INTRODUCTION

It has been a trend over the past decade to look into the relationship between tissue pH and the onset and progression of systemic chronic diseases specifically cancer.

The saliva pH has been studied by some authors in relation to systemic diseases such as in GERD, Bulimia Nervosa and Burning Mouth Syndrome ***(Aframian et al., 2010).*** Also,In chronic renal failure patients on peirtoneal dialysis with and without diabtes mellitus ***(Abubekir Eltas et al., 2011).*** Another example was a study that showed significant changes in salivary pH in asthma patients compared to controls ***(Masanari Watanabe et al., 2010).***

Furthermore, some data also confirms the alteration of salivary pH in individuals on a more acidic (high protein) versus more alkaline (more vegetbales diet) ***(Young, 2002)***, which explains the trend of alkalizing one's body through an alkaline diet or alkaline water to achieve better health and control chronic diseases.

With regards to tissue pH in presence of malignancy, a number of studies using pH-sensitive magnetic resonance imaging contrast agents, microelectrodes, and magnetic resonance spectroscopy with hyperpolarized C-13 have consistently shown that the extracellular pH (pHe) of tumors is significantly lower (6.6–7.0) than that of healthy tissues (7.2–7.4) ***(Gillies et al., 2004), (Gillies et al., 2002), (Helmlinger et al., 1997), (Gallagher Faet et al., 2008)***.

AIM OF THE STUDY

This study aims at exploring salivary pH variations in patients with chronic systemic disease such as liver cirrhosis and liver cancer patients. And to observe whether or not there are discrepancies in salivary pH level in different liver disease spectra.

THE BODY pH AND ITS SIGNIFICANCE

Physiology of the Body pH

pH value denotes to the concentration of $[H^+]$ ions in a solution. The pH of the blood is perhaps the most tightly regulated process in human physiology. life or death depends on this complex regulation. A Hydrogen ion is a single free proton from a hydrogen atom. Molecules containing hydrogen atoms that can release hydrogen ions into a solution are referred to as acids. An example is HCL acid which ionizes in water to form hydrogen ions $[H^+]$ and chloride ions [CL-]. A base is an ion or molecule that can accept an $[H^+]$ for example HCO^{-3} is a base because it can accept $[H^+]$ to form H_2CO_3.

Regulation of the hydrogen ion in the body is similar to other ions in many ways, however, this ion in specific is under a much more tighter control than others. This precise control is essential because the activities of all enzyme systems in the body are influneced by H concentration and any small changes have clinical implications **(Figure 1).** Therefore, changes in the H concentration alter virtually all cells and body functions ***(Guyton & Hall, 2010)***. It is no surprise then that the H concentration of the body fluids normally is kept at low concentration compared to other ions. For example, the concentration of Na in the

Extracellular fluid (145mEq/L) is about 3.5 million as great as H concentration in extracellualr fluid, averages 0.00004mEq/L. Also the normal variation in H concentration in the extracellular fluid is about one millionth as great as that for Na emphasizing the importance of such tight control ***(Decoursey, 2003)***.

As mentioned, since the H concentration and variation is kept under tight control (normal variations are normaly under 3-5nEq/L and 10 to 160 nEq/L in extreme conditions), H concentration is always expressed on a logarithm scale using pH units. pH is related to the actual H concentraion using the following formula: PH= log 1/$[H^+]$= -log $[H^+]$

For example the normal $[H^+]$ is 40nEq/L (0.00000004Eq/L)

Therefore, the normal pH = -log (0.00000004) i.e. pH=7.4

According to this formula pH is inversely proportional to the $[H^+]$ concentration, so a high pH corresponds to a low concentration of H^+ and a low pH corresponds to a high concntration of H^+ (i.e. more acidic solution).

The normal pH of arterial blood is 7.4, whereas the pH of venous blood and interstitial fluids is about 7.35 because of the extra amounts of carbon dioxide (CO2) released from the tissues to form H_2CO^3 in these fluids. Because the normal pH of arterial blood is 7.4, a person is considered to have *acidosis* when the pH falls below this value and to have *alkalosis* when the pH rises above 7.4. The lower limit of pH at which a person can live more than a few hours is about 6.8, and the upper limit is about 8.0. Intracellular pH usually is slightly lower than plasma pH because the metabolism of the cells produces acid, especially H_2CO^3. Depending on the type of cells, the pH of intracellular fluid has been estimated to range between 6.0 and 7.4. Hypoxia of the tissues and poor blood flow to the tissues can cause acid accumulation and decreased intracellular pH ***(Guyton & Hall, 2010).***

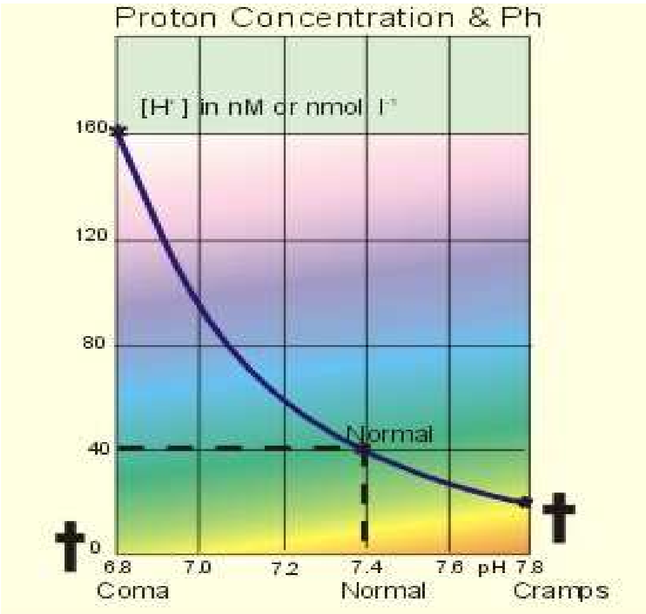

Fig. (1): pH extreme levels and its physiological impact ***(Paulev& Zubieta, 2011).***

Regulation of the pH in the body:

There are three primary systems in the body that regulate H^+ concentration to prevent acidosis or alkalosis:

The chemical acid-base buffer systems:

These systems are the fastest and some of them react within a fraction of a second to changes in H^+ concentrations. Buffer systems do not eliminate or add H^+ to the body but keep them tied up until balance is restored. These include the carbonate buffer system, the phosphate buffer system and the proteins (intracellular buffers).

The Bicarbonate buffer system:

The bicarbonate buffer system is the most powerful extracellular buffer in the body. This is due mainly to the fact that the two elements of the buffer system, HCO_3^- and CO_2, are regulated by two organs, the kidneys and the lungs as discussed later. As a result of this regulation, the pH of the extracellular fluid can be precisely controlled by the relative rate of removal and addition of HCO_3^- by the kidneys **(Figure 2)** and the rate of removal of CO_2 by the lungs ***(Capasso et al., 2002).***

Key players are two ingredients: (1) a weak acid, HCO_3^-, and (2) a bicarbonate salt, such as $NaHCO^3$.

H2CO3 is formed in the body by the reaction of CO_2 with H_2O.

H_2CO_3 ionizes weakly to form small amounts of H and HCO_3^-.

$H_2CO_3 \rightarrow H^+ + HCO_3^-$

$NaHCO_3$ ionizes almost completely to form HCO_3^- and Na^+.

$NaHCO_3 \rightarrow Na + HCO_3$

When an acid or base is added to the body, the buffers just mentioned, bind or release H^+, thereby minimizing the change in pH. Buffering in ECF occurs rapidly, in minutes. Acids or bases also enter cells and bone, but this generally occurs more slowly, over hours, allowing cell buffers and bone to share in buffering as discussed foreward ***(Rhoades and Bell, 2012).***

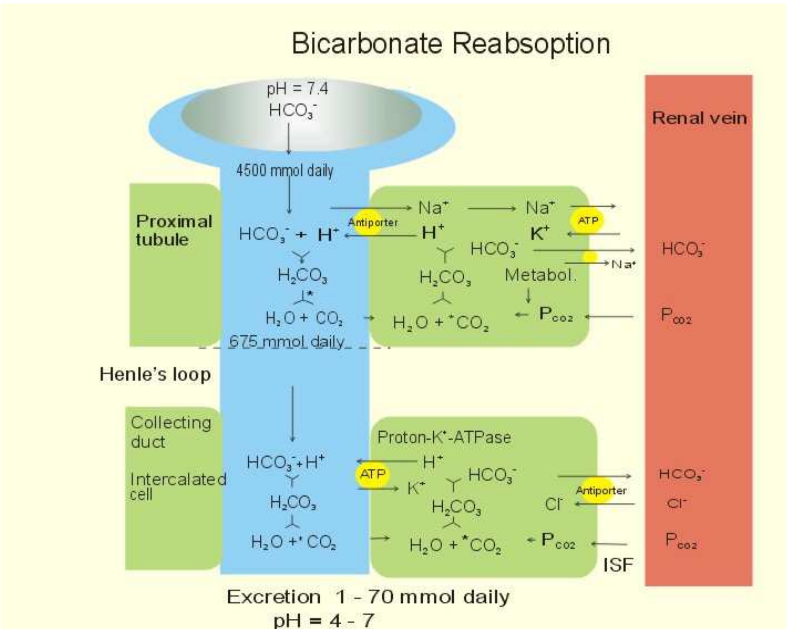

Fig. (2): Bicarbonate Handling in Kidneys, ***(Paulev& Zubieta, 2011).***

The phosphate buffer system

The main elements of the phosphate buffer system are $H_2PO_4^-$ and $HPO_{4,}$ However, its concentration in the extracellular fluid is low, only about 8 % of the concentration of the bicarbonate buffer. Despite it's insignificant role as an extracellular buffer, *the phosphate buffer is especially important as an intracellular buffer in the tubular fluids of the kidneys,* **(Figure 3)** for two reasons: (1) phosphate usually becomes greatly concentrated in the tubules, thereby, increasing the buffering power of the phosphate system. (2) The tubular fluid usually has a considerably lower pH than the extracellular fluid does. *The phosphate buffer system is also important in buffering intracellular fluid* because of another two reasons, First, cells contain large amounts of phosphate in such organic compounds as adenosine triphosphate (ATP), adenosine diphosphate (ADP), and creatine phosphate. Although these compounds primarily function in energy metabolism, they also act as pH buffers. ***(Lemann et al., 2003).*** Second, intracellular pH is generally lower than the pH of ECF and is closer to the pK_a of phosphate. (The cytosol of skeletal muscle, e.g., has a pH of 6.9) Phosphate is thus more effective in this environment than in one with a pH of 7.4. Bone has large phosphate salt stores, which also help in buffering ***(Rhoades and Bell, 2012)***.

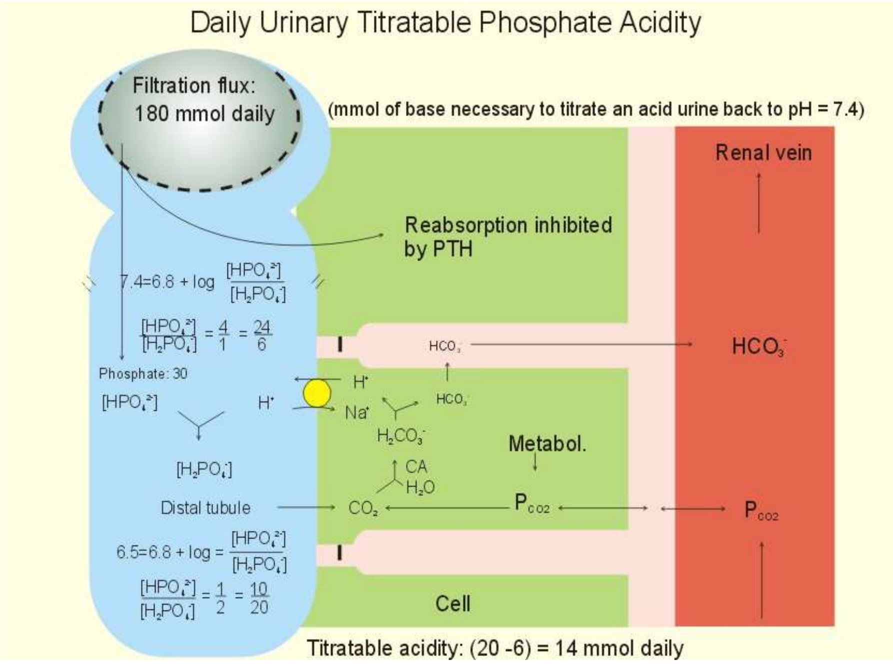

Fig. (3): phospahte control through kidneys, ***(Paulev& Zubieta, 2011)***

<u>The protein intracellular buffer system:</u>

Proteins are among the most plentiful buffers in the body because of their high concentrations, especially within the cells. The pH of the cells, although slightly lower than in the extracellular fluid, changes approximately in proportion to extracellular fluid pH changes. **(Figure 4)** There is a slight amount of diffusion of H^+ and HCO_3^- through the cell membrane, although these ions require several hours to come to equilibrium with the extracellular fluid, (except for rapid equilibrium that occurs in the red blood cells) CO_2, however, can rapidly diffuse through all the cell membranes. *This diffusion* *of the elements of the bicarbonate buffer system causes the pH in intracellular fluid to change when there are changes in extracellular pH.* For this reason, the buffer systems within the cells help prevent changes in the pH of extracellular fluid but may take several hours to become maximally effective ***(Burton, 1992).***

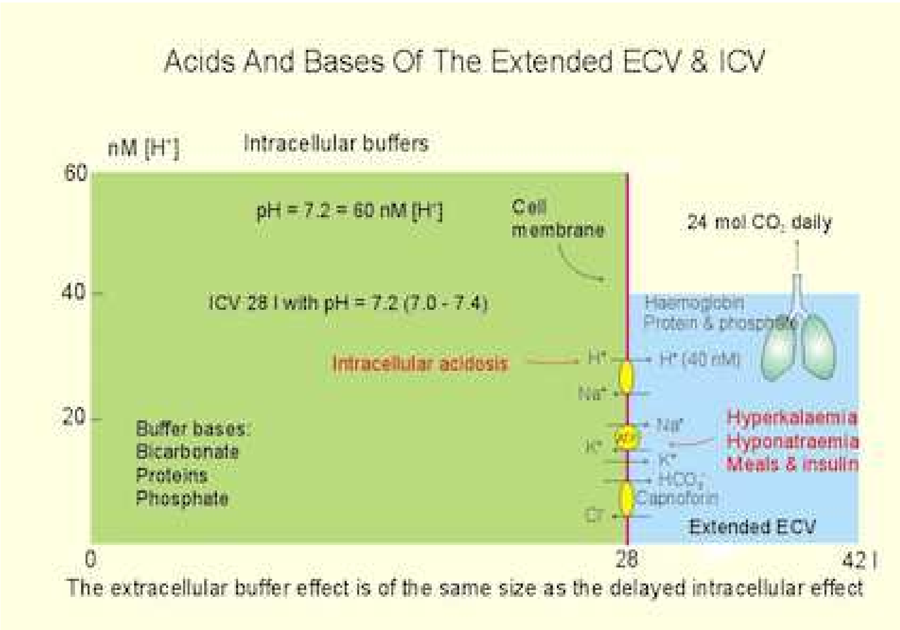

Fig. (4): Extracellular vs Intracellular Buffering effect ***(Paulev& Zubieta, 2011).***

In the red blood cell, hemoglobin (Hb) is an important buffer, as follows: $H^+ + Hb \rightarrow HHb$

Approximately 60 to 70 percent of the total chemical buffering of the body fluids is inside the cells, and most of this results from the intracellular proteins. However, except for the red blood cells, the slowness with which H^+ and HCO_3^- move through the cell membranes often delays for several hours the maximum ability of the intracellular proteins to buffer extracellular acid-base abnormalities ***(Guyton & Hall, 2010).***

The Respiratory regulation of the acid-base system:

CO_2 is formed continually in the body by intracellular metabolic processes. After it is formed, it diffuses from the cells into the interstitial fluids and blood, and the flowing blood transports it to the lungs, where it diffuses into the alveoli and then is transferred to the atmosphere by pulmonary ventilation. About 1.2 mol/ L of dissolved CO_2 normally is in the extracellular fluid, corresponding to a Pco_2 of 40 mm Hg. If the rate of metabolic formation of CO_2 increases, the Pco_2 of the extracellular fluid is likewise increased. Conversely, a decreased metabolic rate lowers the Pco_2. If the rate of pulmonary ventilation is increased, CO_2 is blown off from the lungs, and the Pco_2 in the extracellular fluid decreases. Therefore, changes in either pulmonary ventilation or the rate of CO_2 formation by the tissues can change the extracellular fluid Pco_2. If the metabolic formation of CO_2 remains constant, the only other factor that affects Pco_2 in extracellular fluid is the rate of alveolar ventilation. The higher the alveolar ventilation, the lower the Pco_2; conversely, the lower the alveolar ventilation rate, the higher the Pco_2. As discussed previously, when CO_2

concentration increases, the H_2CO_3 concentration and H^+ concentration also increases, thereby lowering extracellular fluid pH. Not only does the alveolar ventilation rate influence H^+ concentration by changing the Pco of the body fluids, but the H concentration affects the rate of alveolar ventilation. The alveolar ventilation rate increases four to five times normal as the pH decreases from the normal value of 7.4 to the strongly acidic value of 7.0. Conversely, when plasma pH rises above 7.4, this causes a decrease in the ventilation rate ***(Madias & Adrogue, 2003).***

Respiratory control cannot return the H^+ concentration all the way back to normal when a disturbance outside the respiratory system has altered pH. Ordinarily, the respiratory mechanism for controlling H^+ concentration has an effectiveness between 50 and 75 percent. *Respiratory regulation of acid-base balance is a physiologic type of buffer system* because it acts rapidly and keeps the H^+ concentration from changing too much until the slowly responding kidneys can eliminate the imbalance ***(Guyton & Hall, 2010).***

Renal Control of acid-base balance:

The kidneys control acid-base balance by excreting either an acidic or a basic urine. Excreting an acidic urine reduces the amount of acid in extracellular fluid, whereas excreting a basic urine removes base from the extracellular fluid. The overall mechanism by which the kidneys excrete acidic or basic urine is *as follows:*

Large numbers of HCO_3^- are filtered continuously into the tubules, and if they are excreted into the urine, this removes base from the blood. Large numbers of H^+ are also secreted into the tubular lumen by the tubular epithelial cells, thus removing acid from the blood. If more H^+ is secreted than HCO_3^- is filtered, there will be a net loss of acid from the extracellular fluid. Conversely, if more HCO_3^- is filtered than H^+ is secreted, there will be a net loss of base ***(Gennari & Maddox 2000).***

Each day the body produces about 80 milliequivalents of nonvolatile acids, mainly from the metabolism of proteins. These acids are called *nonvolatile* because they are not H_2CO^3 and, therefore, cannot be excreted by the lungs. The primary mechanism for removal of these acids from the body is renal excretion. The kidneys must also prevent the loss of bicarbonate in the urine, a task that is quantitatively more important than the excretion of non volatile acids. Each day the kidneys filter about 4320 milliequivalents of bicarbonate (180 L/day -24 mEq/ L); under normal conditions, almost all this is reabsorbed from the tubules, thereby conserving the primary buffer system of the extracellular fluid ***(Gennari & Maddox, 2000).***

Both the reabsorption of bicarbonate and the excretion of H^+ are accomplished through the process of H^+ secretion by the tubules. Because the HCO_3^- must react with a secreted H^+ to form H_2CO^3 before it can be reabsorbed, 4320 milliequivalents of H^+ must be secreted each day just to reabsorb the filtered bicarbonate. Then an additional 80 milliequivalents of H^+ must be secreted to rid the body of the nonvolatile acids produced each day, for a total of 4400 milliequivalents of H^+ secreted into the tubular fluid each day. When there is a reduction in the extracellular fluid H^+ concentration (alkalosis), the kidneys fail to reabsorb all the filtered bicarbonate, thereby increasing the excretion of bicarbonate. Because HCO_3^- normally buffers hydrogen in the extracellular fluid, this loss of bicarbonate is the same as adding an H^+ to the extracellular fluid. Therefore, in alkalosis, the removal of HCO_3^- raises the extracellular fluid H^+ concentration back toward normal. In acidosis, the kidneys do not excrete bicarbonate into the urine but reabsorb all the filtered bicarbonate and produce new bicarbonate, which is added back to the extracellular fluid. This reduces the extracellular fluid H^+ concentration back toward normal ***(Guyton & Hall, 2010).***

Thus,the kidneys regulate extracellular fluid pH concentration through three fundamental mechanisms:

(1) Secretion of H^+

(2) Reabsorption of filtered HCO_3^-

(3) Production of new HCO_3