forward as altogether insufficient for meeting the traditional requirements of the Society's 'Transactions.' The spirit of the existing statutes of the Society, which were here uncomplied with, guards against such a procedure."

The passage in my first letter to which reference is here made stood as follows :—

"There is another point which I feel in the interests of the position of the Royal Society in its unbiassed relation to Science ought to be considered. As I mention in my criticism to be published of the paper, it works up to, and fits in with, the teaching of the Cambridge Physiological School ('Foster's Text Book of Physiology,' appendix by Sheridan Lea, pp. 58, 98). The Statutes of the Society (chap. xiii, secs. v. vi.) carefully provide for the action of a majority in the reference of papers. In the case of this paper the position stands that in place of the provision under the statutes for the nomination of referees being complied with, action is taken (and as it happens in an unauthorised way) for the Society, contrary to what is considered ethically becoming, by a representative of the School where the particular teaching with which the paper falls in is adopted."

As the Senior Secretary and (with the exception of the Foreign Secretary) the only physiologist amongst the officers, Professor Foster will not, I think, dispute that it is permissible to look on him as responsible for the action that was taken.

No one, for a moment, will call in question his perfect freedom, outside the Royal Society, to act as he may think proper upon any opinion he may entertain. In his capacity as an officer of the Society, however, he is placed in a different position. It is a controversial matter that is involved, and his writings show that he stands in the position of a partisan in connection with it. These circumstances, I consider, should have influenced his course of action. The paper was dealt with during the vacation, and it is not impossible that convenience may have led to the nomination of referees from the Cambridge Physiological School, among whom a community of thought would be likely to exist. Everyone is aware of the influence produced by a biassed mind; and, that a wrong step was here taken is shown by the expression of regret which the President and Council of the Society have deemed it right to tender.

35, Grosvenor Street,
Grosvenor Square;
December, 1895.

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PHYSIOLOGY OF THE CARBO-HYDRATES.

AN EPICRITICISM.

THE 'Edinburgh Medical Journal' for December, 1894, contains a communication by Dr. Noël Paton entitled "The Physiology of the Carbohydrates—our Present Knowledge of their Relations to the Animal Economy."

In his opening sentence Dr. Paton says, "The appearance of Dr. Pavy's 'Physiology of the Carbohydrates' necessitates a reconsideration of the generally adopted views in regard to the part played by sugar in the animal economy, and in regard to the relationship of the liver to the substance."

A further insight into the origin of Dr. Paton's communication is given by what he says at its conclusion. He had presented a "paper" to the Royal Society in 1893, which was published in the 'Transactions' in 1894. The title of this "paper" is "On Hepatic Glycogenesis;" and as a basis it involves the assumption that the liver is endowed with a glycogenic function. As my work is incompatible with this assumption, to make the road clear it was disposed of, as the following words from the 'Edinburgh Medical Journal' show:—"At the beginning of my paper on Hepatic Glycogenesis, published last winter, before the appearance of Dr. Pavy's work, I said, 'That one of the great functions of the liver is to produce sugar will not, at the present time, be denied by any physiologist,' and added in a foot-note, 'The theory of Pavy, repeated in nearly every text-book, that the liver is "a sugar-destroying, and not a sugar-forming organ," rests on so unsubstantial a basis, and has been so completely refuted by the work of Seegen and other investigators, that it need not be considered.'"

As Dr. Paton's views and mine cannot exist together, either one or the other must fall. My book followed Dr. Paton's "paper," and it took the ground from underneath it. As an act, therefore, of self-defence, it became necessary that an effort should be made to overthrow it. But Dr. Paton from his lofty position condescendingly says, "To let these statements [the above-mentioned statements in his "paper"] stand after the publication of Dr. Pavy's book would be marked discourtesy, and I am therefore glad of an opportunity of bringing forward the evidence upon which they were made." This evidence I will take point by point, and by the time I have finished I think I shall have shown that upon no single point does Dr. Paton succeed in touching, much less shaking, my position. His evidence against me consists, as will be seen, of cullings from the works of others, and in many instances of hasty misrepresentations controverted by the book itself. Nowhere does he in reality adduce anything to refute the validity of my experimental results. There is, perhaps, no subject in physiology that demands such rigorous precautions in experiment and such caution in inference as the one in question. It is one in which the experimentalist is beset with exposure to fallacy in every direction, from which it requires a long training to escape. The literature of the subject is loaded with contradictory statements, due to observers having been often too ready to jump at conclusions upon imperfect and erroneous evidence, and it is not difficult to draw from around whatever is wanted to suit the conception entertained. Dr. Paton's own work contained in his paper in the 'Philosophical Transactions,' as I shall demonstrate when I come to speak of it, stands as a conspicuous illustration of unsound work, and of how easily, when fitting in with preconceived ideas, unsound work may be accepted by others and lead to the propagation of error.

It would have been marked discourtesy, Dr. Paton says, not to have noticed my book, and he was glad of an opportunity for bringing forward the evidence upon which his statement was founded that my work stood so completely refuted that it need not be considered. I also have to say that I am glad that Dr. Paton's criticism of my book has given rise to the occasion for expressing myself upon the controversial points raised by him. In my book I have given an exposition of the system of knowledge which my work has led up to. To have discussed at every step the varied results obtained by others would have broken the continuity and introduced confusion. I took it that what is known of me as a steady worker upon the subject for so many years would, with the detailed description I gave of my mode of experimenting, be taken as affording some sort of guarantee of substantiality in my results. I was mistaken, however, as far as Dr. Paton is concerned. Without adducing a particle of evidence derived from an experimental testing of my work, he instead has sat down, and simply through the medium of his pen attacked me on every side with statements drawn from the heterogeneous mass of writings extant. As I have said, I am glad to respond to the call for meeting these statements; and, if I mistake not, what I have to say will serve materially to strengthen the contents of my book.

In the fourth paragraph of his article Dr. Paton says, "The teaching of Bernard has not only been accepted by physiologists; but it has directed the practice of physicians in the treatment of diabetes for the last thirty years." Let us see what amount of truth is contained in the latter part of this sentence. I think it is a point upon which I may lay claim to a right to speak.

As regards the drug treatment of diabetes, it cannot be said that we have obtained aid or suggestion from any view that may be taken with respect to the disease. What we know has been derived purely from the teachings of experience.

As regards the dietetic treatment, it was also experience, and experience only, which started the principle of action that has been adopted by the profession. Bernard's glycogenic doctrine was made known in 1848. Our countryman Dr. Prout, writing previous to this time (he died in 1850), urged that "the first and chief point to be attended to in the treatment of diabetes is diet;" and the principle he advocated is the same as is acted upon now. Bouchardat, who introduced gluten as an article of food for the diabetic, gave cases in the 'Comptes Rendus de l'Académie des Sciences' for 1841 to show the advantage of its employment.

I have shown that experience, and not Bernard's teaching, has constituted the principle upon which the practice of physicians in the treatment of diabetes has been based. I will proceed further, and show that if Bernard's teaching should be allowed to exert an influence upon the mind, as there has been lately a tendency in the French school to do, the consequences arising are inconsistency and harm.

It will be well here to look plainly at what we have to deal with in diabetes.

Essentially it may be said the disease is the result of a mal-application of carbohydrate matter within the system. No one will deny this assertion. Now comes the question of the seat of application. I say that application takes place in the healthy subject in such a manner, and in such a position, that the opportunity is not afforded for carbohydrate matter to reach the general circulation in a free form for functional transport to the systemic capillaries. The advocates of the glycogenic doctrine say that sooner or later the carbohydrate matter of our food is transported in the form of sugar through the general circulation to the systemic capillaries for utilisation; and it is held that this transportation is wanted as an operation to meet the requirements of life. I shall later on be called upon to speak about the blood in relation to sugar, and the relation of sugar in the urine to that present in the blood. An important train of reasoning, bearing upon the glycogenic theory, is permissibly deducible from the relations existing. Suffice it here to say that the elimination of sugar in diabetes constitutes a result of its pre-existence in the blood, and stands in proportion to the extent of such pre-existence. The severity of the symptoms of the disease goes with the elimination of sugar occurring. Check the elimination of sugar, and the patient becomes reinstated from, it may be, an extreme state of weakness, emaciation, and distress, to a state of health, and as long as the elimination of sugar can be kept down the state of health persists. Cases often present themselves in which, under the exclusion of ordinary starchy and saccharine articles of food from the diet, the disease may be held under control, and the patient kept going for an indefinite time in a perfectly satisfactory way. Even further than this: as a result of treatment, in which the exclusion of starchy and saccharine articles constitutes an essential part, the capacity for partaking of carbohydrate food without leading to the escape of sugar, may in the course of time become to a greater or less extent restored.

I think it will not be denied that the principle advocated by Prout has constituted the guiding principle, up to recent times, of dietetic treatment in our own country. In France, if we substitute Bouchardat's name for Prout, the position is the same. Bouchardat, whom I several times met and conversed with, I know, strongly insisted upon the exclusion as far as practicable of carbohydrate matter from the food of the diabetic.

During the last few years, it is true, the proposition has been started in Paris, based upon the glycogenic doctrine, that starchy matter should be specially made to enter into the diabetic diet. Sugar, it is contended, is required for consumption by the tissues, and carbohydrate food, it is said, should therefore be given to meet this requirement. Potato is named as the form of carbohydrate food that should be administered. The influence of this teaching has not altogether escaped extension to this side of the English Channel, and not long ago I heard at one of our Societies in London a distinguished physician not only saying that potatoes should be given, but entering into minute particulars concerning how they should be cooked!

To plain-thinking persons it will seem rather an irreconcilable procedure in the dietetic treatment of the diabetic to take out the starch from his bread, and to give him starch in the form of potato. Yet this in reality is the position in which the matter stands. France is particularly the country in which the extraction of gluten from wheaten flour is undertaken for home use, and likewise for export. In the face of this, the recommendation is given that potato, an article consisting almost purely of starch, should be specially used as an article of diet by the diabetic.

If we look for a moment into the matter we shall see the unreasonable position in which persons are placed who base their action, in the manner I have mentioned, upon the glycogenic doctrine. That the symptoms and various evils attending diabetes go with the extent of elimination of sugar, is shown by the effect produced when the sugar is brought down by restricting the patient with regard to carbohydrate food. It is the abnormal state induced by the passage of sugar through the system which does the harm, and when this is controlled health becomes restored, and continues restored as long as the control is found to be maintained.

In the face of this, upon the plea that sugar should be supplied to the tissues, carbohydrate matter is to be administered. Although evidence exists, with the voidance of sugar with the urine, that more carbohydrate matter is reaching the system than can be tolerated or properly dealt with, carbohydrate matter is to be further given. The effect produced is simply an increased elimination with a proportionate aggravation of the trouble already present. Is not such a proposition a direct affront to common sense? Undoubtedly, if the sugar has been removed from the urine it is consistent with rational procedure to try the administration of a certain amount of carbohydrate food, and wise to give it as far as it is found to be borne without leading to the appearance of sugar in the urine. Given within the capacity existing for properly disposing of it, good is conferred. Indeed, experience shows that what the system has the power of properly disposing of is wanted by it, and that loss of weight and other ill effects follow if the system does not get it.

To give carbohydrate food when it is already running off as sugar in the urine is alike harmful and opposed to the dictates of common sense. To give it as far as, from the state of the urine, it is found to be tolerated, is rational and beneficial. Withheld at the proper time, the recovery of the power of properly disposing of it is promoted, and cases frequently present themselves in which the power is so far restored, that in the course of time what may be spoken of as a considerable amount of carbohydrate matter can be taken without leading to the escape of sugar; whilst, at the beginning, sugar for a while was voided upon the special restricted diet for the diabetic. The restoration of the power of properly making use of carbohydrate food is the achievement which denotes really successful treatment of diabetes. The state associated with the natural non-saccharine condition of the urine promotes this restoration. The state, on the other hand, associated with the voidance of sugar does exactly the reverse. That these two assertions are true is shown by the growth in the severity of a case that is observed before the patient is placed under dietetic treatment, and by what may be found to occur afterwards.

It is not true, then, that the teaching of Bernard "has directed the practice of physicians in the treatment of diabetes for the last thirty years;" but it may be affirmed that when it is allowed to influence the mind the effect produced is positive harm.

In the next paragraph but one Dr. Paton says, "The fact that Dr. Pavy bases upon his conclusions in regard to the physiology of the carbohydrates a pathology and therapeutics of diabetes, the fact that the acceptance or rejection of his theories must modify the mode of treating diabetes, necessitates on the part of the physician a specially searching scrutiny of the work."

I quite agree that it is only right and proper that my work should be subjected to a searching scrutiny. The work not only stands of importance in relation to diabetes, but likewise in relation to the large question connected with the manner in which carbohydrate matter becomes naturally disposed of in the animal system. Upon this point no advance has hitherto been made since the glycogenic theory was first promulgated, upwards of forty-five years ago. Persons are still groping for information regarding the supposed destruction of sugar as a functional operation alleged to take place in the systemic capillaries. The information is not likely to be obtained seeing, I consider it may be said, that in the healthy state the carbohydrate matter of our food is not permitted to reach the general circulation in a free state for transmission to the tissues. In diabetes it does, however, reach the general circulation in the form of sugar, and as a result gives evidence of the fact by appearing in the urine. Here, in truth, lies the difference between health and diabetes. The glycogenic doctrine, implying as it does passage of sugar into the general circulation, means diabetes. To be free from diabetes, the carbohydrate matter of our food must be disposed of before the opportunity is afforded for its reaching the general circulation in the form of sugar. In proportion as this fails to be effected, so will be the degree of diabetic condition existing.

In saying that I base upon my conclusions, in regard to the physiology of the carbohydrates, a pathology and therapeutics of diabetes, Dr. Paton has reversed the actual state of things. In reality, it has been by observing what transpires where the physiological disposal of carbohydrate does not take place, viz. in diabetes, and the effects of treatment of the condition, that I have been materially aided in attaining the physiological knowledge that has been acquired. The hitherto received physiology has stood inconsistent with the facts supplied by experience connected with diabetes and its treatment, and has thus offered obstruction to advance. The new physiology I have propounded, on the other hand, harmonises with, and has been built up with the aid of, pathology. With the touch thus existing, as pathology in its turn has assisted physiology, so it may be hoped that physiology will now lend assistance to carrying us to a deeper penetration into the pathology of diabetes.

Running on from the sentence I have quoted there comes, " Each piece of evidence must be weighed with impartiality, and each link in the chain of reasoning must be individually tested." Then Dr. Paton continues, "This cannot be done by a mere perusal however careful of Dr. Pavy's book. A knowledge of the immense amount of work which has been accomplished by many other observers is essential, and this is not obtained from his book. For Dr. Pavy gives hardly any consideration of the work of others. Whether he has not studied their work, or whether, having studied it, he has come to the conclusion that it is not of sufficient value to require consideration at his hands, we do not know. But much of that work has been accomplished by, and in the laboratories of, physiologists of reputation, and has been accepted by their *confrères* throughout the world, and we consider that Dr. Pavy would have dealt more fairly with his readers who are not professed physiologists had he placed before them the results of other investigators, and had he attempted to overcome the mass of evidence so opposed to his own views."

I am here pretty sharply and unceremoniously taken to task, but what does it all amount to? In reality it simply constitutes disparagement by innuendo. The real point is whether my experimental work is sound or unsound. If sound, it matters not what may have been the results obtained by others. Dr. Paton adduces nothing in the shape of tangible evidence against me. He simply speaks of the work in general performed by others. But the results that have been obtained by different workers present a very conflicting character. In no other physiological subject, perhaps, is there so much conflicting material. The investigator is everywhere confronted with pitfalls, and to escape from these requires the greatest vigilance. Did not my work start with the disclosure of a fallacy which the greatest physiologist of modern times had overlooked? Ever since, experience has taught me the necessity of being on the closest look-out for fallacy, alike in experimentation and in the conclusions drawn from experiments. Without intending disrespect, I do not deny that it has been my principle through life to rely only on my own work. My book is in no sense a compilation of the work of others. I have kept before me the problem of what becomes of carbohydrate matter in the animal system, and have devised my own line of analytical and other procedure to solve it. I have scrupulously taken precautions to safeguard myself, as far as it has been practicable to do, from error, and have purposely given in full detail a description of the processes adopted, so that they might be put into practice by others. This Dr. Paton has not done. In no single instance has he put my assertions to experimental test, but has contented himself with drawing from the conflicting testimony existing the material that has fallen in with what he wanted.

As I have said, I have devised my own line of analytical procedure, which is not only reliable, but gives an extent of information which cannot be obtained without it. I shall show as I proceed that nothing that is said by Dr. Paton invalidates this statement.

Dr. Paton next proceeds, after entering into a few considerations regarding the chemical nature of carbohydrates, to devote attention to the question of "*Proteids as a source of Carbohydrates.*"

Without seemingly intending it, what Dr. Paton here says, in substance, supports my results. "It is now," he remarks, "universally admitted, on the evidence of the formation of glycogen from proteids, that in the laboratory of living protoplasm carbohydrates may be formed from proteids." Substitute "cleaved off from" for "formed from," and the phraseology conforms with what I have myself used. My contention is that the chemical changes taking place in connection with life result in building up on the one hand, and breaking down on the other. The building up is effected by the synthesising power possessed by living protoplasm. Protoplasmic power is the term applied to it. The breaking down is a process effected by ferment action, and different ferments break down in different ways. For instance, as I have suggested, the proteid molecule built up by the synthesising influence of protoplasmic action from carbohydrate and nitrogenous matter may be subsequently broken down by ferment action of different kinds, with the liberation it may be of glycogen, it may be of sugar, or it may be of fat. There is certainly nothing inconsistent with carbohydrate entering the proteid molecule as sugar and coming out in another form. Indeed, it is what is considered to take place in the course of the operations of vegetable life. For instance, cellulose, starch, inulin, &c., are derived from the sugar of the sap. It is through the instrumentality of protoplasm that the transformation is effected, and the view regarded as rationally to be entertained is that the sugar is first synthesised with a nitrogenous compound — asparagin mainly—into proteid, and that from this the carbohydrate is cleaved off in the particular form that is come across. The same kind of reasoning may be applied to the production of fat, the difference simply being that through the influences existing fat instead of carbohydrate is cleaved off from the proteid molecule. There is nothing chemically inconsistent, indeed on the contrary, with the proposition that carbohydate may enter the molecule, and fat be cleaved off. As I suggest in my book, and give grounds for the view, the fat, lactin, and casein of milk may all constitute cleavage products from the proteid or protoplasmic matter of the mammary gland cells. In the section on the glucoside constitution of proteid matter I show that casein differs from albumin, fibrin, &c., in not yielding, under chemical treatment, cleavage carbohydrate, which tallies with its being already a cleavage product.

The facts adduced by Dr. Paton indicating the formation of carbohydrate from proteids, or, as I should express it, the breaking down of proteids with the setting free of a carbohydrate, strictly harmonise with the view advocated in my work. He refers first to von Mering's experiments on phloridzin diabetes, wherein it is shown that diabetes may be produced under conditions to render it presumably evident that the sugar must be derived from a breaking down (disintegration is Dr. Paton's word) of the proteids of the body, and secondly to the appearance of glycogen in the liver and muscles under the influence of chloral in the unfed animal, after disappearance has been caused by strychnine poisoning. He says, "How is this brought about? Does the proteid molecule contain carbohydrate molecules, which are set free when it breaks down; or must the elements of the proteid molecule be more completely broken down, and then, after becoming part of the protoplasm, be changed into the carbohydrate molecule?"

Dr. Paton, then, here distinctly avers upon experimental grounds that proteid is susceptible of breaking down in such a manner as to yield carbohydrate. In phloridzin diabetes sugar is liberated. The fact exists that sugar becomes largely evident, and I suppose it may be assumed that through ferment agency, either directly due to the phloridzin or indirectly created by it, the result is brought about. The condition seems to be comparable to that noticeable in association with diabetes of pathological origin, where sugar is manifestly being derived from a breaking down of the tissues. In these instances it is sugar that is produced; and this, by virtue of the property of diffusibility belonging to it, becomes taken up by the blood and eliminated with the urine. In Dr. Paton's second illustration it is glycogen instead of sugar that is produced, and let us note the difference as regards further result. Sugar with its diffusibility cannot do otherwise than tend to escape. Glycogen, on the other hand, with its colloidal character stands in a diametrically opposite position. As a non-diffusible principle its production is followed by accumulation, and it must be brought under some kind or other of collateral influence for it to be made to disappear. I believe it is this form of carbohydrate which results from the natural breaking down of proteid, and that in association with the diabetic condition above illustrated circumstances happen to exist which lead to the splitting off of sugar instead of glycogen.

Does the proteid molecule, asks Dr. Paton, as I have already cited, contain carbohydrate molecules, which are set free when it breaks down; or must the elements of the proteid molecule be more completely broken down, and then, after becoming part of the protoplasm, be changed into the carbohydrate molecule?

We are led in the latter part of Dr. Paton's sentence into a rather tangled position. Proteid is the chemical basis of protoplasm. The proteid molecule, instead of being broken down and yielding carbohydrate as a cleavage product, in accord with what I hold I have shown to occur as a result of the action alike of chemical agents and ferments (gastric digestion), is supposed to undergo more complete dissolution, and in this imaginary process to yield products which are assumed to go back into protoplasm (living proteid), to be built up into carbohydrate. I fear Dr. Paton has here got not only beyond the range of fact, but into a region too cloudy for the human understanding usefully to attempt to penetrate.

Continuing, Dr. Paton says, "According to Pflüger ('Pflüg. Arch.,' 1888), the latter [supposition expressed in his problem] is the probable explanation. He points to the fact that in the formation of urea, not only is the proteid molecule broken down, but the derived ammonia bodies have to be built up to form urea. In the formation of fats from carbohydrates the same breaking down and reconstruction must occur. The same form of glycogen is produced from dextrose, from lævulose, and from proteids; and hence the protoplasm must either first make these substances part of itself before changing them to glycogen, or without making them part of itself must convert them to some common forerunner of glycogen, like glucose."

The opinion of Pflüger here put forward does not amount at the most to anything more than conjecture. Moreover the facts revealed in what I have recently said about the glucoside constitution of proteid matter were not available at the time the view was expressed. "In the formation of fats from carbohydrates the same breaking down and reconstruction must occur." Not at all so. The carbohydrate may enter the proteid, and the fat afterwards be cleaved off. This, as I have given grounds for believing, is really what occurs. Again, as regards the same form of glycogen being produced from different sugars and proteids, this fits in exactly with the doctrine I have advanced. It is quite consistent that different forms of carbohydrate may enter the proteid molecule whilst a given form is cleaved off. It is further consistent that a given form may enter, and different forms, according to the circumstances existing, be cleaved off,—as, for example, with cellulose, starch, inulin, &c., in the case of the vegetable kingdom.

"But Dr. Pavy attempts to furnish evidence that the proteid molecule does contain a carbohydrate moiety, which is the source of carbohydrates formed from proteids."

"Attempts" to furnish evidence! I nowhere for a moment hint that carbohydrate exists in proteid to the extent of a moiety, but I do something more than attempt to furnish evidence that it is present in what may be spoken of as a not insignificant amount. Dr. Paton in his next paragraph enumerates the cardinal points of evidence I have brought forward in support of my contention. To an open mind the evidence adducible of the product constituting a carbohydrate will be admitted to be sufficient, but Dr. Paton makes endeavours to resist, and I will proceed to show what his endeavours amount to. Before doing so I may here summarily state that the evidence comprises the cleaving off of carbohydrates by potash in the amylose form, and by sulphuric acid and pepsin digestion in the form of sugar. The amylose form agrees with Landwehr's "animal gum" obtainable from mucin. In its conversion into sugar by boiling with sulphuric acid the cupric oxide reducing power becomes gradually raised as with other amyloses, and the character of the crystalline osazone obtainable from the product taken at successive stages of the conversion conforms with the characters of the crystals from the known sugars standing as intermediary bodies between the ordinary amyloses and glucose. The sugar derived alike from the amylose material and directly from the proteid by the action of sulphuric acid agrees in minute detail with sugar as ordinarily met with. The only negative characters presented are as regards fermentability and optical activity.

As regards fermentability, it is known that it is by no means an essential property for a sugar to possess. Neither Landwehr's sugar (gummose) obtained from mucin nor the sugar obtained by Kossel from nuclein is fermentable. It therefore even stands in accord with what might be looked for that the sugar obtained by me should be non-fermentable.

As regards optical activity, this, as I show in my book (pp. 41, 52), is a property in connection with which much diversity exists. It is known that some sugars are absolutely inactive, whilst others may be inactive through the neutralising effect of the presence of varieties with opposite rotatory powers.

In his next remark Dr. Paton shows that he has fallen into serious misconception. He says, apparently oblivious of what he said just previously about the carbohydrate amounting to a moiety, "According to Dr. Pavy the amount [carbohydrate from proteid] is small, only about 2 to 3 per mille." What Dr. Paton here says is drawn from what I have stated (pp. 65, 214) about the difficulty created in the estimation of glycogen as a constituent of the components of the body by the cleavage product from proteid passing with the glycogen in the potash process of analysis. I mention that the amount of cleavage material, expressed as glucose, included in the figures given cannot be computed to stand at more than about two or three per 1000 *of the fresh tissue examined.* But Dr. Paton is talking in his remarks about the carbohydrate obtainable from the proteid molecule, which is quite a different matter. If he refers to p. 31 of my book he will find that egg-albumin in the water-free state yielded me thirty per 1000 of product reckoned as glucose. This was with the employment of 2 per cent. acid for the conversion of the primary non-reducing material into a cupric oxide reducing sugar. At p. 37 I show that with 10 per cent. acid the cupric oxide reducing capacity became nearly doubled, so that to this extent the figures representing actual material have to be raised. This brings them to sixty per 1000, which is very different from what Dr. Paton through misconception has represented. Further, at p. 220 I state that there are grounds for believing that this falls short of representing the whole of the carbohydrate locked up in the proteid taken. It seems to me, with the agency of the direct action of sulphuric acid upon the proteid, that a considerably larger quantity of carbohydrate is obtainable than through the potash process. At all events, with only a moderate quantity of proteid taken osazone crystals settle out in a complete shower when the product after treatment with phenyl-hydrazin is removed from the water-bath and poured into a glass to cool. There is not the slightest difficulty in obtaining any amount of the carbohydrate product that may be required for experimenting with, but strong agents must be brought to bear, alike for splitting up the proteid and the subsequent carrying of the cleavage product towards glucose. In a recent instance I kept the proteid boiling in 10 per cent. sulphuric acid with the employment of the inverted condenser for eight hours, and the product yielded me an osazone which separated out whilst on the water-bath, and in other respects more closely resembled typical glucosazone than any I have previously obtained.

"The fact," continues Dr. Paton, "that the percentage composition of peptones is the same as that of the native proteids from which they are derived is opposed to the view that any molecule rich in carbon is split off in this manner by peptic digestion." To this I reply that there is no fixed or absolutely definite composition given for the group of proteids, and that the range admitted to exist is sufficiently wide to allow of the splitting off of a certain amount of carbohydrate without interfering with the view propounded. Moreover Dr. Paton is confronted with the actual fact that a "something" presenting the characters of a carbohydrate is in reality obtainable.

"The existence of a carbohydrate molecule in mucin has been already demonstrated by Landwehr. But mucin and its allies are really combinations of a proteid with a carbohydrate."

No exception can be taken to this statement reading it as meaning that the carbohydrate is incorporated as an integral part of the molecule. The position to be contended for is clearly expressed in the following quotation from Dr. Sheridan Lea's appendix, published in 1892, to Foster's 'Text-book of Physiology' (p. 76) :—"Notwithstanding the views which have frequently been advanced that mucin is in reality a mixture of proteid and carbohydrate material, it is now known with considerable certainty that it is a unitary substance which, from what has been already said, might be almost regarded as an animal glucoside." I would simply add to this that I believe I am right in saying that opinion has advanced since the above was written and that by common consent it *is* now regarded as an animal glucoside.

"It is *possible* that the proteid molecule in splitting *may* yield a carbohydrate. But we have to consider the question of whether the evidence offered *proves* that such a carbohydrate molecule is yielded. Now, the carbohydrates are not the only bodies which yield crystalline compounds with phenyl-hydrazin. A glance at Beilstein's *Organic Chemistry* shows that a vast number of substances give such crystalline compounds. It is only when an elementary analysis of the compound is made that it can be definitely concluded that a carbohydrate is a constituent part. Such analyses are given by Baisch in his researches on the carbohydrates of the urine, and by Bial in his observations on the sugar produced by the ferment of the blood, and the conclusions of these authors are thus placed beyond doubt. But Dr. Pavy does not give such analyses, and without them we are not justified in accepting his conclusion that the substance is a carbohydrate."

Reference need not be specially made to Beilstein's 'Organic Chemistry' to learn that there are other substances besides the cupric oxide reducing sugars which give crystalline osazones with phenyl-hydrazin. It is now stock knowledge what kind of bodies do so, and that they are restricted to certain groups possessing a particular molecular constitution.

It may be said that neither of them besides the carbohydrate could live under the treatment to which my cleavage product is exposed,—namely, first of all boiling with 10 per cent. potash solution, and then with 10 per cent. sulphuric acid before being brought under the influence of the phenyl-hydrazin. This is a point which perhaps has not been considered by Dr. Paton.

As regards what is said with respect to elementary analysis, I should have thought the whole surroundings would have sufficed to satisfy the mind that the osazone obtained could not be otherwise than one derived from a carbohydrate. However, Dr. Paton's suggestion has led me to seek competent assistance to meet the alleged requirement.

Mr. A. R. Ling, Fellow of the Chemical Society, and of the Institute of Chemistry, &c., who has been for some time past engaged in the study of the carbohydrates has kindly undertaken to perform the task, and I will leave him to speak for himself regarding the results of his work.

Note on a Product of the Hydrolysis of Egg Albumin.

By Arthur R. Ling.

It has been shown by F. W. Pavy ('Proc. Royal Soc.,' 1893, **54**, 53; also 'The Physiology of the Carbohydrates,' 1894, pp. 27—57) that when egg albumin is boiled with a 10 per cent. solution of potassium hydroxide, one of the products is an amorphous substance, closely resembling, if not actually identical with, Lanwehr's so-called animal gum obtained by the action of alkalis on mucin ('Zeit. Physiol. Chem.,' 1881, **6**, 75; 1883, **8**, 122).

As in the case of Landwehr's animal gum, the substance obtained from albumin was found to yield, when heated with 10 per cent. sulphuric acid, a solution having cupric oxide reducing properties, and also possessing several other characters in common with a solution of ordinary glucose. These characters have been already fully described by Dr. Pavy in the work referred to; but, in order to render this portion of the subject quite clear, I propose to give here a brief summary of them.

If the cupric oxide reducing solution obtained from proteid matter is freed from sulphuric acid by means of barium carbonate, and the filtrate evaporated to dryness, a residue is left which presents the appearance of a sugary extractive, and has a pronounced baked-sugar odour. A solution of the substance darkens in colour on being boiled with alkalis (Moore's test); it dissolves hydrated copper oxide in the presence of excess of alkali (Trommer's test); and gives a strong neat sugar reaction (reduction) with Fehling's solution. The substance also gives the furfuraldehyde reaction with concentrated sulphuric acid and phenols.

It should here be mentioned that Dr. Pavy also finds that when egg albumin is boiled with 10 per cent. sulphuric acid direct, the cupric oxide reducing substance may be formed in a single operation.

Foremost among the reactions exhibited by the cupric oxide reducing substance from proteid matter should, however, be mentioned its ability to yield a crystalline compound when heated with phenylhydrazine acetate. This crystalline compound, moreover, bears the closest resemblance to the phenyl-osazones of the sugars, discovered by Emil Fischer.

These facts appear, therefore, to point unmistakably to the view that the cupric oxide reducing substance in question is a sugar, and such indeed is the opinion very justly expressed by Dr. Pavy, who draws inferences from his experiments of great importance from a physiological point of view, which need not, however, be discussed by me.

Despite the above-mentioned positive evidence thus brought forward by Dr. Pavy in support of the view that the cupric oxide reducing substance is a sugar, this conclusion has been called into question and, to some extent, challenged by D. Noël Paton in a polemical paper published by him in the 'Edinburgh Medical Journal' (ibid., December, 1894). One of the complaints raised by Dr. Paton is that no analytical data as to the composition of the osazones have been furnished by Dr. Pavy, and, in the absence of such evidence, he is unable to accept the conclusion that the osazone is derived from a sugar, since many other compounds besides sugars are capable of forming osazones.

It would be encroaching on ground which properly belongs to Dr. Pavy were I to attempt to answer one of the objections raised by Dr. Paton. At Dr. Pavy's suggestion, however, I have undertaken the examination of several samples of the osazone, prepared in his laboratory in the manner already described, with the view of establishing the composition of the substance and of further characterising it. I have also personally studied the mode of formation of the osazone, and have prepared several quantities of it, my preparations being in every way identical with Dr. Pavy's.

I now propose to describe my experiments, and to state the conclusions which I am able to draw from them.

I. *Osazone prepared from the Product of the Successive Action of Potash and Sulphuric Acid on Albumin.*

Sample A.—This consisted of bunches or sheaves of light yellow well-defined needles, and melted at 205° to 206° with decomposition. It contained a minute trace of ash. Analysis of the substance, after being dried at 100°, gave the following results :

(I) 0·1576 gave 0·3474 CO_2 and 0·0856 H_2O.

(II) 0·1101 gave 15·25 c.c. moist nitrogen at 24·5° and 757 mm.

(III) 0·1148 gave 15·95 c.c. moist nitrogen at 23·5° and 752·5 mm.

	Calculated.	Found.			
	$C_6H_{10}O_4(N_2HC_6H_5)_2$	I.	II.	III.	
C	60·33	60·11	—	—	per cent.
H	6·14	6·03	—	—	,,
N	15·64	—	15·41	15·44	,,

A portion was re-crystallised from alcohol in presence of extracted animal charcoal. It was then much brighter in colour, and melted with decomposition at 207° to 208°.

Sample B.—Dark pasty crude substance. After being several times recrystallised from 83 per cent. alcohol, and treated with extracted animal charcoal, it was obtained in a form identical with that of the last-described substance, and in colour of a light canary-yellow. It melted at 207° to 208° with decomposition.

Sample C.—Brownish yellow needles of melting-point 195°. After purification by recrystallisation and treatment with extracted charcoal, it agreed in all respects with the other specimens.

II. *Osazone prepared from the Product of the Direct Action of Sulphuric Action on Albumin.*

Sample A.—This was repeatedly recrystallised from alcohol and treated with extracted charcoal, when it melted at 207° to 208° with decomposition, and was, to all appearance, identical with the specimens above described. Analysis of a specimen dried at 100° yielded the following values:

0·1744 gave 0·3841 CO_2 and 0·0962 H_2O.

	Calculated. $C_6H_{10}O_4(N_2HC_6H_5)_2$.	Found.
C	60·33 .	60·06 per cent.
H	6·14 .	6·13 „

Sample B.—This, after the usual purification, melted at 209° to 210° with decomposition. Analysis gave the following result:

0·1089 gave 15·00 c.c. moist nitrogen at 22·5° and 769·5 mm.

	Calculated. $C_6H_{10}O_4(N_2HC_6H_5)_2$.	Found.
N	15·64 .	15·75 per cent.

III. *Osazone prepared by myself from the Product of the Successive Treatment of Albumin with Potash and Sulphuric Acid.*

The method adopted by me is essentially the same as that described by Dr. Pavy (loc. cit.) thus: Egg albumin (50 grams) is suspended in 10 per cent. potassium hydroxide (300 c.c.), and allowed to remain therewith for about twelve hours, after which, as a rule, the albumin has completely dissolved. The solution is now boiled under a reflux condenser for thirty to forty-five minutes, and subsequently rendered slightly acid with acetic acid and filtered. The filtrate is evaporated to a syrup, which is mixed, while hot, with alcohol, and the somewhat limpid liquid poured in a thin stream into 95 per cent. alcohol (2 litres). A precipitate falls, which is at first flocculent but soon becomes syrupy, and ultimately, after remaining for about twenty-four hours, settles on the bottom of the containing vessel as a brownish glaze. The alcoholic liquid is now decanted, the glaze drained, and taken up with 10 per cent. sulphuric acid (220 c.c.) After boiling for six hours, the liquid is diluted with water, and the sulphuric acid exactly precipitated by barium hydroxide. The filtrate is evaporated and boiled with extracted animal charcoal. A portion on being now titrated with Fehling's solution, is generally found to give an apparent sugar content, for the whole product, calculated as glucose, of 2 grams. The solution is next introduced into a boiling-tube and heated in a bath of boiling water for about two hours, with phenylhydrazine (2 grams) and 50 per cent. acetic acid (2 c.c).

The osazone obtained in this way from a number of experiments agreed in every respect with the substance prepared by Dr. Pavy, and, moreover, when purified by repeated recrystallisations from alcohol in presence of extracted animal charcoal, it melted at 207° to 208° with decomposition. A specimen dried at 100° was analysed with the following result:

0·1779 gave 23·70 c.c. moist nitrogen at 17° and 768 mm.

	Calculated. $C_6H_{10}O_4(N_2HC_6H_5)_2$.	Found.
N	15·64 .	15·65 per cent.

I have submitted all the various preparations of the osazone, including those prepared by Dr. Pavy, to repeated recrystallisations from 80 per cent. alcohol, and find that after this treatment they all melt quite constantly at 210°—212°, at the same time frothing up and decomposing.

The analytical data which I have given prove therefore beyond all doubt that the compound, obtained by treating the cupric oxide reducing substance, produced by the hydrolysis of proteid with phenylhydrazine acetate, has the composition of ordinary glucosazone from glucose. The similarity in general appearance and physical properties of the compound under discussion with those of glucosazone are such that the two must be regarded as belonging to the same class of chemical compounds. So far, therefore, Dr. Pavy's conclusions are completely confirmed. The somewhat higher melting-point, optical inactivity, and slightly different crystalline appearance of the new osazone as compared with glucosazone are sufficient to show that it is not identical with the latter. It exhibits to a marked degree the tendency of forming supersaturated solutions, in which respect it again differs from glucosazone. As far, however, as my own personal experience goes, the osazone derived from galactose also possesses the character of forming supersaturated solutions.

In conclusion, I wish to state that with Dr. Pavy's concurrence I intend to continue the study from a chemical point of view of the apparent carbohydrate compounds produced by the hydrolysis of proteïd. I venture to hope, therefore, that I shall be in a position at no distant date to give a detailed account of further experiments on this most interesting problem.

It is my pleasure to express my thanks to my colleague, Mr. Julian L. Baker, for the assistance he has rendered me during the prosecution of these experiments.

Next, Dr. Paton tries to deprive my views of the support which Hammarsten's and Kossel's researches have been asserted ('Brit. Med. Journ.,' July 14th, 1894) to give to them. He says, "The researches of Hammarsten and Kossel, which at first sight might appear to support this theory [glucoside constitution of proteid matter], are found upon careful examination to have no bearing upon it. Prof. Hammarsten ('Zeitsch. f. phys. Chem.,' Bd. xix, p. 19) has recently described a product of the splitting of the complex nuclein* of the pancreas, which yields a crystalline osazone, which reduces Fehling, and which he considers to be a carbohydrate, although he has been unable to prepare it in sufficient quantity for analysis. Kossel ('Arch. f. Phys.,' 1893, p. 157) [the reference should stand 'Arch. f. Anat. u. Phys.'] shows that the carbohydrate which he is able to obtain from the nuclein of yeast is derived not from the proteid, but from the nucleic acid part of the substance. Thus the researches of these two chemists cannot be considered as giving support to the theory that the proteid molecule contains a carbohydrate part."

My German *confrère* Dr. Grube has looked into this matter very carefully, and disputes Dr. Paton's assertion that the meaning to be attached to Hammarsten and Kossel's writings does not support my work. An epitomised representation approved by Dr. Grube, and framed from the digest made by him of what is said by these authors, may be given in the following terms :—

* "Nucleins are compounds of a complex phosphorus-containing acid, nucleic acid, linked to varying quantities of a proteid."

Hammarsten endeavoured to ascertain if there were other glucoproteids (glucosides) in the body besides the mucins and mucoids, and found that he could isolate a proteid from the mammary gland and pancreas, which on boiling with a dilute mineral acid gave a substance that possessed cupric oxide reducing power, and formed a crystalline osazone with phenyl-hydrazin. Considerations connected with the proteid derived from the pancreas led him to regard it as a body of a higher stage of complexity than an ordinary proteid, and to speak of it as a nucleo-proteid. Kossel says the albuminous substances do not, in the free state, take part in the most important functions of the cell, but constitute components of more complex compounds. In each cell substances can be demonstrated to exist which contain an albuminous base and a conjugated group of atoms. Hoppe-Seyler, he states, first recognised the nature of these substances, and called them "proteids." Nucleins are substances composed of a phosphorus-containing group, nucleic acid, associated with the albuminous base, and are split up by alkalies into these component parts. The nucleic acid derived from yeast, when split up with dilute acids, yields a carbohydrate. If proteids are looked upon from a general point of view as albuminous bodies, it is permissible to say that the formation of sugar (in accord with my own work the sugar is stated to be non-fermentable) from albumin by chemical action (durch Umsetzung) has been in several instances demonstrated, but in all instances in which the demonstration has been accomplished the carbohydrate did not come from the albuminous base (Eiweisskern), but from the associated group (prosthetische Gruppe). The proteid, in the clearest manner possible, is stated by Kossel to consist of an albuminous part *plus* an associated conjugated group.

Looking at what is here said, I have little doubt the reader will dissent from Dr. Paton's suggestion that the researches of Hammarsten and Kossel, although, as he admits, at first sight appearing to support the views I have advanced regarding the glucoside constitution of proteid matter, are found upon careful examination to have no bearing upon them. In his attempt to annul the effect of evidence standing in confirmation of my work, Dr. Paton has brought an amount of zeal into play that has led him to over-reach himself in the process of reasoning adopted.

For example, in his foot-note he says nucleins are compounds of a complex phosphorus-containing acid, nucleic acid, linked to varying quantities of a proteid. I would here ask, does the proteid cease, looked at as a whole, to be a proteid on being raised in complexity by the linking referred to? Such would be contrary to the view entertained by the authors he is arguing upon. Again, Kossel, he says, shows that the carbohydrate which he is able to obtain from the nuclein of yeast is derived not from the proteid, but from the nucleic acid part of the substance. Not from the proteid? What is the proteid here, under the meaning of Kossel, but in truth the nuclein? To have expressed it correctly, Dr. Paton should have said "not from the albuminous part of the proteid," instead of "not from the proteid."

I now put the question, upon what kind of ground does the concluding sentence, "Thus the researches of these two chemists [Hammarsten and Kossel] cannot be considered as giving support to the theory that the proteid molecule contains a carbohydrate part," in Dr. Paton's paragraph stand? I think I may leave the answer in the hands of others.

But there is no longer ground for quibbling upon the point. Dr. Paton in speaking of the osazone that I have obtained from egg-albumin says that the carbohydrates are not the only bodies which yield crystalline compounds with phenylhydrazin, and that it is only when an elementary analysis of the compound has been made that it can be definitely concluded that a carbohydrate is a constituent part. With the analyses given on the preceding pages I presume Dr. Paton will allow that the question he has raised may now be regarded as settled.

Before quitting this subject I should like to take the opportunity of recording my concurrence with the views expressed in the writings of the authors quoted regarding the specifically recognised principles constituting educts of more complex compounds which stand as the agents concerned in the operations of life. However necessary it may be in the case of chemistry unconnected with living operations to insist on isolation and purification of the bodies dealt with, in the case of physiological chemistry it must be borne in mind that the instability of the bodies concerned is such that in the process of treatment for isolation and purification we may be carried away to products of change, and thereby be dealing with these in place of the original body. Upon these grounds a too rigid enforcement of a rule of thumb procedure as a basis of action in connection with the study of the chemistry of life may impede, in the prosecution of research, penetration to the phenomena standing closest to the operations of life. At the same time I do not deny that advantage is derivable from getting, as far as practicable, at the constituent bases of the complex compounds referred to.

The next point dealt with relates to the methods of analysis for the carbohydrates. Dr. Paton says, "By Dr. Pavy's method of extracting the sugar by alcohol, two dangers are run. First, that all the sugar is not removed (Schenk, 'Pflüg. Arch.,' Bd. xlvi); and, second, that by using strong alcohol the sugars less soluble in alcohol, such as maltose, are not completely taken up, or by using a weaker alcohol that some of the lower dextrins are also taken up with the sugar. We believe that this will in part explain the curious results obtained by Dr. Pavy on boiling his so-called sugars with sulphuric acid,—results which lead him to conclusions as to the nature of the sugars in opposition to other chemists. There is also the danger that the alcohol may take up from the blood or tissues reducing substances other than sugar."

"In the estimation of glycogen the researches of Vintschgau and Dietl ('Pflüg. Arch.,' Bd. xiii, p. 253), which show that boiling with potash does destroy glycogen, are ignored (p. 152)."

There is no justification for the remarks that are here made. I plainly state (p. 61) at the commencement of the section devoted to the analytical procedure that either alcohol or methylated spirit is to be used. Not a word is said anywhere to suggest that diluted spirit was employed. As to all the sugar, whether maltose or not, not being taken up: this constitutes a gratuitous innuendo, for I enter in detail into the question of reliably securing the full extraction of the sugar, and show by subsequent aqueous extraction that after the process recommended to be adopted no sugar is left. The matter was carefully considered by me many years ago, and dealt with at length in the 'Proceedings of the Royal Society' for 1881. From this source I quote illustrative examples in my book.

The suggestion about the lower dextrins being taken up: what does this amount to? Strong spirit being used glycogen escapes solution and thus does not enter into the question. It thence comes that the product of alcoholic extraction possessing a cupric oxide reducing power below that of glucose affords unmistakable evidence that there is present carbohydrate matter in an intermediate stage, or in intermediate stages, between glycogen and glucose. More than this, and whether the carbohydrate matter in question falls under what is reckoned as dextrin or as sugar the means of differentiation at present at our disposal do not allow us to determine. The figures show that the product contains carbohydrate other than glucose: whether as sugar or dextrin is not indicated. The remark that alcohol may take up from the blood or tissues reducing substances other than sugar has no more force than it would have in the case of water—perhaps even not so much.

Notwithstanding, then, the attempts of Dr. Paton to represent otherwise, there is nothing to detract from the value of alcohol for service in the manner I have recommended. Indeed, it may be considered to be quite as necessary to definitely know the nature of the sugar that is being dealt with in connection with animal as well as vegetable products. What chemist would allow himself to remain ignorant of whether he was dealing with maltose or glucose in a product before him from the vegetable kingdom? The quantitative estimation by cupric oxide reducing action is entirely dependent upon a knowledge of the nature of the material under examination. The cupric oxide reducing power of maltose and glucose stand as 61 to 100. A given cupric oxide reducing effect obtained, indicating say the presence of 61 milligrammes of carbohydrate, would, if the carbohydrate consisted of glucose, furnish a correct representation of the amount of material present.

Should the carbohydrate happen, however, to consist of maltose, the same cupric oxide reducing effect would in reality be the representative of 100 milligrammes, instead of 61, of carbohydrate material. Hence, unless a process of extraction be adopted permitting of the nature of the carbohydrate present being ascertained, no proper estimation of its amount can be effected. Watery extraction by taking up glycogen would introduce a vitiating factor into the question.

The glycogen, it is true, might be subsequently removed by the Schmidt-Mulheim process of precipitation with perchloride of iron and sodium acetate, but the presence of the sodium acetate would afterwards interfere with the accomplishment of what is wanted by the converting action of sulphuric acid. For the sulphuric acid to act it is necessary that it should be free. Brought into contact with sodium acetate, it would be appropriated by the base and acetic acid set free; and on account of the unknown quantity of the sodium acetate present it would be difficult if not impossible to properly adjust the amount of sulphuric acid to be employed to meet the requirements of the case. We are thus driven to the alcoholic process of extraction as the only suitable one for our purpose, and it fortunately happens that it readily and reliably enables us to secure the attainment of the object desired.

Now, as regards the remark indirectly discrediting the potash process of analysis for the estimation of glycogen by reference to the researches of Vintschgau and Dietl, let me at once state that the work of these authorities—and the same may be said of the work of Külz—does not stand in a position to bear comparison with my own, from the length of time, viz. two to three hours, boiling was carried on. Külz directs two to three hours for liver, and six to eight hours for muscle.

Does Dr. Paton think that I took no steps to previously prove my ground, but proceeded simply in a haphazard way? If so, I will tell him that I investigated for myself the effect of boiling glycogen with different strengths of potash solution and for different lengths of time, and that it was upon the strength of this investigation that my recommendation was founded. The object of the process is to bring the substance under examination into a state of solution, and to sufficiently alter the albuminous matters to prevent their falling as such with the subsequent treatment with spirit. Unless this is accomplished, unchanged albuminous matter would be present during the part of the procedure embracing conversion into glucose by sulphuric acid, and afterwards give rise to the production of the obscuring effect of a biuret reaction in the estimation of the glucose by the ammoniated cupric test.

Minuteness of subdivision is an important factor in aiding solution in potash. In my process, which involves previous extraction with spirit, the material is easily rubbed down in a mortar to a fine powder; and, as I have learnt in the course of my investigations on the glucoside constitution of proteid matter, when really finely reduced a less strength of potash solution and less length of time of boiling than I specified in my directions will suffice for effecting what is wanted. In the absence of the preliminary treatment with alcohol the substance is in a state to be only slowly penetrated and attacked by the potash solution, hence the length of time that other workers have found to be necessary for getting their material dissolved. There is a further point of much importance. Other workers have conducted the operation in an open vessel. I did so many years ago, and then obtained discordant results. This showed that something faulty existed, and in reality led up to the employment of the inverted condenser, which I consider a necessary part of the operation.

To show that there is no sensible or material destruction of glycogen by the application of my process, I may give the particulars of a test experiment recently performed. Some glycogen derived from the liver of a rabbit, which, having been extracted by the potash process, may be spoken of as free from dextrin, was taken and dissolved in water. 40 c.c. of the solution were simply put into about 500 c.c. of methylated spirit for reyielding the glycogen in a precipitated form. Another 40 c.c. were boiled for half an hour, with potash added to bring to 10 per cent. strength, and then poured into a corresponding quantity of spirit, and acetic acid added for the purpose of bringing to a neutral state. The precipitate from the first 40 c.c. of the solution soon settled out in a flocculent form, leaving a perfectly bright supernatant liquid. The precipitate from the other, as is usual after boiling with potash, fell in a finely divided form and took time to settle. In three days' time the liquid was clear, and the precipitates were now collected, treated with sulphuric acid for conversion into glucose, and the glucose estimated. The glycogen shown to have been recovered amounted, in the first case, to ·166, and in the second ·162 grm. The amount of glycogen recovered after subjection to boiling with potash was thus only four milligrammes less than after simple precipitation without exposure to the action of potash. The difference stands at 2·4 per cent.

In another like experiment the glycogen recovered after boiling with potash was ·110 against ·112 grm., which gives a loss of 1·8 per cent.

As I have mentioned in my work (p. 152), the dextrins fail to resist the action of potash in the same way as glycogen and starch, so that should either of these latter principles be to ever so slight an extent changed, a more or less marked loss will be observable on boiling with potash. The glycogen operated upon in the above experiments had been obtained by treating the liver with potash. With glycogen obtained by aqueous extraction and the application of Brücke's method, I found that a large loss occurred under subsequent treatment with potash. Brücke's process involves the use of strong hydrochloric acid, and it is usual in books to find that the process is directed to be conducted at a temperature reduced by the aid of ice, without its being stated whether this is for diminishing the liability to change. I have not had recourse to the employment of ice but without it the effect of the application of Brücke's process to glycogen that has been prepared with potash is to place it in a condition to yield in a decided manner to subsequent boiling with potash. For instance, in one experiment the amount of glycogen recovered after simple precipitation was ·142 against ·114 grm. after boiling with potash, showing a loss of 19·4 per cent. In another experiment the figures showed even a greater loss.

I am glad the occasion has arisen for entering as I have done into these several particulars bearing upon the question of analytical procedure. I and every one must admit that the matter stands in a position of fundamental importance. The difficulty of extracting glycogen simply by the agency of water is acknowledged by all to be very great, and my own experience shows that, however the process may be carried out, it is not allowable to conclude that extraction has been fully effected without the confirmatory evidence afforded by the subsequent application of the potash process to the residue. At the best, aqueous extraction is a lengthy, tedious, and even, it may be said, laborious operation, and one requiring to be almost without break carried on. With the potash process, on the other hand, nothing could be more easy and simple; and there is the further advantage that at any stage the proceeding may be stopped, to suit the convenience of the operator, without fear of harm arising.

I have spoken of the effect of boiling with potash leading to glycogen falling in a very minutely divided form under alcoholic precipitation. This of course requires to receive attention to prevent loss from suspension in the filtrate being mistaken for destruction. It seems as if a molecular alteration occurred. The solution loses much of the lactescence it possessed before subjection to the influence of the potash, and it is also to be remarked that boiling with acetic acid and even prolonged boiling with water give rise to a similar kind of result. When analysis is being conducted of animal products, the associated matter that falls on precipitation with spirit helps to carry down the finely divided glycogen, and thus places matters in a more satisfactory position than where glycogen in a pure form is being operated upon.

Taking now the method of analysis as a whole—that is for both sugar and glycogen—it stands, I think it may be justifiably said, as the only satisfactory procedure yet described for giving reliably and fully the information that may be considered to be required.

Upon the point of what form of sugar is found in the portal blood, Dr. Paton correctly says that I find a sugar which has its cupric oxide reducing power increased by boiling with sulphuric acid. He then proceeds to state, " The fact that in the same experiment this increase of reducing power varied from 10 to 20 or 30 per cent., militates against any definite conclusions being drawn from the observations." There are thirty-eight observations in all given. In five, two portions of blood were collected and analysed, the flow, after the collection of the first, being for a few minutes stopped by compression of the vessel before the collection of the second. Three were after animal food, and the cupric oxide reducing power stood in the first as 83 to 82, in the second as 82 to 73, and in the third as 96 to 85. One was after starchy food, and the reducing power stood as 80 to 70. The last was after the ingestion of cane-sugar. It is not quite comparable with the others, as citric acid was used instead of sulphuric acid, with the view of ascertaining if any cane-sugar was present. The amount of sugar in the second portion was greater than in the first, but the figures before and after citric acid being the same, no cane-sugar existed. The outside variation noted thus amounts to eleven. Compare this with Dr. Paton's statement. Actually, however, the variation observed is exactly what might be looked for. The second portion of blood was blood that had been delayed for a short time in the capillaries, which would give the opportunity for the effect of absorption upon the blood reaching the capillaries from the arteries to be made more manifest, as in reality occurred.

Dr. Paton next discusses the question of whether sugar, as a natural phenomenon of life, is discharged from the liver into the general circulation. He first says, "The flow of blood through the liver is so enormously rapid that a very small increase in the percentage amount of sugar would indicate an enormous production in the twenty-four hours;" and after alluding to the conclusion I have drawn from analyses of the blood on the right side of the heart and the portal vein, proceeds: "Now this method of experiment is not in any way calculated to elucidate the question. In the first place, the analysis of the heart-blood and of the portal blood were not made at the same time, or even in the same animal. Secondly, the animal was dead; and Dr. Pavy has insisted that in the case of the liver post-mortem changes commence at once. Such changes may also occur in the blood. That they do occur later is admitted by Dr. Pavy himself. And lastly, the heart blood is the blood from the liver enormously diluted with blood from the rest of the body; and hence if the blood from the rest of the body is poor in sugar any increase in the sugar in the blood from the liver will not be manifested."

Professor Michael Foster a long time ago broached the view that the amount of sugar escaping from the liver might be so small as to fall within the limits of error of analysis; and thus through the constancy and rapidity of the flow of blood a considerable quantity might be carried away in the course of the twenty-four hours without showing itself in a distinctly manifest form. I know this argument has had great weight with physiologists. It is not so forcible now as when first propounded, owing to the advance that has taken place, and the greater precision that has been reached in blood analysis for sugar. Duplicate analyses show that such closeness in the results is attainable as to justify our regarding analytical evidence as entitled to great consideration as a point amongst the facts before us. There is evidence, however, of another nature—namely, evidence derivable from the state of the urine—that can be adduced standing in opposition to the assumed escape of sugar from the liver. This I will enter upon after I have dealt with the points raised by Dr. Paton concerning the blood.

Dr. Paton says that my experimentation showing that the blood of the right side of the heart contains much less sugar than that of the portal vein is not in any way calculated to elucidate the question.

In the first place he remarks, "The analyses of the heart blood and of the portal blood were not made at the same time or even in the same animal." How, I would ask, could this be done to properly give the information required? Dr. Paton does not seem to have realised the position involved in his suggestion. The collection of the heart-blood in the manner adopted, by diverting the flow of blood from the arteries, would frustrate attempts to collect blood from the portal vein. Again the collection of blood from the portal vein would interfere with its passage through the liver, and the experiment would be open to the objection that the blood on the other side of the liver was not receiving the supposed influence of the stream derivable from the organ. If I had only founded my conclusion upon a few observations some objection might be taken to their full acceptance. As it is, the statement I have given that "the amount of sugar naturally present in the blood of the general circulation may be stated to range from about 0·6 to 1 or a little over 1 per 1000," is based upon upwards of one hundred observations upon different animals. It is to be observed that the representations given constitute *range*, and not *average*, and that it is impossible for the range to be exceeded during life without leading to a recognisable saccharine condition of the urine standing in proportion to the extent to which the limit mentioned is surpassed. Thirty-eight observations are given of the state of the portal blood taken under various conditions, and these, I think it will be admitted, amply suffice to furnish information upon which reliance can be placed.

"Secondly," Dr. Paton says, "the animal was dead, and Dr. Pavy has insisted that in the case of the liver post-mortem changes commence at once. Such changes may also occur in the blood. That they do occur later is admitted by Dr. Pavy himself." This as argument is pure subtlety. Whilst asserting that the post-mortem change sets in quickly, I have nowhere stated that sufficient time is not given in a post-mortem experiment, if expeditiously performed, for a knowledge of the ante-mortem state to be obtained. Indeed, I have distinctly affirmed the reverse. The blood coagulates speedily after death, but is there not a certain time during which it can be obtained in the fluid state belonging to life? An alteration of the state of the blood as a post-mortem effect not only *may*, but *does* occur if time is given, but it is in the direction of an increase of sugar. The change which subsequently occurs in drawn blood gives no grounds for considering that it can be read as having any physiological meaning connected with it.

"And lastly," Dr. Paton continues, "the heart blood is the blood from the liver enormously diluted with blood from the rest of the body; and hence if the blood from the rest of the body is poor in sugar any increase in the sugar in the blood from the liver will not be manifested."

It might be taken from this remark that I had ignored the point to which it refers. On the contrary, it has received full consideration in my work. It is perfectly true that the heart blood is blood from the liver, mixed (this is a more appropriate word under the circumstances than diluted) with the blood flowing through the systemic veins. For the sake of argument, let it be supposed for the moment that sugar is transmitted from the liver into the general circulation, and that the blood of the hepatic veins contains more sugar than that of the systemic veins. The effect of this would be to raise the amount of sugar in the right ventricular blood beyond that in the general systemic venous blood in proportion to the extent to which sugar escaped from the liver. In other words, we are brought to the position that the right ventricular blood should contain more sugar than the blood flowing through the systemic veins. Now, no one contends at the present time that sugar is removed from the blood in its transit through the lungs. This being the case, the arterial blood, as far as sugar is concerned, is the representative of right heart blood. Hence through the medium of arterial blood we can compare in a reliable way right heart blood with general systemic venous blood to see if any sugar is given to the latter by passage through the hepatic veins from the liver. The results derived from exact experiments on the comparative state in the same animal of arterial and systemic venous blood show that no sensible difference is discoverable. But there ought to be a difference if sugar from the liver mingled through the medium of the hepatic venous blood with the general systemic venous blood, and produced the influence that would follow upon the right ventricular, and thence the arterial blood. The absence of sensible difference as regards sugar between arterial and venous blood tells strongly in a further way. It tells us that there cannot be the discharge of sugar from the liver alleged under the glycogenic doctrine to occur, as the consequence would be accumulation, kept, it may be said, within immoderate bounds by elimination with the urine—in other words, diabetes.

In connection with the question of the comparative state of the blood going to and coming from the liver, Dr. Paton next considers that "a discussion of the results of other investigations is required. It is not sufficient to say,—'To obtain such a specimen directly from the hepatic veins is, I consider, a procedure attended with so much liability to the introduction of error as to be unsuited for employment.' Seegen adopts this method, using in many cases an ingenious catheterisation of the hepatic vein, and in a series of sixty-four experiments finds a marked increase in the sugar of the hepatic blood in unæsthetised [*sic*] animals, and in animals anæsthetised with chloroform, morphine, and in those under the action of curare."

In here quoting me Dr. Paton has given a few words away from the context. In the text I am strenuously maintaining that physiological conclusions can only rightly be based upon a knowledge of the physiological state. My original work commenced with pointing out the error that had been committed in taking a state that was not the natural or physiological one to represent the normal living condition. In urging the necessity of having a representation of the physiological state as regards the blood to deal with, I say,—

"How speedily an alteration in the state of the blood, attended with the presence of an abnormal amount of sugar, may take place through the influence of altered conditions connected with the liver I illustrated at the outset of my researches, now upwards of thirty years ago. From the readiness with which the blood flowing from the liver is thus thrown into an altered state, it is necessary that close attention should be given and proper precautions observed to escape being misled; otherwise sugar may be met with more or less greatly in excess of what is ordinarily present, and the inference thence be erroneously drawn that its discharge from the liver takes place in a manner that does not naturally occur."

"In dealing with the problem before us it is, then, a matter of primary importance that the blood taken for examination should be in a state representative of that naturally belonging to life. *To obtain such a specimen directly from the hepatic veins is, I consider, a procedure attended with so much liability to the introduction of error as to be unsuited for employment.*" The italics denote the words quoted, and are not in the text.

Now the experiments of Seegen, referred to by Dr. Paton, may be unhesitatingly affirmed to constitute experiments from which it is absolutely not permissible to draw a physiological deduction. I have personally known Dr. Seegen for many years, and I hold him in high esteem as a friend, but I must speak of his work as utterly devoid of value from a physiological point of view. Let anyone read the description ('Glycogenie animale,' traduction par Dr. Hahn, Paris, 1890) of his *modus operandi* for obtaining his specimens of blood, and it will be seen that his experiments are not in reality representative of physiology. Nothing to place reliance upon could possibly come from experiments of such a nature, and it is a pity so much good intention and labour have been spent in so futile a way. His experiments lead him to deny that glycogen is the source of the sugar asserted to be poured into the circulation by the liver. He is further led into this impossible position: whilst contending that the liver is constantly throwing into the circulation a large quantity of sugar, his experiments on the comparative state of arterial and venous blood show no recognisable difference between the two as regards amount of sugar, which, taking the results as they stand, brings it to there being no exit to meet the continued ingoing taking place. In the results of a further series of experiments given in the 'Centralblatt für Physiologie' of 20th October and 3rd November, 1894, those belonging to the two kinds of blood taken before electrical stimulation of the muscles show no sensible difference. Those with stimulation of the muscles with direct faradisation show a decided diminution of sugar, whilst with those where muscular contraction was excited by faradisation applied to the nerves a marked increase of sugar in the venous blood was as a rule noticed. In all the experiments the figures representing the amount of sugar present stood much higher—in some very much higher—than those belonging to the natural state. They correspond with those associated with diabetes, and simply from this point of view are inadmissible as representative of physiology.

From the last quotation Dr. Paton continues, "In the experiments of Bock and Hoffmann (1874) an increase is also shown; while even the experiments of Bleile (1879) and of Abeles show in some cases an increase. The question of whether Abeles is right in concluding that the increase is the result of injury of the liver as the result of the experiment deserves consideration."

Seegen quotes these authorities fully, and discusses the bearing of their work upon his own, but I cannot find any reference made to experiments on the relative state of portal and hepatic blood by Bock and Hoffmann. Nor are they to be found in the abstract of what they say given in 'Virchow-Hirsch Jahrb.,' 1874. Experiments, however, are mentioned on the effect of isolation of the liver from the circulation, and these will fall under consideration below. Bleile ('Archiv f. Anat. u. Physiol.,' 1879) found that the amount of sugar in his experiments was greater in the hepatic than in the portal blood if the vena cava was stopped, whilst the reverse was observed when the vena cava was left free. The result he considered was attributable either to (1) accident, (2) admixture of blood containing less sugar with that of the hepatic vein when the vena cava was left free, or (3) the effect of the disturbance of the circulation by the operative manipulation necessary. The remark touching Abeles I may allow to stand where Dr. Paton has left it.

We are next taken to the side of the question connected with the isolation of the liver from the circulation : " there is yet further strong evidence that the liver is constantly pouring sugar into the blood. Bock and Hoffmann have succeeded in excluding the liver from the circulation in dogs, and they find that the blood rapidly becomes free from sugar. Minkowski, taking advantage of the communication between the portal vein and the renal vein in the goose, excluded the liver from the circulation without bringing about the vascular disturbance caused in mammals. He, too, found that the blood rapidly became free from sugar. There can be no two interpretations of these experiments. Even Lieblein, who has recently attempted to throw the liver out of action by injecting weak acids into the bile-ducts, and who has thus succeeded in getting the liver free of glycogen, and who finds that the injection of phloridzin still causes glycosuria, concludes from this, not that the liver does not make sugar, but that other organs of the body can also do so."

Bock and Hoffmann's experiments are now reached, and what I said of Seegen's experiments applies here. Can any *physiological* deduction be drawn from such a state as that following the operation performed? We cannot look upon the living system as constituting a mere chemico-physical machine, and be satisfied that we are properly unravelling the normal processes of life by observing what occurs under such extremely abnormal circumstances as those involved in the experiments under consideration. I doubt, with the modern method of analytical procedure (Bock and Hoffmann wrote in 1874), whether the blood would have been in reality found actually free from sugar. I can only say that present evidence is to the effect that sugar to some extent is always to be met with everywhere in the body. But allow this to pass, and let us see what is mentioned by the authors about the time that it took for the disappearance of the sugar. They say that the disappearance took place after forty-four minutes if they isolated the liver from the circulation by blocking the inferior cava above the renal veins, and ligaturing the cœliac axis and mesenteric arteries, and then the portal vein; and after seventy-nine minutes if with the blocking of the inferior cava they tied the aorta above the cœliac axis as a preliminary operation to tying the portal vein. The amount of sugar normally present in the blood they give at ·7 to 1·1 per 1000, an estimate which stands in strict accord with my own observations, and in contrast to the results of Seegen. With the amount of sugar here implied as existing, and the time elapsing for disappearance, it may well be asked what possible significance can these experiments possess in relation to the occurrence of sugar destruction as a necessary part of the assumed glycogenic function.

Minkowski's experiments ('Archiv für experiment. Pathol.,' 1886) on the extirpation of the liver in geese were, as far as their primary object was concerned, conducted in relation to the question of the formation of urea. They were, however, incidentally turned to account for the observations made upon the question of sugar. As the result of the operation, it is stated that the blood in the course of a short time was found free from sugar. Dr. Paton makes bold to assert, "There can be no two interpretations to these experiments;" but what does Minkowski himself say? In speaking of the cause of the disappearance of the sugar, he considers it doubtful whether it was due to the absence of the liver function, or to the general effect of the operation upon the animal.

It is known that phloridzin produces glycosuria in the fasting animal, and therefore quite apart from origin from food through glycogen of the liver. The view entertained of its action is that it leads to the splitting up of the proteids of the body with the liberation of sugar. The quoted experiment of Lieblein simply supplies confirmatory evidence, and the information given shows that glycogenesis as it is understood in its application to the liver is not needed to account for the production of sugar in the animal system. Sugar to a certain extent is met with in all the textures of the body, and under natural circumstances no more is found in the liver than in many parts elsewhere. From the muscles it is possible that sugar may be given to the blood, and the muscles contain glycogen, sometimes in large amount, in the same manner as the liver. As pointed out in my work, there are grounds for considering this glycogen as derived by cleavage from the proteid of the muscle, and not from carbohydrate conveyed in a free form to the tissue by the blood. Instead of glycogen, sugar, as the result of the influence existing, may be split off, or it may happen that the glycogen may be secondarily transformed into sugar.

I have now gone fully through what has been brought forward from the work of other investigators in relation to the question of whether, as a natural phenomenon of life, sugar is discharged into the circulation from the liver, and I leave it to be judged if any of the evidence adduced can for a moment be accepted as authorising such a conclusion.

It happens, as I will next proceed to show, that another aspect presents itself from which the question may be examined; and here, literally speaking, we have simply to read off the revelations of nature for arriving at our goal.

Observation from every point of view shows that the urine affords an index of the state of the blood in relation to sugar. No one will dispute that the sugar of the urine is derived from that previously existing in the blood. Conversely it may be said if sugar exists in the blood it will appear in the urine, and it does so in proportion to the amount that is present. This is a well-established fact in connection with diabetes, the kidney constituting in reality the eliminating organ for the sugar, which is wrongly permitted to reach the general circulation. What applies in diabetes equally applies in health; and if from any circumstance whatsoever sugar reaches the blood of the general circulation, it will in proportion be discoverable in the urine. There is no tolerating capacity against elimination, as was at one time alleged. The erroneous notion arose from the elimination, when minute, being beyond the capacity or sensitiveness of the tests formerly employed to reveal it. With the tests now at our command the sugar in the urine derived from the small quantity naturally existing in the blood is readily brought into view. There is no different kind of action with respect to sugar elimination in health and diabetes. The only difference existing is as regards the amount eliminated, and this, as I have said, is dependent upon the amount previously existing in the blood. It would be anomalous indeed if there were one law for a certain quantity, and another law for a quantity that simply happened to be less.

Upon these premises it follows that it is permissible to look to the urine for affording us knowledge regarding the entry of sugar into the blood of the general circulation. Under natural circumstances the blood contains a certain small amount of sugar, which may be considered as constitutionally belonging to it, just as sugar, it would seem, belongs as an integral part to the other constituents of the body. Authorities are agreed that the amount of sugar in the blood is not a fluctuating one, or associated with the amount of carbohydrates ingested. Within ordinary limits both blood and urine remain uninfluenced by the character of the food in relation to carbohydrates. It matters not whether only the comparatively insignificant amount of carbohydrate existing in an animal diet, or the large amount existing in many kinds of vegetable food, be ingested : the result is the same. But if, in accordance with the glycogenic doctrine, the carbohydrate has to reach the systemic capillaries for disappearance in some unknown way, the position as regards the blood and consecutively as regards the urine cannot be the same. True, it is said that by the transformation into, and storage as, glycogen in the liver, means exist for regulating the transit into the blood, so that only a small quantity passes at a time—a quantity, it is alleged, which may be insusceptible of definite recognition in consequence of falling within the limits of liability to error to which the analytical procedure is open. No matter, however, whether or not there is any validity in this suggestion, the carbohydrate some time or other must undergo transportation. If the carbohydrate is ingested, and if it has to pass to the systemic capillaries for destruction, there must be within a given time a transit under a diet including farinaceous articles altogether out of proportion to that occurring under a diet of animal food.

Now, the state of the urine constitutes, as I have pointed out, a very delicate indicator of the passage of sugar into the general system. My daily experience in connection with diabetes for a large number of years has taught me that the slightest dietetic deviation on the part of the patient is immediately revealed through the condition of the urine. It is surprising with what exactitude the result occurs. Under the restricted diet for diabetes the urine with ordinary testing shall give no indication of the presence of sugar. Let there be a deviation to ever so small an extent, and within a very short time sugar will be discoverable, and discoverable in proportion to the extent to which carbohydrate matter has been taken.

The experiments I have given in my book on the introduction of sugar into the circulation of rabbits by injection into the jugular vein and into the subcutaneous tissue speak exactly in the same way. Even with the employment of half a gramme per kilo. body weight a decided influence is producible.

How does this fit in with the glycogenic theory? The sugar discoverable in the blood during subsistence upon animal food (which in reality stands as representative of the sugar naturally existing as a standard constituent of the blood) reveals itself by the presence of a certain amount of sugar in the urine, but the large amount of sugar derivable from vegetable articles of food, and assumed to pass to the tissues for destruction, fails to reveal itself, inasmuch as no increase is observed in the amount of sugar present in the urine. Whatever goes to the tissues must also go to the kidney. It might only, it is true, pass continuously in small amount, but nevertheless the fact remains that in a given time a certain quantity infinitely larger than that which could be derived from animal food would have to pass. Yet this is to happen without giving evidence of its doing so. Although the passage of a certain quantity is revealed, the additional quantity goes for nothing as far as indication of transit is concerned viewed through the urine.

What this additional quantity may amount to may be ascertained by looking to the carbohydrate contained in the respective diets, or, on the other hand, to the sugar respectively eliminated in a case of diabetes where the carbohydrate taken is not retained, but simply allowed to filter through the system and escape with the urine.

Looking to the latter, I may give an illustration from a case examined in detail in relation to diet, and cited in full in my work on 'Diabetes.' When the patient was on a diet consisting exclusively of food from the animal kingdom, the amount of sugar eliminated during twenty-four hours was 37 grammes, whilst for a corresponding period when the diet included 16 ounces of ordinary bread, and other carbohydrate-containing articles of food, the sugar amounted to 685 grammes. Allowing that some may have possibly been drawn from splitting off from the tissues, the difference still shows the greatly increased amount of carbohydrate that would have to be transported through the circulation in the one case as compared with the other, if, in accordance with the glycogenic doctrine, the carbohydrate matter of our food had to be conveyed as sugar in a free form to the tissues for destruction. Yet, in the healthy state, this has to happen without producing any traceable influence upon the urine.

Any system, it may be said, which involves functional transit of carbohydrate matter as sugar to the tissues for destruction is negatived by the absence of evidence of this transit. When the carbohydrate matter of our food does, indeed, reach the circulation as sugar, it becomes conspicuously revealed by the presence of sugar in proportionate amount in the urine. Such entry, therefore, means glycosuria, and what is wanted for the production of the state belonging to health is that the carbohydrate should be disposed of in a manner to be prevented reaching the general circulation as sugar.

I submit, then, that the evidence to be drawn from a reading of the information supplied by nature is, through the train of reasoning deducible from it, absolutely fatal to the glycogenic theory.

To proceed, we come to the passage, "The well-known facts of the accumulation of glycogen under carbohydrate, and to a lesser extent under proteid diet, are stated by Dr. Pavy." Dr. Paton might have said *were discovered* by Dr. Pavy, for the connection of glycogen accumulation with the ingestion of carbohydrate matter was first pointed out by me in the 'Philosophical Transactions' for 1860.

Afterwards Dr. Paton says, "The idea is advanced by Pavy that the glycogen which is formed by the liver on a proteid diet is derived from the carbohydrate which is supposed to split off from the proteid molecule during digestion. Certainly the amount of glycogen stored under a proteid diet is small, but then if a arbohydrate were liberated, as contended by the author, and to the extent of 0·2 per cent. as suggested by him, it would hardly afford an adequate supply of a material such as glycogen, which is, as he admits, constantly being removed from the liver."

Apart from what I have already said about the figures here given not representing the truth as regards the amount of carbohydrate present in proteid matter the term proteid diet gives an inaccurate impression. There is no such thing as a pure proteid maintenance diet. Animal food which gives the nearest approach to it contains other sources of carbohydrate besides cleavage carbohydrate from proteid matter. I distinctly affirm this in my work, stating under the head of animal food as a source of glycogen in the liver (p. 118), "Flesh and other animal substances consumed as food can be shown by analysis to contain free carbohydrate under the form of sugar, and to some extent also of glycogen. Sugar from these sources will, as the result of the ordinary operations of alimentation, reach the portal vein, and thence the liver."

As regards glycogen being constantly removed from the liver, my view is to the effect that it is applied by protoplasmic action within the hepatic cells to fat and proteid formation as circumstances require ; and looking at the length of time that elapses under absence of food before complete disappearance occurs, there is no ground for considering that there need be more than a slow removal ordinarily taking place, which the carbohydrate derivable from animal food would be quite adequate to meeting. The position as regards removal here conceived as existing is very different from that implied by the glycogenic doctrine. In the case of animals to which the purified proteids have been exclusively administered by way of experiment, glycogen, it is stated, has been found in somewhat greater amount than under starvation, but in smaller amount than under a meat diet. This agrees perfectly with what might be expected.

I now reach the question of glycogen in relation to sugar-production in the liver. Dr. Paton upon this point makes a strong attack upon my views with evidence supplied from his own work.

I have already mentioned that a communication by Dr. Paton on "Hepatic Glycogenesis" appeared in the 'Transactions of the Royal Society' for 1894, which was issued just before the publication of my book. It happens that the views expressed in these two publications are of a nature to render it impossible for both to be right. It was therefore obligatory, in vindication of the position in which he stood before the Royal Society, that Dr. Paton should make endeavours to upset my work. How far he has succeeded in the portion of his criticism I have as yet dealt with I consider I may safely leave to others to decide. I think also it will be found that I may safely do the same with respect to the portion that remains to be examined. Hitherto the line of procedure I have had to adopt has been one of defence. The position is now changed, and in the case of the point standing immediately before us for consideration I shall proceed to show upon what foundation the work of Dr. Paton rests. Before doing so I will set forth in plain and unequivocal terms my own contention, so that it may be seen how the whole question stands.

To start with, we are in presence of the fact that the liver contains glycogen, an amylose form of carbohydrate, which in like manner to starch is readily susceptible of being hydrolysed and transformed into sugar by the agency of amylolytic ferments. Besides this form of carbohydrate there is likewise present a certain amount of sugar which in the living and normal state is only small, although a readily accessible source for it in the shape of glycogen exists more or less abundantly around.

The glycogen, according to my view, is derived from the free carbohydrate conveyed to the liver from the alimentary canal in the portal blood. The amount of this will depend upon the amount of carbohydrate derivable from the food, and further, it may be said, upon the amount that happens to be applied to fat and proteid formation by the cells of the villi. It is the surplus, or that which escapes being disposed of by these operations that reaches the portal blood, and the operation being the result of protoplasmic action, the relation of nitrogenous matter to carbohydrate in the food comes in as an influencing factor. I am under the impression, from what I have seen, that after a certain amount of carbohydrate without a due proportion of nitrogenous matter more sugar will reach the portal vein, and thence more glycogen be found in the liver, than after the same amount of carbohydrate with a larger proportion of nitrogenous matter.

Hitherto it has been supposed that the glycogen met with after animal food is derived from a breaking down of nitrogenous matter effected within the liver itself. A proteolytic action, however, in reality is performed by the digestive secretions, and is not needed to be performed by the liver. I have already referred to the sources of carbohydrate existing in animal food, and stated that they suffice to give sugar to the portal blood from which the glycogen of the liver in animal feeders may be derived.

As to what becomes of the glycogen, I have not far back adduced evidence which may be looked upon as proving that it cannot, in accordance with the glycogenic theory, undergo transformation into sugar for transport through the circulation to the tissues for destruction. I hold that it is disposed of *in situ* by the protoplasmic power of the hepatic cells. The cells of the liver thus play a supplementary office to those of the villi. The carbohydrate which escapes appropriation by the latter becomes conveyed to the sphere of influence of the former, and if the proper amount of power is here exercised an arrest takes place, which prevents the general circulation being reached. If the general circulation is not reached, the opportunity is not afforded for the carbohydrate to pass off as sugar with the urine. If, on the other hand, the proper arrest does not take place sugar reaches the general circulation, and in proportion as it does so passes off with the urine.

The necessity for the supplementary action of the liver becomes apparent when we consider that something is required to meet absorption of sugar from the stomach, and also that provision has to be made for the protoplasmic appropriation of carbohydrate in animals, as the bird, &c., where the villi and lacteal system do not attain the state of development observed in mammals. The *foie gras* of the Strasburg goose may be regarded as an illustration of extensive fat production in the liver arising from the state of things just referred to.

Glycogen, as is known, exists, and it may be in considerable quantity, in different parts of the body. Its origin from the food accounts for the special amount found in the liver. Presumably in other parts it constitutes a cleavage product from proteid matter. Its non-diffusible nature fits it for storage, like starch in the vegetable kingdom, and wherever existing it may be regarded as material designed for being drawn upon as required by the protoplasm within the sphere of influence of which it lies.

As regards sugar, the position in reality of the liver is not *physiologically* different from that of the other structures of the body. If dealt with in a proper manner for ascertaining the condition existing during life in relation to sugar, no more is found than in other parts, and frequently indeed not so much. Moreover the kind of sugar agrees with that generally encountered in the structures of the body in not consisting of glucose.

Whilst there is this accord between the liver and the other structures of the body as a physiological condition, a marked divergence is, on the other hand, perceptible in the state found shortly after death. As a result of the destruction of life a rapid alteration occurs. The liver becomes loaded with sugar in a manner that the other structures do not, and the kind of sugar now discoverable is ordinarily found to be glucose.

We know that the altered condition observable is due to the transformation of glycogen into sugar. Concurrently with the production of the one there is a disappearance of the other. It is contended under the glycogenic doctrine that what demonstrably occurs after death likewise occurs during life; but, as I have pointed out, it may be taken as impossible that it can do so and leave the urine in the state in which it is normally found. If the ferment which comes into operation after death existed and were allowed to have its play during life, it would not take long for the glycogen ordinarily present, especially at a period of fasting, to be made to disappear. The two cannot live together. Either the ferment, if present, must be under some inhibitory influence during life, or it must become developed with the occurrence of death, just as the ferment does which occasions the coagulation of the blood, where the circumstances are such as to render it obvious that the ferment cannot exist in the physiological state.

In this way the *post-mortem* production of sugar is brought into the position of a phenomenon due to the setting free of a ferment which does not give signs of being in existence normally during life. It is true under abnormal conditions the same kind of ferment action that occurs after death may take place during life, and here the urine becomes charged with sugar. The glycogen, also, according to experimental observation, as I have pointed out at p. 145 in my work, disappears from the liver.

Dr. Paton, whilst admitting that ordinary ferment action is in operation after death, endeavours to show that the active production of sugar that is noticeable in the initial *post-mortem* period is attributable to a continuance of molecular life. He says, "I have endeavoured to show ('Philosophical Transactions,' 1894) that the conversion of glycogen to sugar takes place during the life of the liver cells, and as a result of their metabolism. When an animal is killed the cells of the different organs continue to live for some time. I have shown that it is in the first half-hour or so after the animal is killed that the conversion of glycogen to glucose goes on most rapidly; and, further, that by pounding the liver with fine sand so as to destroy the cells this rapid conversion is stopped. Again, I have endeavoured to show that various agents which diminish the activity of protoplasmic changes diminish the rate of this early amylolysis. After the cells have ceased to live, and after they begin to manifest post-mortem changes a slow amylolysis continues as a result of the development of a ferment."

Let us look at what is here contended for, and at the validity of the evidence adduced in its support.

Observation shows that immediately after death an active production of sugar in the liver occurs with a concurrent disappearance of glycogen, and that the process slows down in activity as time proceeds. Sugar is produced and glycogen disappears, and, excluding Seegen, it may be considered to be universally admitted that the glycogen constitutes the source of the sugar. Now what does the transformation of glycogen into sugar, looked at as an operation, consist of? It is simply the transmutation of an amylose carbohydrate into a carbohydrate possessing a higher degree of hydration. In this kind of transmutation we have the occurrence of breaking down and hydration. The composite amylose molecule is split up and hydrated, and as a result the carbohydrate is carried into either the saccharose or glucose group. The process, as is well known, does not require the instrumentality of vitality for its accomplishment. Acids and ferments very readily effect the change. Not so, however, with the opposite kind of transmutation,—that is to say, the conversion of members of the glucose and saccharose groups into an amylose carbohydrate. Here we have really to look to vitality as the mainspring of action. By the synthesising and dehydrating power possessed by living protoplasm, sugar molecules are fabricated into molecules of the more composite amylose type. For synthesis and dehydration the agency of living protoplasm is required, but for breaking down and hydration it may be considered that vitality is not called into requisition except in so far as it is related to the genesis of the enzyme which is the source of the change from ferment action that occurs.

It is common knowledge that sugar-production in the liver takes place more actively during the first period after death than later on. Upon it has been founded the idea that the early more active production is due to vitality, whilst the later is the result of ferment action. The same phenomenon—namely, the transmutation of carbohydrate in the direction of increased hydration—is thus attributed to two sources of action, notwithstanding the one suffices, and that it can be definitely said that vitality is not needed for the accomplishment of the operation.

The idea alluded to is adopted by the Cambridge School of Physiology (Foster's 'Text-book of Physiology,' appendix by Sheridan Lea, pp. 58, 98), and Dr. Paton's work is directed to giving support to it. In the first part of his communication he supplies experimental evidence bearing on the rate of change during given periods. Without any logical right to do so he includes in his argument the sugar found in the liver at the moment of death, reckoning it as belonging to the change occurring in the first period taken. But this is a minor matter, and with the remark that has been made may be allowed to pass.

What is of real—indeed, paramount importance is that Dr. Paton has omitted to ascertain whether the phenomenon observed with respect to rate of change, upon which the conclusion regarding the effect of vitality has been based, is exclusively associated with the early after-death period.

Now it happens that a mode of experimenting is open to us which carries us entirely away from the possible influence of life. It is known that ferments are susceptible of being precipitated by alcohol without having their activity destroyed. The liver treated with alcohol, and subsequently suitably dried, yields a product which may be preserved for an indefinite time. I experimented largely upon this subject with the liver of the rabbit, dog, and cat, long before Dr. Paton's experiments were conducted. The results given in my book are derived from a series planned upon the strength of what I had previously observed. In the one case (p. 150) the coagulated liver was dealt with alone; in the other (p. 154) some spontaneously dried blood was added. The results are arranged in a tabular form to show the progressive change that occurred from half an hour onwards to seven hours. The liver was derived from the dog, and the preliminary analysis of it in the fresh state showed that a good amount of glycogen was present. A representation is given of the progressive loss of glycogen and production of sugar, and, moreover, the figures for the sugar were determined before and after treatment with sulphuric acid. All this information is supplied, and it is perceptible from it that the difference as regards rate of change which Dr. Paton adduces as affording evidence of the operation of vitality is visible in my results.

Dr. Paton says it is in the first half-hour or so after an animal is killed that the conversion of glycogen into glucose proceeds most rapidly. Identically the same is noticeable in dried alcohol-coagulated liver substance treated with water and subjected to a suitable temperature for ferment action to occur. Pretty nearly the full transformation of glycogen occurred in six to seven hours, and about half was accomplished in the first half-hour.

Unless, therefore, it is to be considered that vitality is susceptible of retention in alcohol coagulated liver substance bottled and kept for an indefinite time upon a laboratory shelf, the deduction which has been drawn from the more active initial conversion occurring entirely loses its support.

Dr. Paton in his criticism of my book glides over the experiments with alcohol coagulated liver to which I have been referring with the remark, "No one denies that such a ferment may develop, though Dastre believes that micro-organisms have usually much to do with such a conversion." Such a remark, it may be simply said, has no bearing upon the point.

After fallaciously premising that the more active initial conversion of glycogen into glucose observed to occur in the liver of the recently killed animal is due to the operation of vitality, Dr. Paton proceeds in his communication in the 'Philosophical Transactions' to show that by pounding the liver with fine sand immediately after death so as to destroy the structural integrity of the cells the rapid initial conversion is stopped, and to contend that in the absence of vitality only a slow conversion by ferment action takes place. Let us examine this matter, and see how Dr. Paton's conclusions stand under submission to experimental investigation.

The method of procedure adopted by Dr. Paton consisted of taking three portions of the liver of a rabbit that had just been killed, and submitting them to the following treatment. One was thrown into boiling water, and regarded as a check specimen. Another was finely triturated with sand to destroy the structural integrity of the cells, and placed in about 150 c.c. of ·75 per cent. salt solution. The third was coarsely minced, and in this state placed into a similar quantity of the salt solution. The last two were then put into an incubator with the temperature kept at 37°—40° C.

Two experiments only are supplied. In the one the time of exposure in the incubator was 1 hour 48 minutes, and in the other 4 hours 8 minutes. After exposure in the incubator the two portions were next treated with boiling water in like manner to the check specimen to arrest further change. Finally, the glycogen after extraction was estimated in each case by the employment of Brücke's method and weighing, and taken as a measure of amylolytic change.

The results of the two experiments are given at p. 245 of the volume of the 'Transactions.' Percentage expressions are used, but I will transform them into parts per 1000.

In the first experiment the check portion of liver yielded evidence of containing 59·7 per 1000 of glycogen, the minced 44·1, and the pounded with sand 54·7 *plus* an unknown quantity from, as it is stated, slight loss due to cracking of beaker from bumping caused by sand. The effect of these figures is to represent that 15·6 per 1000 of glycogen was transformed into sugar in the minced portion, or that alleged to have vitality in operation to exert a transforming influence; and taking the accidental loss into consideration, something less than 5·0 per 1000 in the pounded with sand portion, or that in which vitality was presumed to be destroyed.

In the second experiment, from the check portion 52·67 per 1000 of glycogen was obtained, from the minced 23·36, and from the pounded with sand 50·61. According to these figures there was a loss by transformation of 29·31 per 1000 of glycogen in the presence of the assumed play of vitality, and only 2·06 per 1000 in the absence of it.

These two experiments constitute the pivot upon which Dr. Paton's work published in the 'Transactions' rests. Upon the strength of them he says, "The results of these two experiments are so fully confirmatory that it was not considered necessary to extend the series." They stand as the basis of his endeavours to show that the early active transformation of glycogen into glucose is due to vitality. The remainder of his paper is devoted to considerations which only in a collateral way support his contention, and do not afford anything of the nature of direct proof of vitality being concerned in the matter.

Ferment action as the source of the conversion is what Dr. Paton endeavours to refute; and with an operation attended with so much irregularity as regards precise results, and influenced by such slight circumstances — circumstances with which our acquaintance is as yet far from complete—he considers himself at liberty, upon the strength of two experiments, to speak in the manner he has done.

Logically, he has no right to arrive at the conclusion put forward without ascertaining that the different circumstances under which the minced and pounded with sand portions of liver are situated may not have modified ferment action. The position of the two is very different during the exposure to warmth in the incubator. In the one case the liver lies as a compact mass at the bottom of the saline solution, whilst in the other the triturated substance is more or less diffused through the 150 c.c. of salt solution.

Again, he is content with gauging the extent of change by estimating the loss of glycogen, neglecting to obtain the corroborative evidence derivable from estimating the gain of sugar, notwithstanding in the first part of his paper he gives experiments presenting strikingly concordant results showing that the information can be supplied. The difficulties in the way of reliably estimating the glycogen are discussed by him, and in the face of what is said it is nothing less than surprising that not merely should preference be given to the glycogen determination, but that it should be allowed to stand unsupported.

With the adoption of a proper process nothing is more easy than to make a reliable estimation of sugar. As a further point, it may be remarked, no proof is supplied in the details of the experiments themselves that the full extraction of glycogen was effected.

I have repeated Dr. Paton's experiments, and in other ways carefully gone over the ground of experimentation upon the subject. From the lengthy nature of the analytical operations required to be undertaken, anyone who has had experience in the matter will realise the expenditure of time and labour the work has involved.

The first point noticeable in connection with the details to follow is the variability in the results obtained in different experiments. The amylolytic action occurring in the pounded with sand portions as compared with the minced is in all cases considerable; and, evidence is forthcoming, both through the loss of glycogen and the gain of sugar, of its standing greater. Nowhere do the figures at all coincide with those belonging to Dr. Paton's experiments. Where two sets of experiments were conducted upon the same liver, and different processes of analytical procedure adopted, the results vary in such a manner as to suggest complications arising out of the production of dextrins, which are differently influenced by different strengths of alcohol. As is well known, the precise strength of alcohol existing constitutes a nice point in relation to the precipitation of glycogen and the dextrins. Where the carbohydrate is determined by titration with the copper test, with the assistance of treatment with sulphuric acid, we are upon much broader and more reliable ground than where Brücke's process and precipitation and weighing are adopted.

As I have said, my results stand in antagonism with those of Dr. Paton. If the active transformation of glycogen into sugar that is observed ordinarily to take place in the liver of the recently killed animal depends upon a continuance of vitality, and pounding with sand removes this vitality, in not a single instance ought there to have been after the pounding with sand the evidence of the active transformation that was presented. If the power alleged to constitute the cause of a given effect has been destroyed, what, it may be asked, remains to account for the effect that is observed to occur? Again, besides the simple pounding with sand experiments, others will be adduced in which the frozen liver is dealt with, and unless vitality is susceptible of being retained in a piece of frozen liver kept in an ice safe for twenty-four hours vitality cannot play the part suggested.

It will be remembered that of the two experiments given by Dr. Paton, the one which stands without defect showed a loss of glycogen of merely 2·06 per 1000 in the pounded with sand portion, which means that only what may be styled an insignificant amount of transformation of glycogen occurred. If we turn to another part of his paper we come across evidence of a conflicting nature.

He says chloroform accelerates the early active change alleged to be due to vitality, and not the later change assigned to ferment action. Some liver which had been pounded with sand, and therefore was assumed to have its vitality destroyed, was treated with salt solution, and squeezed through calico. Glycogen was added to the liquid extraction obtained, and it was then placed in an incubator as part of an experiment with chloroform vapour. There ought only to have been under the circumstances evidence presented of the insignificant amylolytic change asserted to occur in connection with liver substance that has lost its vitality, but in Exp. 20, p. 256, it is noticeable that no less than 29·1 per cent. of the 0·656 gramme of glycogen present at starting disappeared in eight hours.

If we give attention to the experiment previously referred to in which the pounded with sand portion showed the loss of only 2·06 per 1000, and look at the loss in relation to the glycogen present at starting instead of to the liver, so as to bring the figures of the two experiments into a right position for comparison, we get the loss of 29·1 per cent. in the chloroform experiment standing by the side of 3·8 per cent. in the other. It is true, the time of exposure in the incubator was in the one case eight hours against four in the other, but in the presence of the stream of chloroform vapour passed through the product the effect of micro-organisms would be excluded. In the part of the experiment where a stream of air instead of chloroform vapour was employed the loss stood at 23·9 per cent. How, it may be asked, are these high results to be reconciled with the experimental evidence that constitutes the only direct testimony supplied in support of what is contended for? The presumed influence of vitality having been eliminated from the question by the pounding with sand, whence came the power to produce the marked effect found to occur?

The method of extracting glycogen adopted by Dr. Paton constitutes a very prolonged operation. It involves, according to the directions given, boiling the portions of liver examined in over a litre of water for about twelve hours, the water being renewed as evaporation takes place. During this process there is violent bumping, which incurs the risk of a fruitless result from the vessel breaking. Nothing, it is true, can be said against it as far as the removal of glycogen is concerned, but from the length of time occupied in the boiling and subsequent evaporation it presents inconveniences of no small weight, and it is not unreasonable to consider that the possibility exists of a certain amount of change occurring. Extraction by the repeated performance of boiling in a capsule, rubbing down in a mortar, and straining by squeezing through linen, constitutes a process more in accord with ordinary laboratory procedure. Six, seven, or eight times at least the operation should be repeated. By finally treating the residue with the potash process, information is obtainable of the amount of glycogen that has escaped extraction with water. Without the information thus supplied, doubt must be considered to hang over the results. By the potash process I mean the process I have described, consisting of dissolving in potash solution with heat, precipitating with alcohol, collecting the precipitated glycogen, and after treatment with sulphuric acid estimating it as glucose. Glycogen, as I have shown, like starch, and in contradistinction to the dextrins, has the power of almost completely resisting destruction in the process of boiling with potash according to the plan recommended. I have previously dealt with this point at p. 42, and I consider not only that the process may be accepted for supplying the supplementary information it is designed to do, but that without its employment incompleteness must be looked upon as existing.

The subjoined examples give a view of the results I have obtained. In each case it was the liver of the rabbit, in portions of about 10 grammes, taken immediately after death, that was dealt with. The preliminary steps as regards mincing, pounding with sand, and plunging the check portion into boiling water, were precisely the same as those adopted by Dr. Paton. The sand employed about equalled in weight that of the liver : 150 c.c. constituted the quantity of salt solution used. The time of exposure in the incubator was in each experiment approximately four hours, and the temperature of the incubator 37° C.

For the sake of uniformity the carbohydrate is represented throughout as glucose. We thus get equivalent terms of expression for representing loss of glycogen and gain of sugar. At the same time the figures supply a proper comparative representation of the total carbohydrate in the respective specimens of liver belonging to an experiment. A changed position of the carbohydrate takes place, and if everything were represented with exactitude, and we were quite sure that in every case nothing besides a changed position occurred, the totals should agree. Experience shows that this cannot altogether be looked for, but no very material divergence in properly conducted experiments should be visible; and looking at all the circumstances of the case, including the several steps of analysis to be gone through, it may be regarded as even surprising how close an accord is sometimes obtained.

It is to be understood that the figures throughout the experiments are to be read as expressing parts per 1000.

Experiment A.

Liver experimented upon in the manner described.

Extraction with water and concentration. Treatment with Brücke's process. Precipitation of glycogen from filtrate by four times its volume of methylated spirit. Collection of precipitate and washing according to method described by Dr. Paton, and estimation of glycogen by weighing. Residue from aqueous extraction treated by potash process, and glycogen determined by conversion with sulphuric acid into glucose and titration with the ammoniated cupric test.

	Extracted glycogen expressed as glucose.		Glycogen in residue expressed as glucose.		Total glycogen expressed as glucose.
Check	26·11	...	4·37	...	30·48
Pounded with sand .	9·45	...	4·55	...	14·00
Minced	0·94	...	2·55	...	3·49

Other portions of the same liver after being similarly experimented upon were extracted with alcohol (methylated spirit) for the separation and determination of the sugar. Residue from alcoholic extraction extracted with water in the autoclave for half an hour at 150° C., and afterwards further by three poundings, boilings, washings, and strainings. The glycogen (the sugar, it will be remembered, was previously removed by the extraction with alcohol) was determined — after separation of the albumin by faintly acidifying with acetic acid and boiling—by conversion into glucose, by sulphuric acid in the autoclave, and titrating with the ammoniated cupric test. The amount of glycogen left in the residue from aqueous extraction was ascertained by the employment of the potash process. As was to be expected from what is known about the difficulty of extraction after alcohol coagulation it comes out higher than in the case of the other portions of liver.

		Sugar expressed as glucose.	Extracted glycogen expressed as glucose.	Glycogen in residue expressed as glucose.	Total glycogen expressed as glucose.	Total carbo-hydrate expressed as glucose.
Check	Before sulph. acid . .	11·67	24·24	7·05	31·29	42·69
	After sulph. acid . .	11·40				
Pounded with sand	Before sulph. acid . .	16·23	13·20	11·80	25·00	46·87
	After sulph. acid . .	21·87				
Minced	Before sulph. acid . .	26 07	13·03	4·60	17·63	44·02
	After sulph. acid . .	26·39				

It will be noticed, on looking at these two sets of figures, that whilst those for the check portions stand quite close, those for the portions subjected to the changing influence of exposure in the incubator present, although running in the same order for the minced and pounded with sand, a wide difference. The same kind of difference is elsewhere observable. The production of dextrins as a part of the change occurring during exposure in the incubator presumably accounts for it in conjunction with the effects of the different strengths of spirit employed in the two cases.

Experiment B.

Extraction with water. Liquid obtained concentrated, and treated with ten volumes of methylated spirit to separate sugar and glycogen. The alcoholic liquid evaporated and sugar determined. The precipitated glycogen treated with sulphuric acid in autoclave and estimated as glucose. The residue from aqueous extraction subjected to potash process.

		Sugar expressed as glucose.	Extracted glycogen expressed as glucose.	Glycogen in residue expressed as glucose.	Total glycogen expressed as glucose.	Total carbo-hydrate expressed as glucose.
Check	Before sulph. acid . .	6·99	25·77	3·96	29·73	37·71
	After sulph. acid . .	7·98				
Pounded with sand	Before sulph. acid . .	16·46	10·66	3·19	13·85	34·43
	After sulph. acid . .	20·58				
Minced	Before sulph. acid . .	17·83	5·94	4·94	10·88	36·89
	After sulph. acid . .	26·01				

Portions of the same liver extracted at once with alcohol, and otherwise subjected to the same process as described for the latter series of results in Experiment A.

		Sugar expressed as glucose.	Extracted glycogen expressed as glucose.	Glycogen in residue expressed as glucose.	Total glycogen expressed as glucose.	Total carbo-hydrate expressed as glucose.
Check	Before sulph. acid . .	10·27	31·81	4·36	36·17	46·64
	After sulph. acid . .	9·96				
Pounded with sand	Before sulph. acid . .	23·73	11·80	6·15	17·95	40·56
	After sulph. acid . .	28·61				
Minced	Before sulph. acid . .	22·72	16·15	4·96	21·11	45·58
	After sulph. acid . .	24·47				

Experiment C.

Extraction with water and procedure by method adopted by Dr. Paton described in connection with Experiment A. Glycogen in residue, determined by potash process. In the pounded with sand portion there was a loss from a little escape of the initial extraction liquid from a cracked vessel whilst standing on the laboratory bench, but the amount was ascertained and fully allowed for.

	Extracted glycogen expressed as glucose.	Glycogen in residue expressed as glucose.	Total glycogen expressed as glucose.
Check	43·18	3·69	46·87
Pounded with sand .	28·4	2·34	30·74
Minced . . .	24·15	2·19	26·34

Portions of the same liver after incubation extracted with alcohol and the residue submitted to the process described for the second set of results in Experiment A.

	Extracted glycogen expressed as glucose.		Glycogen in residue expressed as glucose.		Total glycogen expressed as glucose.
Pounded with sand .	21·52	...	9·19	...	30·71
Minced . . .	26·90	...	4·01	...	30·91

Experiment D.

Extraction with water and concentration.

Precipitation of glycogen from concentrated liquid by ten times its volume of methylated spirit, and determination as glucose after conversion in the autoclave by sulphuric acid. Residue from aqueous extraction treated with potash in the usual way.

	Extracted glycogen expressed as glucose.		Glycogen in residue expressed as glucose.		Total glycogen expressed as glucose.
Check . . .	96·09	...	4·38	...	100·47
Pounded with sand .	77·04	...	4·45	...	81·49
Minced . . .	72·06	...	3·75	...	75·81

I will now pass to experiments in which the liver, before being placed in the incubator, was put into a freezing mixture and frozen into a hard mass. I take it that such a proceeding applied to such a structure may be considered to destroy the vitality suggested as constituting the source of the active amylolysis under consideration. Certainly, I imagine, there can be no question of the destruction of vitality by freezing and keeping in an ice safe for twenty-four hours. Experiments of this nature will be adduced.

I have not looked at this matter before from the side of loss of glycogen as a measure of amylolytic action; but I have long known that sugar is produced in a frozen liver that has been allowed to thaw without any apparent difference from what occurs in the unfrozen state, allowing of course for the difference that the sudden reduction of the initial temperature would exert.

In speaking in my work of the suspension and not destruction of the capacity for sugar production by freezing, I say (p. 135), "Observation, in fact, shows that a piece of frozen liver which has been allowed to thaw, and is afterwards set aside at an ordinary temperature, contains in some hours' time about as much sugar as a piece of liver which has not been frozen."

Experiment E.

This experiment, which was confined to the estimation of sugar in frozen and unfrozen portions of liver, illustrates what I have just stated. A rabbit was killed and a piece of liver instantly removed and plunged into a freezing mixture. The other portion was taken in the ordinary way, but speedily, and subjected to experiment. Specimens from both the frozen and non-frozen portions after weighing were treated with alcohol for the extraction of sugar. These constitute the check specimens. The frozen portion supplies a representation of the condition existing previous to the occurrence of *post-mortem* change. In the other portion the higher figures are the result of a certain amount of *post-mortem* change having occurred. The other specimens were placed in the incubator for four hours, and then treated with alcohol and the sugar extracted.

Frozen Liver.

		Sugar expressed as glucose.
Check	Before sulphuric acid	1·85
	After "	2·76
Pounded with sand	Before "	14·29
	After "	17·33
Minced	Before "	15·87
	After "	16·84

Non-frozen Liver.

		Sugar expressed as glucose.
Check	Before sulphuric acid	4·75
	After "	5·44
Pounded with sand	Before "	18·47
	After "	17·10
Minced	Before "	17·90
	After "	16·58

Experiment F.

The liver used for this experiment was from the same rabbit as in Experiment D; a portion of it after having been placed in a freezing mixture was put into an ice safe, and kept there for twenty-four hours, when the specimens were dealt with precisely as in Experiment D. On removal from the ice safe it was found to have lost the hard frozen state existing when put in. It will be seen that as regards percentage of glycogen somewhat lower figures are presented.

The fall from change in the incubator very closely coincided with that noticeable in connection with the part of the liver that had not been frozen.

	Extracted glycogen expressed as glucose.		Glycogen in residue expressed as glucose.		Total glycogen expressed as glucose.
Check . . .	84·34	...	5·81	...	90·35
Pounded with sand .	67·23	...	3·52	...	70·75
Minced . . .	61·15	...	4·04	...	65·19

Experiment G.

In this experiment, after the animal was killed, the whole liver was taken out, well pressed in a cloth, and then placed in a freezing mixture. In half an hour, whilst in a hard frozen state, it was removed, rinsed on the surface, wiped, and put into an empty wide-mouthed stoppered bottle, which was closed and immersed in a mixture of ice and salt. The whole was then placed in an ice safe for twenty-four hours. The design was to keep the liver in a hard frozen state, and with the arrangement adopted it was found in this state at the end of the time. After having become thawed, portions were next taken for experiment. One was minced and put at once into boiling water; another, pounded with sand and mixed with 150 c.c. of salt solution; and a third, minced and put into a similar quantity of the salt solution. The last two were then exposed as in previous experiments for four hours to a temperature of 37° C. in the incubator. The process of extraction of glycogen and sugar with water was next carried out, and the residue afterwards treated as usual with potash for the determination of the unextracted glycogen as glucose. In order to make the analysis as complete as possible the sugar in the product of aqueous extraction was determined as well as the glycogen, and the latter by both Brücke's process and estimation as glucose. To accomplish this the aqueous liquid was evaporated down and brought to 200 c.c. Half was taken for Brücke's process and weighing. The other half was put into ten times its volume of methylated spirit, and the spirit containing the sugar was separated by filtration from the glycogen precipitate, evaporated, and the sugar determined before and after treatment with sulphuric acid. The glycogen precipitate was converted into glucose by sulphuric acid in the autoclave, and the glucose determined by titration.

Estimation of extracted glycogen by Brücke's process and weighing.

		Sugar expressed as glucose.	Extracted glycogen expressed as glucose.	Glycogen in residue expressed as glucose.	Total glycogen expressed as glucose.	Total carbo-hydrate expressed as glucose.
Check	Before sulph. acid . .	3·80	52·15	3·25	55·40	62·98
	After sulph. acid . .	7·58				
Pounded with sand	Before sulph. acid . .	24·08	31·32	2·40	33·72	71·90
	After sulph. acid . .	38·18				
Minced	Before sulph. acid . .	29·46	32·30	3·74	36·04	75·12
	After sulph. acid . .	39·08				

The next table represents the extracted glycogen estimated by conversion into glucose and titration. As it was only this part of the analysis that differed, the figures for the sugar and for the glycogen in the residue serve alike for both tables, and are repeated for the calculation of the total carbohydrate.

Estimation of extracted glycogen by conversion into glucose and titration.

		Sugar expressed as glucose.	Extracted glycogen expressed as glucose.	Glycogen in residue expressed as glucose.	Total glycogen expressed as glucose.	Total carbo-hydrate expressed as glucose.
Check	Before sulph. acid . .	3·80	46·76	3·25	50·01	57·59
	After sulph. acid . .	7·58				
Pounded with sand	Before sulph. acid . .	24·08	20·05	2·40	22·45	60·63
	After sulph. acid . .	38·18				
Minced	Before sulph. acid . .	29·46	18·26	3·74	22·00	61·08
	After sulph. acid . .	39·08				

It happened without its being known at first that the methylated spirit used for this experiment consisted of the commercial article which had been by mistake sent to the laboratory instead of the purer kind ordinarily supplied. The discovery was made in connection with the application of Brücke's process and the precipitation of glycogen for weighing. When the spirit was added to the watery liquid, owing to the extent to which it became diluted, some of the oily impurity separated out and subsequently impeded filtration, and thus may have given rise to the higher figures for the glycogen estimated by weighing, and the discordancy in the total carbohydrate. Taking the experiment as a whole, it speaks in unmistakable language. The glycogen figures obtained by titration, it may be stated, would not be liable in the same way to be affected by the spirit.

Under the vitality view, neither the minced nor the pounded with sand portions of liver should have given any material evidence of change. In reality, both through production of sugar and loss of glycogen, a similar extent of change will be seen to have occurred as is noticeable in the recently removed liver allowed to remain under ordinary circumstances.

By this array of evidence Dr. Paton's work is completely controverted. The view attributing the active production of sugar noticeable in the liver immediately after death to the effects of a prolongation of vitality in the cells is in the strongest manner contradicted. Everything conspires to sustain the view, originally propounded by Bernard and since generally accepted but now attempted to be overthrown, that it is the result purely and simply of ferment action. It is known to be a feature of ferment action in general that the process occurs with much greater activity at first than later on. As time advances it progressively slows down and what is noticeable, therefore, in the case of the liver strictly agrees with what might be looked for.

But, there is an experimental discrepancy which requires to be accounted for. Much variation is traceable in the results derived from different livers. Nothing, however, has been come across at all reconcilable with the evidence on which Dr. Paton has based his conclusion. In search for something to explain the difference between us, I tried the effect of a deviation in the manipulative process with the view of ascertaining what kind of result would be given by an imperfect extraction of glycogen from the check and minced portions. The pounded with sand portion stands, by virtue of what has been done, in a favorable position for the full extraction of glycogen. The other two, however, are not similarly circumstanced and require attention to be given to place them so. Actual experiment gave me results showing an accord with Dr. Paton's figures and thus suggested the possibility of error of manipulation having occurred leading to an imperfect extraction of glycogen in the case of the two coarsely minced portions.

Under the circumstances now presented, I deemed it right to place myself in communication with Dr. Paton and acquaint him of the results I had obtained. I wrote to him and furnished him with a detailed representation of some of my experiments and suggested that probably under the circumstances he might consider it desirable to repeat his own before I published mine.

I received from him a reply, in character with the general tone of his writings, intimating that it was impossible for him at the moment to set aside other work in order to repeat experiments upon a subject which *at present* he did not think required further investigation. He in addition stated, with respect to the question of manipulative error, that the process adopted strictly accorded in all its details with the directions given in a previous part of his paper.

I cannot do otherwise than accept this assurance; but doing so involves the necessity of at the same time saying that Dr. Paton must have happened to drop upon livers in an exceptional state.

I have shown from the experimental evidence already adduced that from different livers very discordant results are obtained. I have carried my work on, and the further evidence obtained substantiates this statement.

The experiments with the imperfect aqueous extraction of glycogen are available for illustration, inasmuch as the glycogen in the residue was also determined, and the total glycogen serves to represent what is wanted. I will give the series in a tabular form.

Total Glycogen per 1000 *from Aqueous Extraction and Treatment of the Residue with Potash.*

	Check.	Pounded with sand.	Minced.
I. (a poor liver) ...	10·79 ...	6·67 ...	7·43
II.	25·00 ...	20·81 ...	18·36
III.	58·84 ...	* ...	48·09
IV.	24·29 ...	7·81 ...	5·06
V.	63·42 ...	51·17 ...	46·81
VI.	80·24 ...	70·90 ...	55·80

The following is an additional experiment in which the ordinary method of procedure by full aqueous extraction was adopted. It also illustrates the variability that is noticeable.

	Glycogen extracted.	Glycogen in residue.	Total glycogen.
Check . . .	71·76 ...	2·25 ...	74·01
Pounded with sand .	68·62 ...	1·3 ...	69·92
Minced . . .	52·54 ...	2·03 ...	54·57

I will now proceed to give some experimental results based on the simple determination of sugar extracted by the agency of water. It is generally admitted, and the view is endorsed by Dr. Paton, that glycogen is the only source of the sugar produced. For the supply of evidence, therefore, on the point in question it does not signify whether it is drawn from the loss of glycogen or gain of sugar. This difference, however, exists. The one is a lengthy and

* Lost by breaking of flask. The figures for the check and minced portions serve to illustrate the variability presented in the case of different livers.

troublesome operation, whilst the other is susceptible of being much more quickly and easily carried out, and, on account of its greater simplicity, is less open to the chance of accidental error occurring. Indeed, all that is necessary is to proceed with successive poundings, extractings with moderate quantities of boiling water, and strainings, to securely remove the sugar, and then, after concentration, to titrate direct with the ammoniated copper solution. With 10 grammes as the quantity of liver experimented with concentration need not be carried further than to about 100 or 150 c.c. If necessary for the easy filtration of the concentrated liquid some sodium sulphate may be used.

It will be understood that in the following experiments what is represented is the cupric oxide reducing power of the product expressed as glucose. It reveals extent of sugar production but whether the product consists of glucose or of a mixture of glucose and a sugar of lower cupric oxide reducing power, is not shown.

Experiments on Rabbits. Exposure in Incubator for Four Hours. Sugar per 1000.

	Check.	Pounded with sand	Minced.
I. The liver was plunged immediately after death into a freezing mixture. It was hence not thought necessary to take a check portion	—	6·59	13·2
II. Do.	—	11·70	13·13
III. Do.	—	11·80	18·00

	Check.		Pounded with sand.		Minced.
IV. Frozen as above, and check portion taken . . .	4·27	...	18·14	...	22·96
V. Taken instantly after death without being frozen . .	6·66	...	26·96	...	20·19
VI. Do. . .	6·32	...	15·66	...	17·82

Experiments on Cats. Sugar per 1000.

	Check.		Pounded with sand.		Minced.
I. Liver treated immediately after death	—	...	44·11	...	26·40
Portions treated 20 minutes after death	18·20	...	36·91	...	—
II. —	11·21	...	45·23	...	26·14
III. —	8·53	...	24·90	...	22·56

In the case of the cat, then, it is to be noted that in each of the three experiments that have been performed the results obtained stand diametrically opposed to what is contended for by Dr. Paton. The same state of things is noticeable in Exp. V upon the rabbit; and it may be further said that a certain number of examples of a like nature presented themselves amongst the experiments included in the group given at the beginning.

What, now, is to be said of the experimental evidence standing as the basis of Dr. Paton's communication contained in the 'Philosophical Transactions'? If we look at it, what does it consist of? One experiment accompanied with a defective one in which owing to accidental loss from cracking of a beaker one of the results has to be expressed by the figures given, followed by + an unknown quantity. Against this association of one experiment and a defective one there are to be found later on in the communication two results, which I have commented upon at p. 80, supplying evidence of a nullifying nature. The position must be pronounced to constitute an anomalous one.

From all sides evidence is forthcoming telling in the strongest manner against what is contended for, and yet, by the force of circumstances, the "paper" has found a place in the Royal Society's 'Transactions.' To clear the course of my work which blocked the way, its author has not stood at scruples but has coolly delivered himself in a foot-note (which I will help him in giving prominence to by here placing in capitals) as follows :

"THE THEORY OF PAVY, REPEATED IN NEARLY EVERY TEXT-BOOK, THAT THE LIVER IS 'A SUGAR DESTROYING, AND NOT A SUGAR-FORMING ORGAN,' RESTS UPON SO UNSUBSTANTIAL A BASIS AND HAS BEEN SO COMPLETELY REFUTED BY THE WORK OF SEEGEN, AND OTHER INVESTIGATORS THAT IT NEED NOT BE CONSIDERED."

For judgment I appeal to the facts and reasonings adduced in my recently published book and in this epicriticism.

There is a factor to which consideration has escaped being given that may serve to account for the *post-*

mortem production of sugar that occurs in the liver. With reference to the source of the difference evidently existing in the *ante-* and *post-mortem* states I suggest in my work (p. 144) that there becomes developed at death an energetic sugar-forming ferment, or that there must be some restraining or inhibitory influence during life preventing the ferment change which occurs after death.

As a result of the attention I have recently been giving to this matter, Heidenhain's discovery relating to zymogens and enzymes has presented itself to my mind as possibly capable of giving us the clue to the required explanation. With the presence of the zymogen during life and not the enzyme, the escape from the production of sugar—I will go further and say our escape from diabetes—is rationally to be accounted for. With the production of enzyme, on the other hand, from the zymogen as a *post-mortem* event, like the event that occurs in the case of the coagulation of the blood, production of sugar may naturally be expected to follow.

I have to some extent experimented upon the subject, and, as far as I have gone, have obtained results that tend to favour the view suggested. To more than this upon the point, however, I will not at present commit myself.

From this review of Dr. Paton's experimental work I will proceed with the examination in hand of his criticism of my book. He says:

"Is hepatic glycogen changed to glucose in the normal state of the animal? Pavy argues against this as follows. In the first ten or twelve minutes after death 10 or 12 parts per 1000 of sugar is formed. At this rate all the glycogen would disappear in three quarters of an hour. Hence, if amylolysis goes on at the same rate in the living animal (we have in the 'Philosophical Transactions' argued against this)," [no, what Dr. Paton has argued against is that in the absence of vitality amylolysis does not go on at the same rate as under the influence of the existence of vitality] "in starvation, sugar would not be found in the liver (he himself contends against the view that it is so found)" [Dr. Paton incorrectly introduces the word "*sugar*" for *glycogen* (pp. 144–5), and then founds a cynical remark upon what he wrongly puts into my mouth] "because the glycogen would all be used up. Surely he is the last person to use this argument, since he himself contends that carbohydrates are yielded by the splitting, of the proteid molecule, and proteids are constantly split up during starvation."

Surely Dr. Paton must have imperfectly grasped the situation. I distinctly contend, in opposition to the prevailing view, that the liver is not a proteolytic organ, saying at p. 121: "Authorities speak of glycogen taking origin from proteid matter within the liver by a proteolytic action exerted by its cells. It has been in this way considered that the presence of glycogen is to be accounted for under subsistence upon animal food. I have shown, however, that another explanation can be given, without looking to the liver for the performance of a proteolytic office. The source of the glycogen is sufficiently to be accounted for by the occurrence of the same kind of action as that which is in operation after the ingestion of vegetable carbohydrate food. It is the province of ferment action to break down and hydrate, and of protoplasmic action to synthesise and dehydrate. The proposition, therefore, which I have advanced stands in accord with the natural order of events, whilst to assume that the protoplasmic liver-cells perform the work of ferment action does not. That they should dehydrate or transmute sugar into glycogen is, on the other hand, consistent with what can be shown to be accomplished by protoplasmic matter elsewhere."

Further, upon the point concerning glycogen and proteid matter, I say, at p. 236: "I have referred to the protoplasmic transmutation of sugar into glycogen in the liver. But the transmutative power by no means exclusively belongs to the liver—indeed, I am disposed to think that it exists as a general property of the protoplasmic matter of the body. Certainly, glycogen is recognisable, and even it may be largely so, in other parts of the body, and it is probably produced at its seat of presence. Every part of the body contains sugar, intrinsically belonging to it, which may come from the proteid matter around. The glycogen may be derived from this sugar, or it may possibly constitute a product of the cleavage process. In the multifarious actions occurring in a living part there is, doubtless, much complexity of result arising from the antagonistic effects of ferment and protoplasmic actions."

Because proteolysis may be going on throughout the body in starvation it by no means follows that the liver should continue as when under the influence of food supply to be the seat of special production of glycogen.

In the next paragraph Dr. Paton says :—" Dr. Pavy (p. 225) also advances the following argument. He takes the case of the rabbit, in which animal the alimentary canal is always more or less full. He says that the portal blood, always contains more sugar than the blood of the general circulation, and argues that therefore glycogen must be changed to something not sugar. We do not accept his assertion :" [Dr. Paton without the support of a single experiment confronts the direct and extensive analytical evidence given at pp. 103–8 on the condition of the portal blood with the presumptuous statement 'we do not accept his assertion'] " but taking his own figures, what is the state of matters? The amount of sugar in the portal blood of the rabbit in full digestion varies enormously—from 1·44 to 4·6 mille." [Yes, it varies with the amount of carbohydrate ingested and absorbed.] " I cannot in his book find any analysis of the sugar in the blood of the general circulation of the rabbit, but in the dog (p. 168) it varies from 0·8 to 1·23 per mille." [Dr. Paton is here at the wrong part of the book. He has picked up the part headed *comparative examination of arterial and venous blood collected simultaneously from the dog during life* and misapplied it. In the proper place, under the head of amount of sugar in blood, at p. 161, I say—From a collection of upwards of 100 observations, conducted at various times over a number of years, upon the dog, cat, rabbit, sheep, ox, horse and pig, the amount of sugar naturally present in the blood of the general circulation may be stated to range from about 0·6 to 1·0, or a little over 1·0, per 1000.] "Now, if sugar is being constantly destroyed or got rid of from the blood by the kidneys, as we shall afterwards see is maintained by Dr. Pavy," [what is maintained by me is that with the small quantity of sugar existing naturally in the blood, as elsewhere throughout the system, there is a corresponding small quantity of sugar present in the urine, but the smallness of the quantity is such as to escape being revealed by ordinary testing and to deprive it therefore from possessing any bearing connected with the physiological application of carbohydrate matter] "the amount of sugar leaving the liver may be sufficiently great to prevent glycogen accumulating without at the same time raising the percentage of sugar in the general circulation."

It is fruitless to argue upon false premises. It is not the fact, as will be obvious to every one else, if not to Dr. Paton, that the sugar existing in healthy urine is sufficient in quantity to play the part argumentatively suggested. The quantity of sugar in healthy urine, regarded from a physiological point of view, is altogether too insignificant to do more than serve as an illustration of the relation that exists between the state of the urine and that of the blood. If sugar reached the blood from the liver to prevent the accumulation of glycogen, so in proportion would it reach the urine and lead to the production of diabetes.

In the next point of criticism Dr. Paton deals with "*Blood in relationship to sugar*" and says "As to the nature of the sugar in the blood, Pavy states (p. 157) that 'the sugar met with is usually one possessing a cupric oxide reducing power more or less below that of glucose.' This statement is in direct opposition to the observations by Seegen and to the recent observations of Pickhart ('Ztsch. f. phys. Chem.,' Bd. xvii), which satisfactorily prove that the sugar of blood in all its reactions corresponds to glucose."

What kind of language is appropriate for the misrepresentation that here occurs? Especially bearing in mind that we are dealing with science will it not be considered an unpardonable offence for the words quoted to be taken from the position they hold and applied as Dr. Paton has done? I will give the paragraph as it stands in my work, placing the quoted words in italics.

"Observation, conducted upon blood derived from different sources, shows that the kind of sugar found in all parts of the circulation, with the exception of the portal system, possesses a cupric oxide reducing power that is not, as a rule, increased, and, if increased, only slightly so, by boiling with sulphuric acid—a character which implies, broadly speaking, the existence of glucose. This, I may state upon the strength of a very large number of observations, is the kind of sugar present under ordinary or natural conditions, but, as will be subsequently shown, under certain deviations from the ordinary state, as, for instance, after the administration of anæsthetics, the inhalation of carbonic oxide, &c., *the sugar met with is usually one possessing a cupric oxide reducing power more or less below that of glucose.*"

Again, at p. 102, I say, "It will be observed in the analytical results to follow that the kind of sugar found in the portal blood is one possessing a cupric oxide reducing power standing ordinarily below, and sometimes considerably below, that of glucose. As will be seen from what will be stated later on, a point of difference here exists between the sugar of the portal blood and that present in the general circulation, which, under natural circumstances, is found to have the cupric oxide reducing character of glucose."

Dr. Paton's next paragraph runs: "Pavy no longer maintains, as he once did, that sugar is absent from the blood. He no longer maintains, as he did at a somewhat later date, that it is only a trace—about 0·5 per mille., but he now gives results more nearly corresponding to those of other observers. His results are still lower than those usually obtained. He gives from 0·6 to 1·0 per mille., while Seegen finds about 1·5 per mille. on an average. It is not sufficient to argue that Seegen's results are erroneous because 'the larger quantities are incompatible with the state presented by healthy tissue.'" The word tissue is here erroneously inserted for urine.

In his efforts to disparage, Dr. Paton does not shrink from assuming a bold front. He writes in this way in the face of what is found at p. 162 of my book, running as follows :—" In former times, when less facility existed for the quantitative determination of the sugar of the blood, the amount corresponding with what I have represented as naturally present during life was usually spoken of as a 'trace,' the expression being founded upon the slight reaction yielded by the copper test. I do not think previous attempts had been made to determine what this 'trace' meant, expressed in figures, but in my communication contained in the 'Transactions' of the Royal Society for 1860, I gave the results I had obtained from the analysis of three specimens of the blood of dogs, and stated that the quantities stood respectively at 0·470, 0·730, and 0·580 per 1000. These amounts it will be seen, although obtained upwards of thirty years ago, coincide closely with those yielded by the present improved methods of analysis."

Thus, what I said thirty-five years ago stands in practical conformity with what I say now. I have no misgiving about the figures mentioned by me being correct. They agree with the figures obtained by modern authorities in general with which Seegen's figures differ. In truth, what has occurred is that the figures formerly given by others as representative of the normal state are no longer accredited, and that those given by modern authorities coincide with mine, instead of mine having undergone any essential alteration.

Under the head of "*The Utilisation or Non-utilisation of Sugar in the Body*" Dr. Paton's remarks are of a general nature until we reach the latter part. In these general remarks there is nothing calling for notice from me except to record that Bock and Hoffmann, Abeles, and Seegen are mentioned as standing in accord with me upon the point of absence of difference between arterial and venous blood with respect to amount of sugar.

Next, the criticism runs :—"The recent interesting work of Lépine and Barral on the destruction of glucose by a ferment in the blood must be considered. It cannot be summarily dismissed as it is by Dr. Pavy (p. 177) by saying, 'Sugar does not reach the general circulation as the glycogenic doctrine implies, the *raison d'être* of a glycolytic ferment in the blood does not exist, and the question about glycolysis in the blood is devoid of the physiological significance that would otherwise belong to it.'" Is this ingenuous criticism ? Is it a criticism with the straightforwardness belonging to it that ought to be expected in the presence of Science ? Dr. Paton puts his quotation as representing so many affirmations, and for the purpose begins with a capital S although he picks the quotation up in the middle of a sentence. He says the work of Lépine and Barral is summarily dismissed by me by the quotation given. Does this stand in accord with truth? In order that the reader may judge I will quote what I say, placing in italics that which Dr. Paton has thought it a becoming action to omit. "*Weighty objections have been raised by various authorities to the experiments and conclusions of Lépine, looked at upon their own merits. There is, however, the further point, which has not been considered by others, that if, as follows from what has been adduced in this work,* sugar does not reach the general circulation as the glycogenic doctrine implies, the *raison d'être* of a glycolytic ferment in the blood does not exist, and the question about glycolysis in the blood is devoid of the physiological significance that would otherwise belong to it."

Continuing from the quotation given Dr. Paton's words run, "Far from this being the case, the recent discovery of the fact that after removal of the pancreas this glycolytic action of the blood diminishes, and the fact that extirpation of the pancreas causes, glycæmia and diabetes, renders these observations of Lépine and Barral of the greatest practical importance. Why sugar should be thus destroyed we know not; but the evidence in favour of its destruction appears satisfactory. Possibly in this ferment we have a means of getting rid of any sugar which has been allowed to pass from the liver in excess of that required by the tissue."

What does this amount to? First, the assertion that certain observations relating to glycolysis in the blood are rendered of the greatest "practical" importance, and then the admission "why sugar should be thus destroyed we know not," followed by the suggestion, "possibly in this ferment we have a means of getting rid of any sugar which has been allowed to pass from the liver in excess of that required by the tissue." What need be said upon this argumentative display, based upon such purely hypothetical premises?

Nothing has hitherto been adduced to place the doctrine of glycolysis in the blood in a position of touch with the requirements of physiology. It is seen Dr. Paton stands forward and suggests that possibly it may be the means of getting rid of any sugar which has been allowed to pass from the liver in excess of that required by the tissue. In what part of the circulatory system does this glycolysis occur? If in the arteries it would deprive the tissues of sugar which is assumed to be required to be transmitted to them. Is it only in operation during the existence of the assumed excess in the blood? If not, again it would be standing in antagonism with the needed supply to the tissues. And all this time the kidney which receives the same blood as the tissues escapes betraying any indication of this hypothetical varying condition, although there is rrefutable evidence to show that the state of the urine moves in a most sensitive manner with that of the blood in relation to sugar. The point of real importance touched upon in the paragraph is the connection of the pancreas with the occurrence of glycosuria. For the explanation of the glycosuria following complete extirpation of the pancreas we need not look in the direction of diminished glycolysis in the blood. The abnormal passage of sugar into the systemic circulation through the wrong state arising from the absence of the influence of the pancreas will just as readily account for it.

"But Dr. Pavy himself admits that sugar is present in the blood, and hence, if it is not to accumulate there, it must be changed or excreted by the kidneys."

Certainly, sugar is present as a constitutional ingredient of the blood just as it is present in all the structural components of the body and in the urinary excretion derived from the body which, neither more nor less, constitutes a participant of the general state. The presence of sugar in the blood from this point of view is a very different thing from sugar reaching the blood for functional transit to the tissues for destruction. Did this happen there would be sugar, in proportion to the extent to which it reached the circulation for functional transit, beyond the ordinary constitutional amount to deal with, and as the blood containing such assumed sugar for functional transit flows alike to the kidney and tissues there should be sugar in the urine beyond that associated with sugar as a constitutional component of the blood. As a matter of fact, however, we find a certain standard condition of blood and urine existing irrespective of nature of food and of fasting or repletion which is inconsistent with the supplementary passage of sugar into the circulation for functional transmission to the systemic capillaries for destruction. In reality, when an extraneous entry does take place, no matter from what cause arising, it is immediately rendered manifest by the production of a cotemporaneous and concordant altered condition of blood and urine, and, as a concurrent event, the entry being met by proportionate elimination with the urine, accumulation is kept down. In connection with the presence of sugar in the blood only as a constitutional component and not for functional transit, the question of elimination for preventing accumulation does not similarly present itself as a required operation.

Next follows—" *The urine in relationship to Sugar:* —Dr. Pavy confirms the results of other observers that sugar in minute quantities occurs in the urine in health. He says that 0·05 per cent. is present. This is in opposition to the recent most careful observations of Baisch ('Ztsch. f. phys. Chem.,' Bd. xviii), who finds that the sugar as ascertained after *separation* estimated by its power on polarised light amounts to at most 0·009 per cent., or estimated by its reducing power to 0·012 per cent. These results correspond closely to those of Seegen."

What does this criticism amount to? A question of *quantity* of sugar present in healthy urine, which at the most is only quite minute. Dr. Paton's assertion, however, that I say that 0·05 per cent. is present is not strictly correct. I intentionally wrote guardedly and, instead of absolutely committing myself upon the point, stated (p. 187), that after precipitating with the neutral followed by the basic acetate of lead "If the cupric oxide reduction found to occur be read off as produced entirely by sugar, the amount of this principle ordinarily existing in healthy urine may be said to stand at about 0·5 or a little over per 1000." Looking at the criticism also from the point of view of Baisch's figures, it is to be noted that they refer to "sugar as ascertained after *separation*." Is proof forthcoming, it may be reasonably asked, that loss did not occur in the process of separation previous to estimation? Seegen, it is to be stated, argues against the presence of sugar in normal urine ('Glycogénie Animale,' Paris, 1890, p. 218), and consequently gives no figures to be dealt with.

"The fact that glucose when injected into the blood appears in the urine has been known for long. Pavy contends that the amount of sugar in the urine is strictly proportionate to that in the blood. His experiments on p. 189 do not substantiate this thesis, and the observations of Seegen on diabetes are opposed to the conclusion."

The experiments in question are on the injection of sugar into the jugular vein and afterwards killing the animal and examining the blood and the urine where urine was procurable. Dr. Paton omits to give consideration to an important factor bearing on the matter and passes over what I say on the following page upon the point. My words run :—"It must not escape notice in experiments of the nature under consideration, that the state of the bladder, as regards fullness or emptiness at the time of the injection, will form an important factor in determining the character of the result obtained, by influencing the extent to which the urine secreted after the injection is diluted by that previously existing."

I dispute the truthfulness of the assertion that the observations of Seegen on diabetes, if the observations are looked at as a whole, are opposed to the conclusion. They are to be found at p. 237 of his 'Glycogénie Animale.' There are irregularities undoubtedly visible in the table given but it must also be said that these irregularities on being looked into are of a nature to cast the gravest suspicion upon the intrinsic correctness of the results with which they are connected. In disagreement with modern accredited authority, Seegen places the amount of sugar present under normal circumstances in the blood as high as 1·7 per 1000. He admits that in diabetes a close relationship exists between sugar in the urine and carbohydrate matter in the food. How is this relationship to exist except through the medium of the blood? No one contends that the sugar in the urine is not drawn from the blood, but the blood is to remain uninfluenced by the ingested carbohydrate whilst the urine is influenced by it—in other words, sugar is to vanish in the circulation and start into reappearance in the urine. Could there be anything more opposed to reason than this? If Seegen's figures and teaching are correct, blood with 1·70 per 1000 of sugar in the healthy subject gives none to the urine. Looking at his diabetic cases, blood with much less sugar, viz. 1·23 per 1000 was associated with traces of sugar. In one case I note that, with a mixed diet, with 1·82 per 1000 in the blood there were 38 per 1000 in the urine, and in the same case after two days of absolute restriction to animal food 1·81 per 1000 in the blood and 6 per 1000 in the urine. In another case, with 35 per 1000 in the urine there was the large amount—the largest to be found in the table—of 4·80 per 1000 in the blood. These are some of the anomalous results to be found, and suffice, I consider, to show that Seegen's observations cannot be accepted without assuming that the blood, although the medium for yielding sugar to the urine, escapes being influenced in the manner that it is admitted the urine is.

Dr. Paton silently passes over the observations that I have given at p. 193 on the point in question. In mine, looking at the fact that the urinary sugar represents the amount passed during the twenty-four hours, and that the blood analysed was collected at a particular period within the twenty-four hours, there is noticeable what may be spoken of as a remarkably close relationship between the condition of the blood and urine in the different cases.

In addition to evidence of this kind, all experience in connection with diabetes conspires to show that the urine is most sensitive to, and speedily and proportionately influenced by, the ingestion of carbohydrate matter. For the urine thus to become influenced, it follows either that the blood must become correspondingly so or that the sugar must reach the urine in some mystic way without undergoing intermediary transit.

With the utmost confidence I re-assert the proposition that according to the sugar encountered in the urine so has sugar pre-existed in the blood; and, conversely, in proportion as sugar is present in the blood so will it reveal itself in the urine, and this alike for the small quantities belonging to health and the larger quantities belonging to diabetes.

Continuing, on the effect of the injection of sugar into the circulation, Dr. Paton says, "The most astonishing part of his experiments on this subject is that in Experiments V and VIII on rabbits, he gets a sugar in the urine after the injection of glucose, the reducing power of which is nearly doubled by boiling! Such a fact is worthy of more than passing comment. What sugar has its reducing power thus increased? Apparently the sugar in these experiments was not isolated before it was estimated, and he has no right to assume that the increased reduction is due to a sugar."

Dr. Paton does not shrink from scoffing at my experimental results with a note of exclamation. Even the young may find that they have something to learn, and notwithstanding his book-lore upon the strength of which he has written with so much arrogance and self-assurance Dr. Paton may live to find that something remains even for him to learn. Certainly he has to learn how to treat with proper respect results which may not happen to fall in with his preconceived views. He has treated with derision the results obtained in Experiments V and VIII, p. 189, in the face of the further experiments recorded and standing plainly before him at p. 238. I will here mention what I state in my book concerning these experiments which speak for themselves in such unequivocal terms as to supply a complete reply to the vague sort of criticism that has been quoted.

The experiments were throughout of the simplest possible nature. They consisted of the injection of glucose derived from honey into the subcutaneous tissue of rabbits to the extent of 1 gram per kilo. of body weight. The urine was obtained about two hours afterwards, and its cupric oxide reducing power determined before and after boiling with 2 per cent. sulphuric acid. Seven experiments upon rabbits taken consecutively are given. The term sugar comprises the cupric oxide reducing product looked at as a whole. The table of results runs as follows :—

Nature and amount of Sugar in the Urine after the subcutaneous injection into the Rabbit of 1 *gram of glucose per kilo. of body weight.*

		Sugar per 1000, expressed as glucose.	Cupric oxide reducing power of the sugar eliminated in relation to that of glucose at 100.
Rabbit I.			
10 c.c. of urine obtained ...	before sulphuric acid	48·08	73
	after „ „	65·79	
Rabbit II.			
8·6 c.c. of urine obtained ...	before sulphuric acid	19·84	38
	after „ „	51·89	
Rabbit III.			
3 c.c. of urine obtained ...	before sulphuric acid	39·68	42
	after „ „	94·46	
Rabbit IV.			
4 c.c. of urine obtained ...	before sulphuric acid	34·72	50
	after „ „	69·44	
Rabbit V.			
Over 15 c.c. of urine obtained	before sulphuric acid	21·74	61
	after „ „	35·72	
Rabbit VI.			
Over 30 c.c. of urine obtained	before sulphuric acid	10·41	54
	after „ „	19·23	
Rabbit VII.			
4·1 c.c. of urine obtained ...	before sulphuric acid	48·78	44
	after „ „	110·85	

With such results, is there any room for quibbling? As a direct effect of the introduction of glucose into the system through the subcutaneous tissue the urine became charged with a large amount of something possessing a cupric oxide reducing power more or less considerably below that of glucose. The circumstances throughout do not permit of its being conceived that this something was other than carbohydrate. At all events, if Dr. Paton contends otherwise, something more is wanted of him than a mere shallow suggestion. In three of the experiments, I treated the urine with absolute alcohol, which is a precipitant of animal gum, and found that the residue from evaporation of the alcoholic filtrate behaved in the same way as the original urine.

Since the publication of my work I have conducted a similar experiment upon a dog. Catheterism was first of all performed to empty the bladder. 7·2 grams of glucose (from honey) in 36 c.c. of water were then injected subcutaneously, giving 1 gram of glucose per kilo. body weight. The urine obtained in two hours' time was found to possess a cupric oxide reducing power expressed as glucose equivalent to 16·28 per 1000 on initial titration and to 35·56 per 1000 after the usual treatment with sulphuric acid. The initial reducing power here stands in the proportion of 45·7 to glucose at 100. Treated with phenyl-hydrazine, stellate crystals were given instead of the characteristic acicular crystals of glucosazone. They were exhibited at the meeting of the Physiological Society held at Oxford in 1894.

In connection with diabetes I have in a large number of instances determined the cupric oxide reducing power of the urine before and after treatment with sulphuric acid with the result of finding that it is not invariably glucose that is present. Now and then I have come across a specimen where the initial titration has shown a cupric oxide reducing power more or less markedly below that existing after subjection to the action of sulphuric acid.

Dr. Paton inquires, "What sugar has its reducing power thus increased?" It does not seem to have occurred to him that a mixture of carbohydrates may exist instead of a single specific sugar.

In the hydrolysis of an amylose carbohydrate, do we not get intermediate or transitional principles formed, giving a mixed assemblage in the product? This is thoroughly recognised in connection with the transformation of starch as a process occurring in the vegetable kingdom. It ought also to be similarly recognised in association with the animal kingdom, but the minds of physiologists seem here to look for the presence only of the ultimate principles of change.

Now, admitting that products are presented to us with the possession of varying cupric oxide reducing powers according to the assemblage of transition principles present derived from change in the direction of increasing hydrolysis, may it not also come to pass that products similarly are presented from change in the opposite direction. In speaking (p. 224 *et seq.*) of the transmutation of carbohydrates by increased and decreased hydration through the agency respectively of ferment and protoplasmic actions I give illustrations of the transmutation in each direction in both the animal and vegetable kingdoms, and I will here cite a few passages from what I say :—

"Whilst ferments and chemical agents transmute by increase of hydration—carrying, for instance, the principles of the amylose into the saccharose and glucose groups—the effect of the influence exerted by living matter is to reduce from the higher to the lower states of hydration."

"A notable illustration in the animal kingdom of transmutation by reduced hydration for storage is supplied by what occurs within the liver. The sugar contained in the portal system, taking origin from the carbohydrate matter ingested, is stopped by the cells of the organ and transformed into glycogen."

"I have referred to the protoplasmic transmutation of sugar into glycogen in the liver. But the transmutative power by no means exclusively belongs to the liver—indeed, I am disposed to think that it exists as a general property of the protoplasmic matter of the body. Certainly, glycogen is recognisable, and even it may be largely so, in other parts of the body, and it is probably produced at its seat of presence. Every part of the body contains sugar intrinsically belonging to it, which may come from the cleavage of the proteid matter around. The glycogen may be derived from this sugar, or may possibly constitute a product of the cleavage process. In the multifarious actions occurring in a living part there is doubtless much complexity of result arising from the antagonistic effects of ferment and protoplasmic actions."

"Further evidence is afforded, through the kind of sugar found in the urine after the direct introduction of glucose into the system, of the carbohydratetransmuting power of living matter in the direction of a diminution of hydration, taken as represented by a diminished cupric oxide reducing capacity."

"It would hardly have been surmised that such results [on the urine after glucose injection] would have been yielded. They agree, however, with the general tenor of experience set forth in this work which is to the effect that by the agency of living matter the carbohydrates are moved within the system in the direction of lessened cupric oxide reducing power instead of in the reverse direction as happens as a consequence of ferment action."

Before quitting this subject I may just make a passing allusion to a further point which fits in with what has been mentioned above. It has been distinctly noticeable that the blood after the glucose injections has yielded a larger amount of amylose carbohydrate than is ordinarily present. The matter is referred to at p. 216 in my work.

"Dr. Pavy considers that any sugar which escapes into the blood is excreted in the urine. Now, during starvation the amount of glucose in the blood remains the same, and if glucose is still being excreted in the urine, this is surely the strongest possible argument for the constant formation of sugar apart from ingested carbohydrates from whatever source derived."

Dr. Paton does not seem to be able to carry his mind beyond the conception of a mechanical ingoing and outgoing. Have not all the parts of the body, the blood included, the power of self maintenance? Sugar is a constitutional component and provision in each case may naturally be presumed to be made as with the other components by intrinsic action for its presence. I do not deny the formation of sugar as a result of the chemical changes going on everywhere. What I deny is that sugar is functionally produced in a given organ—the liver—for subsequent destruction and administration to force-production. Did this happen, its transit through the circulation would give sufficient excretion in the urine to be something to talk about, in place of the insignificant amount actually present, which can be of no import beyond affording an illustration of the relation existing between blood and urine.

The presence of sugar in muscle, spleen and other organs is next referred to. Dr. Paton endorses my results by saying "That carbohydrate is contained in these organs is already known," but he afterwards remarks "Dr. Pavy's observations seem to indicate that glucose is not *the* sugar of muscle. Now, the very careful experiments of Panormoff show that it is glucose."

This remark is on a par with his other criticism. What he adduces fails to stand the test of examination. That Panormoff's experiments, as far as they go, may have been carefully conducted I do not call in question, but, incontestably, it is not permissible to advance them as showing what they have been affirmed to do.

If reference be made to Panormoff's work ('Zeit. f. phys. Chem.,' **17**, 596) it will be seen to rest on the narrow basis of the kind of reaction given by phenyl-hydrazine. The question of difference in the cupric oxide reducing effect observed before and after treatment with sulphuric acid is allowed to pass without consideration. The circumstances, however, demand that this question should be met and satisfactorily disposed of before judgment can be given. This as I have said has not been done and confronting Dr. Paton's assertion that Panormoff's experiments show that the sugar of muscle is glucose is the fact which is not attempted to be denied that from muscle a something is susceptible of extraction by alcohol (glycogen is therefore excluded) which comports itself with sulphuric acid and the copper test in such a manner as only to be accounted for by the presence in it of carbohydrate matter in some other form than that of glucose.

Confronting Dr. Paton also are three photo-engravings in my book of micro-photographs of osazone crystals obtained from muscle. In one the crystals are not those of glucosazone and in the other two whilst there are acicular glucosazone crystals there are also osazone crystals from another sugar.

In reality it may be considered that muscle contains carbohydrates of different forms which are susceptible of solution in alcohol of 95 per cent. strength and therefore exclusive of glycogen and its approximating dextrins. By alcohol a differentiation may be thus far affected which I have availed myself of in the analytical procedure I have adopted. The carbohydrates in the alcoholic extraction I have spoken of under the comprehensive term sugar. Strictly speaking the dextrins approaching maltose would be included. We have not the power at present to correctly discriminate, but if a product should give, in its initial titration, a lower cupric oxide reducing result than after treatment with sulphuric acid, we know conclusively that we have something to deal with other than, or in addition to, glucose. I say in my work (p. 90) in speaking of the sugar found in the portal blood after the ingestion of starchy matter that "in its totality it has a cupric oxide reducing power varying between that of maltose and that of glucose, thus showing that, whether glucose is present or not, sugar other than glucose must to a greater or less extent exist." This is exactly the position of what is likewise comprehended under the term of sugar of muscle. In its totality it has a cupric oxide reducing power more or less considerably below that of glucose. It therefore absolutely follows that it cannot be made up of glucose. As I have previously remarked it is not sufficiently realised by physiologists that animal products may contain, in the same manner as is to be observed in connection with the vegetable kingdom, a mixture of carbohydrate principles instead of simply one.

Glucosazone crystals on account of their greater insolubility much more readily separate out than the osazone crystals of other sugars. Every one knows, who has worked at the osazones, of the uncertainty attending the result when small quantities are dealt with. Impurities interfere with the deposition of crystals. These circumstances may account for Panormoff obtaining only glucosazone crystals. He extracted his muscle with water and then evaporated down with heat and afterwards treated with alcohol. My own results were obtained by treating the muscle at once with alcohol.

The point under consideration does not in the slightest degree affect the general question, but it shows how eagerly Dr. Paton has grasped at anything he could bring forward to disparage my work. Notwithstanding the extensive array of evidence I adduce he, upon the strength of results obtained by a Privat-Docent of the University of Kazan, in an off-hand way disposes of it with the expression "Now, the very careful experiments of Panormoff show that it is glucose." No particulars are supplied to show the foundation upon which this assertion is made, but the impression conveyed is that it is upon work of a nature to be unconditionally accepted.

Next follows a discourse in a haughty tone on "*The Fate of Carbohydrates in the Body.*" Juniority is associated in some with a belief in personal supremacy entitling them to authoritatively set other people right. Nothing occurs in this discourse to demand comment until the point regarding the formation of fat in the villi is reached. Here Dr. Paton's remarks run, "Dr. Pavy further suggests that the epithelial cells of the intestine take up carbohydrates and convert them into fat. This theory *may* be correct, but the experimental evidence on which he bases it is defective. He feeds rabbits on oats which contain 5 per cent. of fat, and because he finds fat in the epithelial cells of the intestines, he concludes that this is derived from the *carbohydrates* of the oats. How can he tell that the fat seen in the cells is not the fat of the oats? Dr. Gulland and I find that in rats, while a diet of fat causes a loading of the epithelium with fat particles, a diet of starch and sugar leaves the epithelium entirely free of fat globules."

Yet another misrepresentation. It is not true that because I find fat in the epithelial cells of the intestines of rabbits fed on oats which contain 5 per cent. of fat I base my conclusion that fat is derived from the carbohydrates of the oats. Nothing could give a more garbled view of the ground of my proposition. I say that after feeding a rabbit with oats I have seen the villi charged with fat globules and the lacteals filled with milky chyle to an extent quite irreconcilable with the fat being derived from the 5 per cent. existing in the oats. I will quote from what is actually contained in my book (p. 248), italicising certain parts.

"It is well known that *after food rich in fatty matter the lacteals are charged with milky chyle, that the cells of the villi are more or less loaded with fat,* *and that fat globules pass from these cells through the centre of the villus to reach the current in the lacteal system.*"

"Observation conducted upon the vegetable feeder after the ingestion of food rich in carbohydrate matter and poor in fat *reveals the existence of a precisely similar state of things.* On taking, for instance, a rabbit about four hours after a meal of oats, killing it, and opening the abdomen, *coils of the small intestine are seen, especially after a few minutes' exposure, to present a white opaque appearance, with milky streaks or lines upon the surface, due to flow of chyle beneath the peritoneum; and the lacteals of the mesentery, owing to the milky character of their contents, are conspicuously visible. The receptaculum chyli is also, from the same cause, readily perceptible, and, if cut into, gives exit to a strongly milky fluid. On the intestine being laid open, a more or less densely white condition of the internal surface presents itself to view, due to the extent to which the mucous membrane is charged with fat, and the villi stand out as opaque projections.*"

"In order that the condition described may be satisfactorily visible, *it is necessary that favorable circumstances should exist. The animal itself must be in a good healthy state. The food must be of a natural kind and sufficiently rich in farinaceous constituents.* Moistened oats, in the case of rabbits, have yielded the most marked results—*after fasting, with unfavorable food, and in ill-conditioned animals the appearance strikingly differs. The intestine is transparent and* *watery, and the lacteals are not perceptible.* Between this condition and that in which the lacteals are fully injected, any intermediate degree of milky character may, of course, be perceptible."

"*From the appearances presented, then, to the naked eye, it is learnt that under suitable food, rich in starchy matter, the same passage of fat occurs as after feeding directly with fat. It seems to me impossible that the quantity of fat observed to be thus entering the system could be derived from that contained in a free state in the food.* Analysis of the oats consumed in my experiments placed the amount of fat present at 5 per cent., which agrees with the estimations made by others."

With such passages contained in my work I will leave it to be judged whether the words "He feeds rabbits on oats which contain 5 per cent. of fat, and because he finds fat in the epithelial cells of the intestines he concludes that this is derived from the *carbohydrates* of the oats" do not stand as an offence to good faith—an inexcusable offence, it may be said, in the presence of science.

As regards the experiments on feeding rats with starch and sugar, I too experimented on rats in 1858 (probably 20 years before Dr. Paton commenced his medical studies) in relation to the same point. Such food constitutes in reality a starvation diet. I found upon one occasion that one of the rats had been devoured by the others. As no animal would fatten on starch and sugar only, how is it to be conceived that fat should be found at the seat of its production, like it is when natural circumstances prevail? The expectation is based upon a wrong conception. I contend that it is by living protoplasm that the work is done; and, for living protoplasm to do its work, it must, as every one knows, be situated in the midst of proper conditions; just as applies to a living object taken in its entirety. Dr. Paton's ideas run in too purely chemical a groove.

Dr. Paton has since published some further matter on this subject but it contains nothing that in reality touches my position. He does not seem to have properly grasped the situation and to realise the want of power in the material put forward, and the line of experimenting adopted, to meet the point before him.

In his final section Dr. Paton deals with the "*Physiology of the Carbohydrates applied to Diabetes.*" He says:—" When Dr. Pavy comes to apply his theory to diabetes it completely fails to explain the phenomena of many forms of the disease. The idea that the intestinal epithelium and the liver are two barriers against the invasion of the system by sugar, the former converting it into fat, the latter converting it into glycogen and then into fat or proteids, and that diabetes is simply a weakening of one or both of these two barriers, leaves many forms of diabetes unexplained. How will this explain cases in which glycosuria continues on a purely proteid" [animal is the word presumably meant] "diet? According to him it may be in part due to the carbohydrates always associated with proteids, but it may also be due to the carbohydrate part of the proteid molecule, which he has attempted to demonstrate. The existence of this carbohydrate element cannot be accepted; but supposing it does occur" &c.

I consider I need not waste time and space by quoting further. The remainder of the paragraph is devoted to an argument bearing on the relation in amount of sugar and urea eliminated in diabetes as hypothetical joint derivatives of the splitting up of proteid matter. For the third time (*vide* pp. 21, 64) Dr. Paton founds a criticism upon an erroneous representation of the amount of carbohydrate stated to have been obtained by me from the splitting up of proteid by the agency of potash.

It is a pure invention by Dr. Paton that when I come to apply my theory to diabetes it completely fails to explain the phenomena of many forms of the disease. I in the strongest manner dissent from the statement. Why, it is through diabetes that I have been led on in the prosecution of my work, and aided in the framing of my views! Whilst the glycogenic theory leaves everything unintelligible, the phenomena of diabetes stand, under the views I have propounded, explicable in a most simple and clearly comprehensible way.

To prevent our being diabetic it is necessary that the carbohydrate matter of our food should not be permitted to reach, in the form of free sugar, the general circulation. Doing so is just what happens in diabetes, and if the glycogenic theory held good the result would be universal diabetes. Assimilated, or disposed of, in the manner I contend for, the carbohydrates of our food are not afforded the opportunity of presenting themselves as sugar in the general circulation; and, with this the case, they cannot get into the urine. One source of sugar in the urine is impairment or failure of the power to stop our alimentary carbohydrate from reaching the general circulation as sugar. My everyday experience in connection with diabetes is showing me how sensitive the urine is to the food. Up to a certain point—that is, within the range of assimilative power existing—carbohydrate matter can be ingested without producing any effect on the urine. If it is effectively stopped whilst on its way to the general circulation, and thus prevented reaching it, of necessity it cannot reach the urine. Let the quantity that is ingested, however, exceed by ever so little the assimilative—in other words, arresting capacity, and sugar will appear in the urine in proportion to the extent the capacity is surpassed. It is truly surprising how finely balanced the urine is to the carbohydrate matter contained in the food taken.

Besides the elimination of sugar that is related to the food ingested, sufficient evidence exists to render it obvious that there may be elimination having its source in the tissues, and standing quite apart from the alimentary origin of glycosuria. At p. 229 in my book I state, "There is, then, a class of case in which the fault consists only of a loss, or, it may be, varying degrees of impairment, of the power of disposing of ingested carbohydrate matter in such a manner as to prevent its reaching the general circulation; and another, in which, in addition to this, a condition within exists attended with the splitting up, with sugar as a cleavage product, of the proteids of the body. The former is completely controllable by dietetic management, the latter only so to a partial extent."

The system of knowledge propounded by me is fully reconcilable with both sources of glycosuria, whilst the glycogenic theory does not intelligibly fit in with either. How, for instance, is any lucid explanation to be drawn under the glycogenic theory of the facts observed regarding the elimination of sugar in diabetes in relation to food ingested. And, certainly, if carbohydrate matter is consumed as is implied under the theory, it becomes hard to rationally account for the origin of sugar from the tissues. I call upon anyone to show, upon sound evidence and reasoning, that the glycogenic theory leads otherwise than into inscrutable darkness—than into a position from which it is impossible to acquire any satisfactory comprehension of the phenomena observable, both in health and diabetes, in relation to the carbohydrates.

On the other hand, viewing carbohydrate matter as contributing to fat- and proteid-formation, we have before us, not only that which harmonises with what is susceptible of recognition as occurring in the vegetable kingdom, but that which agrees with the resultant phenomena of life in the animal kingdom made manifest to us by common observation. If carbohydrate matter becomes applied, as I hold it does, to fat- and proteid-formation in the position assigned, no opportunity is given for its reaching the general circulation and thence the urine. If ingested and not so applied, however, it follows that something else must ensue, and the something else seen to occur is passage into the general circulation as sugar and subsequent elimination as unutilised material with the urine. Thus we have the glycosuria associated with alimentation satisfactorily accounted for. In connection with the glycosuria of tissue origin, things stand on an equally intelligible footing. The carbohydrate matter, the primitive source of which is in the vegetable kingdom, contained in our food is regarded as being in part synthesised into proteid, the basis of our tissues. Being simply locked up under the form of a glucoside, it is ready to be liberated whenever exposed to the requisite influence for bringing the result about. Nothing further than the presence of a suitable ferment is needed to provide for the eliminated sugar taking its origin from the tissues.

But Dr. Paton says at the end of the passage quoted that the existence of a carbohydrate element in the proteid molecule cannot be accepted. He has striven hard, it is true, to upset the evidence that has been adduced in favour of the glucoside constitution of proteid matter. In speaking of the crystalline osazone I have obtained from the cleaved sugar, he says the carbohydrates are not the only bodies which yield crystalline compounds with phenyl-hydrazine, and that "It is only when an elementary analysis of the compound is made that it can be definitely concluded that a carbohydrate is a constituent part." The elementary analysis has now been made and definitely shows the crystalline compound obtained to be a sugar osazone. The details of the work are given in a previous part of this epicriticism. I take it therefore that Dr. Paton will now admit that ground no longer exists for him to hold to his statement that the existence of a carbohydrate element in the proteid molecule cannot be accepted.

Finally, Dr. Paton comments on the interest attaching to some recently published experiments of Kaufmann on section of the nerves of the liver and pancreas in connection with Bernard's diabetic puncture of the fourth ventricle, and in the preceding short paragraph says, "Then how does this theory" [the system of knowledge propounded by me] "explain the diabetes which follows extirpation of the pancreas? Dr. Pavy simply ignores this problem."

No doubt there is interest attaching to the effect of operations on the nervous system in leading to the production of glycosuria. I myself experimented largely upon this subject many years ago, and my results were communicated to the Royal Society in 1859. I was the first to show that lesion of certain parts belonging to the gangliated cord of the sympathetic produced glycosuria. I know the want of constancy attending operations of this kind. Even with Bernard's puncture the attainment of the result cannot be depended upon, and I speak from personal knowledge in saying that in the hands of Bernard himself a want of positive result not unfrequently followed his puncture of the fourth ventricle. I make these remarks to show that caution is needed in framing conclusions from the kind of operation that is under consideration.

As regards the point touching the explanation of the diabetes following extirpation of the pancreas, it is a relief to come across a statement entirely free from misrepresentation; for, as asserted, it is perfectly true that I simply ignore the problem. I did so advisedly, seeing that I was dealing with the physiology of the carbohydrates, and that I did not feel, on account of our imperfect knowledge of the *modus operandi* of the extirpation, that reference to it would do anything towards throwing light upon the matter before me. The question having been raised, however, in such a manner as to suggest that a difficulty is presented I am quite ready to enter into it and can show that under my view nothing irreconcilable exists. It is perfectly compatible that under an absence of the influence of the pancreas a state of blood may be induced (1) which interferes with the proper performance of protoplasmic action in relation to the disposal of carbohydrate matter, thus permitting it to reach the general circulation as sugar and thence the urine; (2) which provides a ferment for transforming the store of glycogen in the liver into sugar; and (3) which further is imbued with the power of leading to the breaking down of proteid matter with the liberation of sugar, as occurs in association with phloridzin administration. These propositions agree entirely with what I have represented as constituting the conditions existing in diabetes as a disease.

I have now patiently and perseveringly waded through all that Dr. Paton has had to say in his communication contained in the 'Edinburgh Medical Journal' against my work and the conclusions deducible from it. I have dealt with his criticism, taking it sentence by sentence, and have temperately and fully discussed every point that has been raised. I am not aware that a single point requiring notice has been left unanswered. Superficially looked at, the article illusively conveys the impression of possessing a validity and fair-dealing character which examination shows do not belong to it. For the foundation for the latter part of this assertion I refer to the contents of the preceding pages.

Professedly one motive prompting Dr. Paton to write his article has been a personal regard for me. He says :

"At the beginning of my paper on Hepatic Glycogenesis, published last winter, before the appearance of Dr. Pavy's work, I said 'That one of the great functions of the liver is to produce sugar will not, at the present time, be denied by any physiologist,' and added in a foot-note, 'the theory of Pavy, repeated in nearly every text-book, that the liver is 'a sugar destroying, and not a sugar forming organ,' rests on so unsubstantial a basis and has been so completely refuted by the work of Seegen and other investigators that it need not be considered.' To let these statements stand after the publication of Dr. Pavy's book would be marked discourtesy, and I am glad of an opportunity of bringing forward the evidence upon which they were made."

The evidence upon which they were made. I have shown what this evidence is worth. In no single instance has anything bearing the stamp of soundness been put forward.

As to the marked discourtesy, from which the article written is stated to be designed to keep Dr. Paton clear, I think it will be conceded, in view of the line of action taken, that "omission" would have stood to better purpose than "commission." It certainly must be regarded as an anomalous course of procedure that endeavours avowedly made to avoid the commission of an act of discourtesy should take the shape of the article I have been dealing with in the preceding pages—an article which, as

I have shown, is freely interspersed with attacks founded on unpardonable misrepresentations.

Read by the light of these misrepresentations and the tone recognisable throughout is it not permissible to say that the article betrays evidence of an attempt, at all hazards, to write down my book? Anything seemingly that has come to hand that could be applied to disparagement has been eagerly grasped at, even to the extent of picking up words from a sentence away from their context and thereby putting into my mouth exactly the reverse of what I have in reality asserted. And, for what? to sustain the renowned foot-note employed to clear the way for work which, according to the experimental evidence herein adduced, stands upon as faulty a foundation as any piece of work could do. Be it noted, this is no mere abstract statement of mine, but an assertion founded on experimental results detailed in the pages of this volume.

I have now completed my undertaking and I submit that nowhere has Dr. Paton's criticism touched even the fringe of my position. The facts of the case stand in open view before the reader and I leave it to him to frame his own judgment upon the matter.

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