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PATHOBIOCHEMISTRY OF METABOLIC DISORDERS

Limits of Inner Maintenance

Jan Pláteník et al.

Pathobiochemistry of Metabolic Disorders

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FOREWORD

Pathobiochemistry means biochemistry of disease. Just like the ‘usual’ biochemistry describes the chemical processes in the healthy organism, pathobiochemistry deals with the yet more varied and complicated molecular mechanisms of disease manifestation and progression. The idea of such application of biochemical knowledge is naturally attractive both for the medical students and their teachers. Accordingly, it is several decades ago when the pathobiochemistry, as a small lecture course following and extending the major biochemistry classes, found its way to the curriculum of our medical school. Obviously, the subject requires a careful definition. As our understanding of diseases everywhere approaches the ‘chemical’, molecular level, nearly any pathogenetic mechanism could be regarded as pathobiochemistry, and we certainly cannot teach everything or just anything. It is better to deal with the general pathogenetic mechanisms, rather than specific diseases. This still leaves a lot of space, and currently there are three alternative variants of the pathobiochemistry course at our faculty.

Our version of these classes has evolved a focus on the ‘degenerative’ pathogenetic mechanisms. The healthy human body is a nearly perfect chemical machine that is equipped with regulatory circuits for countless biochemical and physiological variables, and constantly repairs and renews itself. However, these elaborate mechanisms of self-regulations and renewal do have limits. Exceeding these limits, either by external perturbation or simply by accumulation of errors with time, leads to a dysfunctional state or disease that might not have any evident cause. The doctors know it: once a serious diagnosis is formulated, all too often the patient asks why, what has been done wrong that caused the disease. But sometimes nothing is obviously wrong, and still the disease comes. Many conditions do have definite, well defined causes, but often there is an inherent, innate pathogenetic component acting as an invisible cause, or contributing to the disease progression. Medicine even knows the term disease *sui generis*, i.e. just happening without any evident cause. Undoubtedly, any understanding of such ‘inner’ reasons for diseases can help a lot in the subsequent study of internal medicine. And by exploring the build-in limits of the living organisms we may actually touch some of the very general secrets of life. That is what we try to provide in our pathobiochemistry course. Our lectures are found interesting, even enjoyable, both by the students and teachers who give them. Only a lack of written materials or a suitable textbook has been consistently recognized as the main drawback of our course. This book is intended as an attempt to fill this gap.

The eleven chapters of the book cover the scope of all our seven pathobiochemistry lectures, and include a few related topics that would also deserve a lecture were we had more

time. The chapters are of unequal length, some are brief and provide just basic information, whereas in others the topic demanded a more comprehensive explanation that goes beyond the current scope of our course. Regardless of this variation all the texts should be useful, and it should be noticed how much the topics are interconnected. An adequate knowledge of 'normal' biochemistry is expected, although occasionally the basic principles are explained as well. The list of literature after each chapter is meant both as a tribute to the sources and as a suggestion of further reading for those who would like to learn even more.

This book is intended to complement and support our lectures, not to replace them. We believe it will help us move our course to another level and make the discussions with students even more thought-provoking than before. And despite its obvious imperfections, we also hope that it will be found useful for a wider audience. Let me wish you an enjoyable reading.

Prague June 27th 2023

Jan Pláteník
On behalf of all co-authors

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ACID-BASE EQUILIBRIUM AND ITS MOST COMMON DISORDERS

Martin Vejražka

Acidity of the internal environment of the body has a major impact on most processes that take place in it. Even minor pH changes cause conformational changes of proteins and thus influence their function. As a result, the activity of enzymes, the function of receptors, transport proteins, ion channels and pumps, etc. are altered.

Physiologically, the **pH of plasma** and some other body fluids is maintained within a very narrow range of **7.36 to 7.44**. Buffer systems and regulation of some physiological and metabolic processes contribute to the pH stability of the internal environment.

1 BUFFER SYSTEMS

Buffer systems allow **very fast** reaction to the the excess and loss of acids and bases in the inner environment. The reactions by which the resulting concentrations of H^+ protons return to normal values occur within fractions of a second. At the same time, their buffering capacity is reduced, so without regeneration, these systems would quickly deplete.

About half of the total buffer capacity of whole blood is accounted for by **hydrogen carbonate** (bicarbonate) buffer. The second most important is **hemoglobin** buffer, followed by other **proteins** and organic and inorganic **phosphates**.

1.1 Hydrogen carbonate buffer

Undoubtedly, the most important buffer system in the human body is hydrogen carbonate buffer. In most body fluids, it accounts for more than half of the total capacity of all buffers. However, the particular importance of the hydrogen carbonate buffer is mainly due to the wide **openness of its system**.

The general reaction scheme of the hydrogen carbonate buffer is simple:



Carbon dioxide is continuously supplied by tissue **respiration** and is excreted from the body by the lungs in a controlled way. Similarly, the hydrogen carbonate anion is absorbed from food or is formed in some **metabolic processes**, and at the same time is excreted in the urine

by the kidneys. If the concentration of hydrogen carbonate buffer components changes as a result of the buffering reactions, the organism has the possibility to return these concentrations to normal values quite quickly (depending on the situation within a few minutes to several days) and thus restore the pH of the internal environment. No other buffer in the human body can be replenished as fast as the hydrogen carbonate buffer. Thanks to its openness, the hydrogen carbonate buffer is virtually **inexhaustible under physiological conditions** or even in mild acid-base balance disorders.

Hydrogen carbonate buffer is such an important part of the mechanisms that maintain the acid-base balance in the human body, that in clinical practice the diagnosis of acid-base equilibrium (ABE) disorders is often simplified solely to examining its components.

2 REGULATION OF PHYSIOLOGICAL AND METABOLIC PROCESSES

In addition to buffer systems, the regulation of some processes also helps to maintain the pH stability of the internal environment. These mechanisms are much **slower than buffer** reactions, but allow the buffer composition to be completely restored and thus return the pH of the internal environment back to normal. The most important is regulation of lung **ventilation** and regulation of **transport processes** in the kidneys.

2.1 Respiration

Hydration of carbon dioxide as it dissolves in blood and other body fluids produces carbonic acid. Thus, the partial pressure of carbon dioxide in the blood corresponds to the concentration of carbonic acid, i.e. conjugate acid of the hydrogen carbonate buffer. A change in the ratio of conjugate acid to conjugate base (here a hydrogen carbonate anion) must necessarily be accompanied by a change in the pH of the system. For ideal solutions, this relationship can be described using **Henderson-Hasselbalch equation**:

$$pH = pK_a + \log \frac{c_b}{c_a}$$

where pK_a is dissociation constant of carbonic acid,

c_a is molar concentration of carbonic acid (conjugate acid),

c_b is molar concentration of hydrogen carbonate anion (conjugate base).

Increasing the partial pressure of CO_2 in blood (**hypercapnia**), for example when ventilation decreases, is accompanied by **respiratory acidosis** and *vice versa*, **hypocapnia** is accompanied by **respiratory alkalosis**.

The regulation of the pH of the internal environment by respiration is prompt under physiological conditions. It responds almost immediately to deviations. With greater deviation, respiratory compensation gradually increases, so that maximum efficiency is achieved after a few hours.

2.2 Renal regulation of ABE

In the long term, the renal regulation of acid and alkali excretion is the most important for maintaining a stable pH of the internal environment. In general, the metabolic processes of human (and more generally all heterotrophic organisms) produce excess acids. So it is not surprising that urine pH is more acidic than blood pH in most situations.

Renal tubular cells can regulate the secretion of H^+ cations. Most of these cannot remain free in the urine, but react with bases present in it, especially with hydrogen phosphates to form dihydrogen phosphates (Fig. 1-1). Due to this buffering process, the pH of the urine itself changes relatively little, so it is more accurate to say that the **titratable acidity** of the urine changes significantly.

In addition, renal tubular cells regulate the **reabsorption of hydrogen carbonate anions** (Fig. 1-1) from primitive urine into which they have been filtered from the blood in glomeruli.

The regulation of the pH of the internal environment by the kidneys is very effective, but it responds relatively slowly to more profound changes. This is because the activity of transport mechanisms needs to be altered, including the expression of certain proteins. Therefore, it reaches maximum efficiency in approximately five days from the beginning of the disorder.

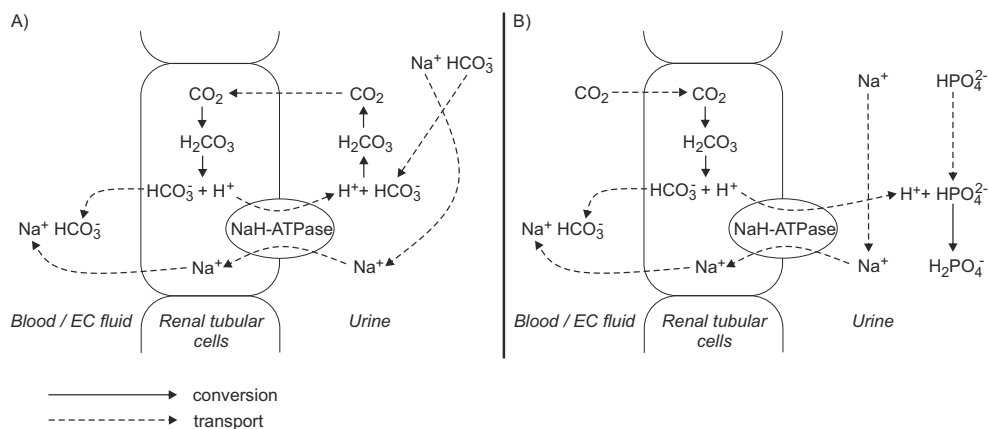


Figure 1-1: Mechanism of urine acidification (simplified).

Hydrogen carbonate anions are reabsorbed from primitive urine to blood (panel A). Sodium cations accompany hydrogen carbonate anions to keep electroneutrality of the process. As a result, hydrogen carbonate concentration is decreased in urine, leading to its acidification, and increased in blood plasma.

Hydrogen carbonate anions can also be produced by hydration of carbon dioxide and subsequent dissociation of carbonic acid (panel B). Protons H^+ are excreted to urine, increasing its titratable acidity. Hydrogen carbonate anions are transported to the blood, followed by sodium cations. The whole process leads to increase in blood plasma hydrogen carbonates, i.e. alkalinisation of the inner environment, and acidification of urine.

EC fluid – extracellular fluid.

3 SELECTED PARAMETERS USED IN THE DIAGNOSIS OF ACID-BASE BALANCE DISORDERS

In practice, acid-base balance examination is performed in conjunction with blood gas testing. The reasons are both historical and due to the fact that severe respiratory disorders are inherently associated with severe ABE disorders, so that such combined testing is useful. Blood gas and acid-base balance tests require the same method of sampling and pre-analytical sample processing, i.e. anaerobic sampling and rapid transport.

A number of indicators are used to assess the acid-base balance. Part consists of directly measured analytes, especially components of buffer systems and the main ions. In addition, a number of calculated parameters have been described which may, in some situations, facilitate the description of ABE failures. At this point, we will focus only on selected parameters, which, however, in the vast majority of situations are sufficient for a full diagnosis of ABE disorders.

3.1 pH of whole blood

The parameter that gives general information about the acid-base balance is the pH of whole blood. The pH of the **arterial blood** is measured in the most ideal case. If arterial blood sampling is not available for any reason, a sample of **arterialized capillary blood** is drawn, i.e. sample of capillary blood from well-blooded tissue in which the blood flow is fast enough to approach the composition of arterial blood. The determination of ABE parameters from **venous blood** has a significantly lower informative value. If venous blood is sampled, it should be collected as much as possible from the center, preferably from a central venous catheter leading to the right atrium.

Blood pH alone reports significant and uncompensated disorders of acid-base balance. If the pH of the blood is lower than normal, we talk about **acidemia**, if it is higher, about **alkalemia**. However, it should be borne in mind that blood pH may be within the reference limits even in some deep ABE disorders, so this parameter should never be interpreted separately.

Reference values: 7.36–7.44

3.2 Partial pressure of carbon dioxide (pCO₂)

In the body, carbon dioxide is readily hydrated to carbonic acid, i.e. conjugate acid of the hydrogen carbonate buffer. The equilibrium of this reaction is established very quickly, also due to catalysis by the enzyme **carbonic anhydrase** (carbonic acid hydro-lyase, E.C. 4.2.1.1).

The partial pressure of carbon dioxide informs us about the role of ventilation in the acid-base balance disorder, i.e. the respiratory component of ABE. Increased losses of carbon dioxide, for example in hyperventilation, lead to a decrease in pCO₂, i.e., **hypocapnia**. Decreasing the concentration of carbonic acid is the cause of **respiratory alkalosis**. Conversely, carbon dioxide retention associated with **hypercapnia** (increase in pCO₂) characterizes **respiratory acidosis**.

Reference values: 4.8–5.8 kPa.

3.3 Concentration of hydrogen carbonates

The hydrogen carbonate anion is the conjugate base of the hydrogen carbonate buffer. The concentration ratio of hydrogen carbonate anion to carbonic acid (and hence $p\text{CO}_2$) is tightly bound to the pH of the internal environment; their relationship can be described by the equation of Henderson and Hasselbalch. However, the **actual value of the hydrogen carbonate concentration** has limited use and is not evaluated in clinical practice. The difficulty is that it cannot be evaluated separately, but only together with $p\text{CO}_2$ within the Henderson and Hasselbalch equation. The actual hydrogen carbonate anion concentration varies both during changes in metabolic processes that affect ABE, and during changes in respiration.

The more commonly used parameter is the so-called **standard concentration of hydrogen carbonate anion**. The term “standard” here, as with other ABE parameters, means that this would be a value that would be achieved with “standard” respiration, i.e. with $p\text{CO}_2$ just 5.3 kPa. We can imagine that instead of measuring the actual concentration of hydrogen carbonate anion in the patient’s blood, we would first equilibrate the blood sample in an atmosphere with $p\text{CO}_2 = 5.3$ kPa and the hydrogen carbonate anion concentration would be determined thereafter. In fact, both actual and standard concentrations of hydrogen carbonate are not measured directly, but rather calculated. Theoretically, both values could be calculated from pH and $p\text{CO}_2$ using the Henderson-Hasselbalch equation. Due to the fact that blood is not an ideal solution and thus the real values differ from the theoretical ones, empirical equations are employed.

It follows from the definition of standard hydrogen carbonate concentration that it is not affected by any possible respiratory disorder of ABE – this is eliminated by setting $p\text{CO}_2$ to 5.3 kPa. Thus, the standard concentration of hydrogen carbonates informs us purely of the **metabolic component** of the acid-base balance. **Metabolic acidosis** is characterized by a decrease in standard hydrogen carbonates and **metabolic alkalosis** by its increase.

Reference limits: 22–26 mmol/l

3.4 Base excess

Similar information as standard hydrogen carbonate concentration gives us another standard parameter, the so-called base excess. This is a calculated parameter, but we can simply imagine that we could measure it by doing the following:

Suppose we first equilibrate a whole blood sample in an atmosphere with $p\text{CO}_2$ of 5.3 kPa. This eliminates any deviation by the respiratory component of the ABE. Then we measure the pH of the blood. Now imagine that the pH of a particular sample is greater than 7.4 – that is, it contains more conjugate buffer bases and since we eliminated the respiratory component of the disorder in the previous step, it is metabolic alkalosis. Now add the acid to the sample until the pH reaches 7.4. The amount of acid that we had to add, expressed in mmol per liter of whole blood, is referred to as base excess (BE).

With more frequent metabolic acidosis, we would have to add a base to adjust the pH of the blood sample. We would then talk about a similar parameter, the **base deficit** (BD). However, it is more common to use a BE with a negative sign.

As already mentioned, the base excess is a standard parameter. If BE is negative, it is metabolic acidosis, positive BE is characteristic of metabolic alkalosis.

Reference limits: –2.5 to +2.5 mmol/l

4 THE MOST COMMON CAUSES OF METABOLIC ACIDOSIS

Metabolic acidosis is characterized by a decrease in the standard concentration of hydrogen carbonate and a negative base excess. It can be triggered by two groups of faults:

1. Increasing the concentration of relatively strong anions.
2. Increased loss of hydrogen carbonate anion.

4.1 Metabolic acidosis caused by increase of the concentration of relatively strong anions

With a physiological concentration of about 24 mmol/l, the hydrogen carbonate anion is an important component of the **ionogram** (Fig. 1-2). Its conjugate acid, carbonic acid (or, assuming some simplification, carbon dioxide) has no charge.

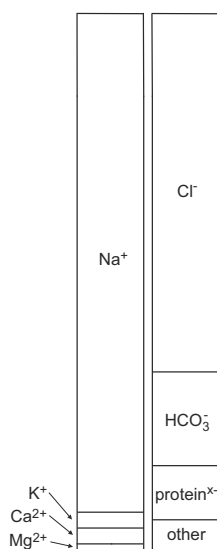


Figure 1-2: Ionogram of blood plasma (simplified).

The concept of electroneutrality of blood plasma assumes that the total concentration of positive charge of all cations (column on the left) equals to the total concentration of negative charge of all anions (column on the right). Every change in concentration of any of the major ions must therefore be balanced by another change, so that electroneutrality is restored.

To put it simply, it is possible to imagine that the body fluids must have approximately the same total concentration of cations and anions, i.e. the body fluid must be electroneutral. If there is a metabolic disorder in which charged particles accumulate or are lost, the disorder must be accompanied by another event that leads to the **maintenance of the electroneutrality** of the internal environment. By adopting this simplified concept, it is conceivable that the deflection of electroneutrality will often be compensated by a corresponding increase or decrease in the hydrogen carbonate anion concentration. If there is no change in respiration and pCO_2 at the same time, the ratio of HCO_3^- to carbonic acid will deviate and thus the pH of the inner environment will change.

The accumulation of relatively strong anions occurs in a number of situations:

– **Lactate accumulation – lactic acidosis**

- Anaerobic metabolism (Chapter VII):

Lactate overproduction occurs in tissue hypoxia, whether caused by impaired perfusion, diffusion and transport of substances between blood and tissue cells, or in whole-body hypoxia in cardiac and respiratory insufficiency.

- Extreme physical load:

Lactate produced by the muscles cannot be utilized in the Cori cycle quickly enough.

- Ethanol consumption (Chapter XI):

Oxidation of ethanol with an alcohol dehydrogenase and aldehyde dehydrogenase system results in the reduction of large amounts of NAD^+ to NADH in hepatocytes. The reduction state of the cells with a relative excess of NADH causes that lactate is not effectively oxidized by the lactate dehydrogenase to pyruvate and, at the same time, any pyruvate is reduced to lactate.

- Other causes:

These include, for example, inherited mitochondrial disorders and other rare metabolic diseases leading to lactic acidosis. Lactate accumulation will also occur due to poisoning of mitochondrial respiratory chain, e.g. with cyanide. Oxidation products of methanol and ethylene glycol have similar effects.

– **Accumulation of ketoacids – ketoacidosis**

In situations where glucose cannot cover the energy demands of cells in usual extent, fatty acid oxidation increases. This results in large amounts of ketone bodies, including anions of acetoacetic acid and hydroxybutyric acid. Conditions leading to ketoacidosis include, for example:

- Decompensated type 1 diabetes mellitus:

Insulin deficiency causes a lack of glucose as an energy source in cells of insulin-dependent tissues.

- Starvation.

- Extreme energy consumption during physical exercise, sepsis, etc.

- Alcohol consumption:

Alcohol inhibits the oxidation of glucose fragments in the Krebs cycle by reducing pyruvate to lactate (see above). Lactic acidosis also decreases urinary excretion of ketone bodies as the small organic anions compete for common transport mechanisms in renal tubular cells.

– **Accumulation of sulfates and phosphates in renal failure**

Sulfates and phosphates, which are formed by oxidation of sulfur and phosphorus-containing organic substances, respectively, are mostly non-metabolized waste products. These are strong anions, which are mainly excreted unchanged by glomerular filtration. Renal insufficiency is typically accompanied by their accumulation and leads to metabolic acidosis.

– **High supply of substances metabolized to anions**

For example, oxidation of simple alcohols leads to the production of large amounts of anions. Oxidation of ethanol gives acetate. In this case, the effect of anion overproduction is rather transient, since acetate in the form of acetyl coenzyme A becomes a substrate for pathways of energetic metabolism and is either consumed in the Krebs cycle or used for fatty acid synthesis. The oxidation of methanol forms formate, and similarly ethylene

glycol is transformed to oxalate. Both products have significant impact. These are anions that can be only slowly metabolized or excreted. Their accumulation leads to severe metabolic acidosis, which plays an essential role in the toxicity of such alcohols.

4.2 Metabolic acidosis from loss of hydrogen carbonate

Metabolic acidosis from loss of hydrogen carbonate is most often the result of increased losses of HCO_3^- from the **digestive tract**. Physiologically, pancreatic juice and fluid secreted by the duodenal mucosa are alkaline due to the high content of hydrogen carbonates. This makes it possible to neutralize the strongly acidic chymus that enters the duodenum from the stomach. Most of the hydrogen carbonates are later reabsorbed in the more distal parts of the intestine. If this reabsorption fails, e.g. in diarrhea or other disorders of the digestive tract, metabolic acidosis develops.

Large losses of hydrogen carbonates can also occur in **tubular function disorders**, since hydrogen carbonates that have passed to primitive urine by glomerular filtration are not sufficiently absorbed.

Rapid **dilution of extracellular fluids** also results in metabolic acidosis, for example when large amounts of infusions are administered. The processes that keep pCO_2 constant are much faster than the processes that keep HCO_3^- constant. Thus, rapid dilution of extracellular fluids results in a more significant decrease in HCO_3^- than pCO_2 , resulting in acidification of the internal environment.

5 THE MOST COMMON CAUSES OF METABOLIC ALKALOSIS

Metabolic alkalosis is less common than metabolic acidosis, but is potentially more dangerous. Acidosis is frequently associated with relatively common situations, such as high physical load, work on oxygen debt, starvation, etc. Basically, scarcity of situations leads to alkalosis, and therefore human compensation mechanisms are not so well developed.

Most often, metabolic alkalosis is caused by the **loss of chloride**, which – simply put – is replaced by a higher concentration of hydrogen carbonates in the ionogram. Chlorides may be lost especially with gastric juice, e.g. in vomiting, or in the urine in some tubular function disorders, or due to overdose with thiazide diuretic drugs.

Other disorders that lead to metabolic alkalosis are **hypoproteinemias**. Proteinuria in nephrotic syndrome, disorders of proteosynthesis in liver failure, protein deficiency in malnutrition all belong to this group. Proteins behave mainly as polyanions, so that hypoproteinemia leads to anion deficiency in the ionogram.

The rapid **loss of water** from the extracellular environment, which increases the concentration of hydrogen carbonates, also leads to metabolic alkalosis. In fact, it is a reversal of the mechanism that leads to metabolic acidosis by rapid fluid administration. This concentration metabolic alkalosis is encountered e.g. in rapid dehydration after administration of higher doses of diuretics, in profuse sweating, large transudation from burns, etc.

Metabolic alkalosis can also be caused by some disruptions in mineral management. Typically, **hyperaldosteronism**, either primary (Conn's syndrome), or secondary to the activation of the renin-angiotensin-aldosterone system, or to a failure of aldosterone degradation by

the liver, results in alkalosis. Hyperaldosteronism leads to a decreased excretion of sodium cations at the expense of potassium ions and protons. Thus, there is a greater acidification of urine and hypokalemia, which further promotes alkalosis.

6 RESPIRATORY DISORDERS

As already mentioned, increased carbon dioxide output by respiration causes **respiratory alkalosis** and conversely carbon dioxide retention leads to **respiratory acidosis**. The exchange of oxygen and carbon dioxide, respectively, between the blood and the air in the lungs is to some extent independent of each other. Therefore, there are relatively frequent situations where hypoxia and hypocapnia occur simultaneously. Hypoxia can lead to lactic metabolic acidosis. At the same time, respiratory efforts are increased, so that respiratory alkalosis develops. This combination can be found, for example, in some conditions associated with pulmonary edema, pneumonia, embolism in smaller branches of the pulmonary artery, etc. Only severe respiratory failure causes hypoxia and hypercapnia at the same time.

7 RELATIONSHIP BETWEEN ACID-BASE BALANCE AND ION METABOLISM

In the sections devoted to metabolic acidosis and alkalosis we have already discussed the **simplified concept of electroneutrality** of body fluids and the relationship between the concentration of the main ions and the composition of the hydrogen carbonate buffer. Generally, a disorder of cation and anion concentrations is always accompanied by an acid-base balance disorder – anion excess or cation deficiency leads to metabolic acidosis and *vice versa*, anion deficiency or cation excess is associated with metabolic alkalosis. However, the relationship between the principal ions equilibrium and ABE is bilateral – every major disorder of acid-base balance is inevitably accompanied by disruption of the mineral levels.

However, there are more links between the main ion concentrations and the buffer composition. From the medical point of view, processes in which major ions are exchanged for protons play an important role.

7.1 Exchange of K^+ and H^+ on cell membranes

Not only extracellular but also **intracellular buffer systems** are involved in maintaining the acidity of the internal environment. Thus, for example, if the extracellular space is acidified, some of the excess protons pass through the plasma membrane of the cells into the intracellular space, where they are taken up by intracellular buffers. As the proton is a positively charged particle, the process cannot remain alone – the membrane potential of the cells would change with all its consequences. Electroneutrality of the process is achieved by passing additional ions across the cell membrane, which compensates for the charge change. The plasma membrane has a relatively high permeability to K^+ cations. In case of acidosis the K^+ will go out of cells so we can talk about the exchange of H^+ for K^+ on the plasma membrane. Acidosis therefore leads to hyperkalemia and *vice versa* alkalosis leads to hypokalemia. However, ion

exchange can also be forced in the opposite direction – hyperkalemia is the cause of metabolic acidosis, hypokalemia leads to metabolic alkalosis.

7.2 Competition between Ca^{2+} and H^+ for protein binding sites

Buffers include protein molecules such as blood plasma proteins. They contain several types of functional groups that act as weak acids and bases and may be in a protonated or deprotonated state according to the pH of the internal environment. These groups include **carboxyl groups** on which a simple acid-base reaction may take place:



The carboxylate anion of the protein $-\text{COO}^-$ can effectively bind some divalent cations, especially Ca^{2+} . Thus, during *in vivo* buffering events, there is no reversible change from the carboxyl group to the carboxylate anion and back, but there is a change from the free carboxyl group to the carboxylate group with a bound calcium.

As a result of these changes, the originally **ionized form of calcium** is converted to non-diffusible, **non-ionized protein-bound calcium** and back. Changes in the availability of ionized calcium have other physiological consequences, as the concentration of Ca^{2+} in ionized form plays an important role in a number of events – for example, it is crucial for regulation of neuromuscular irritability.

7.3 Paradoxic aciduria

In severe alkalosis, which is not due to changes in the transport of substances in the kidneys, we expect that the kidneys will start to secrete more basic urine and alkaliuria develops as a result of compensatory events. This is indeed the case, but only to a limited extent. As mentioned above, heavier alkalosis is accompanied by hypokalemia due to cell membrane events. As the K^+ concentration decreases, the kidneys do not have enough potassium to exchange it with sodium from primitive urine and, instead, sodium is exchanged for proton in the renal tubules. This leads to acidification of urine, the so-called paradoxic aciduria. The loss of acids from the body gets deeper and alkalosis worsens.

Literature/Suggested Reading

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II. PATHOBIOCHEMISTRY OF INFLAMMATION

Marta Kalousová

Inflammation is defined as a reaction of the organism to any pathologic insult. Phylogenetically as well as ontogenetically, inflammation is the oldest defence mechanism.

Inflammation can be caused by **biological factors**, i.e., **infection** – bacteria, viruses or fungi, **chemical factors** – poisons or corrosive substances, **physical factors** – e.g. burns, **mechanical factors** – e.g. injury, or by **tissue ischemia and necrosis**. The pathogenic insult causes an acute local inflammation that in case of adequate reaction should be repaired. However, in case the insult is intense, acute local reaction can escalate to a systemic response; in case of an extensive insult, systemic response can develop even without local reaction (septic shock when the microorganisms enter the bloodstream or anaphylactic shock when the impulse is non-infectious). If the local inflammatory reaction is insufficient, a chronic inflammation can develop instead of reparation (Fig. 2-1).

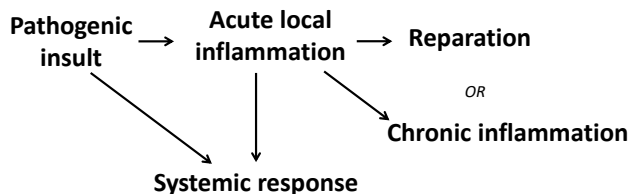


Figure 2-1: The course of inflammation.

1 SIGNS OF INFLAMMATION

Classical signs of inflammation include **calor**, **rubor**, **tumor**, **dolor** and **functio laesa**, which means that the inflamed site can be observed and felt as hot, red, swollen, painful, and with impaired function. This is caused by an increased blood flow through the inflamed site (hyperemia), release of mediators of inflammation, exudation of the fluid to the extracellular space and tissue damage. In **acute local inflammation**, transient vasoconstriction (in case of injury) followed by vasodilation and an increased vascular permeability can be seen. Leukocytes migrate to the site of inflammation, release mediators and can phagocytose. Neutrophils are capable of respiratory burst and the role of reactive oxygen species (ROS) in killing

bacteria should be mentioned (Chapter III). Blood clotting cascade, fibrinolysis as well as the complement and kinin systems are activated. Later, specific immunity becomes involved (B- and T-lymphocytes), encompassing production of antibodies as well as the specific cellular response. The aim is to demarcate, destroy and eliminate the pathogenic agents and to repair the damage. Inflammation is then actively terminated. **Chronic inflammation** is given by persistence of foreign material at the site of inflammation. In this case, rather lymphocytes, monocytes and plasmatic cells are found (compared to neutrophils in acute inflammation), and there is a sustained production of mediators of inflammation. Deposition of collagen and fibrinogen leads to fibrotization (scarring), which is mediated mainly by TGF β (transforming growth factor beta).

Systemic inflammatory response (SIRS) is characterized by either increased or decreased temperature ($> 38\text{ }^{\circ}\text{C}$ or $< 36\text{ }^{\circ}\text{C}$), either increased or decreased leukocyte count ($> 12,000/\mu\text{l}$ or $< 4,000/\mu\text{l}$) or more than 10 % of immature forms based on the intensity of inflammatory response (high or insufficient/exhausted), tachycardia more than 90/min, and tachypnoea more than 20 breaths/min or a decreased $\text{pCO}_2 < 4.3\text{ kPa}$. Systemic inflammatory response is connected to increase in acute phase reactants and release of glucocorticoids. In case of infection, it can further develop to **sepsis** and **septic shock** with hypotension, organ hypoperfusion and multiorgan damage. The definition of sepsis has developed and changed over years with inclusion of some laboratory parameters such as markers of inflammation and signs of organ damage. However, current quick diagnosis of sepsis is solely based on clinical signs: hypotension, tachypnoea, and alteration of consciousness.

Focusing on inflammation mainly from the biochemical point of view, mediators of inflammation, phagocytosis and respiratory burst as well as laboratory and metabolic changes during inflammation will be mentioned.

2 MEDIATORS OF INFLAMMATION

Mediators of inflammation include alarmins (DAMPs – damage-associated molecular patterns); vasoactive amines – histamine and serotonin; eicosanoids – derivatives of arachidonic acid such as prostaglandins, prostacyclins, thromboxanes and leukotrienes; cytokines; reactive oxygen and nitrogen species; as well as complement, coagulation, fibrinolytic and kinin systems.

Alarmins (DAMPs – damage-associated molecular patterns) are released from damaged (necrotic) cells and include e.g. HMGB1 (high mobility group box 1), EN-RAGE (extracellular newly identified receptor for advanced glycation end-products binding protein) – also called S100A12, proteins S100A8/A9, interleukins (IL-1 α), heat shock proteins, nucleosomes or ATP (adenosine triphosphate). Apart from DAMPs, **PAMPs (pathogen-associated molecular patterns)** are also of importance – they are represented by microbial structures such as dsRNA and DNA, bacterial protein flagellin, fungal β -glucan and α -mannan cell wall components, lipopolysaccharide of Gram-negative bacteria, lipoteichoic acid of Gram-positive bacteria or peptidoglycan. Both DAMPs and PAMPs bind to **pattern recognition receptors (PRRs)** that regulate immune response, cell death, or differentiation. Toll-like receptors (TLR) are the best known PRRs. TLR signalling leads to the production of cytokines (e.g. IL-1 β) via activation of nuclear factor-kappa B (NF- κ B). Receptor for advanced glycation end-products (RAGE), another transmembrane receptor, is a multi-ligand receptor