VARIATIONS IN THE FREE HYDROCHLORIC ACID OF THE GASTRIC CONTENTS IN CANCER AND THE SO-CALLED 'PHYSIOLOGICALLY ACTIVE' HYDROCHLORIC ACID

By BENJAMIN MOORE, M.A., D.Sc., *Johnston Professor of Bio-Chemistry, University of Liverpool.*

From the Bio-Chemical Department, University of Liverpool

(Received October 23rd, 1908)

In a paper recently published in the *Proceedings of the Royal Society*,¹ Messrs. Copeman and Hake describe experiments carried out upon the acidity of the gastric contents in mice and rats with transplanted tumours, and also in a number of cases of cancer in man.

The authors state that their results contradict or materially modify the conclusions arrived at by Moore and others,² but I desire no better confirmation of our results than is given by the table of Messrs. Copeman and Hake when using our methods, which are those that have been established by the usage of many previous observers. Because somewhat different results have been given by another method intended to show the so-called 'physiologically active' hydrochloric acid, Copeman and Hake come to the conclusion that this quantity is not materially decreased in cancer in man, and is actually somewhat increased in mice with 'cancerous' growths.

For reasons which will be given later, I am of opinion that this term 'physiologically active hydrochloric acid' is a misnomer, and that the method used shows something quite different from hydrochloric acid in any active form. The method will be criticised after dealing with the other results obtained by Copeman and Hake, but

1. *Proc. Roy. Soc.*, Series B, Vol. LXXX, p. 444, 1908.

2. Moore, Alexander, Kelly and Roaf, *Proc. Roy. Soc.*, Series B, Vol. LXXVI, p. 138, 1905; *Bio-Chem. Journ.*, Vol. I, p. 274, 1906; Wilcox, *Lancet*, June 10, 1905, p. 1566; Morton Palmer, *Bio-Chem. Journ.*, Vol. I, p. 398, 1906; *Guy's Hospital Reports*, Vol. LX, p. 181, 1906.

before passing to this criticism I consider it essential to point out what was really shown regarding the variations in the gastric acidity in malignant disease and other abnormal conditions in the two papers by myself and collaborators, especially since Copeman and Hake have fallen into serious errors regarding our position and results.

Before the appearance of our first paper it was well known that the amount of hydrochloric acid in the stomach contents in cases of carcinoma *of the stomach* was as a general rule much reduced, and in many cases fell to a zero value, and it was equally well known that there were well-marked exceptions to this rule in which the hydrochloric acid was as high or higher than normal.

These facts are supported by a wealth of literature which need not be quoted here.

Our first paper demonstrated by a series of analyses carried out by several different methods, in a number of cases of *cancer in other situations than the stomach*, that exactly the same facts held for the acidity of gastric secretion, no matter where the malignant growth was situated.

The experimental results of our first paper have been confirmed by other authors since then, and, as we shall presently point out, are further confirmed by the table given by Copeman and Hake.

The theory held regarding the diminution in hydrochloric acid in *carcinoma ventriculi* previously to our first paper, was that the cause was local in the stomach itself. In the light of our experiments showing similar results with the growth situated in organs remote from the stomach this view became untenable, and we referred the reduced acidity to a general condition in the blood.

Further experimentation, the results of which were published in a second paper, showed us that *the degree of acidity of the gastric contents is most sensitive in many diseased conditions*, and under an hospital régime; and we gave results showing that in twenty hospital cases, where there was no cancer, the average amount of hydrochloric acid was much reduced below the average normal amount in healthy persons.¹

1. Similar results were obtained independently and practically simultaneously by Morton Palmer (*loc. cit.*).

This point regarding the diminution in hydrochloric acid in non-malignant cases we especially emphasized, and placed as the chief result in the forefront of our conclusions. Reflections on the importance of the hydrochloric acid as a peptonizing agent, as a disinfectant of the alimentary tract and of the food, and as an excitant to the hormone (secretin) for the pancreatic secretion, clearly demonstrate the importance of this depreciation of the hydrochloric acid value as a general factor in disease, and in predisposing to infection; but upon this we need not further dilate here than to remark that we regarded the observation as an important one entirely apart from the question of malignant disease.

So far were we from regarding hydrochloric acid as being absent in *all* cases of malignant disease, or its absence as a diagnostic sign, that we safeguarded ourselves by specially pointing this out in the following words:—

‘It is clear from these figures that the presence or absence of free hydrochloric acid cannot be regarded as a diagnostic sign of any great value. All that can be said is that in the majority of malignant cases it is absent, and in the majority of non-malignant cases it is present. The same statement holds for cancer of the stomach wall itself.’

That the amount of hydrochloric acid is more reduced in malignant than in non-malignant cases is, however, clearly shown by the following summary extracted from our second paper:—

		Malignant		Non-malignant		Ratio
‘Total acidity	=	816	:	1,453	=	0·56
Di-methyl indicator	=	352	:	925	=	0·38
Günzburg’s reagent	=	163	:	502	=	0·32
Methyl-acetate method	=	286	:	630	=	0·45
Mörner-Sjoqvist method	=	637	:	774	=	0·82”

It is entirely an open question whether this greater drop in acidity in the malignant cases is due to the greater severity of the illness and the accompanying cachexia, or is a specific effect in cancer; but in any case it is obvious that it must be referred to a general cause in the system, and not to local conditions in the stomach.

1. The reason for the nearer approach to equality in the Mörner-Sjoqvist method is discussed in our paper and shown to be the presence of organic base in combination with inorganic acid.

Having thus defined the chief experimental results given in our previous papers, I may now turn to the criticisms of Copeman and Hake, and in the first place will reply to certain criticisms of our technique and methods.

It is implied, because we do not explicitly state that no water was added when we were drawing off the test meal at the end of the period, that we probably added water—especially because Copeman and Hake found difficulty in the majority of their cases in getting the stomach contents off without water.

Since the contents were being drawn off in order to make *quantitative* estimations of the hydrochloric acid, it would scarcely seem necessary to a chemist to state that no water was added, but as it appears to be necessary I may now say that the contents were taken without the addition of water. The enormous difficulty experienced by Copeman and Hake was not encountered in doing this, and we believe that we are correct in stating that the withdrawal of stomach contents without adding water has been, and often is, accomplished by others than ourselves. As evidence of the low results for amount of acid in our cases being caused by too much added fluid, Copeman and Hake quoted one case of ours (No. IX) in which, under special circumstances, we gave three pints of test meal instead of one, and withdrew it after one and a half hours, and they state that ‘it is hardly surprising that under these circumstances the free hydrochloric acid obtained was only 0.001 per cent.’ In reply to this I would state that if Messrs. Copeman and Hake will take the trouble to administer a pint of gruel and two pints of water in normal individuals, and withdraw after the same interval of one and a half hours, I should be very much surprised indeed if they did not get more than fifty times as great an amount of hydrochloric acid.

They appear to forget that the physiological experiments show that the acid secretion is regulated by the concentration of acid in the stomach. Fluid added at the time of withdrawal is, of course, a different matter.

With the exception of this one case, in which the amount of fluid

given is definitely stated, the amount given was one pint, and it was withdrawn at the end without adding water.

Messrs. Copeman and Hake next proceed to criticize very adversely the variations in time before we withdrew our test meals for analysis, as follows :—

‘ It is of importance to note, also, that the interval between the administration of the test meals and its withdrawal, as recorded by these observers, varied between the wide ranges of one to two hours, and in a single case was as much as three hours. This absence of uniformity as regards time of withdrawal clearly detracts from the diagnostic value of the results, and it is sufficiently obvious that where a number of experiments are being made under varying conditions of time, whether by one or more observers, no proper comparison will be possible between the results or the deductions drawn from them.’

I regard this statement *as applied to our experiments* as an unfair criticism, which is rendered all the more misleading by the abstract truth of its concluding sentence.

In the first place we did not claim ‘ diagnostic value ’ for our results, and explicitly say so in our discussion (p. 155) ; but this is a minor matter. The obvious meaning that any reader would draw from the above quotation is that we collected our test meals at very varying intervals indiscriminately from one hour to two hours, and in one case up to three hours, so rendering them quite valueless. Let us consider first the single extreme three-hour interval. No mention is made by our critics of the fact that this observation is bracketed in our table with another observation and series of analyses done *on this same patient after an exact interval of one hour*. This is the only case in the entire series where two observations are taken on the same patient, a fact which could scarcely escape the attention even of a casual reader of our table. We may now further draw our critics’ attention to the important fact, which also appears to have escaped their notice, that the two sets of analyses in this case practically coincide after one hour and after three hours respectively, the figures for total acidity being 0·2665 per cent. after three hours, and 0·2847

per cent. after one hour; Günzburg reaction negative in both analyses; free hydrogen ion concentration by methyl-acetate method = 0.00067 per cent. HCl for three hours, and zero HCl for one hour.

Turning to the more serious statement that our times of collection ranged from one to two hours, a glance at the times recorded in our table¹ shows that of thirty-three cases the test meal was siphoned off in *exactly* one hour in twenty cases; in five cases the period was one and a quarter hours; in six cases one and a half hours; while *in two cases only was the interval two hours*. Considering that these are clinical experiments, where the meal is administered and the withdrawal carried out in the ward, I am inclined to think that they were carried out as regards time in a fairly uniform and scientific manner; nor do the results show any variations such as would warrant the criticism quoted above. Within these time limits there is no obvious variation, related to time, visible in the results.

With this I may conclude my reply to Messrs. Copeman and Hake's criticism of our previous papers, and turn now to a criticism of their paper and results.

The first section of the paper deals with the acidity of the stomachs and stomach contents of mice which had carried implanted tumours, and demands only one or two passing references.

It is not surprising to me to find the result stated in this section that the amount of hydrochloric acid in the gastric contents in mice bearing implanted tumours is not diminished, as this adds but one more proof, if such were needed, that these interesting growths, even if carcinomata, are carcinomata situated in healthy animals where the conditions are such that the animal reacts against the effects of the growth, and so produces an immunity which is shown by the absence of general effects in the organism.

Anyone who has stood and watched mice, as I have done, with tumours of this class nearly as large as themselves, must have been struck with the obvious fact that there are none of those clinical characteristics of cancer as observed in man to be seen in the behaviour of the animals.

1. *Bio-Chem. Journ.*, Vol. I, pp. 276 and 277.

The mice are to all appearances perfectly well ; they eat with all the vigour of the healthy mouse, they run about and clean themselves like the normal mice ; there is no cachexia or wasting unless the tumour breaks down owing to its size or to injury.

Attention has been drawn to the absence of cachexia in mice with implanted tumours by Dr. Bashford, the Director of the Imperial Cancer Research, who, as quoted by Copeman and Hake in the very paper we are criticizing, states that on the basis of observations made on almost 3,000 mice with propagated cancerous tumours during two years ' the conclusion has been arrived at that the presence of a tumour even of greater weight than the mouse itself does not necessarily involve a disturbance of the normal nutrition which could be regarded as comparable to the cachexia frequently associated with malignant new growths in the human subject.'¹

In view of these important differences between experimentally implanted tumours in mice and malignant growths in man, I desire to enter a protest against their being described as ' cancerous ' tumours and ' cancerous ' growths, and the mice being described as ' cancer ' mice, in the same sense as these terms are used in regard to the human subject, until the causes of the differences have been cleared up.

It is only in a small percentage of cases that these experimental tumours give rise to metastatic growths, viz., 4 to 5 per cent., whereas in cancer it is only a small percentage which escape metastatic growths.

We are told by Copeman and Hake that ' recent extensive statistics, collected by the Imperial Cancer Research Fund from various London hospitals, have shown that cachexia is not a constant accompaniment of cancer in man.'

Perhaps cachexia is not a *constant* accompaniment of cancer in man any more than the diminution or absence of hydrochloric acid in the gastric contents is a *constant* accompaniment—few or no clinical symptoms or signs are absolutely constant accompaniments of any disease. But a research into the cause of death in cancer unaccompanied by cachexia would be of high scientific interest,

1. *Scientific Reports on the Investigations of the Imperial Cancer Research Fund*, No. 2, p. 40, 1905.

and any such highly interesting cases should be watched to the end. Loss of appetite, wasting and anaemia, with their related conditions of mind and body, are prominent symptoms in many early cases of internal cancer long before a tumour can be located, and certainly before there is ulcerative breaking down of tissue, and ordinary septic toxaemia. In such cases it is possible that the normal tissue cells are being poisoned by toxic substances derived from the cancer cells, and chemical research is much needed to clear up this and similar problems.

In our second paper we showed that there was evidence, from certain of the methods which we employed, that the gastric contents in cancer cases contained organic bases.

The results obtained by Copeman and Hake with the Volhard-Lüttke method prove, as we shall presently point out, that chlorides of such bases existed in the gastric contents from their cases.

Further, over 10 per cent. of the nitrogen of the urine in cancer patients is present as extractive nitrogen, an amount far in excess of what is ever found in health.

Experimental work on the subject of these organic bases is at present being carried out in the Bio-Chemical Laboratory, Liverpool.

It may well be that the absence or diminution of free hydrochloric acid in cancer is closely associated with the cachectic condition, although it often precedes its obvious onset, and if this is the case, it is easy to understand why there is no decrease in tumour mice, because they are never cachectic, and seem to suffer no change in their general health from the presence of the tumour.

We may now turn to the results obtained by Copeman and Hake in the cases in which quantitative determinations of acidity were made in human cases.

Only thirteen cases are described in their table in which the test meal was obtained without dilution with water, and as details are not given regarding the other cases of their thirty-four, we may confine our attention to these tabulated cases.

Three cases of the thirteen are shown by the results to contain

about a normal amount of hydrochloric acid as compared with the single normal control, viz., those numbered V, VII, and XXXI; in the other ten, in my opinion, the figures show that the free hydrochloric acid is very considerably decreased, and in several it is practically absent.

Taking the methods in order, the total acidity to phenol-phthalëin is not markedly reduced, the average of the thirteen cases showing 0.1636 per cent., while the single normal case gave 0.1861; but it is peculiar that while the total acidity to an indicator which will show the faintest trace even of a feeble organic acid is 0.1861 per cent., the 'physiologically active hydrochloric acid' alone amounts to 0.2336 per cent.; that is to very considerably more than the total acid. It may be suggested that there is something wrong with a method which shows 'physiologically active' hydrochloric acid that cannot affect phenol-phthalëin. This result is also given in no less than nine of the thirteen cancer cases. Surely it would be more appropriate, if chlorine which is so firmly attached to base is to be termed hydrochloric acid at all, that it should be designated as physiologically *inactive* hydrochloric acid—it is just about as physiologically active as sodium chloride.

When we pass to the column of the table which shows the reaction to the Günsburg reagent, it becomes obvious that the amount of free hydrochloric acid in the majority of the cases is quite inappreciable. Thus in four of the cases the result is absolutely negative, being marked in the table by 'nil'; in three other cases it is noted as 'faint'; and since the authors state in the text that 'where a very marked reaction was obtained we also made the test quantitatively,' it may be taken that in these seven of the thirteen cases the amount of free hydrochloric acid shown by this most reliable test was too infinitesimal to estimate. That is to say, in more than half the cases there was practically no free hydrochloric acid present. In two other cases the result is given as 'very marked' and 'marked,' but no estimation is made—these two belong to the three cases which we have admitted above as exceptions out of the total thirteen cases. In only four of the thirteen cases is a quantitative determination of

the free hydrochloric acid made by the Günsburg reagent, and from the above quotation from the text it may be taken that these were 'very marked' reactions; the average of the four works out to 0.0496 per cent., the control being 0.1314 per cent. If we add the seven cases where the amount was too low to estimate, the average for these eleven cases has the value 0.0180 per cent., and I do not desire any better confirmation of our figures than this.

If this does not mean a marked reduction in free hydrochloric acid in these cancer cases, I entirely fail to understand what it does mean.

The concentration of free hydrogen ions as shown by velocity of inversion of methyl-acetate is worked out in all of the thirteen cases. This method gives quantitatively the effective acidity for physiological work of the gastric contents, and that which would be most closely connected with its digestive and bactericidal properties, and hence might much more appropriately be taken as showing 'physiologically active hydrochloric acid' in the true sense of the words. The results obtained by Copeman and Hake by the use of it, again entirely support our conclusion as to the depression in acidity in cancer. Thus the average for the whole thirteen cases is 0.0407 per cent. as against 0.1195 per cent. in the control case, or about one-third of the value, and if we take out the three cases which are admitted exceptions, we obtain for the remaining ten cases the average value of 0.0153 per cent., which I regard again as a marked reduction from the normal percentage.

The two remaining methods used by Copeman and Hake are the Mörner-Sjoqvist and the Volhard-Lüttke methods; these two methods are almost identical in principle, and are subject to the same errors.

The Mörner-Sjoqvist method consists essentially in incinerating in presence of excess of barium carbonate, when any free hydrochloric acid *plus* any hydrochloric acid, however firmly combined with organic bases, is converted into barium chloride, and the amount of this estimated in the solution obtained from the incinerated mass. The method introduced by Lüttke consists in making two estima-

tions of chlorides in two incinerations, in one of which acid free or organically combined is driven off on the incineration, and thus does not appear in the analysis, and in the second incineration the addition of alkali to neutralization to phenol-phthalein prevents the loss of any chloride save such as disappears owing to volatilization in the removal of the charred organic matter. The difference in the two figures hence is supposed here as in the Mörner-Sjoqvist method to give free hydrochloric acid *plus* hydrochloric acid combined with organic bases.

The Mörner-Sjoqvist method is by far the better of these two, since there is much less danger of loss due to volatilization of chlorine on incinerating such a small amount of sodium chloride in presence of such an excess of organic matter. It was for this reason, and on account of the great similarity of the two that we used the Mörner-Sjoqvist method in our work.

In our paper we drew attention to the fact that the Mörner-Sjoqvist method, as well as another method of an allied nature which we used to attempt to determine the amount of organic acid, viz., incinerating with excess of alkali, gave figures which indicated a considerable amount of organic base in the gastric contents, and we drew special attention to the importance of the presence of this amount of organic bases.

Thus we found that incinerating with alkali, instead of giving rise to positive figures owing to destruction of organic acids, gave often negative figures indicating that alkali was used up, and having shown that ammonia was practically absent by Schlössing's method, as well as other volatile nitrogenous bases, we were driven to the conclusion that fixed organic bases were present in more than sufficient amount to preponderate and mask any organic acids which might be simultaneously present.

This very important result has not, in my opinion, yet received the attention which it deserves, and is worthy of further investigation to determine the nature and physiological properties of these basic bodies. That they are not proteins or first products of the hydrolytic cleavage of proteins by digestion in the stomach, is shown by the fact

that they neutralize the hydrochloric acid to methyl orange and di-methyl, and even to phenol-phthalein, as also by the fact that only a very weak biuret reaction is given by such samples of stomach contents upon a test meal.

To speak of such firmly combined chlorides as 'physiologically active hydrochloric acid' is, for the reasons above mentioned, peculiarly absurd. Nor does it matter that the acid so combined may previously have been free, as suggested by Copeman and Hake, for after combination it is no longer either chemically or physiologically free or active. Further, as has been pointed out already, under normal conditions of gastric secretion, acid is secreted until the free acid present (or rather the hydrogen ion concentration) approximates to a normal value, so that any alkali would be neutralized, and then free acid would go on being secreted until the normal value of acidity was once more attained.

Hence the figures obtained by Copeman and Hake by the Lüttke method chiefly show firmly combined organic chlorides, and not physiologically active hydrochloric acid, and serve to confirm the results previously obtained by us using the Mörner-Sjoqvist method, and that of incineration after neutralization or with excess of alkali.

Since our principal results have now received confirmation by Morton Palmer, by Willcox, and, in my opinion, by the figures of Copeman and Hake themselves, I still adhere to them, and in conclusion would definitely state them as follows :—

1. The free hydrochloric acid of the gastric contents after a test meal is extremely sensitive in most diseased or debilitated conditions, and the average of a series of cases taken at random from a hospital ward is lower than a similar series from healthy individuals.
2. The depression is particularly well marked in cases of cancer, and this is found to hold wherever the cancer growth happens to be situated. This shows that the very low concentration of hydrochloric acid in cancer is not due, as formerly supposed, to local conditions in the stomach, but must be referred to some general change in the blood affecting the hydrochloric acid secretion. In this connection it may be added that there is a small but distinct increase in the

alkalinity of the salts of the serum in cases of cancer (Moore and Wilson).¹

3. It is at present indeterminate whether the greater drop in cancer cases is due to the greater degree of cachexia and general ill-health in such cases, or whether it is due to something specific in cancer.

4. In addition to the depression in free hydrochloric acid, there is also a considerable amount of bases in combination as chlorides, and these bases do not consist of ammonia or other volatile nitrogenous bases, or of proteins or primary products of protein cleavage, but of organic bases.

5. The depression in free hydrochloric acid occurs in the majority of cases, but there are exceptions to the rule, and for this reason, and also because hydrochloric acid may be absent or reduced in other conditions than cancer, the absence or diminution can not be taken as a diagnostic sign of any very great value.

1. *Bio-Chem. Journ.*, Vol. I, p. 297, 1906.