The effect of sodium bicarbonate supplementation on the decline in gross efficiency during a 2000-m cycling time trial

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Original Investigation

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Abstract
Gross efficiency (GE) declines during high-intensity exercise. Increasing the extracellular buffer capacity might diminish the decline in GE, and thereby improve performance. **Purpose** To examine if sodium bicarbonate (NaHCO₃) supplementation diminishes the decline in GE experienced during a 2000-m cycling time trial. **Methods** Sixteen male cyclists and sixteen female cyclists completed four testing sessions, including a maximal incremental test, a familiarization trial and two 2000-m GE tests. The 2000-m GE tests were performed after ingestion of either NaHCO₃ supplements (0.3 g/kg body mass) or placebo supplements (amylopectin, magnesium stearate, and sunflower oil capsules). The GE tests were conducted using a double-blind, randomized, crossover design. Power output, gas exchange and time to complete the 2000-m time trials were recorded. Capillary blood samples were analyzed for blood HCO₃⁻, pH, and lactate concentration ([La⁻]). Data were analyzed using the magnitude-based inference approach. **Results** The decrement in GE found after the 2000-m time trial was possibly smaller within the male and female groups following NaHCO₃ compared to placebo ingestion, with the effect in both groups combined being unclear. The effect on performance was likely trivial for males (placebo 164.2±5.0 s, NaHCO₃ 164.3±5.0 s, Δ0.1; ±0.6%) and unclear for females (placebo 178.6±4.8 s, NaHCO₃ 178.0±4.3 s, Δ-0.3; ±0.5%), and very likely trivial when effects were combined. Blood HCO₃⁻, pH, and [La⁻] were substantially elevated from rest to pretest following NaHCO₃ ingestion. **Conclusions** NaHCO₃ supplementation results in an unclear effect on the decrease in GE during high-intensity exercise and in a very likely trivial effect on performance.

**Keywords:** performance, fatigue, alkalosis, extracellular buffering, economy
Introduction

The performance oxygen uptake ($\dot{V}O_2$), determined by the maximal oxygen uptake ($\dot{V}O_{2\text{max}}$) and $\dot{V}O_2$ at the lactate threshold, the performance $O_2$ deficit (i.e., anaerobic capacity) and gross mechanical efficiency are the main physiological performance-determining variables.\(^1\) As it has been shown that $\dot{V}O_{2\text{max}}$, remains relatively stable during an athletic career in elite endurance athletes, but that exercise economy or efficiency improved substantially during the same time frame,\(^2\) it seems that reducing submaximal oxygen consumption is necessary to achieve performance improvements. Therefore, it would be desirable to know which variables positively influence exercise economy and/or efficiency.

One of the variables that seems to negatively influence exercise efficiency is fatigue.\(^3\) Gross efficiency (GE), the most valid definition of exercise efficiency,\(^4\) has been shown to decline during prolonged submaximal and (supra)maximal exercise.\(^5\)–\(^9\) If GE declines, more metabolic energy is required to maintain a certain mechanical power output (PO). As it has been stated by Grassi et al.\(^3\) that “the energy yield of the muscle system is by definition limited, the rate of progression of this inefficiency is a major determinant of task failure”, it is of interest to know the underlying cause of fatigue and the reduced GE after prolonged submaximal or (supra)maximal exercise.

The underlying cause of the reduced GE after prolonged submaximal or (supra)maximal exercise seems to be related to the intensity at which the exercise bout is performed, as Noordhof et al.\(^5\) showed that the decrement in GE was largest after relatively shorter time trials. During shorter time trials the mechanical PO is higher, resulting in greater homeostatic disturbances, which could be the cause of the larger decrement in GE after the shorter time trials. The lower running economy found by Hoff et al.\(^10\) when blood lactate concentration ([La\(^{-}\)]) increased from 3 to 5 mmol/L, supports this notion.

During high-intensity exercise there is a significant energy contribution from anaerobic glycolysis.\(^11\) Anaerobic energy production results in the accumulation of several metabolic byproducts, such as hydrogen cations (H\(^{+}\)) and lactate anions (La\(^{-}\)).\(^11,12\) La\(^{-}\) can either be removed by oxidation within the muscle fiber, which increases $\dot{V}O_2$, or it can be released to the blood and removed by other cells.\(^12\) Although the role of acidification in muscle fatigue is still not completely unraveled,\(^12\) it seems quite clear that a high demand on the anaerobic glycolytic energy system is associated with fatigue\(^3\) and a reduction in economy/efficiency.\(^10\) However, it remains to be elucidated if the two (i.e., acidification and inefficiency) are causally related.

The ability of the body to prevent or delay the onset of muscle fatigue due to acidification depends, among others, on the capacity of its buffering systems. During exercise, the acid-base balance (pH) in the working muscles is regulated by intracellular, extracellular and dynamic buffering systems.\(^13\) Bicarbonate (HCO\(_3^-\)) is an extracellular buffer that plays an important role in maintaining extracellular and intracellular pH. The blood [HCO\(_3^-\)], and thus the extracellular buffering capacity, can be increased by ingesting sodium bicarbonate (NaHCO\(_3\)).\(^13\) Due to the extracellular buffering of H\(^{+}\) the H\(^{+}\)/La\(^{-}\) efflux from exercising muscle fibers is stimulated.\(^14\) Ingestion of NaHCO\(_3\) increases the rate at which H\(^{+}\) and La\(^{-}\) can be removed from working muscles during high-intensity exercise, contributing to intramuscular pH maintenance. A meta-analysis from Carr et al.\(^15\) revealed an acute performance enhancement of 1.7% ($\pm$ 95% confidence limit (CL), 2.0%) in male athletes during a single 1-min sprint, when NaHCO\(_3\) was ingested in a dose of 0.3 g/kg body mass prior to exercise. Also, in slightly longer events, such as a 4000-m cycling time trial ergogenic effects of NaHCO\(_3\) have been found.\(^16,17\) So, although it seems clear that a moderate performance benefit of NaHCO\(_3\) ingestion can be expected, it is unclear if NaHCO\(_3\) supplementation diminishes the decrement in GE during time-trial exercise. Therefore, the current study was designed to examine if NaHCO\(_3\) supplementation diminishes the decline in GE experienced during a 2000-m cycling time trial. A 2000-m time trial was chosen, as previous research showed that GE
substantially declines during this event, and because it is expected that the effect of NaHCO₃ supplementation will be larger on a 2000-m time trial, compared to a 4000-m time trial, which is an official event in track cycling. We expected NaHCO₃ ingestion would increase blood [HCO₃⁻] and blood pH and thereby reduces the decrement in GE attained during time-trial exercise. The meta-analysis of Carr et al. also showed that the effect of NaHCO₃ ingestion on mean power was smaller for females than for males, although the difference in the effect between males and females was found to be unclear. As there are currently no studies that investigated possible differences in the effect of NaHCO₃ supplementation between sexes, it is of interest to study the effect of NaHCO₃ supplementation on the decline in GE between male and female cyclists. Therefore, the secondary aim of the current study was to investigate if the effect NaHCO₃ supplementation on the decline in GE differs between male and female cyclists.

**Methods**

**Subjects**

Sixteen trained male competitive cyclists (mean ± standard deviation (SD): age 27.6 ± 6.9 y, training volume 7.0 ± 2.7 h/wk, VO₂max 61.8 ± 4.3 ml·kg⁻¹·min⁻¹), and sixteen trained female competitive cyclists (age 26.3 ± 6.0 y, training volume 5.5 ± 3.2 h/wk, VO₂max 52.3 ± 2.4 ml·kg⁻¹·min⁻¹) participated in this study. Inclusion criteria were: 1) age between 18 and 45 years, 2) experience with cycling time trials, and 3) a low risk profile based on a health-history form. Subjects were excluded if they used intra- or extracellular buffers in the form of supplements (creatine monohydrate, β-alanine or sodium bicarbonate) during the three months preceding the study. Subjects were instructed to avoid strenuous exercise and alcohol consumption during the 24 h before each test and were asked to consume their last meal at least 3 h prior to each test. Subjects were fully informed about the nature and potential discomforts associated with the study, before providing written informed consent. The study was approved by the local ethics committee.

**Experimental design**

Subjects visited the laboratory on four separate occasions. During the first occasion subjects completed a maximal incremental exercise test. After at least 24 h, subjects completed a familiarization trial to become acquainted with the experimental protocol of the GE test, and to minimize the learning effect on pacing strategy. In addition, the familiarization trial gave the subjects the opportunity to select the best gear ratio. Instructions on nutritional intake and exercise the day before the test also applied to the familiarization trial. The remaining two visits to the laboratory consisted of completing a GE test after either NaHCO₃ supplementation (0.3 g/kg body mass (BM), Virtuoos Com B.V., Amsterdam, Netherlands, packed into HPMC capsules) or ingestion of placebo supplements (amylum solani, magnesium stearate, sunflower oil, Virtuoos Com B.V., Amsterdam, Netherlands, packed into HPMC capsules) 150 min prior to the start of the time trial. Based on their results, Carr et al. concluded that the ingestion of NaHCO₃, in a dose of 0.3 g/kg BM, should commence 120-150 min before the start of exercise, which is why the above described dose and timing were chosen. In addition, it has been shown that the delivery method HPMC (i.e. delayed-release) capsules vs. an aqueous solution also elongated the time to peak pH and peak HCO₃ to about 120-130 min after ingestion. The GE tests were conducted using a double-blind, randomized, crossover design. Previous research reported that large variations in dietary intake prior to a test can influence acid-base status, pre-exercise muscle buffering capacity, and consequently subsequent exercise performance. Therefore, subjects were instructed to report their dietary intake in the 24-h preceding the first GE test, and to repeat this before the subsequent test. The minimal time required between the two tests was 48 h, in order to ensure adequate NaHCO₃ washout and to prevent fatigue from the previous GE test. Both time trials were performed at the same time of the day (± 1 h).
All testing sessions were performed in a climate-controlled room (18.0 ± 1.2°C, relative humidity 44.9 ± 7.2%), on a custom-made, electronically braked cycle ergometer (VU-MTO, Amsterdam, Netherlands). The optimal saddle and handle-bar height were determined before the first test and were replicated during subsequent tests. Torque, pedaling frequency, and PO data of the cycle ergometer were sampled at 100 Hz. During the maximal incremental exercise test and the submaximal exercise bouts performed before and after the 2000-m time trial, subjects received visual feedback about their pedaling frequency and elapsed time. During the time trial, subjects only received feedback about the distance covered.

Maximal incremental exercise test
The maximal incremental exercise test was conducted to determine the PO at which subjects reached their maximal oxygen uptake (\(\text{VO}_{2\max}\), \(\text{PVO}_{2\max}\)). The same protocol was used as in previous research.5 The test was ended when pedaling frequency dropped below 80 revolutions per minute (rpm), despite strong verbal encouragement.

Gross efficiency tests
The same protocol is used as in previous research (see Figure 1).5 Subjects had to maintain a pedaling frequency of 90 rpm during the submaximal bouts of the test. During the time trial subjects were instructed to complete the TT as quickly as possible. Subjects ingested either \(\text{NaHCO}_3\) capsules or color-matched placebo capsules 150 min before the start of the time-trial part of the GE tests. The supplements were co-ingested with a meal containing 1.5 g carbohydrate/kg BM and 7 ml/kg BM of fluid, which optimizes blood alkalosis and diminishes the incidence of gastrointestinal (GI) symptoms.22 Before ingestion of the meal and the supplements, subjects completed a validated GI-distress questionnaire.26 The same questionnaire was completed 60 min post ingestion, 5 min prior to the start of the GE test, and 5 min after completion of the test. Capillary blood samples were collected before ingestion of the meal and the supplement (150 min before the start of the time trial, rest), before the start of the GE test (pre-test), before the start of the time trial (pre-TT), and immediately after completion of the time trial (post-TT; see Figure 1).

Data collection and analysis
During all tests, gas exchange data were collected breath-by-breath using open-circuit spirometry (Quark CPET, Cosmed S.R.L., Rome, Italy). Prior to each test the gas analyzer was calibrated according to the manufacturer’s instructions. Heart rate was measured during the entire test using a Garmin heart rate monitor (Soft Strap Premium Heart Rate Monitor, Garmin, Eemnes, The Netherlands).

Breath-by-breath respiratory data were converted to second-by-second data using interpolation. Subsequently, the second-by-second respiratory data was smoothed with a 6-s moving average filter. Values deviating more than two standard deviations from the local mean, were replaced by the local mean. \(\text{VO}_{2\max}\) and maximal heart rate (HR\(_{\max}\)) were defined as the highest \(\text{VO}_2\) and heart rate over a 30-s moving average. Mean \(\text{VO}_2\) and RER values were determined over the dark shaded areas in Figure 1 in order to calculate GE (GE\(_{\text{pre}}\), GE\(_{\text{post1}}\), and GE\(_{\text{post2}}\), respectively).27 In order to calculate GE, the mean RER had to be ≤ 1.0 and \(\text{VO}_2\) needed to be in steady state.5 When these two criteria were not met, corresponding GE values were removed from further analysis. Back-extrapolation was used to determine GE at the end of the 2000-m time trial (GE\(_{\text{extrap}}\)).5

During both GE tests capillary blood samples were collected to measure blood pH, [HCO\(_3^-\)] and [La\(^+\)]. After cleaning the fingertip with alcohol, the fingertip was pierced with a sterile 2.25 mm retracted lancet (Hemocue Safety Lancets, Angelholm, Sweden). After removing the first drop of blood, 100 µL of blood was collected in a lithium heparine coated
minivette (Minivette POCT, Sarstedt, Numbrecht, Germany). Blood samples were immediately analyzed after collection using a blood gas analyzer (i-STAT portable analyzer, Abbott Point of Care, Illinios, USA). Previous measurements with the i-STAT analyzer were found to be reliable and accurate.\textsuperscript{28} To analyze blood pH and [HCO\textsubscript{3}\textsuperscript{-}] EC8+ cartridges (Abbot Point of Care, Hoofddorp, The Netherlands) were used. [La\textsuperscript{+}] was analyzed using the Lactate Pro 2 (Arkray, Kyoto, Japan).

**Statistical analysis**

Data were analyzed using the magnitude-based inference approach. Before analysis, data were log-transformed. Data are therefore reported as back-transformed means and the standard deviation (SD) is therefore expressed as a coefficient of variation (%).\textsuperscript{29} The effect of high-intensity exercise on GE was tested using a post-only spreadsheet.\textsuperscript{30} The effect of NaHCO\textsubscript{3} supplementation on the decrement in GE during time-trial exercise, performance, and blood variables was assessed using a pre-post crossover spreadsheet.\textsuperscript{30} The difference in effect between male and female cyclists was assessed using the combined groups spreadsheet.\textsuperscript{31} The magnitude of the difference in 2000-m performance time was assessed using the smallest worthwhile change obtained from Flyger\textsuperscript{32} (0.5%). Magnitude of differences in GE and blood variables were determined by standardization with the SD of the placebo trial. Magnitudes of differences were interpreted based on the following scale: 0.20, small effect; 0.60, moderate effect; 1.20, large effect; 2.0, very large effect; 4.0, extremely large effect.\textsuperscript{29} Data that represent the differences between conditions are reported as mean effect ±90% confidence limit (CL). Probabilities that an effect is negative, trivial, or positive was based on the following scale: <0.5%, most unlikely; 0.5% to 5%, very unlikely; 5% to 25%, unlikely; 25% to 75%, possibly; 75% to 95%, likely; 95% to 99.5%, very likely; >99.5%, most likely. When the chance of being both beneficial and harmful was >5%, the effect was considered unclear. The clinical inference was reported, except for the difference in effect between male and female cyclists, in that case the non-clinical inference was reported.

**Results**

**Gross efficiency**

Mean GE data of the NaHCO\textsubscript{3} trial and the placebo trial (men and women combined) are shown in Figure 2. Due to missing GE\textsubscript{pre} values of two males and four females, GE data of 27 subjects were analyzed. The effect of NaHCO\textsubscript{3} supplementation on the change in GE during the time trial (GE\textsubscript{extrap} – GE\textsubscript{pre}) was considered possibly beneficial in both males and females (males 0.7; ±1.8%; females 0.6; ±2.1%; i.e. a possibly smaller decline in GE). The difference in the effect of NaHCO\textsubscript{3} between males and females was unclear.

**2000-m performance**

Data of 32 subjects, 16 males and 16 females, were used for analysis. The effect of NaHCO\textsubscript{3} supplementation on performance times was likely trivial for males (placebo trial 164.2 ± 5.0 s, NaHCO\textsubscript{3} 164.3 ± 5.0 s, NaHCO\textsubscript{3} – placebo 0.1; ±0.6%) and unclear for females (placebo trial 178.6 ± 4.8 s, NaHCO\textsubscript{3} 178.0 ± 4.3 s, difference -0.3; ±0.5%). The difference in the effect between males and females was considered possibly trivial. When the effect of NaHCO\textsubscript{3} supplementation on performance times for males and females were combined a very likely trivial effect of NaHCO\textsubscript{3} supplementation was found.

**Blood analysis**

Mean [HCO\textsubscript{3}\textsuperscript{-}], blood pH, and [La\textsuperscript{+}] during each treatment condition are shown in Figure 3. The effect of NaHCO\textsubscript{3} supplementation compared to placebo supplementation on the difference in blood [HCO\textsubscript{3}\textsuperscript{-}] between rest and pre-test differed between men and women, with
men showing a likely larger increase compared to women during the NaHCO₃ trial compared to the placebo trial. However, the effect of NaHCO₃ supplementation on the difference in blood [HCO₃⁻] between rest and pre-test was most likely beneficial in both groups. There were no other substantial differences between male and female cyclists in blood concentrations, and therefore the effect of NaHCO₃ supplementation compared to placebo supplementation of both groups were combined and summarized in Figure 3.

Discussion

The purpose of the current study was to examine if NaHCO₃ supplementation diminishes the decline in GE experienced during a 2000-m cycling time trial. The main finding of this study was that ingestion of NaHCO₃ resulted in a small but possible beneficial effect on the decrement in GE found after a 2000-m time trial in males and females. However, when the effects of males and females were combined an unclear effect was found. The effect of NaHCO₃ on performance time was likely trivial for males and unclear for females, with the difference in effect between males and females being considered possibly trivial and the combined effect being very likely trivial.

GE declined substantially during both the NaHCO₃ and the placebo trial. The substantial decline in GE during the placebo trial is supported by the findings of Noordhof et al.⁵ and Groot et al.¹⁸ The present study revealed that the decline in GE found after a 2000-m time trial was possibly diminished after NaHCO₃ ingestion, which might be explained by the increased pH and [HCO₃⁻]. Several studies have reported that acidosis reduces the efficiency of oxidative phosphorylation.³³,³⁴ Walsh et al.³⁵ showed that a lower oxidative flux was produced with a muscle pH of 6.6 compared to a muscle pH of 7.0, which might explain the smaller reduction in GE at the end of the 2000-m time trial in the NaHCO₃ condition when average blood pH values were 7.28 compared to the placebo condition when average blood pH values were 7.20. However, when the effects of NaHCO₃ supplementation on the decline in GE for males and females were combined, an unclear effect was found, suggesting that more data is needed to come to a final conclusion. It is clear that ingestion of NaHCO₃ prior to time-trial exercise could not prevent the decrement in GE, even though, NaHCO₃ ingestion resulted in a very likely larger decline in blood [HCO₃⁻] during the time trial, suggesting that more H⁺ was buffered, and therefore the H⁺/La⁻ efflux from the exercising muscles was stimulated more than during the placebo trial. So, it seems that other factors besides muscle acidosis might play a role in the decrement in GE during high-intensity exercise. Hoff et al.¹⁰ concluded that increased [La⁻] deteriorates running economy, but increased La⁻ levels and decreased pH are not per se the cause of this decrement. On top of that, muscle acidification is being questioned as the major reason of muscle fatigue.¹²

A meta-analysis from Carr et al.¹⁵ revealed that the effect of NaHCO₃ ingestion on mean power was smaller for females than for males, although the difference in the effect between males and females was found to be unclear. As to date there were no studies in which the effect of NaHCO₃ supplementation on performance was compared between males and females, the current study included both male and female subjects to analyze the differences in the response to NaHCO₃ supplementation. NaHCO₃ ingestion before the 2000-m time trial resulted in an unclear effect on performance in females, with a 1% chance on a harmful effect, a 72% chance on a trivial effect, and a 27% chance on a beneficial effect. The effect of NaHCO₃ ingestion on 2000-m time trial performance in males was likely trivial and when the effects for males and females were combined a very likely trivial effect was found. The lack of a substantial performance benefit in both males and females (and combined) is in agreement with previous studies using exercise protocols of similar or longer duration, but is in contrast with others using exercise protocols of shorter duration.¹⁵ suggesting that NaHCO₃ ingestion might be effective in time trials shorter than 2000 m, such as the 1000-m individual time trial for male
track cyclists and the 500-m time trial for female track cyclists, in which a similar decline in GE can be expected as in the current study.\(^6\) However, recent studies also found a performance benefit of NaHCO\(_3\) ingestion during 4000-m cycling time trials performed by trained male cyclists.\(^{16,17}\) The cause of the difference in effect might be due to the individual variation in time to peak alkalosis.\(^6\) Gough et al.\(^{16,17}\) used an individualized supplementation strategy, in which NaHCO\(_3\) supplements are ingested at a time point which results in peak pH and/or HCO\(_3^-\) concentrations at the start of the time trial for each individual. In the current study an individualized supplementation strategy was not used, which possibly limited the ergogenic effect on cycling performance in the current study. An increase in HCO\(_3^-\) of +6 mmol/L from rest to pretest was obtained in thirteen of the sixteen males and twelve of the fifteen females (of one female blood data was missing), which has been suggested to be a threshold elevation that is necessary before ergogenic effects will be found.\(^{36}\) However, it has also been shown that an almost identical increase in HCO\(_3^-\) did not necessarily result in the same effect on performance.\(^{37}\) So, an individualized supplementation strategy instead of standardized supplementation could have resulted in a substantial performance effect, although this remains to be investigated, as to date no study directly compared the effect on performance of both supplementation strategies.\(^{36}\)

NaHCO\(_3\) ingestion can cause GI discomfort which could interfere with performance.\(^{13}\) To minimize the chance on GI discomfort, we chose to co-ingest the supplements with a meal containing 1.5 g carbohydrate/kg BM and 7 ml/kg BM of fluid, as previous research showed that this optimizes blood alkalosis and diminishes the incidence of gastrointestinal (GI) symptoms.\(^{22}\) Two of the subjects in this study reported one of the GI symptoms to be $\geq 5$, however they were able to complete the time-trial. One subject needed to be excluded from the study because of GI side-effects. It is therefore expected that GI discomfort had a minimal effect on the results of the current study.

**Practical applications**

The current study showed that NaHCO\(_3\) ingestion prior to high-intensity exercise has an unclear effect on the decrement in GE and a very likely trivial effect on performance. Based on the current results, we would therefore not recommend trained male and female cyclists to use NaHCO\(_3\) supplementation during time trial competitions of around 2000 m (~164 s).

**Conclusions**

NaHCO\(_3\) ingestion (dose 0.3 g/kg BM) 150 minutes before a 2000-m time trial results in an unclear effect on the decline in GE, when the effects of NaHCO\(_3\) ingestion on GE for male and female cyclists are combined. The difference in effects between male and female cyclists was also considered unclear. Moreover, the effect of NaHCO\(_3\) supplementation on 2000-m time trial performance was very likely trivial. It seems that other factors besides muscle acidosis are involved in the decrement in GE during high-intensity exercise.
References


Figures

Figure 1 — Experimental protocol of the gross efficiency (GE) tests. GE was determined during the dark shaded areas. The light shaded area represents the time trial of 2000 m. Capillary blood samples were collected before ingestion [rest], before the start of the test [pre-test], before the start of the time trial [pre-TT], and directly following the time trial [post-TT]. The figure is adapted from Noordhof et al.5 Abbreviations: $\dot{V}O_2^{\text{max}}$, PO at which $\dot{V}O_2^{\text{max}}$ was attained; ft, finish time; TT, time trial.

Figure 2 — Means of raw data ± SD of gross efficiency (GE) before and after the 2000-m time trial during the NaHCO$_3$ trial (solid line) and the placebo trial (dotted line). The magnitude of the effects shown, are the combined effects (males and females combined) of NaHCO$_3$ supplementation compared to placebo supplementation on GE and are displayed using the following signs: $\Delta$ unclear; * possibly trivial, ** likely trivial. Pre, before the time trial; extrap, immediately after the time trial; post1, 4-7 minutes after the time trial; post 2, 7:30-10:30 after the time trial (see Figure 1).

Figure 3 — Means of raw data ± SD of blood [HCO$_3^-$] (A), pH (B), and [La$^-$] (C) measured after ingestion of either NaHCO$_3$ supplements (solid line) or placebo supplements (dotted line). The magnitude of the effects shown, are the combined effects (males and females combined) of NaHCO$_3$ supplementation on blood [HCO$_3^-$] (A), pH (B), and [La$^-$] (C), and are displayed using the following signs: $\Delta$ unclear; * possibly trivial, +++ most likely beneficial; † possibly harmful, †† likely harmful, ††† very likely harmful.