Technical note: Assessing gas equilibration systems for continuous pCO_2 measurements in inland waters

T. K. Yoon[1,a], H. Jin[1], N.-H. Oh[2], J.-H. Park[1]

[1]Department of Environmental Science and Engineering, Ewha Womans University, Seoul 03760, Republic of Korea
[2]Graduate School of Environmental Studies, Seoul National University, Seoul 08826, Republic of Korea
[a]Current affiliation: Environmental Planning Institute, Seoul National University, Seoul 08826, Republic of Korea

Correspondence to: J. H. Park (jhp@ewha.ac.kr)

Abstract. High-frequency continuous measurements of the partial pressure of CO_2 (pCO_2) are crucial to constraining the spatiotemporal dynamics of CO_2 emissions from inland water systems. However, direct measurements of pCO_2 are scarce, and no systematic comparisons have been conducted on the suitability of the widely used measurement systems for continuous underway or long-term deployment in various field conditions. We compared spray- and marble-type equilibrators and a membrane-enclosed CO_2 sensor to assess their suitability to continuous long-term or underway pCO_2 measurements in an urbanized river system in Korea. Both equilibrators had a shorter response time compared with the membrane-enclosed sensor, and could capture large spatial variations of pCO_2 during a transect study along a highly urbanized river reach. The membrane-enclosed sensor based on passive equilibration provided comparable underway measurements along the river sections where pCO_2 varied within the sensor detection range. When deployed in a eutrophic river site, the membrane-enclosed sensor was able to detect large diel variations in pCO_2. However, biofouling on the membrane could reduce the accuracy of the measurement during long deployments exceeding several days. The overall results suggest that the fast response of the equilibrator systems facilitates capturing large spatial variations in pCO_2 during short underway measurements. However, the attendant technical challenges of these systems, such as clogging and desiccant maintenance, have to be addressed carefully to enable their long-term deployment. The membrane-enclosed sensor would be suitable as an alternative tool for long-term continuous measurements, if membrane biofouling could be overcome by appropriate anti-fouling measures such as copper-mesh coverings.

1 Introduction

Recent synthesis efforts have highlighted the importance of carbon dioxide (CO_2) emissions from inland waters in the global carbon cycle (Cole et al., 2007; Battin et al., 2009; Butman and Raymond, 2011; Raymond et al., 2013; Borges et al., 2015). Various methods have been employed over the years to measure the partial pressure of CO_2 (pCO_2) in a wide range of aquatic systems (Takahashi 1961; Keeling et al., 1965; Park et al., 1969: Smethie et al., 1985; Kling et al., 1992). Recent studies on ocean pCO_2, such as the International Ocean Carbon Coordination Project and the Surface Ocean CO_2 Atlas

(Feely et al., 1998; Dickson et al., 2007; Pierrot et al., 2009; Pfeil et al., 2013) have stimulated technical advances, including the development of automated equilibration systems and standardized measurement protocols. Compared with the relatively narrow pCO_2 ranges in the oceans (~100–700 µatm) (Valsala and Maksyutov, 2010), the pCO_2 in inland waters ranges from values < 100 to > 10,000 µatm (Abril et al., 2015). Moreover, the range of temporal and spatial variations in freshwater pCO_2 is much wider than those in the oceans because of the substantial variations in environmental conditions, complex C transformation, and emission processes, as well as anthropogenic disturbances (Cole et al., 2007). For instance, the temporal dynamics of phytoplankton metabolisms could cause large diurnal fluctuations of pCO_2 (Nimick et al., 2011). Moreover, turbulence enhances the CO_2 emissions from rapidly flowing waters, which can result in steep downstream gradients of pCO_2 from the upstream sources (Dawson et al., 2001; Abril et al., 2014). Furthermore, the contribution from labile organic matter of anthropogenic origin can enhance the emission of CO_2 from urbanized inland waters (Frankignoulle et al., 1998; Zhai et al., 2005; Griffith and Raymond, 2011). In view of these unique conditions, specific to inland waters, vigorous field tests have to be conducted before new methods can be deployed for continuous underway or long-term measurements of pCO_2 in inland water systems.

The CO_2 emission rate can be determined either by directly measuring the transfer of CO_2 across the water–air interface, or by estimating the flux based on (1) differences in the pCO_2 between the water and air, and (2) the gas-transfer velocity. Floating chambers have been used often to measure the amount of CO_2 released from a fixed area of the water surface during a relatively short measurement period (Podgrajsek et al., 2014; Lorke et al., 2015). However, the attendant technical challenges include the difficulty of deploying the floating chamber stably over often turbulent water surfaces and the disrupted natural turbulence inside the floating chamber that could result in overestimations of the CO_2 flux (Vachon et al., 2010), especially when the chamber is anchored at a fixed spot (Lorke et al., 2015). Recently, Lorke et al. (2015) have proposed improved designs for floating chambers that minimize the bias of the gas-transfer velocity, including a freely drifting chamber on running water, or an anchored chamber with a close contact over the water surface. However, validation and further technical improvements are needed before these proposed chamber systems could be applied in practice. Eddy-covariance flux measurements can be used as an alternative method for direct flux measurement; however, this technique has been applied only in a small number of aquatic systems (Huotari et al., 2011; Polsenaere et al., 2013; Podgrajsek et al., 2014). The indirect measurement approach based on pCO_2 has been used more widely compared with the direct flux measurement method. Moreover, the performance reliability of this method has been evaluated across a wide range of aquatic systems, where the gas-transfer velocity can be estimated with supplementary environmental data (Raymond et al., 2012). The water-air difference in pCO_2 is determined from the pCO_2 measurements in the water and air and subsequently incorporated into an air–water gas-transfer model (Liss and Slaster, 1974; Deacon, 1977; Wanninkhof, 1992; Raymond and Cole, 2001; Wanninkhof et al., 2009). In addition, the pCO_2 can be estimated from two of the three variables in the carbonate equilibrium model, namely, pH, alkalinity, and dissolved inorganic C (DIC) (Lewis et al., 1998; Dickson et al., 2007). As the calculated pCO_2 data can be obtained easily from existing water quality databases, these data have been used widely in estimating the CO_2 emissions from local to global inland water systems (e.g., Li et al., 2013; Lauerwald et al., 2013; Raymond et al., 2013).

However, Abril et al. (2015) have recently warned that the calculated pCO_2 in acidic, organic-rich inland waters could be overestimated by 50 to 300 %, relative to direct pCO_2 measurements. Such overestimation is ascribed to the combined effect of an unaccounted contribution of organic acids to alkalinity and the limited buffering capacity of the carbonate system in these waters.

Various methods have been developed successfully for continuous pCO_2 monitoring in order to address the large spatiotemporal variability in pCO_2 across a wide range of inland water systems (Frankignoulle et al., 2001; Johnson et al., 2010; Crawford et al., 2015). In addition, a small number of sensor-based studies have been successful in resolving large temporal variations of pCO_2 in inland water systems (Johnson et al., 2010; Huotari et al., 2013; Bastviken et al., 2015). Usually, these monitoring techniques were tested in headwater watersheds; however, they have not been applied yet to the long-term monitoring of pCO_2 in larger river systems, where human impact, such as high loads of organic pollutants is severe. Equilibrators have been deployed successfully for continuous underway pCO_2 measurements in large rivers and estuaries (Frankignoulle et al., 1998; Griffith and Raymond, 2011; Bianchi et al., 2013; Abril et al., 2014). However, these efforts have been focused on the spatial variability of pCO_2 rather than on integrating both spatial and temporal variations to provide accurate estimates of CO_2 emission. Moreover, the individual systems developed for continuous pCO_2 measurements have not been compared of consistent measurement accuracy over long-term deployments. Therefore, this study aims (1) to compare the accuracy and maintenance requirements of three widely used gas equilibration systems for field application in a series of laboratory and field cross-validation tests, and (2) to provide recommendations to address the technical problems and maintenance requirements that could hamper continuous long-term or underway measurements of pCO_2 in inland waters. The three systems we compared are a spray-type equilibrator, a marble-type equilibrator, and a membrane-enclosed CO_2 sensor.

2 Gas equilibration systems used for continuous pCO_2 measurements in inland waters

Numerous gas equilibration methods have been used to measure the pCO_2 in inland waters. Gases dissolved in water need to be equilibrated between the liquid and an artificially created *headspace*, after which the gas concentration in the headspace air can be analyzed with a gas analyzer (Swinnerton et al., 1962). Equilibration methods can be grouped into three categories, namely, *manual headspace equilibration*, *equilibrators*, and *membrane-based equilibration* (Table 1, and Figs 1). In addition, these equilibration methods can be classified based on combinations of *manual* vs. *automatic* system operations and *active* vs. *passive* equilibration mechanisms. The system operations include water sampling and circulation, water–air equilibration, and air flow circulation. Active equilibration differs from passive equilibration in that gas transfer across the water–air interface is facilitated by an external supply of energy. Gas analysis is usually conducted by a gas chromatograph (GC) or an infrared gas analyzer (IRGA). Additional to such methods, an isotope ratio mass spectrometer (IRMS) or a cavity-enhanced absorption spectrometer can be used to analyze methane (CH_4) or the stable C isotopes of CO_2 and CH_4 as well (Friedrichs et al., 2010; Maher et al., 2013; Gonzalez-Valencia et al., 2014; Webb et al., 2016). Manual headspace equilibration, by

shaking a collected water sample in a closed bottle or syringe, has long been used as a standard method for measuring pCO_2 in inland waters (Kling et al., 1992; Hope et al., 1995; Raymond and Cole, 2001). Here, we focus on automated gas equilibration systems that can be used for continuous underway or long-term measurements of pCO_2 in inland waters.

Various automated equilibrator systems have been used for both discrete and continuous measurements of pCO_2 in inland waters and oceans (Takahashi, 1961; Keeling et al., 1965; Park et al., 1969; Feely et al., 1998; Frankignoule et al., 2001). The spray- and marble-type equilibrators are the two most widely used such systems (Fig. 1; Table 1). The equilibrator automates the manual shaking used in the headspace equilibration method by using a spray, nozzle, or showerhead (as termed in different papers) and marbles that increase the water–air interface for gas exchange. In the spray-type equilibrator, the pumped water is sprayed from a nozzle and the pCO_2 in the droplets is subsequently equilibrated with the headspace air within the chamber of the equilibrator (Takahashi, 1961). Spray-type equilibrators have been used as a standard method in oceanic pCO_2 monitoring studies since their introduction in the late 1950s (Takahashi, 1961; Keeling et al., 1965; Feely et al., 1998; Dickson et al., 2007; Pierrot et al., 2009), with several commercial versions being currently available (e.g., GO8050, General Oceanics, USA). Spray-type equilibrators have been used in diverse inland water systems (Raymond and Hopkinson, 2003; Zhai et al., 2005; Maher et al., 2013; Crawford et al., 2015; Joesoef et al., 2015). The marble-type equilibrators were developed to address the monitoring conditions specific to inland waters, such as high loads of sediments and organic matter (Frankignoulle et al., 2001; Abril et al., 2006). In the marble-type equilibrator, the pumped water flows over the surface of the marbles, which increase the air–water interface and reduce the volume of headspace air, enabling gas exchange between the flowing water and the headspace air (Frankignoulle et al., 2001; Abril et al., 2006). The equilibrated air continuously circulates in a closed loop linking the equilibrator headspace to a gas analyzer, usually an IRGA, or a cavity-enhanced absorption spectrometer (Friedrichs et al., 2010; Gonzalez-Valencia et al., 2014; Webb et al., 2016). These *automatic* and *active* equilibration systems integrate water sampling, equilibration, and gas analysis in a loop by using water or air pumps powered by external sources (Table 1). This implies that factors such as power supply and maintenance could limit the application of these equilibrator systems to continuous monitoring of pCO_2 in certain inland water systems. For instance, long-term observation at a remote site could be hampered by maintaining power supply for a sustained period. In addition, the components of such an equilibrator system, namely, water pumps, tubing, nozzles, and marbles could become clogged up by small particles and large debris when the system is deployed for long-term monitoring in turbid or eutrophic waters (Santos et al., 2012).

A membrane-based equilibration system can be established when a diffusion-type IRGA CO_2 sensor, enclosed in a water-impermeable but gas-permeable membrane, is placed directly in water (Figs. 1, S1; Table 1; Johnson et al., 2010). Gas equilibration occurs between the inside (headspace over the sensor) and the outside (water) of the membrane. The CO_2 concentration in the equilibrated air inside the membrane is detected by the sensor and the data can be stored by a connected data logger. A number of commercial membrane-enclosed sensor systems are available (e.g., eosGP, Eosense Inc., Canada; Mini-Pro CO_2, Pro-Oceanus Systems Inc., Canada). Some of the CO_2 sensors used in these commercial systems can detect a wide range of CO_2, covering the usual range of pCO_2 found in inland waters. This *automatic* but *passive* equilibration system

does not require extra energy for equilibration or for water and air pumping, therefore, this system has a significant advantage over other equilibrator systems with higher energy demands. This relatively compact "all-in-one" system (from equilibration to detection), together with the relatively low cost, facilitates easy field deployment that can be replicated at multiple locations. In addition, a wide range of applicability allows a cross-comparison of C transfer across various watershed compartments, including sediments, soils, and dead wood (Johnson et al., 2010; Leith et al., 2015; Troxler et al., 2015). The potential problems of the membrane-enclosed sensor system, including long response time and biofouling, have not been adequately investigated yet. More time is usually required for equilibration by passive diffusion, with the typical response times ranging from several to dozens of minutes (Santos et al., 2012; Webb et al., 2016). Longer response times could hamper the detection of large spatial or temporal variations of pCO_2. Moreover, biofouling on the membrane surface can result in over- or underestimation of the pCO_2 over long-term deployment. However, previous studies have not reported any significant effects of biofouling during long-term deployment in headwater streams with relatively low ranges of pCO_2 (Johnson et al., 2010; Crawford et al., 2013; Peter et al., 2014; Leith et al., 2015). No investigation has been conducted on whether the membrane-enclosed sensor could be used for continuous underway measurements of pCO_2 in large rivers and estuaries

Other membrane-based and hybrid equilibration systems, such as membrane contactors, have been used as alternative membrane-based equilibration methods and these systems are available commercially for industrial applications (e.g., Liqui-Cel®). The membrane contactors allow *automatic* and *active* measurement of pCO_2 when coupled with an automated CO_2 analyzer system (Hales et al., 2004; Santos et al., 2012). Such membrane contactors have been used in various inland waters, including boreal (Teodoru et al., 2011) and tropical waters (Abril et al., 2015; Teodoru et al., 2015). However, the potential clogging and biofouling problems remain unresolved, hampering the long-term deployment of these systems in eutrophic waters. Bastviken et al. (2015) have presented a hybrid system that combines floating chambers and low-cost CO_2 sensor modules to detect changes in the CO_2 concentration in the chamber headspace. The use of low-cost detectors enables replicated monitoring at multiple sites, which is essential to detecting the spatiotemporal variations of pCO_2.

Abril et al. (2015) have found general agreement in the measurements obtained from the manual headspace-equilibration method and the marble-type and contactor equilibrators over a wide range of pCO_2 (0–15,000 μatm) and other water chemical properties in various inland waters, ranging from temperate to tropical systems. An earlier comparison had established good agreement between a marble-type equilibrator and the manual headspace equilibration method (Abril et al., 2006). Johnson et al. (2010) have established moderate agreement between a membrane-enclosed sensor and a manual headspace equilibration relevant to the pCO_2 in four boreal inland waters. Santos et al. (2012) have compared various systems, such as a spray-type, a marble-type, three membrane contactor equilibrators (Liqui-Cel), and a passive polypropylene membrane system (ACCUREL® PP, Membrana GmbH, Germany) with the aim of establishing a system for coupled ^{222}Rn and pCO_2 measurements in the groundwater discharged into coastal waters. Although all the systems produced similar results for pCO_2 in the laboratory tests with a groundwater sample (~12,000 μatm), the response times of these equilibration methods differed markedly. The polypropylene membrane exhibited a particularly long response time (82 min)

compared with those of the other systems, which ranged from 4–18 min. Empirical evidence to support the agreement between the membrane-enclosed sensor and the other equilibration systems is still insufficient. In sum, the response time and biofouling of the equilibration systems, together with other maintenance requirements such as power supply and consumable replacement, need to be assessed in various field conditions to enable the successful deployment of such systems in continuous underway or long-term measurements of pCO_2.

3 Materials and methods

3.1 Equilibration systems

To assess the applicability of the three selected equilibration systems to continuous pCO_2 measurements in inland waters, we compared their performance with each other and validated their performance against that of the manual headspace equilibration (Kling et al., 1992; Hope et al., 1995). Manual headspace equilibration was conducted using a polypropylene syringe (60 ml; HSW Norm-Ject Luer Lock Tip; Henke-Sass Wolf GmbH, Germany) to collect a 30 ml water sample and a 30 ml sample of the ambient air. The syringe was shaken manually for 2 min, after which a subsample of the equilibrated air was collected in a 50 ml gas-tight syringe (Swastik Enterprise, Gujarat, India). The gas sample and an additional 30 ml sample of ambient air were injected directly into a GC (7890A, Agilent, USA), fitted with a Supelco Hayesep Q 12 ft. 1/8 inch column during the laboratory tests. In the field tests, the gas sample collected in the syringe was transferred to a pre-evacuated vial for gas analysis in the laboratory, usually within three days. The pCO_2 was calculated from the CO_2 concentrations of the equilibrated air and ambient air samples, water temperature, and barometric pressure, based on Henry's law (Hudson, 2004).

The spray-type equilibrator included a spray nozzle (GG 3/8 - SS 15, Spraying System Co., USA) in an acrylic tube (inner diameter 40 mm, outer diameter 48 mm, and height 200 mm), based on the designs commonly used in previous studies (Figs. 1, S1; Table S1; Keeling et al., 1965; Feely et al., 1998; Raymond and Hopkinson, 2003). A marble-type equilibrator, smaller than those used in previous studies (Frankignoulle et al., 2001; Abril et al., 2006), was designed, based on laboratory tests to enhance the portability of the device without compromising the measurement accuracy. The device consisted of an acrylic tube (inner diameter 40 mm, outer diameter 48 mm, and height 300 mm) filled with glass marbles (diameter 10 mm) (Figs. 1, S1; Table S1). Water was continuously pumped into both equilibrators with a bilge pump (Tsunami T800, Attwood Co., USA) at ~ 2.5 L min^{-1} for the spray-type equilibrator and at ~ 1.5 L min^{-1} for the marble-type equilibrator. Different optimal water flow rates were determined in preceding laboratory tests to enable fast equilibration without causing chamber overflow. A diaphragm pump was used to circulate the equilibrated air through an air filter and a desiccant (Drierite) column between the equilibrator chamber and an IRGA (LI820, Li-Cor, USA) at 700 ml min^{-1}. The collected data were logged every second in a laptop computer. Potential changes in the water temperature inside the equilibrator chamber were checked during field deployments by comparing the in-stream water temperature with that of the chamber outflow in various weather conditions. The temperature differences between the river water and the equilibrator outflow water were usually within

0.3°C. The differences in barometric pressure between the inside and outside of the equilibrator chamber were lower than 5 μatm when the chamber vent was closed. The vent was closed during all the measurements after preliminary laboratory tests had confirmed that the small increase in the barometric pressure would not affect the accuracy of the pCO_2 measurement. The small initial pressure build-up immediately after turning on the water pump was relieved during ventilation for a few seconds through a vent channel that was established by using a three-way cock on the air-flow circuit. In addition, the integral pressure compensation function of the IRGA (LI820) we used was able to reduce any potential risk of inaccurate CO_2 analysis being induced by pressure changes.

The membrane-enclosed sensor system consisted of a CO_2 transmitter, containing a CARBOCAP® sensor (GMP222; GMT222, Vaisala, Finland) and a data logger (CR10X; CR1000, Campbell Scientific Inc., USA), as has been described in detail by Johnson et al. (2010). The sensor probe was enclosed in polytetrafluoroethylene (PTFE) membrane tubing (200-07, International Polymer Engineering, USA) (Figs. 1, S1; Table S1). The open end of the membrane tubing was sealed with a rubberizing compound (Plasti Dip, Plasti Dip International, USA). The membrane-enclosed sensor was placed directly underwater at the desired depth. When necessary, the membrane-enclosed sensor was covered by metal housing to protect the membrane from underwater obstacles such as large floating debris. Power and electric signals were delivered through a cable between the underwater sensor, the transmitter, and the data logger stored in a console box on the ground. Three CO_2 sensors, a data logger, and two batteries (12 V 7 AH, Rocket, Korea; 12 V 100 AH, ATLASBX, Korea) were placed in two portable, custom-made plastic containers. Since the CO_2 sensor does not integrate any compensation function for variations in temperature and barometric pressure, the outputs of the sensor were corrected by separately collected data on temperature and barometric pressure (Johnson et al., 2010).

All the CO_2 analyzers and sensors were calibrated in the laboratory, using CO_2 gases of known concentrations (0, 500, 5,000, and 10,000 ppm) immediately before each laboratory or field test. When the CO_2 sensors were deployed over several weeks, they were checked for measurement accuracy during the maintenance intervals of 1–3 months and, if required, were recalibrated against the same set of standards. The IRGA used for the two equilibrators (LI820) was spanned with a pure N_2 gas (>99.9%) and a CO_2 standard gas at ~10,000 ppm. Linearity check was performed with two additional CO_2 standard gases (500 and 5,000 ppm). During discrete and underway measurements, measurements of air temperature and barometric pressure were recorded by a portable data logger (Watchdog 1650 Micro Station, Spectrum Technologies Inc., USA) that included an integral air temperature/humidity sensor and an external barometric pressure sensor. For long-term deployment tests, additional air temperature/humidity and water temperature measurements were recorded in a Campbell data logger. The water temperature was measured with a portable pH meter (Orion 5-Star Portable, Thermo Scientific, USA) or a multi-parameter water quality sonde (6820 V2, YSI Inc., USA).

3.2 Laboratory tests

The response time and measurement accuracy of the three equilibration systems were compared by using tap and distilled water that were continuously flowing into a 6 L container that was exposed to the ambient air to maintain a constant pCO_2.

Both equilibrators and the membrane-enclosed sensor were first placed in a tap water container (pCO_2: ~2,500 μatm) and subsequently moved to a distilled water container (~600 μatm) to determine the response times. The response time (t_{95}) was determined as the time required to reach the 95 % level of the final stabilized pCO_2 values that exhibited variations smaller than 1 % of the coefficients of variation (CV) for 2 min. In addition, the response time can be assessed by calculating the time constant (τ) of the exponential or e-folding curve fitting of varying pCO_2 values during high-to-low equilibration (Johnson, 1999). As the various response patterns observed for the membrane-enclosed sensor could not be fitted by the ideal exponential decay curve, we present only t_{95} results.

3.3 Field tests of the spray-type equilibrator and the membrane-enclosed sensor

In May 2015, we compared the measurement accuracy and response times of the spray-type equilibrators and the membrane-enclosed sensor at 12 sites. These sites, ranged from forested headwater streams (38°15' N, 128°7' E, 582 m.a.s.l. through stream and river locations, blocked by dams or weirs, to the tidal reach along the Metropolitan Seoul (37°41' N, 126°39' E, 1 m.a.s.l.) of the Han River in South Korea. Additional measurements, using the manual headspace equilibration method and the membrane-enclosed sensor, were performed at 6 of the 12 sites every month from July 2014 to July 2015, in order to obtain more response time data under various field conditions. To compare simultaneously the performance of the three equilibration systems, another field campaign was conducted in May 2016 in a tributary watershed and along the tidal reach of the Han River. The 14 survey sites ranged from a forested headwater stream (37°48' N 127°1' E, 148 m.a.s.l.) through the urbanized stream locations (Joongnang Stream) to the tidal reach of the Han River. The Han River is intensively dammed, with more than ten large dams and several old and newly built weirs. The predominant flow condition of each site was determined as either standing or flowing water, based on the distance from the closest up- or downstream dam or weir, and the specific flow conditions during the field study. The level of pCO_2 and other environmental conditions at the sites were heterogeneous enough to allow a cross-validation test. For example, the ranges of the dissolved oxygen (DO), pH, and dissolved organic carbon (DOC) were 3.5–11.6 mg L^{-1}, 6.8–9.0 and 1.0–5.1 mg C L^{-1} in the 2015 field campaign, and 6.5–10.7 mg L^{-1}, 5.3–7.4, and 1.0–5.0 mg C L^{-1} in the 2016 campaign, respectively.

The water pCO_2 at a depth of 20 cm was determined by using simultaneously the manual headspace equilibration, membrane-enclosed sensor, and spray-type equilibrator systems. The membrane-enclosed sensor was placed directly at 20 cm below the surface. A peristaltic pump (Masterflex E/S portable sampler, Cole-Parmer Instrument Co., USA) was used to collect water into a sampling bottle for the manual headspace equilibration measurement, while a bilge pump was used for the spray- or marble-type equilibrator. The same measurement procedures and instrumental set-ups as in the laboratory tests were used for all three equilibration systems.

3.4 Continuous underway measurements of pCO_2

To test the applicability of the three equilibration systems to continuous underway measurements of pCO_2, a boating expedition was undertaken on 11 May 2015 along the tidal reach of the Han River, which receives varying loads of organic

matter and CO_2 via its tributaries (37°31' N, 127°1' E, 7 m.a.s.l.). The surveyed river reach is influenced strongly by the inflow from several urban streams, including Tan Stream and Joongnang Stream, draining from the Seoul metropolitan area. We assumed that the significant spatial variations in pCO_2 and other water quality components along the confluence with the urban streams would create ideal conditions for cross validation of the three equilibration systems. Prior tests had showed that three equilibration systems would perform well at moderate boat speeds around 10 km h^{-1}. During underway measurements, the speed was maintained at ~10 km h^{-1} over the distance of ~30 km. This speed is consistent with the usual boat speed range used for other continuous underway measurements (Abril et al., 2014; Crawford et al., 2015). The boat was stopped for ~10 min at each of the nine discrete-sampling locations to collect water samples and to measure pCO_2 by using the manual headspace-equilibration method.

The water pCO_2 at 20 cm below the surface was continuously measured at intervals of 1 or 5 s with the three equilibration systems. One membrane-enclosed sensor, together with a bilge pump delivering the collected water into the spray- and marble-type equilibrators, and another on-board membrane-enclosed sensor were attached to a pole and placed 20 cm below the water surface on one side of the boat. Two flow-through IRGAs (LI-820, Licor, USA; GMP343, Vaisala, Finland) were coupled with the spray- and marble-type equilibrators, respectively. A portable multi-parameter pH meter (Orion 5-Star Portable, Thermo Scientific, USA) was used to measure simultaneously the water temperature, pH, electrical conductivity, and dissolved oxygen in the continuously collected water on-board. The air temperature and barometric pressure were recorded in a micro-logger (Watchdog 1650 Micro Station, Spectrum Technologies Inc., USA). The two membrane-enclosed sensors (one placed in the river water and the other immersed in the pumped water on-board) had upper detection limits of 10,000 and 7,000 ppm, respectively.

3.5 Continuous long-term measurements of pCO_2

Several laboratory and field tests were conducted to examine the application potential of the three equilibration systems to continuous long-term monitoring in the tidal reach of the Han River. The spray-type equilibrator was selected for use, as the preliminary tests had shown that it was easier to maintain the power supply and air-flow dehydration with this type of equilibrator than with the marble-type. The long-term measurement stability of the spray-type equilibrator was tested in comparison with that of the membrane-enclosed sensor in a series of unmanned field deployments. However, the significant power consumption from the pumping and the gradual clogging of the nozzle resulted in repeated failures of the system. The resulting pCO_2 data exhibited abnormal patterns 2–3 d after the start of the monitoring. Therefore, we focus here on the long-term performance and the relevant antifouling measures of the membrane-enclosed sensor.

As part of the long-term monitoring project, a membrane-enclosed sensor (“bulk membrane” sensor) was deployed at a depth of 20 cm below the surface, along an uninhabited island on the downstream reach of the Han River, near the city center of Seoul (37°32' N, 126°55' E, 5 m.a.s.l.). The deployment period lasted one year, starting in July 2014. To examine the effectiveness of the copper-mesh screening, intended to reduce biofouling on the membrane surface, another membrane-enclosed sensor, covered with copper mesh (“membrane+Cu” sensor), was deployed at the same site for 43 d from 31 May

to 12 July 2015. The bulk membrane and membrane+Cu sensors were attached to a buoy, ~3 m off a dock constructed along the island shore. Two automobile batteries (12 V 100 AH) were placed in a series on the island to power the sensors, together with the CO_2 transmitter and a data logger. The two parallel batteries supplied power for two weeks to two in-stream sensors and an additional sensor used for the concurrent measurement of the air pCO_2 1 m above the water surface. During routine biweekly maintenance visits, the membrane surface was cleaned with a soft cloth and brushes and subsequently rinsed with deionized water, after which the copper-mesh screen was replaced. In addition to CO_2, the pH, DO, water temperature, conductivity, and turbidity were monitored by using a multi-parameter water quality sonde (6820 V2, YSI Inc., USA). The accuracy of the pH probe was regularly verified on site with pH buffers and concurrent pH measurements, using a portable pH meter (Orion 5-Star, Thermo Scientific, USA). All the collected data were logged at 10 min intervals.

3.5 Data analysis

We assessed the agreement of the pCO_2 measurements among the equilibration systems by linear regression analysis and by examining the CV values across the monitoring sites, after the normal distribution of data had been confirmed by the Shapiro–Wilk test. We used the *t*-test to compare the differences in response time (t_{95}) between the equilibration systems. The relationships between the response time and ΔpCO_2 (defined as the difference between the initial and the stabilized final pCO_2 during deployment) were established for the flowing and standing water types. For the continuous long-term measurements, the relative difference of pCO_2 was calculated from the natural log-transformed ratio between the values of the membrane sensor and the membrane+Cu sensor. The pH–pCO_2 relationships were described by locally weighted scatterplot smoothing (LOESS; Cleveland and Devlin, 1988) to examine the viability of the pCO_2 measurements arising from the biofouling in the time series data. The analysis was based on the assumption that robust pH–pCO_2 relationships could be expected from the carbonate equilibrium model if there were no artifact effects such as sensor biofouling. The temporal changes in the pH–pCO_2 relationships were examined to assess the biofouling-induced deviations from the robust pH–pCO_2 relationship. All statistical analyses, including the descriptive statistics, t-test, regression analyses, and LOESS, were conducted on the R software environment for statistical computing and graphics (R Development Core Team, 2011).

4 Results and discussion

4.1 Cross validation of system performance

A series of laboratory tests established good agreements in the measurement accuracy of the three compared equilibration systems, as demonstrated by the correspondence among the three systems at a given pCO_2 (Fig. 2). The CV of the measurements of the three equilibration systems was 3.0 % for the tap water and 6.2 % for the deionized water. The response time test indicated the fast response of the spray- and marble-type equilibrators (t_{95}: ~ 1 min 45 s for both the low-to-high

and the high-to-low equilibrations) in comparison with the slow response of the membrane-enclosed sensor, which exhibited different values of t_{95}: 16 min 30 s and 19 min for the low-to-high and high-to-low transitions, respectively (Fig. 2).

In addition, the short-term continuous measurements of pCO_2 for 30–60 min at various field sites showed general agreement between the three equilibration systems and the manual headspace equilibration measurements over a wide range of pCO_2, from 152 to 10,150 μatm (Fig. 3). The measurement results of the three equilibration systems and the manual equilibration method showed strongly positive pairwise relationships, with all the comparisons indicating $R^2 > 0.99$ and slope (β) within 0.97–1.02 (Fig. 3). The excellent agreement found between the compared methods is consistent with the results of other studies that have demonstrated the accuracy of the equilibrators (Abril et al., 2006; Santos et al., 2012; Abril et al., 2015), or the membrane-enclosed sensor (Johnson et al., 2010), although these previous comparisons were conducted separately for each equilibration system.

Consistent with the results of the laboratory test, the spray- and marble-type equilibrators exhibited response times that were shorter than those of the membrane-enclosed sensor during the field tests (*t*-test: $P < 0.001$; Fig. 4). Both equilibrators usually reached the level of pCO_2 equilibration within a few minutes (Fig. 4a); whereas the membrane-enclosed sensor required a longer time to reach the same pCO_2 level (Fig. 4b). The mean t_{95} was 1 min 45 s for the spray-type equilibrator and 2 min 5 s for the marble-type equilibrator, without showing noticeable differences between the standing and flowing waters. The small difference in response time between the equilibrators might have resulted from different operational conditions including the difference in water flow (2.5 and 1.5 L min^{-1} for the spray- and marble-type equilibrator, respectively). The response time of the spray-type equilibrator was shorter than the response times reported by another study (8 min; Santos et al., 2012), but were similar to the response times of the marble-type equilibrators reported by other studies (2–3 min; Frankignoulle et al., 2001; Abril et al., 2014). The differences in the response time of the spray-type equilibrators could be ascribed to various factors, including the different levels of pCO_2 (~100–10,000 μatm in this study vs. > 10,000 μatm in the study by Santos et al., 2012), the equilibrator size (251 vs. 1963 cm^{-3}), the length of the air circuit, and the performance of the spray nozzle. The mean t_{95} for the membrane-enclosed sensor was 6 min 58 s and 14 min 49 s for flowing and standing waters, respectively. This result suggests that whereas the response time is mainly controlled by the difference in pCO_2 between the air and water, different degrees of turbulence in different water-flow conditions could affect the gas diffusion velocity significantly, which could be described by the diffusion coefficient of Fick's laws. The longer response time of the membrane-enclosed sensor can be explained by the fact that passive equilibration occurs without any physical process to facilitate equilibration underwater (Santos et al., 2012). Moreover, the diffusion-type IRGA of the membrane-enclosed sensor generally exhibited longer response times compared with those of the flow-through IRGAs of the equilibrator systems. In addition, the gas diffusivity across the water–membrane interface could differ from the diffusivity between the water–air interfaces within the equilibrator chambers. Since water turbulence could enhance the equilibration efficiency of the membrane-enclosed sensor, the deployment time in flowing water could be shortened compared with the longer time required for deployment in standing water.

The response time increased logarithmically for both equilibration systems with ΔpCO_2 (Fig. 4; note a log scale for the x-axis). The response time increased with increasing ΔpCO_2 (i.e., high water pCO_2), with steeper increases being observed for the membrane-enclosed sensor, particularly in standing water. The slope of the relationship was in the descending order: the membrane-enclosed sensor in standing water (7.7), the membrane-enclosed sensor in flowing water (0.8), the marble-type equilibrator (0.6), and the spray-type equilibrator (0.5). There was no clear difference in the response time of both equilibrators across a wide range of ΔpCO_2. Only small portions of the variations in the response time observed for the membrane-enclosed sensor were accounted for by ΔpCO_2 in flowing water ($R^2 = 0.18$; $P < 0.05$). The relationship between ΔpCO_2 and the response time measured in flowing water was not statistically significant ($R^2 = 0.04$; $P = 0.29$), suggesting that the water flow and other in-stream processes could have additional effects on the response time. The temperature could also affect response time, although regression analysis did not indicate any significant relationship between the temperature and response time, probably because of the relatively narrow range of temperature variations among the sampling sites.

The results suggest that the deployment time of the membrane-enclosed sensor for short-term (< 1 h) deployments, as part of multi-site discrete monitoring, should be carefully determined, based on the water-flow conditions and expected range of pCO_2 levels. It is crucial that sufficient time be allowed for underwater deployment to ensure the accurate measurement of pCO_2 with the membrane-enclosed sensor. Therefore, we suggest a minimum deployment time of 10 min for flowing water and 30 min for standing water, which would cover a range of t_{95} that was determined in various flow conditions (Fig. 4). Where the long response time poses an obstacle to multiple discrete measurements in a wide range of locations within a limited space of time, employing the equilibrators could be a quicker alternative with the same level of measurement accuracy.

4.2 Continuous underway measurements

The continuous underway measurements of the two equilibrators and the two membrane-enclosed sensors were generally in good agreement, namely, within 10 % CV, except for the river sections for which drastic changes in pCO_2 were observed (Fig. 5). The two equilibrators produced almost the same results across the monitored reach. This comparison corroborates the accuracy of the previously obtained underway pCO_2 measurements, which have compared separately the performance of each of the equilibrator types with those of the manual headspace equilibration measurements (Abril et al., 2006; Griffith and Raymond, 2011; Abril et al., 2014). The pCO_2 measurements of the membrane-enclosed sensors generally corresponded well to those of the equilibrator and headspace equilibration measurements. However, after 12 o'clock the measurements deviated substantially as the boat entered the river segments where the inflow from a highly polluted tributary enriched in pCO_2 elevated the pCO_2 of the main stem above the upper detection limits of the two different sensors (~10,000 and 7,000 ppm) (Fig. 5). The sensor measurements also deviated noticeably from the measurements obtained with the spray- and marble-type equilibrators for the period from 12:20 to 12:35, during which the pCO_2 changed abruptly. In contrast with the long response times observed for discrete measurements at the 26 sites (Fig. 4), the membrane-enclosed sensors exhibited good agreements with the other results across most of the river sections where pCO_2 changed relatively gradually (Fig. 5). The increased

turbulence from the movement of the boat could have enhanced the equilibration of the membrane-enclosed sensor. In addition, there was little difference in the pCO_2 values measured by the in-stream sensor and another sensor immersed in the pumped water on-board the vessel. The relatively high flow rate of the water pump (1.5–2.5 L min^{-1}) could have generated sufficient mixing for rapid equilibration.

The test results suggest that both the spray- and marble-type equilibrators can be used for underway measurements along waterways with significant spatial variations of pCO_2. However, it remains unclear how long the measurement accuracy could be maintained during an extended cruise along high-CO_2 waterways, without maintenance of the replaceable items, including nozzles, marbles, and desiccants. Bakker et al. (1996) reported on frequent blockages of their showerhead equilibrator with particulate materials derived from algal blooms in Dutch coastal waters. Long deployments of the spray-type equilibrator in eutrophic freshwaters could also result in similar clogging problems. Despite the increasing use of membrane-enclosed sensors for long-term continuous pCO_2 measurements in freshwater systems (Johnson et al., 2010; Huotari et al., 2013; Peter et al., 2014; Leith et al., 2015), previous studies have rarely examined the spatial variations in pCO_2 across a wide range of environmental conditions. Our transect results demonstrate that the membrane-enclosed sensor could provide reliable continuous underway measurements in the inland water systems that show large spatial variations of pCO_2 such as the monitored river reach. Proper calibration of the sensor for a high range of pCO_2 should be done before the sensor is deployed in the high-CO_2 water. As the in-stream and on-board sensors produced almost the same measurement results, we suggest that the on-board measurements with pumped water could be used as a safer method for concurrent measurements of pCO_2 and other water quality components. On-board measurements could be a way to avoid damage by unknown underwater obstacles such as large floating debris.

4.3 Continuous long-term measurements

The pCO_2 measurements from the membrane-enclosed sensors with and without the copper-mesh screen started to diverge substantially 3–5 d after the biweekly maintenance (Figs 6, S2a). During the later phases of the biweekly monitoring intervals, the pCO_2 measurements from the sensor without the copper mesh screen (bulk membrane sensor) exhibited larger diurnal fluctuations than those from the sensor protected with the copper-mesh screen (membrane+Cu sensor). When the daily averages were compared to reduce the diurnal fluctuations, the pCO_2 measurements of the bulk membrane sensor were higher than those of the membrane+Cu sensor. Furthermore, these differences increased with time from the day that maintenance was done. The relative differences in the daily mean pCO_2 between the two sensors remained within 10 % for 5, 2, and 7 d after the routine maintenance on the 153th, 169th, and 182th day of the year, respectively. During the monitoring period, extraordinary algal blooms occurred that were ascribed to a combination of factors, such as severe drought, warm temperatures, and high loads of nutrients discharged from water treatment facilities and the polluted tributaries draining the Seoul metropolitan area. The chlorophyll-a concentration increased from 21.1 mg m^{-3} on 2 June to 46.7 mg m^{-3} on 2 July (Water Information System of Korea; http://water.nier.go.kr). The bulk membrane sensor could have been more prone to biofouling by planktonic and associated bacterial communities than the membrane+Cu sensor was. Enhanced production or

consumption of CO_2 around the sensor membrane apparently amplified the diurnal fluctuations of pCO_2, leading to considerable divergence between the two sensor measurements with increasing time after maintenance.

The relationships between the pH and pCO_2 were used to examine the increasing biofouling effects with time after the maintenance day (Fig. 7). The pH–pCO_2 relationships for the bulk membrane sensor shifted upward with time after maintenance, whereas those for the membrane+Cu sensor remained consistent over time (Fig. 7a). If additional CO_2 molecules were produced or consumed by the biofilms formed on the membrane sensor, it could disturb the usual pH–pCO_2 relationship, which could be explained by the carbonate equilibrium model (Nimick et al., 2011). In addition, the relationships between the daily CVs of pH and pCO_2 were stronger for the membrane+Cu sensor (R^2 = 0.91) than for the bulk membrane sensor (R^2 = 0.51) (Fig. 7b). The consistent pH–pCO_2 relationships observed for the membrane+Cu sensor indicated the reliability of the measured pCO_2 values. However, validating the method would require concomitant pCO_2 measurements using other equilibration methods across a wide range of pCO_2.

The test results suggest that the membrane-enclosed sensor could be vulnerable to biofouling in polluted waters similar to the studied site, which could amplify the diurnal fluctuations of pCO_2. The ever-present problem of biofouling must be taken into account in the long-term deployment of any pCO_2 equilibration system. If appropriate antifouling measures are not undertaken, repeated maintenance visits at short intervals of 3–5 d could be required for such long-term deployments in eutrophic rivers with high levels of and large diurnal fluctuations in pCO_2. Daily average pCO_2 values could be used as representative pCO_2 levels within a week from the maintenance day, but the uncertainty level cannot be determined without concomitant measurements using other discrete or continuous measurements that are not significantly influenced by biofouling. We recommend that the copper-mesh screen be used to minimize the biofouling effects as a cost- and energy-efficient measure. Antifouling techniques can be classified into various categories, including mechanical (e.g., wiper, brush, water jet, and ultrasonic sound) and biocidal (e.g., copper, chlorine, and UV) approaches (Delauney et al., 2010). Currently, wipers and copper-based materials are commonly applied to various water quality probes. For instance, the YSI company supplies antifouling kits for water quality sondes, including wipers, copper-mesh screens, copper-alloy guards, and copper tapes, given that these practices have been found effective in various inland and marine environments (YSI Incorporated, 2010). Compared with other biocides, the relatively low toxicity of copper ensures effective application in aquatic environmental monitoring (Manov et al., 2004). Other mechanical antifouling techniques (e.g., brushing and wiping) could be applied to the membrane-enclosed sensor system, but it should be noted that these additional measures would consume more energy. Biofouling could be a negligible problem in oligotrophic waters. For instance, we deployed membrane-enclosed sensors without copper-mesh screening at a forest headstream for one week and at a reservoir for two weeks in June–July 2015, during the same season the antifouling test was being conducted. Following the 2-week deployment at the oligotrophic reservoir surface water, the membrane surface did not exhibit any visible sign of biofouling (Fig. S2) and the pH–pCO_2 relationship remained stable, showing no significant deviations as time progressed (Fig. S3).

Stable power supply is another important factor for successful long-term and continuous observation. A membrane-enclosed sensor consumes approximately 30 times less power than does a single-bilge pump for equilibrators. The two parallel

automobile batteries generally lasted two weeks, maintaining the power supply to the three membrane-enclosed sensors. Using an analog timer or relay system, the power of the membrane-enclosed sensor could be switched on and off at a pre-set interval. We estimate that the two automobile batteries (2×12 V 100 AH in series) could power one membrane-enclosed sensor for up to 3 or 6 months, assuming 30 min measurement operations at 2 or 4 h intervals. By measuring pCO_2 at intervals of 2 or 4 h, enough data could be provided for daily average values, accounting for 92 or 85 % of daily pCO_2 variations, respectively, as compared with high-frequency pCO_2 measurements at 10 min intervals (Fig. 8). In inland waters, with low risk of biofouling, the membrane+Cu sensor could withstand an extended monitoring time up to several months, if the temporal resolution were set at hourly scales, considering the trade-off between the time resolution and the increasing power demand.

5 Conclusions

In the laboratory tests and field comparisons at sites encompassing headwater stream, lacustrine, riverine, and estuarine waters with a wide range of pCO_2, the pCO_2 measurements of the three tested equilibration systems agreed well with each other and with the manual headspace equilibration measurements. Both the discrete measurements at 26 sites and the underway measurements along the tidal river reach demonstrated the rapid and accurate responses of the two equilibrator systems to large spatial variations in pCO_2. These results suggest that both equilibrators can perform well during short underway measurements of pCO_2 in isolation, or in combination with cavity-enhanced spectrometric measurements of $\delta^{13}C$ in CO_2 and CH_4. However, further tests are required to determine how long the marbles and the nozzle could remain unaffected by biofouling or clogging during continuous deployments over several hours to days. To address potential clogging and blockage problems of the equilibrators, spare sets of the equilibrator chamber should be prepared during underway measurements. An automated switching between replicate equilibrator chambers at pre-fixed intervals could help to extend the monitoring duration. Although technical challenges, such as power supply and the limited capacity of the desiccant, prevented our equilibrator systems from performing properly during long-term deployment over several days, future studies could explore other types of CO_2 sensors that consumed less power and were more moisture resistant.

The membrane-enclosed sensor exhibited longer response times compared with those of the equilibrators, especially at slow water flow, which is a disadvantage for observing rapid and/or large pCO_2 variations. Nevertheless, this sensor captured the spatial variations of pCO_2 reasonably well within its upper detection limit during the underway measurements along the highly urbanized river reach. This result demonstrates the applicability of the membrane-enclosed sensor for underway pCO_2 measurements, particularly in inland water systems where the spatial variability of pCO_2 is relatively small or gradual. The copper-mesh screening was found efficient for reducing the inaccuracy of the pCO_2 measurements, attributed to the biofouling on the membrane surface, which results from extended deployment in eutrophic water. We suggest that the membrane-enclosed sensor would be suitable for long-term continuous measurements if the sensor had a proper detection range and could be protected by a biofouling-resistant covering.

Although studies on inland water pCO_2 are advancing toward fine-resolution and broad-extent observation, no single approach was able to unveil fully the high spatiotemporal variability encountered in various inland water systems. As the multidisciplinary approach of macrosystems ecology calls for coordinated multiple approaches, in view of the spatiotemporal variability in complex systems (Levy et al., 2014), the results shown here indicate the limitations of the individual monitoring methods. Furthermore, our results suggest that a three-pronged approach should be established to studies on pCO_2 in river systems with strong human influence, namely, coordinated monitoring, involving repeated discrete samplings at multiple sites, long-term monitoring in a few selected sites, and continuous underway measurements along river reaches that have highly variable levels of pCO_2. To better constrain both the natural and the anthropogenic factors that determine spatiotemporal dynamics of CO_2 in diverse inland water systems, equilibration systems need to resolve the high temporal and spatial variability of pCO_2. Although the accuracy of the tested equilibration systems has been validated by our tests and other studies, their applicability to long-term deployment in difficult field conditions, such as limited power supply and biofouling, still requires further rigorous tests.

Acknowledgements

This work was supported by the National Research Foundation of Korea, funded by the Korean Government (2014R1A2A2A01006577). We thank Borami Park and Most Shirina Begum for their assistance with fieldwork and sample analysis. We would like to give special thanks to Gwenaël Abril, David Butman, Will Gagne-Maynard, Mark Johnson, Peter A. Raymond, Jeffrey Richey, and Enrique Sawakuchi for providing us with information on their equilibration systems. We gratefully acknowledge the logistical support provided by the Han River Management Headquarters, Seoul Metropolitan Government.

References

Abril, G., Richard, S., and Guérin, F.: In situ measurements of dissolved gases (CO_2 and CH_4) in a wide range of concentrations in a tropical reservoir using an equilibrator, Sci. Total Environ., 354, 246–251, 2006.Abril, G., Martinez, J.-M., Artigas, L. F., Moreira-Turcq, P., Benedetti, M. F., Vidal, L., Meziane, T., Kim, J.-H., Bernardes, M. C., Savoye, N., Deborde, J., Souza, E. L., Alberic, P., Landim de Souza, M. F., and Roland, F.: Amazon River carbon dioxide outgassing fueled by wetlands, Nature, 505, 395–398, 2014.

Abril, G., Bouillon, S., Darchambeau, F., Teodoru, C. R., Marwick, T. R., Tamooh, F., Ochieng Omengo, F., Geeraert, N., Deirmendjian, L., Polsenaere, P., and Borges, A. V.: Technical Note: Large overestimation of pCO_2 calculated from pH and alkalinity in acidic, organic-rich freshwaters, Biogeosciences, 12, 67–78, 2015.Bastviken, D., Sundgren, I., Natchimuthu, S., Reyier, H., and Gålfalk, M.: Technical Note: Cost-efficient approaches to measure carbon dioxide

(CO_2) fluxes and concentrations in terrestrial and aquatic environments using mini loggers, Biogeosciences, 12, 3849–3859, 2015.

Bakker D. C. E., De Baar H. J. W. and de Wilde H. P. J. Dissolved carbon dioxide in Dutch coastal waters. Mar. Chem., 55, 247–263, 1996.

Battin, T. J., Luyssaert, S., Kaplan, L. A., Aufdenkampe, A. K., Richter, A., and Tranvik, L. J.: The boundless carbon cycle, Nature Geosci., 2, 598–600, 2009.

Bianchi, T. S., Garcia-Tigreros, F., Yvon-Lewis, S. A., Shields, M., Mills, H. J., Butman, D., Osburn, C., Raymond, P., Shank, G. C., DiMarco, S. F., Walker, N., Reese, B. K., Mullins-Perry, R. Quigg, A., Aiken, G. R., and Grossman, E. L.: Enhanced transfer of terrestrially derived carbon to the atmosphere in a flooding event, Geophys. Res. Lett., 40, 116–122, 2013.

Borges, A. V., Darchambeau, F., Teodoru, C. R., Marwick, T. R., Tamooh, F., Geeraert, N., Omengo, F. O., Guerin, F., Lambert, T., Morana, C., Okuku, E., and Bouillon, S.: Globally significant greenhouse-gas emissions from African inland waters, Nature Geosci., 8, 637–642, 2015.

Butman, D. and Raymond, P.A.: Significant efflux of carbon dioxide from streams and rivers in the United States, Nature Geosci., 4, 839–842, 2011.

Cleveland, W. S., and Devlin, S. J.: Locally weighted regression: An approach to regression analysis by local fitting, J. Amer. Statist. Assoc., 83, 596–610, 1988.

Cole, J. J., Prairie, Y. T., Caraco, N. F., McDowell, W. H., Tranvik, L. J., Striegl, R. G., Duarte, C. M., Kortelainen, P., Downing, J. A., Middelburg, J. J., and Melack, J.: Plumbing the global carbon cycle: Integrating inland waters into the terrestrial carbon budget, Ecosystems, 10, 172–185, 2007.

Crawford, J. T., Striegl, R. G., Wickland, K. P., Dornblaser, M. M., and Stanley, E. H.: Emissions of carbon dioxide and methane from a headwater stream network of interior Alaska, J. Geophys. Res. Biogeosci., 118, 482–494, 2013.

Crawford, J. T., Loken, L. C., Casson, N. J., Smith, C., Stone, A. G., and Winslow, L. A.: High-speed limnology: Using advanced sensors to investigate spatial variability in biogeochemistry and hydrology, Environ. Sci. Technol., 49, 442–450, 2015.Dawson, J. J. C., Bakewell, C., and Billett, M. F.: Is in-stream processing an important control on spatial changes in carbon fluxes in headwater catchments?, Sci. Total Environ., 265, 153–167, 2001.

Deacon, E. L.: Gas transfer to and across an air-water interface, Tellus, 29, 363–374, 1977.

Delauney, L., Compère, C., and Lehaitre, M.: Biofouling protection for marine environmental sensors, Ocean Sci., 6, 503–511, 2010.

Dickson, A. G., Sabine, C. L., and Christian, J. R. (Eds.): Guide to best practices for ocean CO_2 measurements, PICES Special Publication 3, Carbon Dioxide Information Analysis Center, Oak Ridge, USA, 2007.

Feely, R. A., Wanninkhof, R., Milburn, H. B., Cosca, C. E., Stapp, M., and Murphy, P.: A new automated underway system for making high precision pCO_2 measurements onboard research ships, Analytica Chimica Acta, 377, 185–191, 1998.

Frankignoulle, M., Abril, G., Borges, A., Bourge, I., Canon, C., Delille, B., Libert, E., and Théate, J.-M.: Carbon dioxide emission from European estuaries. Science, 282: 434–436, 1998.

Frankignoulle, M., Borges, A., and Biondo, R.: A new design of equilibrator to monitor carbon dioxide in highly dynamic and turbid environments, Water Res., 35, 1344–1347, 2001.

Friedrichs, G., Bock, J., Temps, F., Fietzek, P., Körtzinger, A., and Wallace, D. W. R.: Toward continuous monitoring of seawater $^{13}CO_2/^{12}CO_2$ isotope ratio and pCO_2: Performance of cavity ringdown spectroscopy and gas matrix effects, Limnol. Oceanogr. Methods, 8, 539–551, 2010.

Gonzalez-Valencia, R., Magana-Rodriguez, F., Gerardo-Nieto, O., Sepulveda-Jauregui, A., Martinez-Cruz, K., Walter Anthony, K., Baer, D., and Thalasso, F.: In situ measurement of dissolved methane and carbon dioxide in freshwater ecosystems by off-axis integrated cavity output spectroscopy, Environ. Sci. Technol., 48, 11421–11428, 2014.

Griffith, D. R., and Raymond, P. A.: Multiple-source heterotrophy fueled by aged organic carbon in an urbanized estuary, Mar. Chem., 124, 14–22, 2011.

Hales, B., Chipman, D., and Takahashi, T.: High-frequency measurement of partial pressure and total concentration of carbon dioxide in seawater using microporous hydrophobic membrane contactors, Limnol. Oceanogr. Methods, 2, 356–364, 2004.

Hope, D., Dawson, J. J. C., Cresser, M. S., and Billett, M. F.: A method for measuring free CO_2 in upland streamwater using headspace analysis, J. Hydrol., 166, 1–14, 1995.

Hudson, F.: Sample preparation and calculations for dissolved gas analysis in water samples using GC headspace equilibration technique, RSKSOP-175, Revision No. 2, U.S. Environmental Protection Agency, USA, 2004.

Huotari, J., Ojala, A., Peltomaa, E., Nordbo, A., Launiainen, S., Pumpanen, J., Rasilo, T., Hari, P., and Vesala, T.: Long-term direct CO_2 flux measurements over a boreal lake: Five years of eddy covariance data, Geophys. Res. Lett., 38, L18401, 2011.

Huotari, J., Haapanala, S., Pumpanen, J., Vesala, T., and Ojala, A.: Efficient gas exchange between a boreal river and the atmosphere, Geophys. Res. Lett., 40, 5683–5686, 2013.Joesoef, A., Huang, W. J., Gao, Y., and Cai, W. J.: Air–water fluxes and sources of carbon dioxide in the Delaware Estuary: spatial and seasonal variability, Biogeosciences, 12, 6085–6101, 2015.

Johnson, J. E.: Evaluation of a seawater equilibrator for shipboard analysis of dissolved oceanic trace gases, Anal. Chim. Acta., 395, 119–132, 1999.

Johnson, M. S., Billett, M. F., Dinsmore, K. J., Wallin, M., Dyson, K. E., and Jassal, R. S.: Direct and continuous measurement of dissolved carbon dioxide in freshwater aquatic systems – method and applications, Ecohydrology, 3, 68–78, 2010.

Keeling, C. D., Rakestraw, N. W., and Waterman, L. S.: Carbon dioxide in surface waters of the Pacific Ocean: 1. Measurements of the distribution, J. Geophys. Res., 70, 6087–6097, 1965.

Kling, G., Kipphut, G., and Miller, M.: The flux of CO_2 and CH_4 from lakes and rivers in arctic Alaska, Hydrobiologia, 1992.

Lauerwald, R., Hartmann, J., Moosdorf, N., Kempe, S., and Raymond, P. A.: What controls the spatial patterns of the riverine carbonate system? – A case study for North America, Chem. Geol., 337-338, 114–127, 2013.

Leith, F. I., Dinsmore, K. J., Wallin, M. B., Billett, M. F., Heal, K. V., Laudon, H., Ö quist, M. G., and Bishop, K.: Carbon dioxide transport across the hillslope–riparian–stream continuum in a boreal headwater catchment, Biogeosciences, 12, 1881–1892, 2015.

Levy, O., Ball, B. A., Bond-Lamberty, B., Cheruvelil, K. S., Finley, A. O., Lottig, N. R., Punyasena, S. W., Xiao, J., Zhou, J., Buckley, L. B., Filstrup, C. T., Keitt, T. H., Kellner, J. R., Knapp, A. K., Richardson, A. D., Tcheng, D., Toomey, M., Vargas, R., Voordeckers, J. W., Wagner, T., and Williams, J. W.: Approaches to advance scientific understanding of macrosystems ecology, Front. Ecol. Environ., 12, 15–23, 2014.

Lewis, E., Wallace, D., and Allison, L. J.: Program developed for CO_2 system calculations, Carbon Dioxide Information Analysis Center, Lockheed Martin Energy Research Corporation for the US Department of Energy: Tennessee, USA, 1998.

Li, S., Lu, X. X., and Bush, R. T.: CO_2 partial pressure and CO_2 emission in the Lower Mekong River, J. Hydrol., 504, 40–56, 2013.

Liss, P. S., and Slater, P. G.: Flux of gases across the air-sea interface, Nature, 247, 181–184, 1974.

Lorke, A., Bodmer, P., Noss, C., Alshboul, Z., Koschorreck, M., Somlai-Haase, C., Bastviken, D., Flury, S., McGinnis, D. F., Maeck, A., Müller, D., and Premke, K.: Technical Note: Drifting versus anchored flux chambers for measuring greenhouse gas emissions from running waters, Biogeosciences, 12, 7013–7024, 2015.

Maher, D. T., Santos, I. R., Leuven, J. R. F. W., Oakes, J. M., Erler, D. V., Carvalho, M. C., and Eyre, B. D.: Novel use of cavity ring-down spectrometry to investigate aquatic carbon cycling from microbial to ecosystem scales. Environ. Sci. Technol., 47, 12938–12945, 2013.

Manov, D. V., Chang, G. C., and Dickey, T. D.: Methods for reducing biofouling of moored optical sensors, J. Atmos. Oceanic Tech., 21, 958–968, 2004.

Nimick, D. A., Gammons, C. H., and Parker, S. R.: Diel biogeochemical processes and their effect on the aqueous chemistry of streams: A review, Chem. Geol., 283, 3–17, 2011.

Park, P. K., Gordon, L. I., Hager, S. W., and Cissell, M. C.: Carbon dioxide partial pressure in the Columbia River, Science, 166, 867–868, 1969.

Peter, H., Singer, G. A., Preiler, C., Chifflard, P., Steniczka, G., and Battin, T. J.: Scales and drivers of temporal pCO_2 dynamics in an alpine stream, J. Geophys. Res. Biogeosci., 119, 1078–1091, 2014.

Pfeil, B., Olsen, A., Bakker, D. C. E., Hankin, S., Koyuk, H., Kozyr, A., Malczyk, J., Manke, A., Metzl, N., Sabine, C. L., Akl, J., Alin, S. R., Bates, N., Bellerby, R. G. J., Borges, A., Boutin, J., Brown, P. J., Cai, W. J., Chavez, F. P., Chen, A., Cosca, C., Fassbender, A. J., Feely, R. A., González-Dávila, M., Goyet, C., Hales, B., Hardman-Mountford, N., Heinze, C., Hood, M., Hoppema, M., Hunt, C. W., Hydes, D., Ishii, M., Johannessen, T., Jones, S. D., Key, R. M., Körtzinger, A., Landschützer, P., Lauvset, S. K., Lefèvre, N., Lenton, A., Lourantou, A., Merlivat, L., Midorikawa, T., Mintrop, L.,

Miyazaki, C., Murata, A., Nakadate, A., Nakano, Y., Nakaoka, S., Nojiri, Y., Omar, A. M., Padin, X. A., Park, G. H., Paterson, K., Perez, F. F., Pierrot, D., Poisson, A., Ríos, A. F., Santana-Casiano, J. M., Salisbury, J., Sarma, V. V. S. S., Schlitzer, R., Schneider, B., Schuster, U., Sieger, R., Skjelvan, I., Steinhoff, T., Suzuki, T., Takahashi, T., Tedesco, K., Telszewski, M., Thomas, H., Tilbrook, B., Tjiputra, J., Vandemark, D., Veness, T., Wanninkhof, R., Watson, A. J., Weiss, R., Wong, C. S., and Yoshikawa-Inoue, H.: A uniform, quality controlled Surface Ocean CO_2 Atlas (SOCAT), Earth Syst. Sci. Data, 5, 125–143, 2013.

Pierrot, D., Neill, C., Sullivan, K., Castle, R., Wanninkhof, R., Lüger, H., Johannessen, T., Olsen, A., Feely, R. A., and Cosca, C. E.: Recommendations for autonomous underway pCO_2 measuring systems and data-reduction routines, Deep-sea Res. PT II, 56, 512–522, 2009.

Podgrajsek, E., Sahlée, E., Bastviken, D., Holst, J., Lindroth, A., Tranvik, L., and Rutgersson, A.: Comparison of floating chamber and eddy covariance measurements of lake greenhouse gas fluxes, Biogeosciences, 11, 4225–4233, 2014.

Polsenaere, P., Deborde, J., Detandt, G., Vidal, L. O., Pérez, M. A. P., Marieu, V., and Abril, G.: Thermal enhancement of gas transfer velocity of CO_2 in an Amazon floodplain lake revealed by eddy covariance measurements, Geophys. Res. Lett., 40, 1734–1740, 2013.

R Development Core Team: R: A language and environment for statistical computing; R Foundation for Statistical Computing: Vienna, Austria, 2011, available at: http://www.r-project.org.

Raymond, P. A., and Cole, J. J.: Gas exchange in rivers and estuaries: Choosing a gas transfer velocity, Estuaries, 24, 312–317, 2001.

Raymond, P. A., and Hopkinson, C. S.: Ecosystem modulation of dissolved carbon age in a temperate marsh-dominated estuary, Ecosystems, 6, 694–705, 2003.

Raymond, P. A., Zappa, C. J., Butman, D., Bott, T. L., Potter, J., Mulholland, P., Laursen, A. E., McDowell, W. H., and Newbold, D.: Scaling the gas transfer velocity and hydraulic geometry in streams and small rivers, Limnol. Oceanogr. Fluids Environ., 2, 41–53, 2012.

Raymond, P. A., Hartmann, J., Lauerwald, R., Sobek, S., McDonald, C., Hoover, M., Butman, D., Striegl, R., Mayorga, E., Humborg, C., Kortelainen, P., Durr, H., Meybeck, M., Ciais, P., and Guth, P.: Global carbon dioxide emissions from inland waters, Nature, 503, 355–359, 2013Santos, I. R., Maher, D. T., and Eyre, B. D.: Coupling automated radon and carbon dioxide measurements in coastal waters, Environ. Sci. Technol., 46, 7685–7691, 2012.

Smethie, W. M., Takahashi, T., Chipman, D. W., and Ledwell, J. R.: Gas exchange and CO_2 flux in the tropical Atlantic Ocean determined from ^{222}Rn and pCO_2 measurements, J. Geophys. Res. Oceans, 90, 7005–7022, 1985.

Swinnerton, J. W., Cheek, C. H., and Linnenbom, V. J.: Determination of dissolved gases in aqueous solutions by gas chromatography. Anal. Chem., 34, 483−485, 1962.

Takahashi, T.: Carbon dioxide in the atmosphere and in Atlantic Ocean water, J. Geophys. Res., 66, 477–494, 1961.Teodoru, C., Prairie, Y., and del Giorgio, P.: Spatial heterogeneity of surface CO_2 fluxes in a newly created Eastmain-1 reservoir in northern Quebec, Canada, Ecosystems, 14, 28–46, 2011.

Teodoru, C. R., Nyoni, F. C., Borges, A. V., Darchambeau, F., Nyambe, I., and Bouillon, S.: Dynamics of greenhouse gases (CO_2, CH_4, N_2O) along the Zambezi River and major tributaries, and their importance in the riverine carbon budget, Biogeosciences, 12, 2431–2453, 2015.

Troxler, T. G., Barr, J. G., Fuentes, J. D., Engel, V., Anderson, G., Sanchez, C., Lagomasino, D., Price, R., and Davis, S. E.: Component-specific dynamics of riverine mangrove CO_2 efflux in the Florida Coastal Everglades, Agr. For. Meteorol., 213, 273–282, 2015.

Vachon, D., Prairie, Y. T., and Cole, J. J.: The relationship between near-surface turbulence and gas transfer velocity in freshwater systems and its implications for floating chamber measurements of gas exchange, Limnol. Oceanogra., 55, 1723–1732, 2010.

Valsala, V., and Maksyutov, S.: Simulation and assimilation of global ocean pCO_2 and air–sea CO_2 fluxes using ship observations of surface ocean pCO_2 in a simplified biogeochemical offline model, Tellus B, 62, 16640, 2010.

Wanninkhof, R.: Relationship between wind speed and gas exchange over the ocean, J. Geophys. Res. Oceans, 97, 7373–7382, 1992.

Wanninkhof, R., Asher, W. E., Ho, D. T., Sweeney, C., and McGillis, W. R.: Advances in quantifying air-sea gas exchange and environmental forcing, Ann. Rev. Mar. Sci., 1, 213–244, 2009.

Webb, J. R., Maher, D. T., and Santos, I. R.: Automated, in situ measurements of dissolved CO_2, CH_4, and $\delta^{13}C$ values using cavity enhanced laser absorption spectrometry: Comparing response times of air-water equilibrators, Limnol. Oceanogr. Methods, 14, 323–337, 2016.

YSI Incorporated: Using a copper-alloy based system for effective biofouling deterrence, A583 0410, YSI Incorporated, Yellow Springs, USA, 2010.

Zhai, W., Dai, M., Cai, W.-J., Wang, Y., and Wang, Z.: High partial pressure of CO_2 and its maintaining mechanism in a subtropical estuary: the Pearl River estuary, China, Mar. Chem., 93, 21–32, 2005.

Tables

Table 1. Summary of manual headspace equilibration and three gas equilibration systems

System	Principle	Equilibration method	Equilibration time (min)	References
Manual equilibration	Manual; active	Gas equilibration in the headspace over the water sample collected in a bottle or syringe by manual shaking	< 2	Kling et al., 1992; Hope et al., 1995
Spray-type equilibrator	Automatic; active	Enhanced gas equilibration by spraying gas-containing water droplets	1–12	Freely et al., 1998; Webb et al., 2016
Marble-type equilibrator	Automatic; active	Enhanced gas exchange over the large cumulative surface of marbles	1–13	Frankignoulle et al., 2001; Abril et al., 2006
Membrane-enclosed sensor	Automatic; passive	Diffusion-based "passive" equilibration between the inside and outside of the water-impermeable, gas-permeable membrane	> 10	Johnson et al., 2010

Figures

Figure 1. Schematic diagrams of four equilibration methods: (a) manual headspace equilibration; (b) marble-type equilibrator; (c) spray-type equilibrator; (d) membrane-enclosed sensor. Refer to Table 1 for descriptions of basic operation principles, and Figures S1 and S2 for pictures showing the three gas equilibration systems employed during field tests.

Figure 2. Laboratory cross-validation tests of three gas equilibration systems during (a) low-to-high and (b) high-to-low equilibration.

Figure 3. Comparison of pCO_2 measurements of three gas equilibration systems (spray- and marble-type equilibrators and membrane-enclosed sensor) with the manual headspace equilibration at various inland waters, ranging from forested headwater streams to the estuary of the Han River.

Figure 4. Relationship between the response time (t_{100}) and $\Delta\ pCO_2$, as the difference between the initial and the stabilized final pCO_2 measurement, for the (a) spray-type equilibrator, (b) marble-type equilibrator, and (c) membrane-enclosed sensor. Note that the X-axis has a log scale. The solid and dashed lines indicate significant and insignificant relationships, respectively.

Figure 5. Continuous underway measurements of pCO_2, using a spray-type equilibrator, a marble-type equilibrator, and a membrane-enclosed sensor along the tidal reach of the Han River from 11 am to 3 pm, 11 May 2015. The inflow of urban streams containing high loads of organic matter, inorganic nutrients, and CO_2 is indicated by brown arrows. The pCO_2 measurements, using manual headspace equilibration (yellow circle) were performed on-board. Note that the membrane-enclosed sensors did not capture drastic increases in pCO_2 after midday because of the upper detection limit of the sensors (7,000 or 10,000 ppm).

Figure 6. Continuous pCO_2 measurements at a tidal reach of the Han River, using membrane-enclosed sensors without ("membrane" sensor) and with copper-mesh screening ("membrane+Cu" sensor), with (a) Original measurements (gray arrows indicate maintenance timing); (b) Relative differences between the log-transformed measurements by the two sensors; (c) The relationship between the daily means of pCO_2 measurements by the two sensors.

Figure 7. (a) The relationship between pH and pCO_2 during successive four-day monitoring periods following maintenance; (b) the relationship between coefficient of variations (CVs) of daily means of pH and pCO_2. Membrane-enclosed sensors without and with copper-mesh screening are indicated by "membrane" and "membrane+Cu", respectively. Curves were fitted using LOESS (locally weighted scatterplot smoothing).

Figure 8. Temporal resolution effects on pCO_2 measurements by a membrane-enclosed sensor with a copper-mesh covering. The data obtained from the copper-mesh-wrapped sensor in Figure 7 are presented by modifying temporal resolutions from 10 min to 4 h (a). The bottom panel shows the daily mean, minimum, and maximum pCO_2 values normalized to the mean of the four calculations (b).

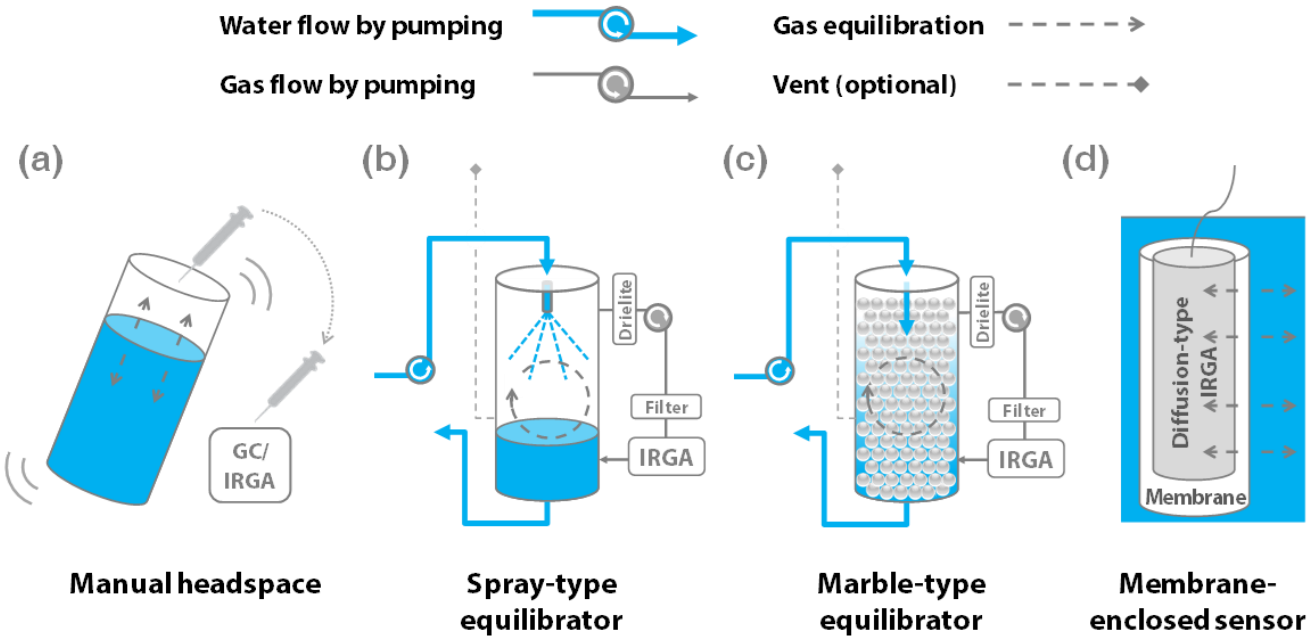
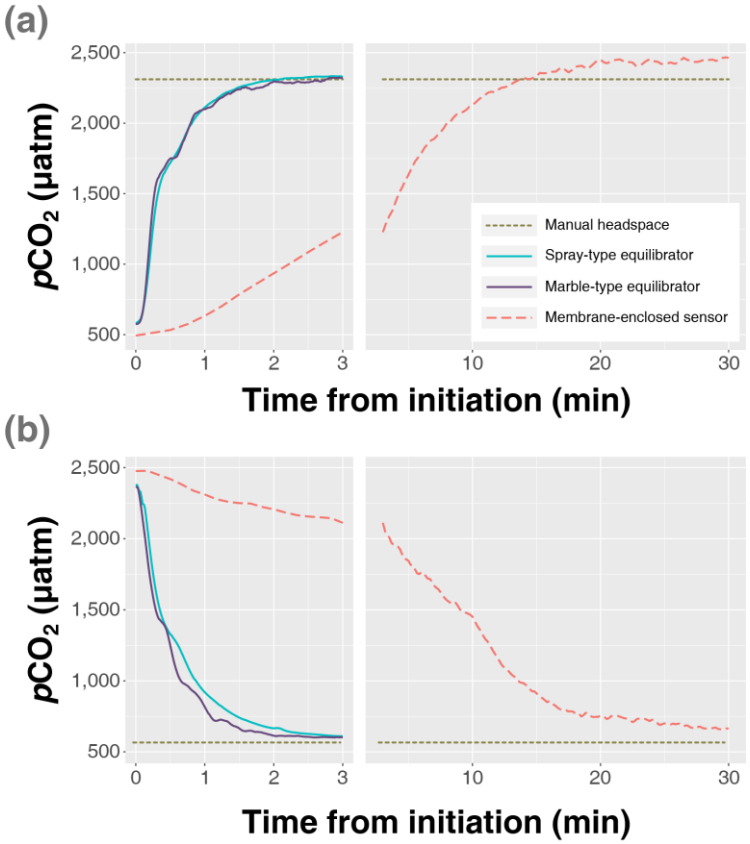

Water flow by pumping
Gas flow by pumping
Gas equilibration
Vent (optional)
(a)
(b)
(c)
(d)
GC/
IRGA
Drielite
Filter
IRGA
Drielite
Filter
IRGA
Diffusion-type
IRGA
Membrane
Manual headspace
Spray-type
equilibrator
Marble-type
equilibrator
Membrane-
enclosed sensor

Figure 1.


(a)
2,500
2,000
1,500
1,000
500
pCO2 (µatm)
0
1
2
3
10
20
30
Time from initiation (min)
Manual headspace
Spray-type equilibrator
Marble-type equilibrator
Membrane-enclosed sensor
(b)
2,500
2,000
1,500
1,000
500
pCO2 (µatm)
0
1
2
3
10
20
30
Time from initiation (min)

715

Figure 2.

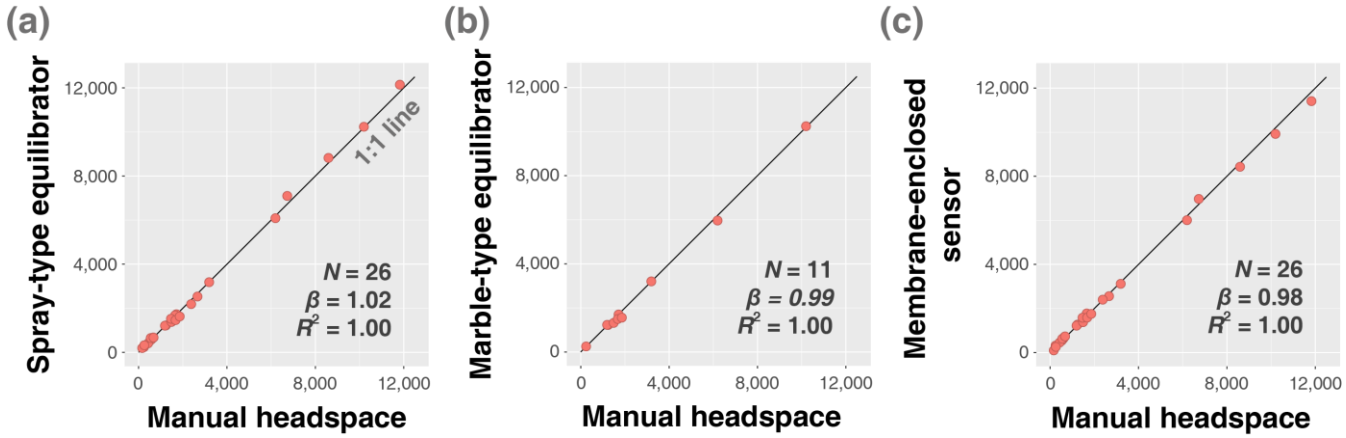

(a)
Spray-type equilibrator
12,000
8,000
4,000
0
1:1 line
N = 26
β = 1.02
R2 = 1.00
0
4,000
8,000
12,000
Manual headspace
(b)
Marble-type equilibrator
12,000
8,000
4,000
0
N = 11
β = 0.99
R2 = 1.00
0
4,000
8,000
12,000
Manual headspace
(c)
Membrane-enclosed sensor
12,000
8,000
4,000
0
N = 26
β = 0.98
R2 = 1.00
0
4,000
8,000
12,000
Manual headspace

Figure 3.

720

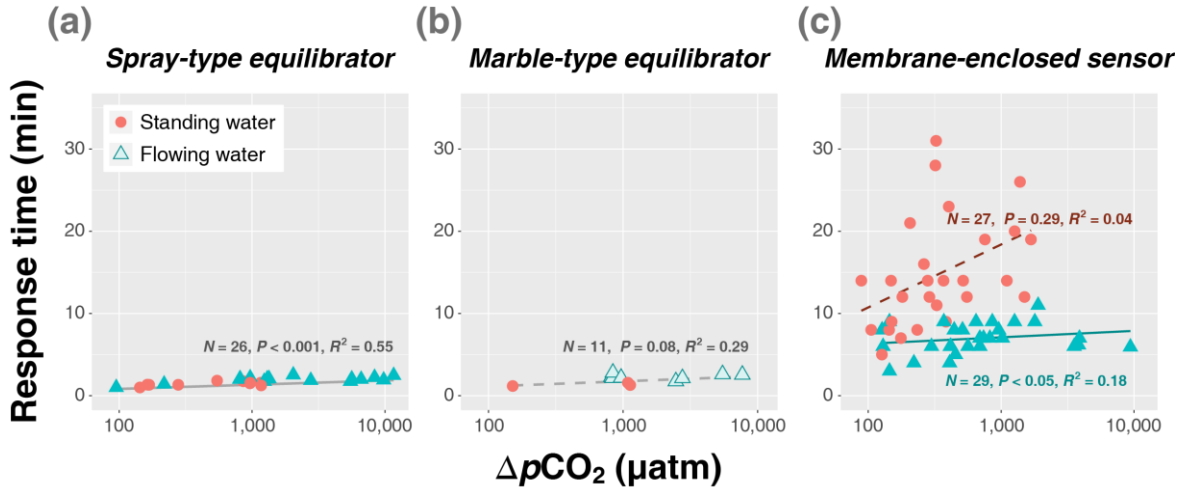
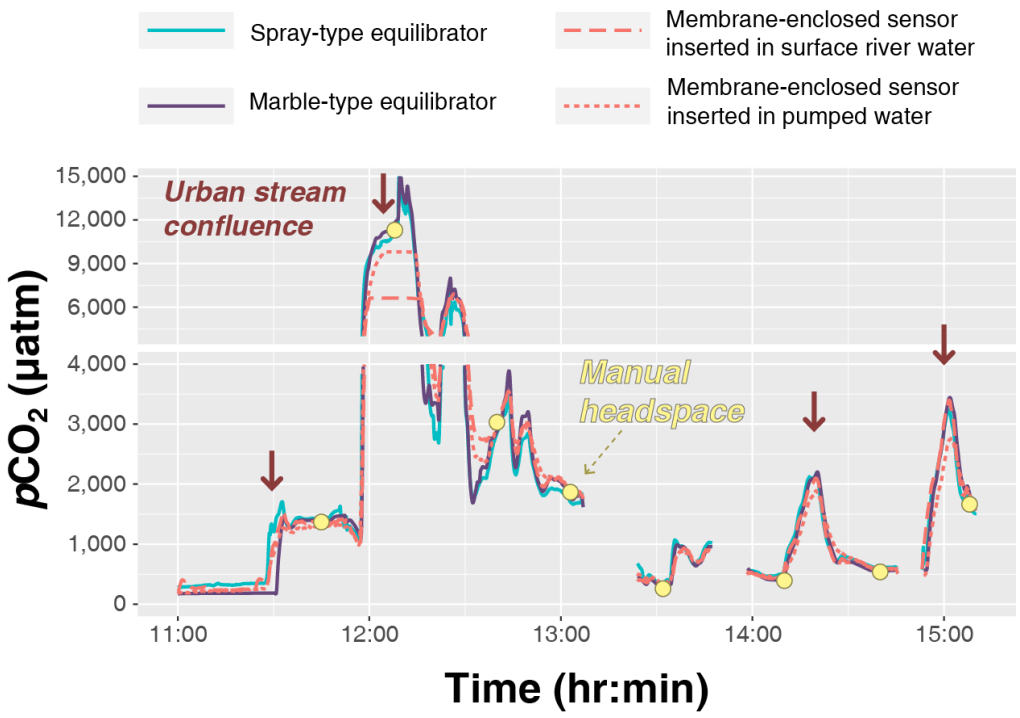

(a)
Spray-type equilibrator
(b)
Marble-type equilibrator
(c)
Membrane-enclosed sensor
Response time (min)
Standing water
Flowing water
N = 26, P < 0.001, R² = 0.55
N = 11, P = 0.08, R² = 0.29
N = 27, P = 0.29, R² = 0.04
N = 29, P < 0.05, R² = 0.18
0
10
20
30
100
1,000
10,000
ΔpCO₂ (µatm)

Figure 4.


Spray-type equilibrator
Marble-type equilibrator
Membrane-enclosed sensor inserted in surface river water
Membrane-enclosed sensor inserted in pumped water
Urban stream confluence
Manual headspace
pCO2 (µatm)
15,000
12,000
9,000
6,000
4,000
3,000
2,000
1,000
0
11:00
12:00
13:00
14:00
15:00
Time (hr:min)

725 **Figure 5.**

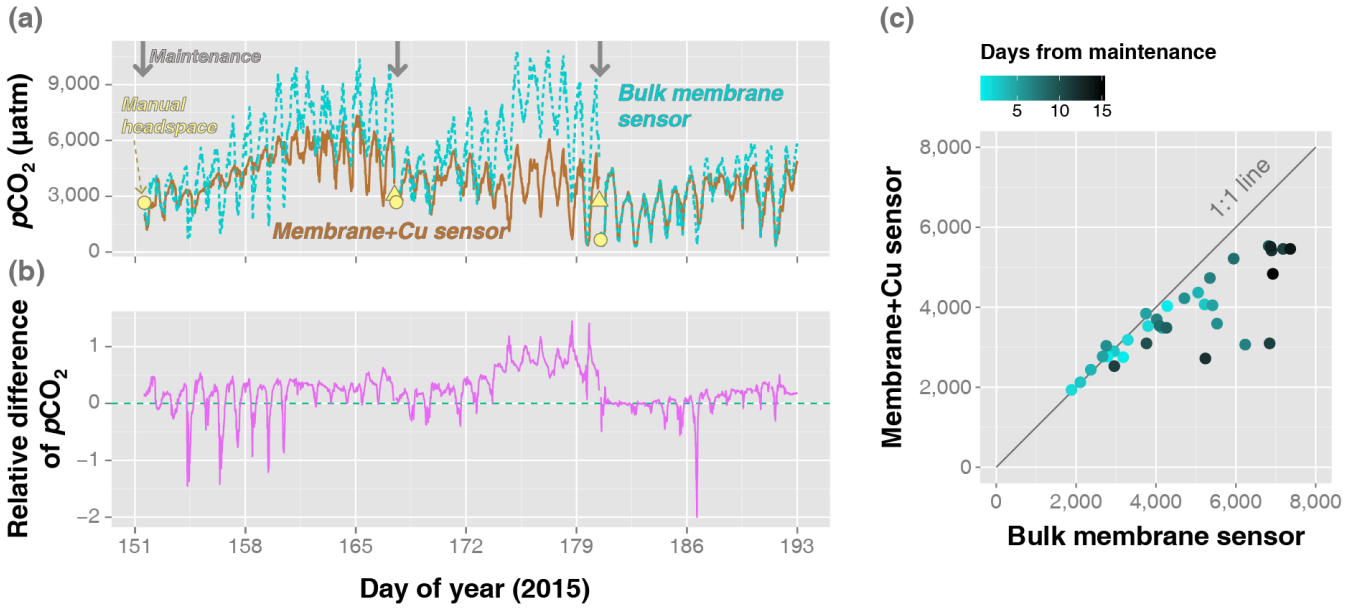
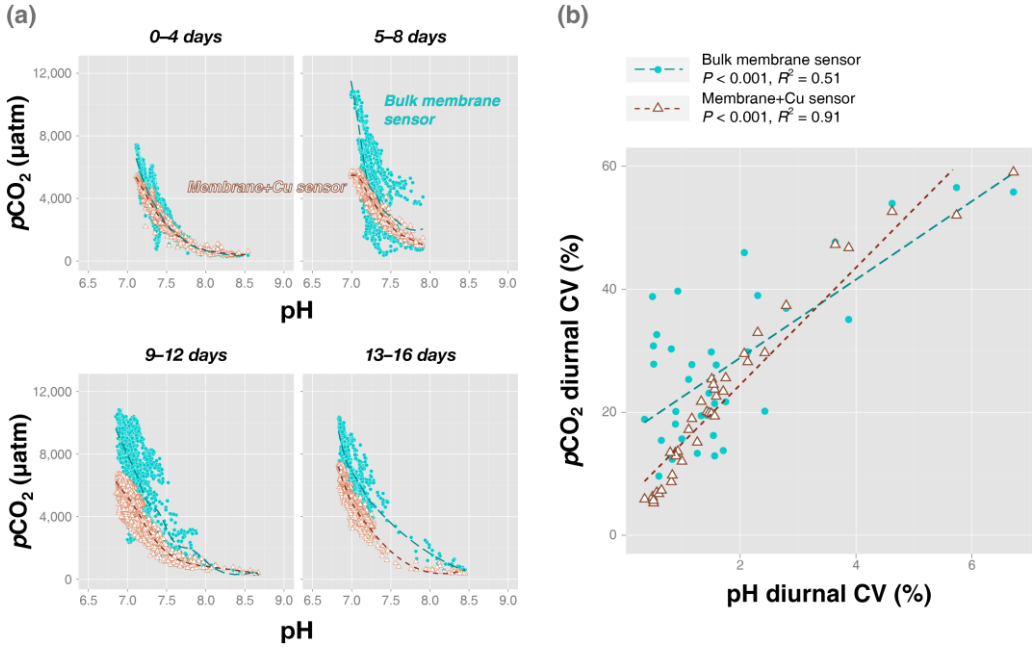

(a)
Maintenance
Manual headspace
Bulk membrane sensor
Membrane+Cu sensor
pCO2 (µatm)
9,000
6,000
3,000
0
(b)
Relative difference of pCO2
1
0
−1
−2
151
158
165
172
179
186
193
Day of year (2015)
(c)
Days from maintenance
5
10
15
Membrane+Cu sensor
8,000
6,000
4,000
2,000
0
1:1 line
0
2,000
4,000
6,000
8,000
Bulk membrane sensor

Figure 6.


(a)
0–4 days
5–8 days
9–12 days
13–16 days
Bulk membrane sensor
Membrane+Cu sensor
pCO2 (µatm)
pH
(b)
Bulk membrane sensor
P < 0.001, R2 = 0.51
Membrane+Cu sensor
P < 0.001, R2 = 0.91
pCO2 diurnal CV (%)
pH diurnal CV (%)

730

Figure 7.

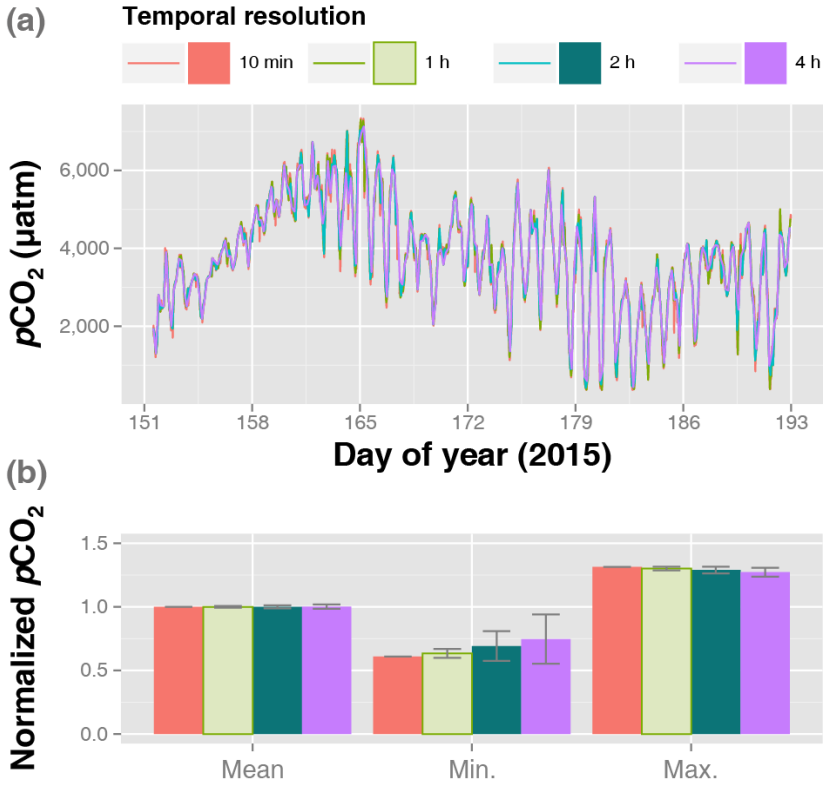

(a)
Temporal resolution
10 min
1 h
2 h
4 h
6,000
4,000
2,000
pCO2 (µatm)
151
158
165
172
179
186
193
Day of year (2015)
(b)
Normalized pCO2
1.5
1.0
0.5
0.0
Mean
Min.
Max.

Figure 8.