

# THE HYDROGEN ION CONCENTRATION OF THE BLOOD IN CARCINOMA.

## I. FROM THE COLORIMETRIC DETERMINATION OF THE BLOOD DIALYSATE.

By WILLIAM H. CHAMBERS.

*(From the Research Laboratories of the Barnard Free Skin and Cancer  
Hospital, and the Department of Surgery, Washington University  
School of Medicine, Saint Louis.)*

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Several investigators have reported a difference in reaction between the blood of patients with malignant tumors and the blood of normal individuals. Moore, Alexander, Kelly, and Roaf (1906) noted a marked decrease in the total acidity of the gastric juice in cancer. The free hydrochloric acid was negative in two-thirds of the cases, regardless of the situation of the tumor. These results were confirmed by Palmer (1906). This led Moore and Wilson (1906) to titrate the serum and inorganic salts of the serum ash against standard  $H_2SO_4$ , and they found that both the serum and the serum ash of cancer patients required slightly more acid to neutralize them to a definite end-point than those of normal persons or those of cases with diseases other than cancer. Watson (1909) extended the work of Moore and Wilson with practically the same results. Menten (1917) determined the hydrogen ion concentration of the serum and the whole blood in normals, cancer cases, and other diseases by the Michaelis gas chain method. In general a greater alkalinity was found in the serum of cancer patients, but this difference did not appear in the whole blood. The reaction of the whole blood seemed to vary with the barometric pressure. As no account was taken of the loss of carbon dioxide from the blood the hydrogen ion concentration was lowered appreciably in all cases, and thus a variable factor was introduced, the value of which is difficult to estimate. All these results, however, indicate in different ways a more alkaline

reaction of the blood in cancer. Therefore, a study of the actual hydrogen ion concentration of the blood was undertaken, using the more recently developed methods and avoiding as far as known any essential changes in the blood after it was drawn. The value of a test which would give the patient with internal cancer an early diagnosis and thus the same advantage of early treatment as the one with an external cancer has often been emphasized. Toward this end the indicator method was chosen because of its adaptability as a clinical test. The results, however, do not show a sufficient difference in the early cases to warrant much diagnostic application.

#### *Method.*

The colorimetric method of determining the hydrogen ion concentration was applied to blood by Levy, Rowntree, and Marriott (1915). They dialyzed the blood in collodion sacs against neutral physiological salt solution, but did not prevent the loss of carbon dioxide from the blood or the dialysate. Scott (1917) and Dale and Evans (1920) have used essentially the same method, modified to minimize the exchange of gases between the blood and the atmosphere. A simplified combination of the two methods was employed in this experimental work. One ounce of "pyroxylin" was dissolved in 500 cc. of ether and ethyl alcohol mixed in equal volumes. The sacs were made in small Pyrex test-tubes (inside diameter 8 mm. and about 60 mm. long). These test-tubes were filled with collodion and emptied immediately, three times in succession, then inverted and drained for 15 minutes. They were then rinsed well with distilled water, the sacs were removed from the tubes and rinsed again on the outside and inside, and left in saline solution until used. Dialysis was carried out in Pyrex test-tubes (inside measurements  $10 \times 100$  mm.) against 3 cc. of neutral 0.8 per cent NaCl.

The blood was drawn from the median vein into a glass syringe without exposure to the air and immediately discharged through the needle into a tube containing approximately 0.2 per cent crystalline potassium oxalate under a layer of pure petrolatum oil. The collodion sac was filled with petrolatum oil and placed in the dialyzing test-tube containing 3 cc. of saline solution. 1.5 to 2 cc. of blood were pipetted into the sac under the oil, forcing

the oil out above it in a layer over the whole tube and completely excluding aeration of the dialysate or blood.

The viscosity of the oil holds the sac suspended in the saline solution (Fig. 1). After dialyzing for 15 minutes the sac was removed with forceps, still leaving a sufficient layer of oil over the dialysate, and 0.2 cc. of a 0.02 per cent solution of cresol red

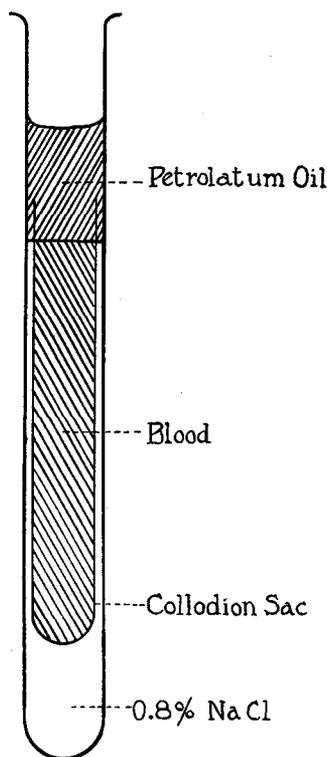


FIG. 1. The dialysis of whole blood under oil.

was added. The pH was read by comparison with standard buffer solutions which were covered by an equal layer of oil. After a comparison of the three indicators, neutral red, phenol red (phenolsulfonephthalein), and cresol red (*o*-cresolsulfonephthalein (Clark, 1920)), it was found that cresol red gave the most distinct color changes between pH 7.0 and 7.8. The  $\text{KH}_2\text{PO}_4$ —

NaOH buffer mixtures of Clark and Lubs (Clark, 1920) were used for the standard comparisons. The reagents were of highest purity and recrystallized several times. The standards varied by pH 0.10 which allowed interpolation to the second decimal.

The blood was taken between 2 and 4 hours after a meal to avoid any effect of HCl secretion on the reaction. A rest of 15 to 20 minutes was required of all normal subjects and patients not confined to bed, as Christiansen, Douglas, and Haldane (1914) have shown that exercise decreases the height of the blood CO<sub>2</sub> absorption curve.

*Correction Factors for Temperature.*

The dialysis of the blood in these experiments was carried on at room temperature which ranged between 20 and 30°C. Clark's standard phosphate solutions are prepared to give the designated pH at a temperature of 20°C. When the comparison with the dialysate is made at any temperature other than 20°C., a correction must be applied to the standards as well as to the dialysate to obtain the true pH at 20°C. The pH of all the blood dialysates in these data were corrected to 20°C.

Henderson (1908) has shown that the ionization constants of carbonic acid and the ion H<sub>2</sub>PO<sub>4</sub><sup>-</sup> in the equilibria  $\frac{\text{NaH}_2\text{PO}_4}{\text{Na}_2\text{HPO}_4}$

and  $\frac{\text{H}_2\text{CO}_3}{\text{NaHCO}_3}$  increase upon raising the temperature from 18 to 38°C.

However, the ionization constant of water increases much more rapidly than that of carbonic acid, which produces a large increase in hydroxyl ion concentration with very little change in the concentration of hydrogen ions. Therefore, solutions containing bicarbonate and carbonic acid at a constant carbonic acid concentration increase in alkalinity with a rise in temperature almost as much as the water constant itself, provided there is no loss of bicarbonate by reaction with other substances in the solution. He concludes that this applies to blood. Disregarding the unknown effect of the change of temperature on the dissociation of the indicator, the data of Table I on the standard phosphate solutions agree in the direction of the change with Henderson's work. Triplicate series of standards under oil as used in the blood

determinations were brought to the indicated temperatures and compared. The results are given in Table I.

Changing the tubes from 37 to 10°C. and *vice versa* gave the same results. Therefore, a temperature correction of pH 0.004 per 1°C. was applied to all readings of the standards at temperatures other than 20°C.

Hasselbalch (1917) published extensive data on the effect of changes of temperature on  $\text{NaHCO}_3$  solutions and blood. He determined the pH of  $\text{NaHCO}_3$  solutions at 38 and 18°C. at various  $\text{CO}_2$  tensions by the electrometric method. The curves plotted as pH against millimeters of tension of  $\text{CO}_2$  show a difference between the two temperatures of pH 0.12 at the same  $\text{CO}_2$  tension, or a difference of about 12 mm. of  $\text{CO}_2$  tension at the

TABLE I.  
*Temperature Changes of Standard Phosphate Buffers.*

Buffer No.	Temperature.		
	10°C.	23°C.	37°C.
	<i>pH</i>	<i>pH</i>	<i>pH</i>
1	7.15	7.20	7.25
2	7.25	7.30	7.35
3	7.35	7.40	7.45
4	7.45	7.50	7.55

same pH. He ascribed this more alkaline reaction at 38°C. to the difference in solubility of  $\text{CO}_2$  (0.928 at 18°C. and 0.560 at 38°C.) being greater than the change in ionization constant of the carbonic acid. Serum gave results similar to the bicarbonate. At the same  $\text{CO}_2$  tension it was pH 7.39 at 18°C. and pH 7.49 at 38°C., a difference of pH 0.10. The results on whole blood differed from those of the serum and bicarbonate in that the same reaction, pH 7.35, was found at both temperatures. Hasselbalch surmised a greater acid dissociation of oxy-hemoglobin at 38 than at 18° as the explanation of this phenomenon in the whole blood. Another factor, however, is suggested by the following calculations. Plotting the curves at 38 and 18° from Hasselbalch's figures<sup>1</sup> and calculating the pH at 40 mm. tension, the following figures are obtained:

<sup>1</sup> Hasselbalch (1917), Blood I, Table II, p. 123.

$$\text{pH at } 38^\circ = 6.09 + \log \frac{40.31}{2.69} = 7.27$$

$$\text{pH at } 18^\circ = 6.19 + \log \frac{49.0}{4.50} = 7.23$$

The equations show that there were 49.0 volumes per cent of combined  $\text{CO}_2$  at  $18^\circ$  and 40.31 volumes per cent at  $38^\circ$  or an increase of 8.69 volumes per cent of combined  $\text{CO}_2$  with the decrease of  $20^\circ$  in temperature. If the pH at  $18^\circ$  is calculated with the same amount of combined  $\text{CO}_2$  as was found at  $38^\circ$  (40.31 volumes per cent), the

$$\text{pH at } 18^\circ = 6.19 + \log \frac{40.31}{4.50} = 7.14$$

or pH 0.13 more acid at 18 than at  $38^\circ$ . This difference of pH 0.13 for whole blood is about the same as that found with bicarbonate and serum between 18 and  $38^\circ$ . These calculations would indicate that the red blood cells made available a larger amount of alkali at 18 than at  $38^\circ$ , which was probably due to the increased amount of dissolved  $\text{CO}_2$  causing a shift of  $\text{Cl}^-$  from the plasma into the cells (Van Slyke, 1921). In this way the increase in available alkali which accompanied the decrease in temperature balanced the increase in  $\text{CO}_2$  solubility and the decrease in acid dissociation so that the reactions at the two temperatures were nearly the same.

Other investigators disagree with Hasselbalch. McClendon (1917) reported that if the  $\text{CO}_2$  tension remained constant, the pH of bicarbonate solutions, sea water, or blood varied with the temperature, pH 0.01 for each degree Centigrade. Evans (1921) concluded that at the same  $\text{CO}_2$  pressure human blood was pH 0.20 more alkaline at 38 than at  $20^\circ$ .

Our data on the temperature factor for correcting the pH of the dialysate of whole blood are not exactly comparable to those quoted above as regards a constant tension of  $\text{CO}_2$ . Where the change in temperature in a solution under oil containing  $\text{CO}_2$  is a cooling one, as in the case of blood from body to room temperature, the solubility of the  $\text{CO}_2$  is increased, which should minimize any loss of dissolved  $\text{CO}_2$  from the solution. It seems probable that there is no change in amount of dissolved  $\text{CO}_2$  with a decrease in temperature from 38 to  $20^\circ\text{C}$ ., but that the increased solubility of  $\text{CO}_2$  at  $20^\circ$  makes the change comparable to a decrease in

tension. This is illustrated by the following results from a pure  $\text{NaHCO}_3$  solution. A solution of  $\text{NaHCO}_3$  (0.0267 M) was equilibrated with 46.0 mm. of  $\text{CO}_2$  at  $37^\circ\text{C}$ . and run into tubes under oil without exposure to the atmosphere. One-half of the solution was kept in the equilibrating room at  $37^\circ$  and the other cooled to  $20^\circ$ . The pH of each was determined by adding the indicator directly to the clear solution. Their colorimetric reaction was pH 7.39 at  $37^\circ\text{C}$ . and pH 7.37 at  $20^\circ\text{C}$ . The total  $\text{CO}_2$  content of the solution was determined in the Van Slyke apparatus and the following data obtained for  $37^\circ\text{C}$ .

Total $\text{CO}_2$ content.....	<i>vol.</i> <i>per cent</i>	62.2
Dissolved $\text{CO}_2$ ( $\frac{46}{760} \times 0.560$ ).....		3.39
$\text{NaHCO}_3$ .....		58.81
pH = 7.31 (Hasselbalch's value for $\text{pK}_1$ ).		
pH = 7.39 (Warburg's " " " ).		

If it is assumed that on cooling no change in amount of dissolved  $\text{CO}_2$  is involved, then at  $20^\circ\text{C}$ . the solution would contain 3.39 volumes per cent of dissolved  $\text{CO}_2$  with a solubility of approximately 0.900 (interpolated from Hasselbalch, 0.928 at  $18^\circ$ ). The  $\text{CO}_2$  tension at  $20^\circ$ , calculated from the equation

$$\frac{\text{CO}_2 \text{ tension}}{760} \times 0.900 = 3.39$$

equals 28.6 mm. or 17.4 mm. lower than the  $\text{CO}_2$  tension of the same solution at  $37^\circ\text{C}$ . According to Hasselbalch's equation, the reduction in  $\text{CO}_2$  tension between pH 7.39 at  $38^\circ\text{C}$ . and pH 7.37 at  $20^\circ$  should be approximately 8 mm. instead of 17.4 mm. Whether this discrepancy was due to technical error or was a valid one was not determined. These data would indicate, however, that if a bicarbonate solution with a molar concentration and  $\text{CO}_2$  tension of the order of blood is kept under oil, a reduction in temperature from 38 to  $20^\circ\text{C}$ . produces a reduction in  $\text{CO}_2$  tension which tends to counteract the change in ionization of the carbonic acid and involves but little change in the hydrogen ion concentration.

To determine the temperature correction factor for the pH of the dialysate of whole blood the following experiments were conducted. Duplicate sets of dialyzing tubes were prepared and

brought to the two desired temperatures, incubator and room temperature or room temperature and ice bath. Immediately after drawing the venous blood, equivalent dialyses were made at the two temperatures. The color comparisons with the standards were made at the dialyzing temperatures. The results are given in Table II.

Each pH figure represents the average of triplicate dialyses. In the last determination, Blood 186, the blood was drawn in the 38° incubator room and the 38° sample was not exposed to any change of temperature. The table shows that dialysis at the lower temperatures, 13 to 28°C., always gave a more acid reaction than dialysis at the higher temperatures, 26 to 38°C. The difference was fairly consistent with five different bloods and averaged

TABLE II.  
*Dialysis of Blood at Different Temperatures.*

Blood No.	Temperature in degrees Centigrade.										Difference in temperature. °C.	Difference in pH.	
	38	32	31	28	27	26	20	19	16	13			
163	pH	pH	7.50	pH	pH	pH	7.38	pH	pH	pH		11	0.12
164		7.43						7.33				13	0.10
181						7.40			7.29			10	0.11
183					7.28					7.10		14	0.18
186	7.16			7.10								10	0.06
Total.....												58	0.57

approximately pH 0.01 for each degree Centigrade or pH 0.18 more acid at 20°C. than at 38°C.

The explanation of these results and comparison with other data on changes in whole blood according to variations in temperature is complicated by two factors of unknown value: first, a possible change in CO<sub>2</sub> tension when the blood is cooled in the test-tube under oil, and second, the effect on the reaction of the dialysis and the accompanying dilution with physiological salt solution. In regard to the first factor, there is probably little if any loss of total CO<sub>2</sub> from the blood when cooled under oil (Joffe and Poulton, 1920). If the decrease of pH 0.20 (Table II) between 38 and 18° is applied to Hasselbalch's blood which has a total CO<sub>2</sub> at 38° of 43.0 volumes per cent at 40 mm. tension,

the distribution of  $\text{CO}_2$  between dissolved and combined  $\text{CO}_2$  at  $18^\circ$  can be calculated as follows:

$$\text{pH at } 38^\circ = 7.27 = 6.09 + \log \frac{43.00 - 2.69}{\frac{40}{760} \times 0.511}$$

$$\text{pH at } 18^\circ = 7.07 = 6.19 + \log \frac{43.00 - 5.00}{\frac{44.5}{760} \times 0.854}$$

The equations show that in changing from pH 7.27 at  $38^\circ$  to pH 7.07 at  $18^\circ$  without changing the total  $\text{CO}_2$ , there would be a decrease in  $\text{NaHCO}_3$  from 40.31 to 38.0 volumes per cent and an increase in dissolved  $\text{CO}_2$  from 2.69 to 5.00 volumes per cent with an increase in  $\text{CO}_2$  tension from 40 to 44.5 mm. Such a change might represent the final equilibrium attained after a shift of HCl between cells and plasma, initiated by a tendency to decrease in  $\text{CO}_2$  tension with decrease in temperature as illustrated in the pure  $\text{NaHCO}_3$  solution. It seems more probable that the second factor, dialysis, plays some part in the change. A discussion of Donnan's membrane equilibrium theory as applied to the dialysis of whole blood is given in the succeeding article.<sup>2</sup> The difference between the pH of the plasma and that of the dialysate is explained by the theory as due to the effect of the non-diffusible ions on the distribution of the diffusible ions between the three phases, red corpuscle, plasma, and dialysate. Inasmuch as no experimental data have been found on the influence of temperature changes on the equilibrium, it is deemed sufficient to note here that in the change from  $37$  to  $20^\circ$  involving a change in the dissociation constants of the salts and colloids of the blood it is quite probable that the equilibria between diffusible and non-diffusible ions are altered.

A third factor, the loss of  $\text{CO}_2$  by diffusion through the oil, was determined on the 0.0267M bicarbonate solution as follows. One sample of the equilibrated  $\text{NaHCO}_3$  was run by replacement with oil directly from the equilibrator through a small bore tube into a comparator tube containing the indicator, and the pH read immediately. This sample, unexposed to oil, gave a reaction at  $37^\circ$  of pH 7.39. Duplicate sets of tubes containing the  $\text{NaHCO}_3$

<sup>2</sup> Chambers, W. H., and Kleinschmidt, R. E., *J. Biol. Chem.*, 1923, 1v, 257.

under oil, without exposure to the air, were kept at 37 and 20°C. for 1 hour and the pH was read at intervals as indicated.

Time after equilibration.	37°C.	20°C.
	<i>pH</i>	<i>pH</i>
Immediately.....	7.39	7.37
15 minutes.....	7.42	7.37
30 ".....	7.45	7.39
60 ".....	7.47	7.39

From these figures it is apparent that an appreciable loss of CO<sub>2</sub> from the solution takes place at 37°C. but no loss was detected at 20°C. for 15 minutes, which was the time and approximate temperature used in the blood dialyses.

The data of Table II do not substantiate the statement of Evans (1921) that if the blood is equilibrated at 38° the dialysis gives the same results as at room temperature. Evans cites Joffe and Poulton (1920) who state that cooling the blood under oil before centrifuging caused no definite change in the total CO<sub>2</sub> of the plasma. However, no change in the total CO<sub>2</sub> of the plasma does not necessarily mean no change in pH, for the pH of the blood is not a function of the distribution of total CO<sub>2</sub> between cells and plasma but of the ratio of combined CO<sub>2</sub> to dissolved CO<sub>2</sub>.

The temperature change for whole blood of pH 0.01 per degree Centigrade determined from the data of Table II is used as a correction factor in the calculation of all samples of whole blood not dialyzed at 20°C. The factor of pH 0.004 per degree Centigrade was applied to the standards when they were read at a temperature other than 20°C. The salt error of the cresol red indicator which was applied to all dialyses was determined for 0.8 per cent NaCl as +pH 0.06. Therefore, the corrections to 20°C. for the dialysate of whole blood dialyzed and read at 25°C. as pH 7.35 would be 7.35 + 0.06 (salt error) - 0.02 (correction for K<sub>2</sub>HPO<sub>4</sub> standards) - 0.05 (temperature correction for whole blood dialysis) = pH 7.34 at 20°C.

#### EXPERIMENTAL DATA.

The main experimental work consists of the hydrogen ion concentration determination of the dialysate from 92 specimens of venous blood. The blood was obtained from patients with car-

cinoma, from those with other diseases, such as non-malignant growths, anemia, tuberculosis, and lues, and from normal subjects.

The normal control bloods were given for the most part by students of the Medical School. Table III shows the results obtained on twenty-three specimens from fourteen normal subjects. The blood samples were numbered consecutively when drawn, so that the first column (blood number) in each table indicates the order in which the bloods were taken. As the numbers of Column 1 show, the normal controls were selected at intervals during the entire period of investigation of the other

TABLE III.  
*Hydrogen Ion Concentration of the Dialysate of the Blood of Normal Subjects.*

Blood No.	Subject.	pH at 20°C.	Blood No.	Subject.	pH at 20°C.
53	J.	7.30	127	K.	7.45
54	W.	7.25	137	"	7.21
55	S.	7.32	144	"	7.38
56	H.	7.35	152	"	7.33
143	B.	7.39	82	C.	7.30
157	D.	7.30	125	"	7.43
174	M.	7.36	138	"	7.10
175	A.	7.37	139	"	7.28
177	R.	7.18	153	"	7.34
178	V.	7.20	164	"	7.34
179	N.	7.30	167	"	7.32
182	R.	7.35			
Average.....					7.31

cases, so that any slight improvement in technique or variation in other factors during the course of the work is checked in this way. The average of the twenty-three determinations listed in Table III is pH 7.31, with a variation of pH 7.10 to 7.45. The variation in a single individual is shown by the four different specimens from Subject K and seven from Subject C. Five of the seven tests of Subject C show a pH of  $7.31 \pm 0.03$ . The other two, Nos. 125 and 138, might be considered as temporary or local deviations from the normal zone, magnified by experimental error.

For convenience in presentation, the carcinoma cases are divided into five tables according to the location of the initial lesion. The following items concerning each case are tabulated: blood num-

TABLE IV.  
*Hydrogen Ion Concentration of the Dialysate of the Blood in Carcinoma of the Head Region.*

Blood No.	Clinical diagnosis.	Pathological diagnosis.	Extent or duration of disease.	Treatment prior to test.	Course.	pH at 20°C.
181	Carcinoma of left ear.	Prickle cell carcinoma.	Anterior auditory canal, no glands involved.	None.	Prognosis good.	7.38
87	Carcinoma of tongue.	Malignant squamous cell carcinoma.	Lesion 1×1.5 cm. Lymph glands not involved (hyperplasia).	Wassermann ++ ++ Antitubercular, intensive for 3 weeks.		7.40
172	Carcinoma of lower lip.	Prickle cell carcinoma, lymph glands negative.	Ulcer 3 cm. in diameter.	None.	Prognosis good.	7.47
106	Carcinoma of right maxilla.	Malignant squamous cell carcinoma.	Lesion 2×4 cm.	None.	Erysipelas 2 weeks later.	7.45
88	Carcinoma of left ear.	Malignant squamous cell carcinoma.	Lesion 4×7 cm. of 10 years duration.	None.	Improved.	7.50
96	Carcinoma of left antrum.	Malignant squamous cell carcinoma.	Entire left upper jaw involved, extensive scarring.	Wassermann ++ ++ Antitubercular, 2 months.		7.30
77	Carcinoma of left antrum.	Malignant squamous cell carcinoma.	Left maxilla and left cervical glands. Glands 8×5×3 cm.	Glands only, removed 11 days prior.	Died 3 months later.	7.51

105	Carcinoma of lower lip.	Malignant squamous cell carcinoma with prickles.	Lip 3×6 cm. Glands 5×8×4 cm. and 3×4 cm.	None.	Died of broncho-pneumonia 2 months later.	7.40
70	Carcinoma of left tonsil.	Malignant squamous cell carcinoma.	Involving regional lymph glands.	None.	Died 14 days later.	7.35
Average.....						7.42

TABLE V.  
Hydrogen Ion Concentration of the Dialysate of the Blood in Carcinoma of the Breast.

Blood No.	Clinical diagnosis.	Pathological diagnosis.	Extent or duration of disease.	Treatment prior to test.	Course.	pH at 20°C.
180	Carcinoma of right breast.	Medullary carcinoma, glands negative.	10 months duration. Breast enlarged twice.	None.	Favorable.	7.26
151 154	Carcinoma of left breast.	Medullary carcinoma, metastases to axillary glands.	Breast tumor 12×8×6 cm. Gland 4×4×4 cm. 5 years duration.	None.		7.32 7.42
165	Carcinoma of left breast.	Adenocarcinoma, metastases to lymph glands.	Breast tumor about 2 cm. in diameter. Axillary gland 3×3×2 cm. 3 months duration.	None.	Favorable.	7.47
128	Carcinoma of right breast.	Adenocarcinoma.	Metastases to axilla.	Right breast entirely gone from plasters.		7.45
58	Carcinoma of right breast.		Axillary and mediastinal lymph glands. Elephantiasis of right arm.		Died 2 weeks later.	7.48
115	Carcinoma of left breast.		Tumor 5×3 cm. extending into axilla. Metastases to vertebræ and pelvis.	Radical right breast operation 7 months previous.		7.44

80	Carcinoma of right breast.	Medullary carcinoma.	Metastases to axilla and under clavicle.	None.	4 weeks later many nodules in axilla and on arms.	7.49
84				7 days after radical breast operation.		7.50
110				1 month after radical breast operation.		7.45
133				2 months after radical breast operation. 1 week after x-ray.		7.51
59	Bilateral carcinoma breast.		Skeleton, lungs, and pleura involved.	1 x-ray treatment.	Died 2 months later.	7.55
113				5 " treatments to thighs.		7.47
Average.....						7.45

TABLE VI.  
*Hydrogen Ion Concentration of the Dialysate of the Blood in Carcinoma of the Thoracic and Abdominal Organs.*

Blood No.	Clinical diagnosis.	Pathological diagnosis.	Extent or duration of disease.	Treatment prior to test.	Course.	pH at 20°C.
150	Carcinoma of sigmoid.	Adenocarcinoma of sigmoid.	Papilloma 1 cm. in diameter. Musculature not involved.		Favorable.	7.32
119	Carcinoma of rectum.	Adenocarcinoma of rectum.	Ulcerous mass of tumor tissue 1 cm. thick.	Colostomy 14 days and radiatum 6 days prior.	Favorable.	7.37
65	Carcinoma of intestine.	Annular adenocarcinoma of intestine.	Small mass in sigmoid. Glands not involved.	Tested during saline hypodermoclysis.		7.33
112	Carcinoma of pancreas.		No lymphatic enlargement. Jaundice.		Died 2 weeks later, cardiac failure.	7.51
92	Recurrent carcinoma of left arm.	Squamous cell carcinoma with metastases to lymph glands.	Ulcer deltoid region 4×8 cm.		Questionable.	7.45
142	Recurrent carcinoma of rectum.	Adenocarcinoma of rectum, chronic inflammation at anus.	Mass filling pelvis, inguinal and femoral glands.	Colostomy and excision of rectum 1 year prior.	Died 3 months later.	7.34
66	Carcinoma of intestine.	Adenocarcinoma.	Occupies entire hepatic flexure, part of ascending colon, glands, and right kidney. Not in liver.		Died 18 days later.	7.38

132	Carcinoma of pancreas and liver.		Head of pancreas, wall of duodenum. Liver studded with nodules. Jaundice.	None.	Died 3 months later.	7.41
155				Cholecystectomy 1 month prior.		7.35
85	Carcinoma of pancreas and liver.	Adenocarcinoma.	Pancreas, liver, retroperitoneal glands. 3 months duration.			7.65
72	Carcinoma of ascending colon.	Colloid carcinoma.	Ascending colon, large mass in omentum, liver studded. 6 months duration.			7.60
131	Carcinoma of stomach.		Entire stomach involved.		Died 19 days later.	7.53
122	Carcinoma of lung.	Carcinoma of right lung, pleura, chest wall, and diaphragm.	Right lung primary, metastases to left pleura, liver, adrenals, and ovary.		Died 2 months later.	7.45
Average.....						7.44

TABLE VII.  
Hydrogen Ion Concentration of the Dialysate of the Blood in Carcinoma of the Pelvic Organs.

Blood No.	Clinical diagnosis.	Pathological diagnosis.	Extent or duration of disease.	Treatment prior to test.	Course.	pH at 20°C.
71	Carcinoma of cervix uteri.	Gelatinous degeneration of carcinoma of the uterus.	Limited to cervix.	Radium 6 days prior.		7.40
173	Carcinoma of uterus.	Carcinoma.	Parametria, anterior and posterior fornix involved.	Radium and x-ray 3 months prior.	Favorable.	7.43
121	Carcinoma of peritoneum, ovarian cyst.	Malignant papillary cyst,—adenoma of ovary.	Most of peritoneum. Cystic ovary and pelvic organs removed 6 years previous.	X-ray during 2 years prior.		7.55
162				2 gallons fluid drained from peritoneum 3 days prior.		7.36
130	Carcinoma of prostate.	Adenocarcinoma of prostate gland.	Complete prostate involved.	Radium and cystostomy 7 days prior.	Prostactectomy. Died 2 days later.	7.53
126	Carcinoma of clitoris.	Prickle cell carcinoma of clitoris with glandular metastases.	Initial tumor 1X2 cm. Inguinal and femoral glands involved later.	None.	Died 8 months later.	7.58
Average.....						7.47

TABLE VIII.  
*Hydrogen Ion Concentration of the Dialysate of the Blood in Inoperable Carcinoma of the Cervix.*

Blood No.	Clinical diagnosis.	Pathological diagnosis.	Extent or duration of disease.	Treatment prior to test.	Course.	pH at 20°C.
63	Inoperable carcinoma of uterus.		Recurrent 9 months after Percy operation.	Radium 1 day prior.		7.63
134	Inoperable carcinoma of cervix uteri.	Adenocarcinoma, glandular hyperplasia, chronic endometritis.	Very extensive.		Favorable with radium.	7.45
146	Inoperable carcinoma of cervix uteri.		Very extensive.		Questionable.	7.46
124	Inoperable carcinoma of cervix uteri.		Metastases to parametria very extensive.	Radium 6 days prior, x-ray 4 days prior.	Died 5 months later.	7.53
Average.....						7.52

TABLE IX.  
*Hydrogen Ion Concentration of the Dialysate of the Blood in Diseases Other than Carcinoma.*

Blood No.	Clinical diagnosis.	Pathological diagnosis.	Description.	pH at 29°C.
114	Pernicious anemia.		R.B.C. (after transfusion) 2,740,000. Hb. 65 per cent.	7.37
73	Simple anemia, myelogenous leucemia.		R.B.C. 3,520,000. Hb. 65 per cent.	7.35
135	Myelogenous leucemia.		W.B.C. 165,400. R.B.C. 2,890,000. Hb. 65 per cent.	7.35
64	Traumatic hematoma.			7.34
60	Fibroid uterus.			7.33
161	"			7.35
160	"			7.33
109	Verruca on left buttock.			7.40
74	Hyperplasia of epithelium.			7.40
163	Carcinoma of cervix.			7.40
183	Epithelioma.			7.24
129	Chronic pulmonary tuberculosis.			7.40
145}	Tuberculosis of rectum and lungs.*			7.43
156}				7.41
117	Ulcer of duodenum.			7.42
140}				7.14
141}	Luës.		Wassermann ++++	7.27
			Lower lip.	

107	Lues.			7.32
83	Exfoliative dermatitis.			7.40
98				7.37
104	Exfoliative dermatitis.			7.35
111				7.35
108	Adhesions of peritoneum.			7.48
81				7.50
Average.....				7.36

\* Numbers enclosed in a bracket are separate determinations on different days on the same patient.

ber, clinical diagnosis, pathological diagnosis, comparative size of the cancer or its duration as an indication of its malignancy, remarks concerning any treatment or condition of the patient which might influence the reaction of the blood, the course of the disease if known, and the pH of the dialysate of the whole blood at 20°C. Clinical data were taken from the hospital records of the patients.

Nine cases of carcinoma of the head region are reported in Table IV. The variation in the pH of the dialysate is quite wide, pH 7.30 to 7.51. The average is pH 7.42, which is pH 0.11 more alkaline than the normal average of Table III. Table V gives the results from thirteen determinations on eight cases of carcinoma of the breast. The range of reaction is greater in this table, pH 7.26 to 7.55, and the average more alkaline, pH 7.45 or pH 0.14 higher than the normal. Table VI shows thirteen determinations on eleven cases of carcinoma of the thoracic and abdominal organs including the intestinal tract, and one carcinoma of the arm. The variation is from pH 7.32 to 7.65, with an average of pH 7.44. Tables VII and VIII contain the cases of carcinoma of the pelvic organs, those in Table VIII being inoperable carcinoma of the cervix and uterus. The average of the six cases of Table VII is pH 7.47. However, it should be noted that Blood 162 was drawn 3 days after 2 gallons of fluid had been drained from the cysts in the peritoneal cavity. As the effect on the blood of this loss is unknown, this determination might be excluded, giving an average for the five cases of pH 7.50, and a range of pH 7.40 to 7.58. The average of the four cases in Table VIII is pH 7.52.

The miscellaneous pathological cases, or diseases other than carcinoma are grouped in Table IX. These include three cases of anemia, several of non-malignant growths and inflammation, two cases of tuberculosis, and several cases from the Dermatological Service. The average of the twenty-four cases reported in Table IX is pH 7.36, only pH 0.05 more alkaline than the normal average. The variation is similar to the normal, being pH 7.14 to 7.50.

The averages for the different groups are assembled in Table X.

Table X shows that the averages for the carcinoma cases are distinctly more alkaline than those of the normal and miscellaneous cases.

The comparison of the carcinoma cases with the cases having other diseases (miscellaneous pathological) and with the normal subjects is graphically shown in Fig. 2. Each point represents a

TABLE X.  
*Average Hydrogen Ion Concentration of the Dialysate of the Blood.*

Table No.	Disease.	No. of determinations.	Average pH at 20°C.
III	Normal.	23	7.31
IX	Miscellaneous pathological.	24	7.36
IV	Carcinoma of head.	9	7.42
VI	“ “ abdominal organs.	13	7.44
V	“ “ breast.	13	7.45
VII	“ “ pelvic organs.	6	7.47
VIII	Inoperable carcinoma of cervix.	4	7.52

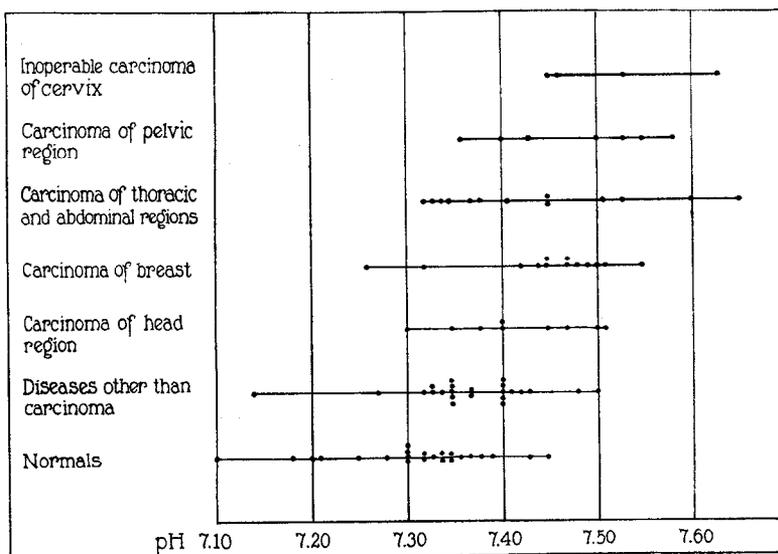


FIG. 2. The hydrogen ion concentration of the dialysate of the venous blood.

pH determination, given in the data of Tables III to IX. The expression of the pH is a linear one but the cases are grouped along several lines, divided according to tables, to avoid the confusion of crowding the points onto one line. In this way

Fig. 2 shows at a glance that the majority (two-thirds) of the normal and miscellaneous cases lie in the zone between pH 7.30 and 7.40, whereas the majority (two-thirds) of the carcinoma cases have a pH above 7.40. It is evident then, that with some exceptions the dialysate of the whole blood of the patients with cancer is more alkaline than that of normal persons or patients with the other diseases studied.

#### DISCUSSION.

Menten (1917) found no discernible relationship between the alkalinity of the blood serum and the degree of anemia in cancer patients. Barr and Peters (1921) report six cases of anemia in which the arterial blood was markedly alkaline, but the venous blood had a normal reaction varying between pH 7.32 and 7.38. Similar results are found in the dialysate from the venous blood of the three cases of anemia reported in Table IX (Nos. 114, 73, and 135) which gave respectively, pH 7.37, 7.35, and 7.35. Many of the carcinoma cases were anemic, however, no correlation was found between the red corpuscle count or hemoglobin determination and the reaction of the dialysate. It seems from this evidence that the anemia of cancer patients is not a factor in producing the more alkaline reaction of the blood dialysate.

The cases of non-malignant growths given in Table IX show no definite change in pH from the normal zone. The three determinations on two cases of tuberculosis were pH 7.40, 7.43, and 7.41. The data indicate that the more alkaline reaction found in the majority of the cases with malignant growth is associated definitely with the carcinoma.

Some indication of the nature of the relationship between the tumor and the hydrogen ion concentration of the blood dialysate is found in a study of the data of Tables IV to VIII. The cases have been arranged in the tables in the order of increasing size or malignancy of the tumor. For example, in Table V, in the first case (No. 180) the carcinoma was confined to the breast and the glands were not involved. In the next three cases (Bloods 151, 154, 165, and 128) there were metastases to the axillary glands, and in the cases following there were more extensive metastases to other parts of the body. The last case (Bloods 59 and 113) was carcinoma of both breasts with metastases through-

out the body. This arrangement of cases applies to all of the carcinoma tables. Its accuracy is, of course, only approximate, for the actual area of the growing tumor can be only estimated, but it is based on a careful compilation of the clinical and pathological examinations. Reviewing all of the data, there seems to be an increase in alkalinity with an increase in the size and extent of the tumor growth. This is particularly well illustrated in Table V. The first case in Table V shows a pH of 7.26, with the intermediate cases between pH 7.42 and 7.50 and the last case pH 7.55. In Table VI, considering only the cases of carcinoma of the intestinal tract, the small tumors show no increase in alkalinity above the normal zone; the cases with metastases, a distinct alkalinity. Blood 66 appears to be an exception, for the extent of the metastases points toward a higher pH than was found. In general the same increase in alkalinity with increased involvement of the different glands or organs is found in the cases of Tables VII and VIII. Table IV, the cases of carcinoma of the head region, presents the greatest number of exceptions. Blood 172 with only a small carcinoma has a pH of 7.47 while Nos. 96, 105, and 70 have quite extensive involvement with reactions of pH 7.30, 7.40, and 7.35, respectively. No explanation of these disagreements has been found. However, Peters, Barr, and Rule (1921) have called attention to the wide variation in venous blood which they found in three normals, pH 7.22, 7.30, and 7.40, and point out that a normal figure for an individual should be established before a change toward the acid or alkaline can be defined, unless it falls distinctly outside the normal zone. Thus a pH of 7.35 or 7.40 might be an alkalosis for certain individuals who have a normal pH of 7.20 or 7.25.

Data on the effect of operative removal of the cancer are meager, for cases in which all of the carcinoma was removed were most often the early cases without metastases which showed no marked alkalinity. One case of carcinoma of the breast gave a reaction of pH 7.37 before operation and 6 days postoperative gave a pH of 7.27, another case of carcinoma of the lip had a pH of 7.47 before operation and a pH of 7.18, 30 days after complete excision of the tumor. Other cases which showed no reduction after operative treatment were later found to have metastases.

These results indicate that with the increasing size of the cancer there is a progressive change in the reaction of the blood dialysate. This may have a very definite clinical value in the prognosis of operative treatment of advanced cases of carcinoma.

## SUMMARY.

The hydrogen ion concentration of the dialysate from the venous blood of forty-five cases of carcinoma averaged pH 7.45 at 20°C. This is distinctly more alkaline than that of the normal subjects which averaged pH 7.31.

The pathological cases other than carcinoma gave slightly more alkaline results than the normals, averaging pH 7.36 at 20°C.

No association of anemia with this increase in alkalinity in the venous blood dialysate in cancer was established.

In general the degree of alkalinity corresponded to the size and extent of the tumor growth. Small tumors without metastases showed little or no increase above the normal zone.

Some modifications to prevent the loss of CO<sub>2</sub> were made in the method of dialyzing blood for the colorimetric determination of the hydrogen ion concentration.

Data are given on the changes in pH and CO<sub>2</sub> tension in cooling a bicarbonate solution under oil and on the loss of CO<sub>2</sub> through the oil at different temperatures.

The factors for correcting the temperature to 20°C. were found to be pH 0.004 per degree Centigrade for the phosphate standards and pH 0.01 per degree Centigrade for the blood.

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