CVII. THE HYPERGLYCAEMIA OF CANCER¹.

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AN essential difference between tumour tissue and normal tissue is the increased ability of the former to glycolyse carbohydrate. Warburg *et al.* [1923, 1924; Minami, 1923] have shown that isolated but "surviving" carcinoma tissue transforms glucose completely into lactic acid. This sugar cleavage is a source of energy which enables the cancer cell to live anaerobically while under similar conditions the normal cell dies. Depriving the cancer cell of this source of energy results in retarded growth; increasing it, by increasing the concentration of glucose, results in accelerated growth. Cori and Cori [1925] have correlated these observations of Warburg with the glucose metabolism of the cancer cell in the living body. They found that blood, in passing through a tumour *in situ*, gains an amount of lactic acid almost equivalent to the quantity of sugar disappearing. Thus cancer cells *in vivo* also utilise sugar differently from the normal. Fundamental studies such as these emphasise the need for a thorough investigation of the carbohydrate metabolism of the cancer cell both in the isolated tissue and in the living body.

A significant fact in this connection is that the energy which the cancer cell obtains by glycolysis depends upon the concentration of glucose in the medium in which it lives. Cori and Cori [1928] have shown that at a normal blood-sugar level the tumour derives 15 % of its energy from glycolysis. At higher blood-sugar levels there is a greater rate of glycolysis. The experiments of Warburg, Wind and Negelein [1926] on isolated tumour material show that a slight increase over the normal glucose concentration has a marked influence on glycolysis. Doubling the sugar concentration by increasing it from 50 to 100 mg./100 cc. more than doubled the rate of lactic acid formation. This important relationship existing between sugar concentration and cellular activity is further brought out by our observation in collaboration with Hueper [1931] that there is a correlation between the amount of blood-sugar of a cancer patient and the malignancy of the tumour as expressed by the number of mitoses in a histological section. The higher the blood-sugar, the greater was the number of cell divisions.

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Numerous investigations carried out on the carbohydrate metabolism of cancer patients have been concerned with sugar tolerance. The results in general show that the blood-sugar level is raised to a point considerably higher in the cancer patient than in a normal individual; that this maximum is reached more slowly; and that the return to the original level is considerably retarded. The figures of Slosse and Reding [1927] show that the average increase in blood-sugar in a cancer patient after ingestion of 50 g. of glucose is 108 mg./100 cc., thus bringing the hyperglycaemia generally to over 200 mg./100 cc. It requires 2 to 3 hours for the original level to be attained again. Thus it is possible that during one-third of the time throughout a day the cancer patient's blood-sugar level is over 150 mg./100 cc. D'Halluin [1930] agrees with Slosse and Reding in that ingestion of 50 g. of glucose always produces, in the cancer patient, an increase greater than 45 mg./100 cc. in the blood-sugar, but his maximum is not as high.

In regard to the fasting blood-sugar level of cancer patients, however, much less information is available nor is there such good agreement. Slosse and Reding's figures, taken from their tolerance curves, show that the fasting blood-sugar level of cancer patients is definitely above normal—if averaged, 18 mg./100 cc.—although they themselves do not draw this conclusion. D'Halluin [1930] states that the fasting blood-sugar level is normal in 75 % of his cases. Chatain [1930] likewise concludes it to be normal.

In view of the disagreement existing on this question, and because of the possible clinical significance attached to it, we undertook to determine the fasting blood-sugar value of a large number of treated and untreated cancer patients. As controls, we also determined the values on a series of normals, a series of benign tumour cases, and a series of medical (non-cancerous and non-diabetic) cases. Our results clearly indicate that the fasting blood-sugar values in untreated cancer cases tend to be high, averaging 20 mg./100 cc. above normal. The values for treated cancer cases and benign tumour cases are also considerably above normal, whereas the medical cases average only slightly above the normal values.

That it has not previously been generally accepted that the fasting blood-sugar value of cancer patients tends to be high appears to be due to two factors.

- (1) A distinction has not been made between a real normal value and a high normal.

- (2) Most sugar methods, especially those ordinarily used in hospital laboratories, are not specific enough for sugar to bring out small differences.

Blood-sugar values ranging from 70 to 120 mg./100 cc. of blood have generally been accepted as normal values. The blood-sugar value obtained necessarily depends upon the method used in determining it, since none of the blood-sugar methods commonly used is absolutely specific for glucose. Other blood constituents contribute toward the "blood-sugar" value. Using a single method, the range covered can be narrowed. Moreover when a distri-

but ion curve is plotted from these figures, the majority of them will be found grouped in a still narrower range. Those values which occur most frequently are the real normal values. Considering the distribution in our normal group

Fig. 1. Blood-sugar determinations. (Shaded area represents values below 85. Plain area represents values above 85.)

(Fig. 1) the majority of the figures (76 %) are found within the range 70 to 85 mg./100 cc. and such will be considered the normal range in discussing the results reported in this paper.

The Hagedorn-Jensen sugar-method.

In the sugar values here reported the Hagedorn-Jensen [1923] sugar-method on oxalated venous blood has been used. The chief reason for adopting this method was that it expresses very nearly the "true" sugar values, being at the same time a micro-method.

In view of the confusion existing as regards the merits of the various copper reagents, these were avoided. Moreover, blood-filtrates of the tungstic or molybdic acid type (Folin-Wu; Benedict) are not suitable for detecting small differences in blood-sugar values. Such filtrates contain, in addition to glucose, many non-sugar reducing substances which act on the reagents used in estimating glucose, and so contribute to the determined value. This non-sugar material has been reported to be very constant in normal as well as in pathological conditions regardless of the blood-sugar level [Somogyi, 1927]. One might suppose, then, that sugar values by any one method would be comparable. If, however, there were a slight variation in this non-sugar fraction, no difference in the final figure would be observed between a blood-filtrate which contained more sugar and less non-sugar reduction and one which contained less sugar but correspondingly more non-sugar reduction. Results which we have obtained on a small number of cancer patients indicate that this non-sugar fraction is less in the blood of cancer patients. Therefore, methods which include amounts of non-sugar reduction in the "blood-sugar" figure could not be expected to bring out slight differences in the "true" sugar values. The Hagedorn-Jensen filtrate is nearly free from these interfering substances and was therefore quite suitable for this purpose, as was also the Somogyi filtrate [1930]. Since our work was started before the latter procedure was available to us it has been used only as a check on some of the results.

Somogyi has recently criticised the Hagedorn-Jensen method by pointing out that the filtrate contains a small amount of non-sugar reducing material which acts on the reagents used for the sugar determination, thus giving a slightly too high sugar value. By modifying the reagents so that the filtrate would be slightly alkaline, he produced a filtrate which contained only traces of non-sugar material. More recently Herbert and Bourne [1930] compared the two methods and found that the Hagedorn-Jensen filtrate gave values on an average only 4 mg. higher than that of Somogyi, hardly outside the experimental error. We have likewise compared the two methods and have found similar results. The agreement was almost always within the limits of experimental error, the Somogyi values in fact averaging 1 mg. higher. Since, according to Somogyi, the complete removal of the non-sugar reducing substances depends upon the alkalinity of the filtrates, we investigated the p_H of our filtrates. Herbert *et al.* [1929, 1930] have reported p_H values of 6.6 and 6.8 for the Hagedorn-Jensen and 7.4 for the Somogyi filtrate. Using Somogyi precipitating reagents which titrated 12.1 cc. of 0.0997 N NaOH to

10 cc. of 1.8 % ZnSO_4 , we found the filtrates to have a p_{H} of 7.1 to 7.2. In the original Hagedorn-Jensen method the authors did not make a titration between their reagents, but stated that the excess of zinc sulphate was so calculated that the solution was never alkaline. However, the effect of the blood-buffers on this mixture was not taken into consideration. In our experiments the Hagedorn-Jensen and Somogyi zinc sulphate reagents were made from the same stock zinc sulphate (45 %), and the same 0.0997 *N* sodium hydroxide was used. The p_{H} of Hagedorn-Jensen filtrates prepared with these reagents was 7.0 to 7.1. The similarity in the p_{H} values of these filtrates may explain the close agreement in the sugar values which we obtained in comparing the two methods. We are able to state that glutathione, believed by Somogyi to be the chief cause of non-sugar reduction, was absent from or negligible in our Hagedorn-Jensen filtrate, for we have added glutathione to plasma in the proportion of 100 mg./100 cc. and practically none (0 to 5 mg. calculated as glucose) was found in the filtrates as an increment over the original sugar figure. With a minimum of non-sugar materials present, it is evident that the results obtained express very closely the true sugar figure unless there is a hitherto unknown reducing substance in cancer blood which comes through in this filtrate.

Kramer and Steiner [1931] have recently criticised the Hagedorn-Jensen method because of errors which occur through the use of cotton in filtration. These authors state that "the technique of filtration is the essential source of inaccuracy which renders the Hagedorn-Jensen method altogether inadequate for analytical work." They claim that 4–22 mg. of glucose are lost by retention in the cotton plug and, moreover, to have confirmed the figures of Folin and Malmros [1929] who found 3–19 mg. lost when 100 mg. of cotton was used. However, the latter pointed out that 100 mg. of cotton were too much; that if only 20 mg. were used there was no error from this cause. At the time we adopted this method for our use two years ago, it seemed that filter-paper would be more convenient. A comparison was made at that time between sugar values obtained on duplicate blood-filtrates, one filtered through exactly 4 mg. of cotton and the other through Whatman No. 1 filter-paper rinsed with distilled water before use. Almost exact agreement was obtained as seen from the following figures:

	Filter-paper mg./100 cc.	Cotton mg./100 cc.
Exp. A (blood sample)	104	106
Exp. B (blood sample)	120	120
(blank)	<u>16</u>	<u>15</u>
	104	105

This is in keeping with Kramer and Steiner's suggestion of the use of filter-paper as an improvement in preparing the Hagedorn-Jensen filtrate.

Further observations on this sugar method have been made as follows.

(1) It is important that the dilute zinc sulphate be fresh. The original authors recommended that this be made from a concentrated stock solution

every 8 days. If the zinc sulphate has deteriorated it produces an increase in the blank value while an increase in the value from the blood-filtrate, if any, is slight. Too high a value being thus obtained in the blank, too low a blood-sugar value will result.

(2) The potassium ferricyanide solution must be checked occasionally. This keeps 6 months at least when stoppered and in the dark; but, if the bottle is in frequent use, a slight deterioration will be noted. When such takes place, it should be discarded and fresh solution taken.

(3) Water which has been kept in the ice-box after being freshly distilled is not necessary. No difference was observed between this and distilled water which had been kept at room temperature for a few days.

(4) It is not necessary to boil the tubes 3 minutes to precipitate the proteins. One minute or until the coagulum rises is sufficient.

RESULTS.

The accompanying diagram, Fig. 1, combines the results which we have obtained in determining the fasting blood-sugar of 37 normals, 52 untreated cancer cases, 67 treated cancer cases, 35 cases of non-malignant growths (untreated) and 32 medical cases.

The normal values cover the range 70–95 mg. of glucose per 100 cc. of blood; but, as discussed previously, owing to the distribution we may consider values between 70 and 85 as the normal values, 76 % of these being within this range. Since such a large proportion of the group is under the figure of 85, we will consider values between 85 and 95 as high normals. The average blood-sugar of the entire group is 80.

The group of untreated cancer cases comprises those cases who had received no medical treatment for the cancerous condition before our determination of their blood-sugar. Diagnosis of malignancy was later confirmed histologically. Any case diagnosed by the physician as also being diabetic was omitted from this and the following groups. None of the cases included showed glycosuria under a normal diet, nor were there any other symptoms of diabetes. An observed hyperglycaemia was, therefore, not due to diabetes mellitus. This group, the untreated cancer cases, shows the highest values for blood-sugar, 90 % having a figure higher than 85, in contrast to the normals, 76 % of which were under this figure. From the graph it is at once apparent that there is a decided shift to the right. The blood-sugars range from 74 to 135 with the peak at 90–100. The average for the group is 20 mg./100 cc. higher than that of the normal group.

By treated cancer cases are meant those who have received one or more X-ray or radium treatments previous to our observation of their blood-sugar. As a group, these cases show almost as high blood-sugar values as the untreated cases. The average value is 98, with the range covered, 66–142. Treatment has not, on the whole, lowered the blood-sugar of this group, although it has done so in certain individual cases.

The non-malignant group includes cases of benign tumours and chronic lesions, all of which are abnormal growths and many of which may be considered precancerous. These cases also have a tendency toward a higher sugar than normal; range, 75–123; average, 95; not, however, quite as high as in the cancer cases.

In contrast to the three latter groups, purely medical cases (diabetes being excluded) do not, in general, show this hyperglycaemic disturbance. The sugar level is quite normal; range, 66–115; average, 85. Since a high blood-sugar is not found in most of these purely medical cases, it is evident that the hyperglycaemia is a characteristic of malignant or precancerous conditions.

DISCUSSION.

This increase in the fasting blood-sugar level, as well as the increased tolerance curve, is in our opinion an expression of the hyperglycaemia of cancer, and since the fasting value is generally determined on hospital patients, it seems worth while to point out its significance. While sugar tolerance curves may give a more complete picture of the ability to metabolise carbohydrate, still the fasting blood-sugar gives a very valuable criterion and is much less difficult to obtain than a complete tolerance test. Since the fasting blood-sugar determination is usually a routine procedure in hospitals and clinics, the value is readily available to the physician. Hitherto, the physician has not been concerned when values of 100 were obtained in cancer patients (100 by the Hagedorn-Jensen method corresponding to about 120 by the Folin-Wu method which is generally used in hospital laboratories). Our results show that such a figure is about 15 mg./100 cc. higher than normal.

It should not be concluded from these results that a high blood-sugar is of any value in the diagnosis of cancer patients because it is not a constant phenomenon. Its importance lies in the prognosis and treatment of the patient. Patients with a high blood-sugar, as Jackson [1929] has pointed out, have a poor prognosis; those with a low blood-sugar respond to almost any form of treatment.

In view of the effect of sugar concentration on the activity of tumours, Cori and Cori [1928] have discussed the question as to whether patients suffering from malignancy would derive any benefit from a low-carbohydrate diet. Their conclusion was that, since the hyperglycaemia following an ordinary meal rarely exceeded 150 mg./100 cc., a severe restriction of carbohydrate seemed unnecessary, but that food which tended to raise the blood-sugar above this level should be avoided. However, as we have pointed out, it is quite probable that in the cancer patient the hyperglycaemia is greater than 150 mg./100 cc. during a considerable part of the day. This fact becomes still more significant when considered in the light of our findings that even the fasting blood-sugar values tend to be considerably above normal.

In our experience, the clinical condition of the patient seems to run

parallel with the variations of the fasting blood-sugar level. If any type of treatment benefits him, the hyperglycaemic level is reduced; if the clinical condition is aggravated, the blood-sugar level is increased. Reduction of the blood-sugar level should, therefore, be beneficial to the patient. Reding (private communication) and Jackson have both reported this to be the case. In any event, blood-sugar values should be carefully watched in connection with treatment in order that the clinical condition may not be aggravated.

On the other hand, does lowering of the blood-sugar in a cancer patient make him respond to that particular type of treatment which aggravates his condition at the higher blood-sugar level? This phase of the problem is being investigated in our laboratories. Thorough and well-controlled clinical observations will be needed to exclude the possibility of any untoward effects before therapeutic measures tending to lower the blood-sugar can be recommended as a supporting therapy.

The fact that the treated cancer cases as a group show practically as high sugar values as untreated cases, may mean that treatment has had no effect on the blood-sugar, or it may mean that while in some of the cases the blood-sugar has been lowered in a corresponding number it has been increased. This indicates the importance, however, of carefully observing the effect of treatment on the blood-sugar of the individual patient.

SUMMARY.

1. The average blood-sugar level in 52 untreated cancer cases, as determined by the Hagedorn-Jensen method, is 20 mg./100 cc. above normal.
2. Treated cancer cases and cases of non-malignant (benign) growths also show blood-sugar values higher than normal, the treated cases being, on the whole, almost as high as the untreated.
3. Purely medical cases do not, in general, show an increase in the blood-sugar level.
4. Relations between blood-sugar, prognosis and treatment are discussed.

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