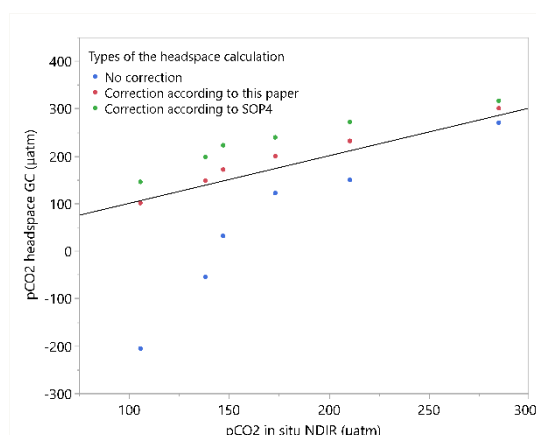


Anonymous Referee #1

The importance of this paper is to point out a major problem in the processing of CO₂ measurements made by headspace equilibration due to the equilibration of CO₂ with HCO₃⁻ to the wider community working on CO₂ dynamics in freshwater. This has been known for decades by the marine CO₂ community, related to the buffering capacity of water due to the presence of HCO₃⁻ that in fact strongly affects all aspects of CO₂ dynamics in marine and freshwater environments. This problem was possibly less acknowledged by the freshwater CO₂ community due to the dominance of soft-water lakes in northern North America and Scandinavia where the very large majority of studies of inland water CO₂ studies have been carried out so far. That said, the authors reinvent the wheel by proposing a “tool for exact CO₂ calculation” because the marine CO₂ community has established for decades a method to correct the CO₂ data from measurements of headspace. This is the SOP N°4 (“Determination of pCO₂ in air that is in equilibrium with a discrete sample of sea”) of the two versions of the “CO₂ Handbook” (DOE 1994; Dickson et al. 2007). This method can be also applied to the type of data reported by the authors by computing DIC from TA and pCO₂_After_eq, correcting DIC for the CO₂ loss or gain during equilibration in the headspace (based on pCO₂_After_eq and pCO₂_Before_eq and using the law of perfect gases), and re-computing “correct” pCO₂ (pCO₂_water) from TA and corrected DIC.

The SOP4 method also allows to correct for water temperature changes between in-situ water and water sample after equilibration. This change of temperature can be substantial (depending on the difference between air temperature and in-situ water temperature) and will lead to a strong bias of pCO₂ values. For the first step of the computation of DIC the water temperature of sample after equilibration is used. For the final step of the computation of pCO₂ (from corrected DIC) the in-situ temperature is used giving corrected pCO₂ at in-situ temperature. I suggest that the authors should mention SOP4 in the ms and compare both “tools”.

As stated in the introduction, we are aware that carbon speciation is routinely considered in the marine literature and have cited the widely used SOP no.4 in our manuscript. We have highlighted this in the revised manuscript with the sentence “A procedure to correct headspace CO₂ data using pH and alkalinity is already available in the SOP N°4 in Dickson et. al. (2007) for marine samples and could be adapted to freshwater samples as well. For convenience, we provide here a modified procedure when the alkalinity of the sample is known by introducing an analytical solution to the equilibrium problem to facilitate calculations (iterative in SOP no. 4) and by using dissociation constants that may be more appropriate to freshwaters.” We directly compared our correction with SOP no4 and got very similar values when the whole range of samples was considered. We feel such comparison actually tackles the more difficult question of whether freshwaters can be assimilated to very dilute seawater and therefore justify the use of dissociation constant formulas that were



determined by sequential dilutions of seawater. For example, when we compared the SOP no.4 method with our procedure on the system where accounting for the shifting equilibrium was most needed (highly undersaturated system using ambient air as headspace), we found that the NDIR values were closer to our estimation than to the SOP no.4, essentially because of the difference in dissociation constants used (see figure). While the question of the most adequate dissociation constants for freshwaters is definitely relevant, it is considerably beyond the

scope of this technical note. Our main purpose here is instead to alert the freshwater community of the importance of accounting for the shift in chemical equilibrium during headspace equilibration and under what circumstances it is most problematic. However, the reviewer's comments have prompted us to better articulate this main message and we have now included a completely new section directly addressing the question of how relevant it is to the limnological community. To this end, we have compared the results of pCO₂ determinations with and without correction from a large dataset of 377 lakes from across Canada for which we had complete ancillary data and precise headspace measurements of CO₂ (<5% error between duplicates). These results show that ignoring the correction would have resulted errors >20% in about 50% of the data. This new paragraph is now towards the end of section 3.4.

Regarding the effect of equilibration temperature vs in situ temperature, it was already included in our procedure and also considered in all our data but we had failed to mention it. After Equ 1 we write " $K_{h \text{ Eq}}$ and $K_{h \text{ Sample}}$ = gas solubility at the equilibration temperature and at the sampling temperature". We also added a figure to the appendix which shows the sensitivity of the results towards errors in the equilibration temperature and address the importance of using the correct temperature by writing "However, care must be taken to make sure that the exact equilibration temperature is known. For example, an error of 1°C in the equilibration temperature results in a 2 % different pCO₂ value (TA=1 mmol L⁻¹, pCO₂ = 1000 µatm, HR = 1) (Figure A1a)".

Finally, I find it regretful that the authors did not reach out to the community for additional data-sets that would have made their case more compelling by extending the range of pCO₂ and Total alkalinity values, and thus more representative of lakes glob-ally. Several groups have obtained similar data-sets of direct pCO₂ measurements by equilibration coupled to NDIR detectors in parallel with pCO₂ measurements based headspace equilibration, and could have been contacted.

The reviewer is right and we agree that Figure 4 can benefit from more data expanding the geographical coverage and range of water chemistry. To this end, we contacted a number of colleagues and have added data from 3 reservoirs and 3 streams in Germany as well as from a Malaysian reservoir. Thus, our dataset now contains 266 observations a variety of systems types in different continents covering a large range in pH and alkalinity. We added to the method section: "We sampled water in 4 reservoirs and 3 streams in Germany, 10 Canadian lakes, and a Malaysian reservoir exhibiting a wide range of TA between 0.03 and 2.4 meq L⁻¹ and pH between 5.2 and 9.8.". As written above we also applied our correction to a large dataset of 377 lakes from across Canada to demonstrate the need for proper correction.

References

Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.) 2007. Guide to best practices for ocean CO₂ measurements. PICES Special Publication 3, 191 pp. DOE. 1994. Handbook of methods for the analysis of the various parameters of the carbon dioxide system in sea water; version 2, A.G. Dickson and C. Goyet, Eds. ORNL/CDIAC-74

Anonymous Referee #2

The authors describe a method to correct pCO₂ measurements using the headspace method with discrete water samples for the effect of the CO₂ equilibrium in freshwater samples. They compare different approaches of the headspace technique with measurements using a membrane equilibrator directly in the field. I mostly agree with the comments from Reviewer #1 that most of the work is described in Dickson et al (2007) and I will not repeat the arguments here. I understand that the authors want to use this technical note to raise awareness in the community. I can't judge if it is worth to publish an article repeating knowledge that is well documented (as the authors state

themselves), or if there are better ways to raise the awareness. But I assume a technical note might be the right way. The authors talk a lot about errors in %. As an outsider of the fresh water community, I was missing a short introduction into the field given the aimed accuracies/precisions of pCO₂ measurements. Also the analytical errors of their methods are not stated. This makes it hard to evaluate the benefit of any correction. An error of 100% means to me that the measurement is not very useful, but I might be wrong here.

We added information about analytical errors at the end of the method section: "Analysis of certified calibration gases showed that the analytical error of both the NDIR instrument and GC was <0.37% at 1000 ppm. Analysis of 7 replicate samples by our GC-headspace method gave a standard deviation of 6%. This includes all random errors due to sampling, sample handling and analysis." We now also assess the error resulting from wrong equilibration temperature by writing "For example an error of 1°C in the equilibration temperature results in a 2% different pCO₂ value (TA=1 meq L⁻¹, pCO₂=1000 μatm, HR = 1).".

As reviewer #1 said, the manuscript would benefit of comparing the two correction methods, the one described here with the one presented in Dickson et al.(2007).

We have already addressed the comparison between SOP4 in our response to reviewer #1.

More specific comments:

I. 41: a space is missing between UNESCO/IHA,2010; and Cawley...)

corrected

I. 58: dissolved

corrected

I. 61: alkalinity (TA); TA is already introduced here, so it can be skipped in line 69

corrected

I.74: What is stable? Can you give the range?

We added "fluctuating ± 5 ppm around the mean".

I.87: Is the equation correct? The units do not cancel out to a pressure.

Yes - the equation is correct. Confusion with the units arose because of the misleading notation of pCO₂ which in fact is a dimensionless mole fraction. This is corrected in the equation and text.

I.88: pCO₂ should be given in pressure units not in ppm

Since gas chromatograph results are typically reported as mixing ratios we would like to keep ppm here. Instead, we renamed pCO₂ to mCO₂ as explained above.

I.92: What do you mean with the "two methods"? NDIR vs. headspace method?

Yes. We changed the sentence to "The difference between headspace and NDIR method...".

I.101: Please give a range and not only "quite".

We specified to "below 10%".

I.107: what is acceptable?

Good question. What is acceptable depends on the precision wanted and thus can be different for different studies. We removed the “acceptable” and replaced it by a more detailed description of the error: “While the fit between the simple headspace calculation and NDIR values over the whole range of values can be considered adequate overall (Figure 2a, $R^2 = 0.92$), it is clear that the deviations can become very large (up to about 300%), particularly at water $p\text{CO}_2$ values $<600\mu\text{atm}$ (Figure 2b). As expected from the simulations, the error in undersaturated samples is positive when using CO_2 -free gas as headspace and negative (sometimes impossibly so) using ambient air (Figure 2b).”

I.116ff and Fig. 3: I don't get the text and the Figure together. Do you mean “using a smaller headspace”? When I look at Fig. 3b at 20°C and $\text{HR}=1$ and move to $\text{HR}=5$ the error increases to 300%.

Correct and thank you for pointing out this error. As written in the manuscript any measure reducing the gas exchange between water and headspace should reduce the error – and with a smaller headspace less gas is exchanged. We changed the sentence to: “In high alkalinity samples, the error can be significantly reduced by using a smaller headspace to water ratio (Figure 3). By lowering the headspace ratio from 1 to 0.25 at 20° the error can be reduced from about 50% to about 10%.”

reference

Dickson, A.G., Sabine, C.L. and Christian, J.R. (Eds.) 2007. Guide to best practices for ocean CO_2 measurements. PICES Special Publication 3, 191 pp

Comment: Freshwater CO_2 Headspace Equilibration Calculations

M. T. Trentman, J. R. Blaszczak, R. O. Hall, Jr.

Koschorreck et al. describe a method for calculating the original CO_2 concentration of a water sample when using a headspace equilibration to extract dissolved CO_2 from water. This manuscript contributes a correction to the final measured CO_2 concentration to account for the near-instantaneous equilibration between CO_2 and HCO_3^- in water during equilibration with the headspace. While the HCO_3^- equilibration correction exists in the chemical oceanography literature (e.g., Dickson et al. 2007), this correction is used less frequently in the freshwater literature. We have multiple ongoing projects in freshwater streams involving data using headspace equilibrations for estimating CO_2 concentrations, and until reading this work, we had not considered correcting CO_2 concentrations for the HCO_3^-

equilibrium despite always having a copy of Dickson et al. (2007) nearby. Thus, we believe this manuscript is a welcome addition to the literature. Given the importance of accurate calculations for estimating CO_2 concentrations and our previous experience with these calculations, our objectives in this comment are to:

1. Use a different mathematical formulation than in Koschorreck et al. to estimate the HCO_3^- equilibrium correction, and compare the magnitude of correction between the two approaches.
2. Evaluate the correction magnitude with samples where headspace equilibration may bias estimates of $p\text{CO}_2$: high alkalinity and a large volume CO_2 -free headspace relative to the water volume.

We used our own grab sample data to compare the full set of headspace equilibration calculations as presented by Koschorreck et al. with the same calculations derived from our group. We note that in parts of our calculations we use different equations than Koschorreck et al. but the approaches are based on the same theory. Our code and a detailed comparison of our calculations with that of Koschorreck et al. are available at https://github.com/jrblaszczak/CO2_headspace_code. First, we added the HCO_3^- equilibrium correction to our existing headspace equilibration calculations using equations 4 and 5 in Dickson et al. (2007) (SOP 4). We then compared the calculated CO_2 concentration from the code provided by Koschorreck et al. with our code using diel grab sample data (sampled every 2-4 hr over a 24 hr period) from streams in northwestern Montana and central Arizona. For these samples, we performed headspace equilibrations in the field using 40 mL of streamwater and 70 mL of injected CO_2 -free air in 140 mL syringes. We equilibrated the headspace by shaking the syringes for 3 minutes, after which we flushed the water from the syringe and stored the remaining gas sample in the syringe until analysis within 48 hours on a Picarro G2131-i analyzer (Picarro, Santa Clara, CA, USA). We measured stream temperature and barometric pressure at the time of water sampling and collected an unfiltered streamwater sample for total alkalinity, which was measured by titration to a pH of 4.5. Stream temperature of our samples was between 7 and 22°C and the total alkalinity was between 1440 and $2060\mu\text{eq L}^{-1}$.

There was minimal variation of the HCO_3^- equilibrium corrections and corrected CO_2 concentrations between the calculations provided by Koschorreck et al. and the calculations we independently compiled. The HCO_3^- equilibrium corrections between the approaches were similar to the hundredth decimal place (expressed as $\mu\text{mol L}^{-1}$ DIC), which we consider to be functionally the same. The average percent deviation of corrected CO_2 concentrations between approaches was 0.9% (SD = 0.6%) which is likewise a small difference. As noted above, the code we provide for this comparison uses different calculations than Koschorreck et al., but is based on the same theory. Thus, we see this comparison as an independent verification of their calculations.

Thank you for this independent check of our procedure

The deviation of HCO_3^- equilibrium corrected and uncorrected CO_2 concentrations for our samples ranged between (2-23%, reported in ppmv). The error in our samples is not surprising given the relatively high total alkalinity, low headspace ratio (1.75), and that we used a CO_2 -free headspace. We are limited in our ability to change our methods to reduce this error given our protocol for measuring gases on the Picarro G2131-I analyzer requires 70 mL of gas per sample, and using CO_2 -free headspace is logistically easier than adding a measurement of air for each sample during a diel. Additionally, equilibration air containing CO_2 would bias estimates of ^{13}C -DIC. By adding the HCO_3^- equilibrium correction we can increase the accuracy of our grab sample CO_2 concentrations without altering our sampling protocol.

While our approaches result in the same outcome we note one difference that is worth mentioning by Koschorreck et al.. This difference involves assumptions we make about calculating the H^+ concentration (eqn. 2, Koschorreck et al.) from the equation for total alkalinity (A_T , eqn. 1) that leads to an algebraically simpler equation and therefore may enable more efficient computation and incorporation of these calculations into larger process models.

$$A_T = [\text{HCO}_3^-] + 2[\text{CO}_3^{2-}] + [\text{OH}^-] - [\text{H}^+] \quad (1)$$

We assume that the H^+ and OH^- concentrations are zero in eqn. 1 for two reasons. First, these concentrations are tiny compared to HCO_3^- and CO_3^{2-} and likely within the range of the error associated with the measured alkalinity via titration or charge balance. Thus, we contend that assuming H^+ and OH^- concentrations are zero will have a negligible effect on the calculation of the overall H^+ concentration. Second, this assumption allows us to solve a 2nd-order polynomial rather than a 3rd-order polynomial equation. While the authors incorporate an elegant solution to solve the 3rd-order polynomial using the polyroot function in R, the use of this function may limit downstream incorporation of these calculations in stochastic simulations. For example, we use adaptations of Markov chain Monte Carlo methods (e.g., the general purpose Bayesian modeling software Stan, Carpenter et al. 2017) to simulate posterior distributions in Bayesian hierarchical models to estimate stream metabolism from diel patterns of CO_2 . In this case, the simpler mathematical solution for solving for the H^+ concentration will facilitate computational speed without sacrificing accuracy.

Given the occasional need for mathematically simpler solutions, we suggest the authors discuss the complexity of their approach, and note that the calculations can be simplified by assuming $[\text{H}^+] \text{ and } [\text{OH}^-] \sim 0$.

As we already commented in our short reply during the discussion phase, the advantages of using an approximation are not clear given current computing power and, more importantly, given that our analytical solution can easily be integrated in any process-based stochastic models (more easily than an iterative solution would be). Our script takes 0.00086 s to run a sample, or 14 minutes for one million runs. It is unlikely that these running times limit the applicability of our method. Also, while the assumption of negligible H^+ and OH^- might be true for high pH and alkalinity waters, it doesn't necessarily hold under low alkalinity conditions typically found in boreal environments where, for example, $[\text{OH}^-]$ can easily be of the same magnitude as $[\text{CO}_3^{2-}]$. We see the strength of our approach in the fact that it can equally be applied to all types of freshwaters.

Overall, we feel that Koschorreck et al. provide a useful contribution to the literature that will lead to more accurate measurements of CO_2 concentrations, particularly in freshwaters. The extensive analyses by Koschorreck et al. of the deviation of corrected and uncorrected CO_2 concentrations across geochemical and methodological scenarios provide necessary context to this issue. Likewise, Koschorreck et al. are the first group to our knowledge to combine calculations for CO_2 concentrations from headspace equilibrations into a streamlined and publicly available R script. We commend the authors for pointing out a commonly neglected correction and for providing the code with which the community can easily overcome this additional step to estimating accurate dissolved CO_2 concentrations.

We acknowledge this point in the abstract by modifying the last sentence to “We provide a convenient direct method implemented in a R-script or a JMP add-in to correct CO_2 headspace results using separately measured alkalinity.

We have a few minor comments on the manuscript:

1. We suggest that the authors carefully re-examine and edit the text in the abstract and conclusion sections that highlights the general importance of factors that may impact the deviation between corrected and uncorrected CO_2 concentrations for consistency. For example, in the conclusion it is noted that samples with pH below 7.5 and pCO_2 above 1000 μatm will have a small error, but in the abstract only the pH is noted. We also note that the abstract and conclusion sections differ in the suggested content of gas used in the headspace to reduce error. The

abstract states CO₂ free gas should be used while the conclusion states that air should be used instead of an N₂ headspace. Consistency of the messaging of these factors would provide more clarity to the manuscript.

We apologize for these inaccuracies. We removed the statement about which headspace gas should be used from the abstract since the potential error is similar for both. We also specified the conditions for low error in the abstract by writing “By analysing the potential error for different types of water and experimental conditions we show that the error incurred by headspace analysis of CO₂ is less than 5% for typical samples from boreal systems which have low alkalinity (<1700 μmol L⁻¹), low pH (with pH <7.5), and high pCO₂ (>1000 μatm).

2. We suggest the authors use more consistent notations, particularly for pCO₂. The authors could note both the 'location' of the pCO₂ measurement (i.e., headspace vs. water) as well as before or after equilibrium in their notation. This caused some confusion for us when evaluating the text and code. A few examples include: line 141- pCO₂ is used while in line 87 pCO_{2water} is used, line 87- pCO_{2Aftereq} is used while in line 146 pCO_{2HSAfter} is used. We believe consistent annotation will improve understanding of these calculations.

We improved the notations as recommended by the reviewer. Notably we now use “mCO₂” for molar mixing ratios and “pCO₂” for partial pressure.

3. Many researchers, including our group, prefer to report our data in units of μmol L⁻¹, especially when we compare data of different molecules (e.g., CO₂ vs O₂ for metabolism). Thus, we suggest that the authors include in the R code the reporting of CO₂ concentrations in μmol L⁻¹. This should not be much trouble given the calculations are already conducted in molar units.

We agree. The JMP script and add-in already provided corrected values in both partial pressure and molar unit. The R script now provides the same.

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