Received: 26 July 2020 | Revised: 8 September 2020 | Accepted: 8 September 2020

DOI: 10.1111/apha.13559

EDITORIAL

ACTA PHYSIOLOGICA

Strong ion difference in urine: A measure of proton excretion or of the net plasma charge alteration?

In the presence of a metabolic acid-base disorder, the kidney is called upon to compensate appropriately, altering the net acid excretion in urine. Metabolic acidosis is normally associated with urine acidification. This process can be appreciated through two different explanatory models.

On the one hand, according to the widely adopted approach, pH regulation is achieved with proton (H^+) removal through the urine, mainly in the form of ammonium cation (NH_4^+).[1] Other buffers, such as phosphate, also contribute to H^+ excretion although to a lesser degree. NH_4^+ excretion results in the regeneration of bicarbonate (HCO_3^-), which increases extracellular fluid pH. Specifically, the process of NH_4^+ production by itself is considered to have no bearing in total acid-base balance, in case the NH_4^+produced in the kidney is not excreted but, being absorbed in blood, is transferred to the liver where it participates in urea production. There, one HCO_3^- is consumed along with one NH_4^+in the process of ureagenesis. Thus, it has been proposed that the net effect on extracellular fluid pH is because of NH_4^+ being excreted rather than absorbed in blood and converted to urea in the liver; excretion of NH_4^+results in net HCO_3^- gain.

Urine [NH_4^+] is indirectly assessed in clinical practice by measurement of the urine anion gap (U_{AG}).[2] [U_{AG}] calculation formula reflects the difference between urine unmeasured cation (UCs) and unmeasured anion (UAs) concentrations. From the law of electrical neutrality, applying to aqueous solutions, it follows:

$$[Cl^-]+[UAs]=\left(\left[Na^+\right]+\left[K^+\right]\right)+[UCs]\rightarrow\left(\left[Na^+\right]+\left[K^+\right]\right)$$
$$-[Cl^-]=[UAs]-[UCs]=\left[U_{AG}\right]$$

Quantitatively the main unmeasured cation in urine isNH_4^+. In this setting, increase of NH_4^+excretion, with more severe acidosis, leads to reduced/negative [U_{AG}] values.

In hyperchloraemic acidosis such a negative correlation between NH_4^+excretion and [U_{AG}] has long been demonstrated. In fact, [U_{AG}] is used as a differential marker for non-anion gap metabolic acidosis, being largely negative in case of diarrhoea with normal kidney function and positive in case of distal renal tubular acidosis.[2]

[UAG] might offer a relatively accurate approximation of [NH_4^+]. However, the association between [NH_4^+] in urine and [UAG] is disturbed in settings such as chronic renal failure, where the increased concentration of other unmeasured ions (sulphate and phosphate) creates discrepancies, making direct [NH_4^+] measurement in the urine seemingly necessary, in order to draw correct conclusions; nevertheless, these discrepancies may be overcome when urine sulphate and phosphate are accounted for in the [UAG] equation.[2]

The concept of changing the [H^+] in a body fluid compartment by removing H^+ from it contradicts Stewart's viewpoint.[3] According to Stewart, what determines [H^+] in a body fluid is mainly [SID], that is the charge space created by the concentration difference of the strong anions and cations in it. The movement of H^+/HCO_3^- in the aquatic environment of various compartments in the body cannot change their concentration; in fact, it does not really exist. Following Stewart's argument, when the [SID] in plasma/extracellular space ($[SID]_{ECV}$) decreases (metabolic acidosis), its gradual restoration to its original value (increase) is mediated by the kidney, that is by excretion of urine with [SID] ($[SID]_u$) $<[SID]_{ECV}$. Gattinoni et al[4] proposed a comprehensive model for the restoration of $[SID]_{ECV}$, utilizing Stewart's physicochemical approach. They suggest that a change in the net electrical charge in extracellular space can be restored by a corresponding change in the net electrical charge in urine, taking into account the volume of body fluid where this change takes place and the rate of urinary excretion. Based on this model, in case of an acidosis disorder, considering that the $[SID]_{ECV}$ is not affected by other modifiers (infusion of electrolyte solutions and metabolic disorders), each elementary part of plasma, which is filtered in the glomeruli and excreted by the kidneys as urine with lower [SID], will augment $[SID]_{ECV}$. Integration of these elementary changes over time will carry out the desired $[SID]_{ECV}$ increase:

$$\int_a^b d([SID]_{ECV(t)}\cdot V_{(t)}-[SID]_u\cdot V_{(t)})*$$

*Riemann-Stieltjes integral; a and b represent the corresponding time points; $V_{(t)}$ could be replaced by the urine production rate.

For a given urine production rate, the smaller the $[SID]_u$ the greater the difference, that is the correction (increase) of $[SID]_{ECV}$. In fact, if $[SID]_u$ becomes negative, the correction will be even greater, since the absolute value of $[SID]_u$ will be added to that of $[SID]_{ECV}$. This translates into enhanced effectiveness of renal function to restore acidosis.

It is important to understand that, in solutions of strong ions with weak electrolytes, when the [SID] is positive (eg normal [SID] value in the extracellular space and plasma), and for a [SID] range from zero to $[A_{tot}]$ (total concentration of weak, non-volatile acids), changes of $[H^+]$ are very small, only a minuscule percentage of the [SID] change (provided that $[A_{tot}]$ does not appreciably alter).[3] However, in solutions with a negative [SID] value, positively charged weak ions are needed to maintain electrical neutrality, and the only ones available are H^+. Therefore, the following equality holds: $[H^+] = -[SID]$. Thus, adding strong acid to a solution with negative [SID] will increase $[H^+]$ as much as [SID] decreases.

$[SID]_u$ is calculated by an equation identical to that of $[U_{AG}]$:

$$[SID]_u = [Na^+]_u + [K^+]_u - [Cl^-]_u$$

Urine acidity change is accomplished by an increase of urinary $[Cl^-]$, or of any strong anion concentration, that is not accompanied by a similar increase of urine $[Na^+]$, leading to a decrease of $[SID]_u$ (Figure 1).

From Stewart's point of view, it makes no sense to refer to $[SID]_u$ as indicative of the presence of some molecules that carry H^+ in the urine, for example, NH_4^+. As stated, $[SID]_u$ determines the extent of weak electrolyte and water dissociation in the excreted urine and therefore $[H^+]$ there. For Stewart, NH_4^+ does not function as a H^+ transporter or, at least, it could function as such as any molecule of H_3O^+ in urine would.[3]

Similarly, the commonly reported trade-off between strong ions and H^+ (in single-digit integer proportions) across membranes separating body fluids and the resulting stoichiometric $[H^+]$ changes on them is questioned. As Stewart notices, the [SID] buffer value for human plasma equals $\Delta[SID]/\Delta[H^+] = -6.9 \times 10^5$, meaning that [SID] should change by hundreds of thousands of Eq/L to produce a change of $[H^+]$ by 1 Eq/L.[3] Only in solutions with negative [SID], a linear relationship between [SID] and $[H^+]$ changes can be observed.

NH_4^+ is a weak cation (pKa = 9.24) that is formed from an add-on base, that is NH_3. NH_3 that dissolves in water combines with H^+ to form NH_4^+. The total concentration of NH_3 (NH_4^+) in urine, affecting urine pH, may be of particular importance for the functionality of the strong ion channels that are pH sensitive.[5]

Apart from the one mentioned above, if we wanted to allocate a role in the NH_4^+ in urine, unifying somehow the translational aspects that are presented, it would be that its presence there, spares the strong cations that, otherwise, would be excreted in urine, along with the strong acid anions (ie NH_4^+ in urine prevents the loss of strong cations). This, consequently, would positively affect the [SID] in plasma and, generally, the extracellular space.

Nevertheless, the transfer of H^+/NH_4^+ through membrane carriers and ion channels has been described in detail. An example is Na^+/H^+ exchanger 3 (NHE-3), an antiporter that is

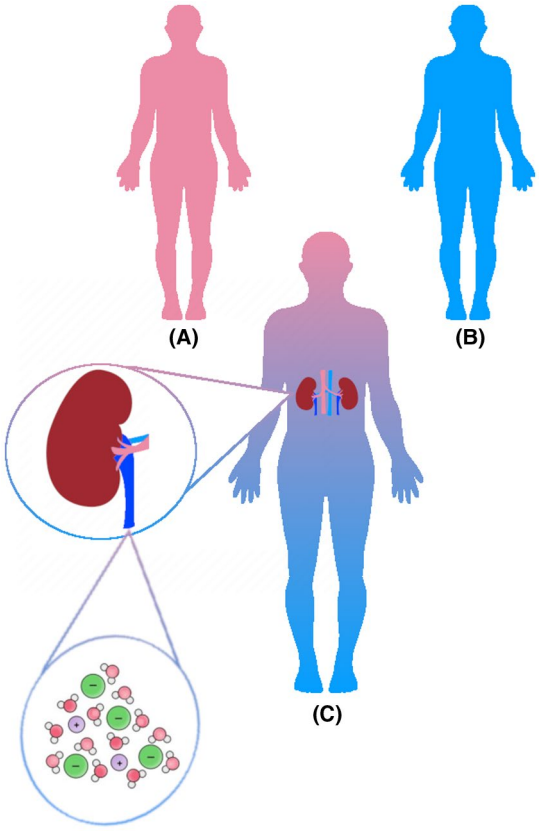

FIGURE 1 A, A patient in an acid-base equilibrium state is shown (colour pink). B, Acute metabolic acidosis caused by an anionic electrical load in the patient's extracellular fluid (colour blue), reducing SID_{ECF}. C, Renal compensation occurs with the excretion of strong anions, a process which normally leads to a low or even negative SID urinary condensate allowing for the gradual increase of SID_{ECF} towards normal levels and correction of metabolic acidosis (gradual restoration of pink colour). In the magnified image of the kidney, the darker blue of the ureter corresponds to the higher urine concentration of the negatively charged strong ions, compared to that in plasma; the elimination of strong anions will gradually correct the metabolic acidosis disorder. The renal artery (colour blue, ie corresponding to the acidic blood that reaches the kidney) and the renal vein (colour pink, ie corresponding to blood free from the strong anion excess that has been excreted in the urine) are also demonstrated. The magnified image of the ureter content shows the dispersion of strong ions among the water molecules; increased concentration of the negatively charged strong ions, for example, Cl^-, increases the dissociation of water and, thus, the proton concentration in urine, lowering pH

thought to mediate H^+ secretion across the apical membrane of epithelial cells in the proximal convoluted tubules.[6] However, investigators have questioned the ability of individual H^+ to participate in transport processes and biochemical reactions. For example, H^+ generation and transport during lactate production, in glycolysis.[7] A main reason the investigators invoke is the restricted mobility of H^+ in aqueous solutions.

Indeed, studies indicate that H^+ diffusion might be much more restricted than we previously thought. Their fleeting existence in water (their lifetime measured in picoseconds)[8] as well as their tiny concentration in the aqueous solutions of the body ($\approx 10^{-7}$M), which would make the conventional diffusion process extremely slow, raise reasonable doubts as to whether each individual H^+ could, actually, participate in transmembranic transport. To account for the observed pH changes, the proponents of the physicochemical approach provide a simple argument: strong ion transport across membranes changes [SID] in the relevant fluid compartments. The [SID] change automatically alters [H^+]/pH in the solution; this is misperceived as being created by the transfer of H^+

In fact, a unique transport mechanism, known as the Grotthuss mechanism (also referred to as structural diffusion) has long been proposed in place of H^+ movement.[9] According to this mechanism, H^+ transfer occurs in a stepwise manner in water or any chain of hydrogen-bonded molecules. Interconversion of cationic complexes and charge re-localization in the hydrogen bond network occur. Each hydrogen-bonded molecule acts simultaneously as a charge donor and acceptor; that is, when an excess H^+ is added to one end of the chain, the adjacent hydrogen bond in the chain releases another H^+. The charge distribution in the network is highly polarizable and may be greatly distorted by changing the magnitude of electrostatic forces.[10] Such a stimulus, for example, can be produced by the alteration of SID into a solution. In addition to the bulk water mass, a Grotthuss mechanism has also been proposed for the transfer of H^+ through ion channels.[9]

These physicochemical observations seem to support the Stewart's notion. On the whole, Stewart's approach may provide a more solid explanatory footing, as it is based on fundamental physicochemical principles (electrical neutrality, conservation of mass and Guldberg-Waage mass action law) claiming universal validity, and do not result from the individual interpretation of separate experimental data. Nevertheless, while both methods approach pathophysiologically the issue of metabolic acidosis from entirely different angles, there seems to be no clear benefit in clinical practice over one another when they are used correctly. All in all, whichever approach one chooses, the concomitant decrease of [SID]u/[UAG] values along with $[SID]_{ECV}$ in the setting of metabolic acidosis demonstrates proper renal response, while greater [SID]u/[UAG] values in the same setting signify impaired urine acidification.

CONFLICT OF INTEREST

None declared.

Emmanouil Alevrakis[1]
Nikolaos Gialelis[2]
Ioannis Vasileiadis[3]

[1]*4th Department of Respiratory Medicine, Sotiria Hospital, Athens, Greece*
[2]*Department of Mathematics, National and Kapodistrian University of Athens, Athens, Greece*
[3]*Intensive Care Unit, 1st Department of Respiratory Medicine, National and Kapodistrian University of Athens, Sotiria Hospital, Athens, Greece*

Correspondence
Ioannis Vasileiadis, Intensive Care Unit, 1st Department of Respiratory Medicine, National and Kapodistrian University of Athens, Sotiria Hospital, 152 Mesogion Ave, 115 27, Athens, Greece.
Email: ioannisvmed@yahoo.gr, isvasileiadis@med.uoa.gr

ORCID

Ioannis Vasileiadis https://orcid.org/0000-0002-9529-9361

REFERENCES

1. Weiner ID, Verlander JW. Renal ammonia metabolism and transport. *Compr Physiol*. 2013;3:201-220.
2. Raphael KL, Gilligan S, Ix JH. Urine anion gap to predict urine ammonium and related outcomes in kidney disease. *Clin J Am Soc Nephrol*. 2018;13:205-212.
3. Stewart PA. *Stewart's Textbook of Acid-Base*, 2nd ed. In: Kellum JA, Elbers PWG, Egham, UK: Lulu Enterprises Ltd; 2009;31-179.
4. Gattinoni L, Carlesso E, Cadringher P, Caironi P. Strong ion difference in urine: new perspectives in acid-base assessment. *Crit Care*. 2006;10:137.
5. Dennis Brown D, Wagner CA. Molecular mechanisms of acid-base sensing by the kidney. *J Am Soc Nephrol*. 2012;23:774-780.
6. Soleimani M, Rastegar A. Pathophysiology of renal tubular acidosis: core curriculum 2016. *Am J Kidney Dis*. 2016;68:488-498.
7. Lindinger MI, Kowalchuk JM, Heigenhauser GJF. Applying physicochemical principles to skeletal muscle acid-base status. *Am J Physiol Regul Integr Comp Physiol*. 2005;289:R891-894.
8. Agmon N, Bakker HJ, Campen RK, et al. Protons and hydroxide ions in aqueous systems. *Chem Rev*. 2016;116:7642-7672.
9. Wraight CA. Chance and design-Proton transfer in water, channels and bioenergetic proteins. *Biochim Biophys Acta*. 2006;1757:886-912.
10. Zundel G. Hydrogen bonds with large proton polarizability and proton transfer processes in electrochemistry and biology. *Adv Chem Phys*. 2000;111:1-217.

How to cite this article: Alevrakis E, Gialelis N, Vasileiadis I. Strong ion difference in urine: A measure of proton excretion or of the net plasma charge alteration?. *Acta Physiol.*2020;230:e13559. https://doi.org/10.1111/apha.13559