



# Development and evaluation of a self-cleaning custom-built auto sampler controlled by a low-cost RaspberryPi microcomputer for online enzymatic activity measurements



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## ABSTRACT

A fully automated on-site device (SAMP-FIL) that enables water sampling with simultaneous filtration and effective cleaning procedures of the device's components was developed and field-tested. The SAMP-FIL was custom-built using commercially available components and was controlled by a RaspberryPi single-board computer operating open-source software. SAMP-FIL was designed for sample pre-treatment with minimal sample alteration to meet the requirements of on-site measurement devices that cannot handle coarse suspended solids within the measurement procedure or cycle. A highly effective cleaning procedure provides a fresh and minimally altered sample for the connected measurement device. The construction and programmed software facilitates the use of SAMP-FIL for different connected measurement devices. The SAMP-FIL sample pretreatment was tested for over one year for rapid and on-site enzymatic activity (beta-D-glucuronidase, GLUC) determination (BACTcontrol) in sediment-laden stream water. The formerly used proprietary sampling set-up was assumed to lead to significant damping of the measurement signal due to its susceptibility to clogging, debris accumulation and bio-film accumulation. The implementation of SAMP-FIL considerably increased the error-free running time and measurement accuracy of BACTcontrol devices. This paper describes how low-cost microcomputers, such as the RaspberryPi, can be used by operators to substantially improve established measuring systems via effective sampling devices. Furthermore, the results of this study highlight the importance of adequate sample pretreatment for the quality of on-site measurements.

## 1. Introduction

On-site monitoring of chemo-physical and bio-chemical parameters in surface waters are currently standard procedures in various fields, such as hydrology, limnology and civil engineering. Although technological progress and scientific questions have advanced on-site monitoring of water resources to higher temporal and spatial resolutions, measurement systems are still technically challenged by common environmental factors, such as suspended solid concentrations. Emerging parameters to be monitored on-site (e.g., enzymatic activity) require measurement devices with complex construction design, including valves and hoses with diminutive apertures [16,23,26,27,32]. The effect of suspended organic and inorganic matter on the accuracy of measurement results and the device running time of such methods

are particularly high in stream draining catchments susceptible to soil erosion (e.g., agricultural catchments). In such cases, sample pretreatment becomes an unavoidable component of the procedure [27] to meet the technical requirements of the measuring devices and prevent valves or tubing from clogging. The required sample pretreatment procedure includes filtering of the natural water sample and filling a vessel from which the connected instruments draw the sample for the intended measurement. As manufacturers may have limited insight and comprehension into the determining factors of specific monitoring locations, proprietary solutions occasionally do not meet the specific demands necessary to enable the optimal operation of the respective measurement instruments. Pre-assembled and commercially available modules for filtration, which allow back flushing and filter cleaning in combination with adequate pumping, can be used for on-site sample

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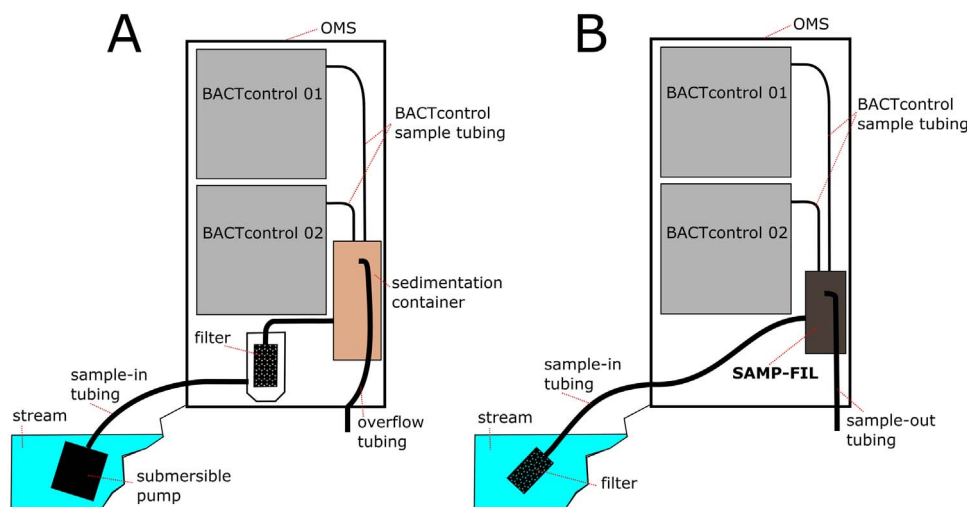
<sup>2</sup> <http://www.waterandhealth.at>

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**Fig. 1.** Schematic of the basic sample pretreatment set-ups and fundamental components of the outdoor monitoring station (OMS). A: The proprietary set-up with the constantly operated submersible pump located in the stream, filter (cartridge enclosed in housing) and sedimentation container. B: Set-up with SAMP-FIL, where the filter cartridge is located in the stream and sample water is provided to the BACTcontrol devices by SAMP-FIL in coordination with their measurement intervals.

pretreatment. These modules are often limited in terms of sampling volume and filter size or have a fixed filter mesh width and thus have strong constraints regarding the conversion of the set-up for other purposes (e.g., connection to another measurement instrument). Furthermore, such modules may provide considerable inside surfaces and cavities for debris and bio-film accumulation, leading to potential adulteration of the measurement signal.

Several types of low-cost microcomputers (e.g., Arduino, RaspberryPi) were released during the last decade and are currently often used within a wide user area of the open-source community [1,14,18,22,29]. The possibility to connect and control peripheral devices (e.g., sensors, relays) due to embedded ports (e.g., USB) or general-purpose input/output (GPIO) and the ability to operate self-programmed scripts (e.g., Python) have great potential within the field of environmental science, particularly regarding measurement engineering and monitoring. Recent studies have described the application of low-cost single-board computers for implementation in monitoring systems, e.g., as a strategic interface or data-logger [14,24,28,30,31]. However, there is a lack of scientific literature describing the selection and assembly of hardware and software in a comprehensive manner to enable the reader to reproduce the same or similar set-ups.

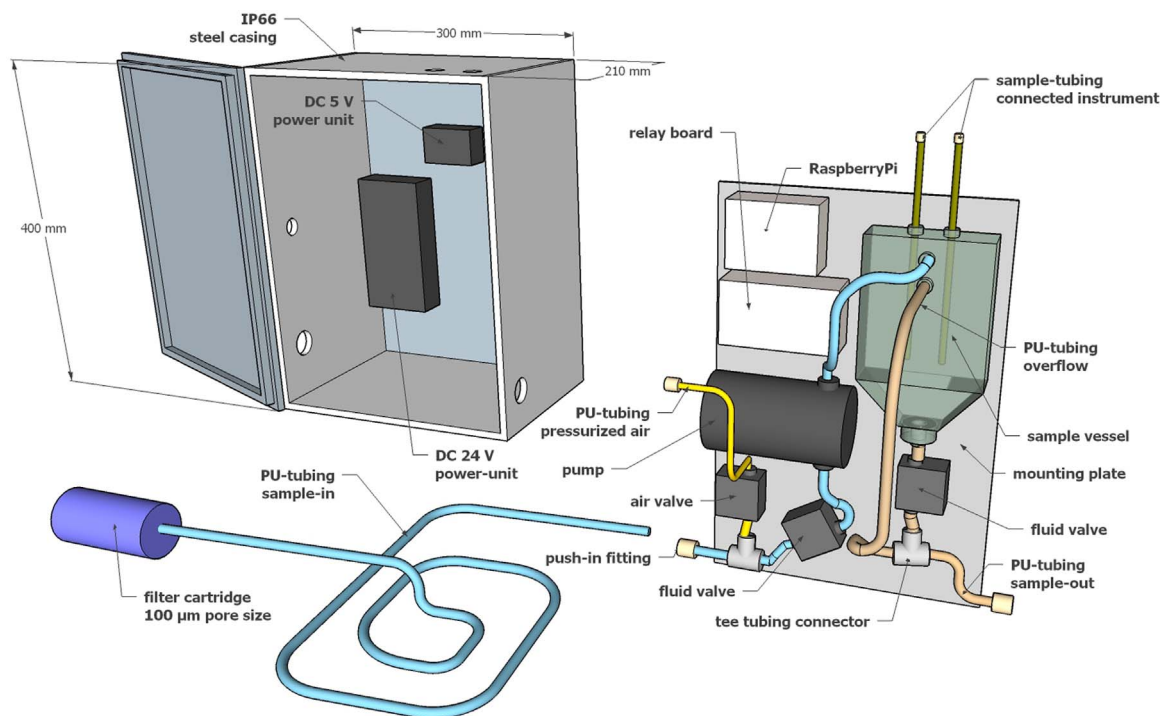
The authors of this paper describe how a robust and effective RaspberryPi controlled autosampler for sample pretreatment, necessary for the on-site enzymatic activity determination in surface water, was developed in a step-by-step manner. Instructions from open-source platforms (e.g., [www.raspberrypi.org/forums](http://www.raspberrypi.org/forums) [20]) have been adapted to this special demand, and the construction was designed following the authors' operating experience of automated on-site enzymatic activity determination in surface water.

The main objective was to construct a programmable sample pretreatment set-up that has an insignificant impact on the natural water sample and is resistant against in-device debris and bio-film accumulation. The hardware components and programmed software are selected and constructed in a manner that easily enables the connection of the SAMP-FIL to another on-site instrument by modification of the sample volume, filter size and operation cycles. A 16-month field campaign was conducted to test the constructed device (SAMP-FIL) in terms of its robustness for continuous long-term automated operation, its impact on measurement accuracy, and the running time of the connected instruments (BACTcontrol).

## 2. Material and methods

On-site detection of enzymatic activities has been suggested as a rapid surrogate for the monitoring of microbiological pollution in water resources [5,9,10,12]. Due to the potential short measuring intervals, this method has high potential as a near-real-time water quality monitoring tool and can contribute important information for identifying faecal contamination. To understand the dynamics and transport processes of faecal-associated contamination in stream water, two devices (BACTcontrol, MicroLAN, Netherlands) for the automated and rapid determination of enzymatic activity (beta-D-glucuronidase) were operated for stream water monitoring at the catchment outlet since 2012. During the measurement process, the sample mixed with specific assay reagents generated an increasing fluorescence signal that reflected the level of enzymatic activity, which was monitored over time. The construction design and sampling and measurement procedures of the BACTcontrol devices have been described in detail by [23,32]. Although the measurement principle yielded consistent long-term data from automated on-site operation in ground water monitoring [23], the use of these instruments for surface water monitoring is a technical challenge due to the high suspended solid concentrations during event run off conditions. Outages of BACTcontrol devices due to technical failure or service are substantially higher when operated in surface water monitoring compared to ground water monitoring [23,27]. To establish long-term on-site operation, in this case, sample pretreatment (filtration through 100  $\mu\text{m}$ ) was an unavoidable step [27]. The BACTcontrol instruments conducted measurements every three hours and were housed in an air-conditioned measurement station situated at the brookside, one meter above the water level.

The proprietary sample pretreatment set-up (Fig. 1A) was based on a submersible pump that was placed in the stream and constantly pumping water with a flow rate of 1.5 l/min through a flow-through housing with a 100  $\mu\text{m}$  filter into a 10 l sample container. The sample container was equipped with an overflow, enabling a constant flow-through and complete exchange of the pre-treated water within less than 7 min. The dimensions and flow-through rates were designed to enable sedimentation of fine material (< 100  $\mu\text{m}$ ) within the container. The BACTcontrol devices obtained the required sample volume (100 ml) for the GLUC measurements from this sample container every 180 min (Fig. 1A). The filter cartridge and hoses had to be manually cleaned on a biweekly basis. The components of this set-up were designed for long-term usage and proved technically robust, with failure-free running times up to 12 months. Nevertheless, evaluation of



**Fig. 2.** Construction plan of the SAMP-FIL hardware. Core components are installed on a detachable mounting plate (right). The IP66 casing houses this plate and the power units. Connections for the sample-in tubing, sample-out tubing, pressurized air tubing and main power are made on the casing. Sample tubing from the connected devices (BACTcontrol) is similarly linked via push-in fittings through the casing.

the continuously measured GLUC signal showed that a majority of GLUC measurements were delayed up to several hours from the hydrological parameters monitored in parallel, such as the stream discharge and turbidity. In particular, during event runoff conditions, the GLUC activity is assumed to be strongly correlated with discharge and turbidity, as these parameters indicate the potential input and transport of surface-associated faecal pollution in the stream. Furthermore, the monitored peaks of GLUC activity appeared to be significantly damped in many cases. The described phenomena occurred despite regular cleaning of the filter cartridge and hoses, indicating that substantial parts of the inner surfaces of the components (e.g., pump, fittings, tubing) are not accessible for the on-site and manual cleaning procedure and that the constant water flow led to bio-film growth, deposition of debris and sintering, causing the retention and delayed release of beta-D-glucuronidase-producing organisms (e.g., *E. coli*) into the measuring device.

### 2.1. Test site

The Hydrological Open Air Laboratory (HOAL, [2,3]) in Petzenkirchen (Lower Austria) is operated and maintained by the Institute for Land and Water Management Research (*Federal Agency for Water Management, Austria*) and the Vienna Doctoral Programme of Water Resource Systems (*Center for Water Resource Systems, Vienna University of Technology, Austria*). The catchment is 0.66 km<sup>2</sup> in area and is drained by a 620 m stream. Twelve point discharges contribute to the discharge of the stream. These include tile drains, springs and surface tributaries (Exner-Kittridge et al., [33]). The mean annual precipitation is 823 mm/yr (1990–2014). The land use of the catchment area is dominated by agriculture, consisting of 83% arable land, 7% grassland, 7% forested area, and 3% paved surface. The hydrogeology is characterized by porous and fissured aquifers consisting of clay, marl and sand. Soils show medium to limited infiltration capacities. The annual sediment erosion is approximately 1 t/ha (Eder et al., [35]). The monitored stream shows high discharge dynamics (minimum discharge 2014: 0.5 l/s, maximum discharge 2014: 73 l/s)

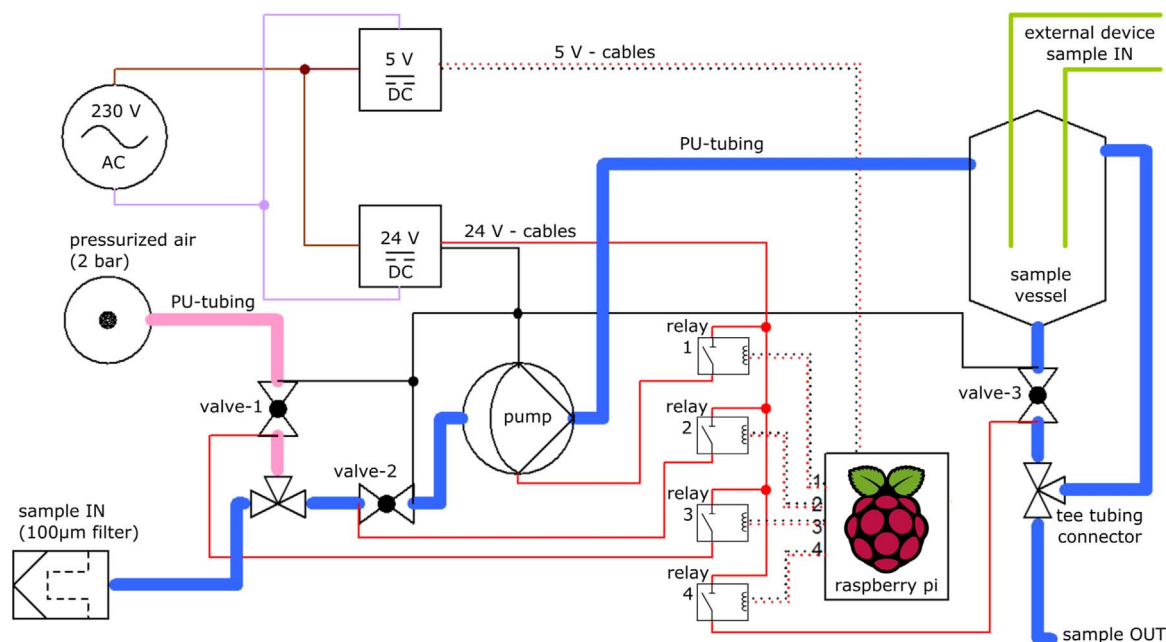
with rapid reactions during rain events. The turbidity in the monitored stream is highly event-linked, as rain events promptly cause an increase of suspended solids (TSS) in the stream water. Maximum TSS concentrations of over 3 g/l were recorded during the test period in July 2014 and January 2015. Depending on the hydrologic condition of the catchment, the stream water turbidity is a diverging combination of eroded sediment flushed into the stream by surface runoff and re-suspended riverbed sediment (Eder et al., [34]). The main source of faecal contamination of ground and surface water is swine manure applied periodically to the fields.

### 2.2. Design and construction

To overcome the problem of suboptimal sample pretreatment, a compact and robust auto sampler (SAMP-FIL) that samples and provides a filtered water sample scheduled to the measurement intervals of the connected BACTcontrol devices was constructed (Fig. 1B). The objective was to conduct the sampling and filtering procedure as quickly as possible to minimize the retention time and adulteration of the sampled water. The flow-through rates were kept high to achieve turbulent pipe-flow to minimize debris and bio-film accumulation within the device. Flushing out the complete equipment with pressurized air enabled filter cleaning and allowed the equipment to dry while idle. To schedule the time sequence of the array of required components, a RaspberryPi single-board computer was chosen because the guidance support by the on-line community is high, it allows on-device programming and it has been reported to be robust for numerous long-term operations [13]. All components of the SAMP-FIL are mounted within a casing of manageable size, where the sampling tubing with the filter, outlet and BACTcontrol devices are connected (Fig. 2).

### 2.3. Hardware

The SAMP-FIL (Fig. 2) was housed in a commercial compact steel enclosure (Rittal, AE 1034.5, width: 300 mm, height: 400 mm, depth:



**Fig. 3.** Schematic diagram of the current lines, tubing and core components of the SAMP-FIL. Fluid tubing is marked in blue, pressurized air tubing is marked in pink and the sample-in tubing of the connected BACTcontrol devices is marked in green. Dashed red and black lines indicate 5 V cables, and solid red and black lines indicate 24 V cables. Brown and purple lines show 230 V main power connections.

210 mm) with protection category IP 66. The sampling was conducted using a rotary diaphragm pump (Charles Austen, RD5 DC, suction pump, flow rate: 5 l/min, empty lift: 8 m, DC: 24 V). Sample inlets and outlets as well as pressurized air inlets were controlled by an array (Fig. 3) of two fluid control valves (SMC Pneumatics, VX 232, 2-port solenoid valve, normally closed, nominal diameter: 8 mm, DC: 24 V) and one air control valve (SMC Pneumatics, VX 220, 2-port solenoid valve, normally closed, nominal diameter: 6 mm, DC: 24 V).

Both fluid- and air control valves operate on DC 24 V. Pressurized air (2 bar) was delivered from a compressor (Jun Air, 6–15) and used to clean the auto-sampler and achieve dry conditions during idle. Pressurized air was also used for the automated cleaning of in situ probes (s::can spectrolyser and Nadler ion-sensitive probe) at the same monitoring station. The sample tubing extended from the SAMP-FIL into the stream. A commercial filter cartridge (Acqua SAN, cartridge size: 1 in., stainless steel mesh) with a pore size of 100 µm was mounted at the end of this tubing (Figs. 2 and 3). All fluid and air tubings are polyurethane (PU) hoses (fluid: SMC Pneumatis, TU 1065, outer diameter: 10 mm, inner diameter: 6.5 mm, air: SMC Pneumatis, TU 0604, outer diameter: 6 mm, inner diameter: 4 mm). The tubing was connected with the filter, valves, pump and sample vessel via one-touch (push-in) fittings (SMC Pneumatics, KQ 2).

For each operation cycle, a filtered sample was delivered into the sample vessel (volume: 500 ml, material: HDPE, format: square). The required sample volume (100 ml) was taken from this sample vessel for on-site GLUC measurement by the BACTcontrol devices.

Operation of the aforementioned components (all sourced DC 24 V) was controlled by a relay board (SainSmart, 8-channel DC 5 V relay module, high current relay: AC 250 V, 10 A or DC 30 V, 10 A, driver current: 15–20 mA, indication of relay output status: LED), which was triggered by the RaspberryPi (Raspberry Pi 1 model B, DC 5 V) via GPIO (general purpose input/output) pins. The RaspberryPi has no default real-time clock (RTC) and retrieves the system time from a network time protocol (NTP) server (time server) whenever it is connected to the Internet. When operated off-line, in cases of power outages (e.g., due to thunderstorms), the RTC is required such that the RaspberryPi reboots with the correct system time after it is reconnected to power. Therefore, a peripheral RTC (DS 1307 RTC) was connected to the RaspberryPi via GPIO to access an accurate and current system

time during off-line operation (as described in: [21]).

The RaspberryPi and relay board (both sourced by DC 5 V) are connected (Fig. 3) to a 5 V commercial power supply unit (2 A). All other components operate on DC 24 V (Fig. 3) and are connected to an adequate commercial power supply unit (6 A). Both 5 V and 24 V power supply units are affiliated with AC 230 V main power connections (Fig. 3).

The BACTcontrol devices and SAMP-FIL were mounted in an air-conditioned, weatherproof outdoor monitoring station (OMS, Fig. 1). The option to mount the SAMP-FIL within the OMS was chosen as the least elaborate from a technical perspective because the length of the tubing was kept minimal and frost-proof housing was assured.

In addition to the robustness of the components, the main focus was set on a design enabling highly turbulent sample-water flow within the tubing and valves to reduce debris, bio-film and sinter deposition on the inner surface of the equipment. The flow rate of the high-performance pump (5 l/min) and the chosen inner diameter of the fluid tubing (6.5 mm) theoretically enabled at straight intercepts of the tubing a flow velocity ( $v$ ) of 2.5 m/s, with a Reynolds number ( $Re$ ) of over 12,500. A flow velocity of 1.7 m/s and a Reynolds number over 10,500 were calculated for the inside of the fluid valves (cylindrical aperture with a nominal diameter of 8 mm). Although this is an estimation and friction due to specific material roughness, bent tubing sections and intersections between components is neglected, highly turbulent flow behaviour within the tubing and valves can still be assumed [17]. Calculations were made for a water temperature of 10 °C, which is close to the annual mean stream water temperature of 10.3 °C, using the following equations [17]:

$$v = \frac{4Q}{\pi D^2} \quad (1)$$

$$Re = \frac{DV\rho}{\mu} \quad (2)$$

where  $Q$  is the volumetric flow rate,  $D$  is the pipe diameter,  $\rho$  is the density of the fluid and  $\mu$  is the dynamic viscosity.



## 2.4. Software

The operating system (OS) of choice was the Linux-based Raspbian, which was installed using the “New Out Of The Box Software” package [15] on the RaspberryPi’s secure digital memory (SD) card. The Raspbian image includes an integrated developer environment (IDE) used for Python programming (IDLE). Four GPIO pins of the RaspberryPi board are used as output to trigger the corresponding relay on the relay-board and are controlled by an executable Python script. The core features of the script are a file-type access to the GPIOs (no additional libraries and packages are required) and the Python *time.sleep* command, which suspends execution for a given number of seconds. This allows for coding a simple time-sequenced state machine that iteratively activates the peripheral devices for a certain time, as specified in the script, with the *time.sleep* command. Following this, the script schedules all necessary operations for one complete sampling cycle. *Cron* was used to schedule the sampling cycle to the measurements of the connected BACTcontrol devices (fixed measurement interval of 180 min). *Cron* is a software-tool to configure scheduled tasks using the system time on Unix systems, e.g., to schedule executable scripts to run at a fixed interval. The programmed Python script was set to be executed by *Cron* shortly before each BACTcontrol measurement starts. To keep track of the working steps conducted by SAMP-FIL, the Python logging module is used within the script. Thus, each programmed operation was logged together with a timestamp in a text file, allowing potential failures to be traced and aligned to erroneous BACTcontrol measurements.

## 2.5. Function

Each sampling cycle of the SAMP-FIL includes seven steps (Table 1). During the “CLEANING” step, the pump, “sample-in” fluid valve, “sample-out” fluid valve and pressurized air valve are activated. All equipment, including the filter cartridge, tubing, fluid valves, pump and sample vessel, are flushed through by pressurized air (Table 1). During the “FLUSHING” step, the valve for pressurized air is closed and all of the equipment is flushed through with recent sample water (Table 1). The “SAMPLING” step enables the filling of the sample vessel by closing the “sample-out” valve while keeping the pump and “sample-in” fluid valve activated (Table 1). During the “READY” step, the pump is deactivated, all valves stay closed and the sampled water remains in the sample vessel from where it is retrieved by the BACTcontrol devices (Table 1). When the abstraction of the water sample by the BACTcontrol devices is completed, the “sample-out” valve is opened to empty the sample vessel (Table 1). After draining the sample vessel, the “sample-out” valve stays open, and the “sample-in” valve, pump and pressurized air valve are activated (Table 1); this “CLEANING” step is identical to step 1 (Table 1), i.e., the residuals of the sample water are flushed out from the tubing, pump, valves and sample vessel, and the filter mesh is cleaned. After these six steps, the SAMP-FIL goes into “IDLE” mode, where no peripherals are activated, and the equipment remains dry (Table 1). This sequence of steps assures the residence of sample water within the device only when it is

**Table 1**  
Scheduled sequences for one complete operation cycle of SAMP-FIL.

Sequence No.	task	function	Status pump (relay 1)	Status valve-1 (relay 3)	Status valve-2 (relay 2)	Status valve-3 (relay 4)
1	Cleaning	Pressurized airflow through the complete system	active	active	active	active
2	Flushing	Flushing the system with recent sample water	active	inactive	active	active
3	Sampling	Filling of sample vessel	active	inactive	active	inactive
4	Ready	Connected devices abstract sample	inactive	inactive	inactive	inactive
5	Emptying	Emptying of sample-vessel	inactive	inactive	inactive	inactive
6	Cleaning	Pressurized airflow through the complete system	active	active	active	active
7	Idle	All components remain dry until next cycle	inactive	inactive	inactive	inactive

required and enhances long-term and continuous operation before and after each sampling cycle as the SAMP-FIL is flushed completely by pressurized air to prevent the filter cartridge from clogging, and any residual water is discarded from the previous cycle.

## 2.6. Field test

A field test with 3 phases was conducted to test the influence of proprietary sample-pretreatment and SAMP-FIL on the on-site measured GLUC activity. During “Phase 1” (23 March 2014 to 23 April 2014), both devices for on-site GLUC measurements (BACTcontrol 01 and BACTcontrol 02) were connected to the proprietary sample pretreatment. During “Phase 2” (27 July 2014 to 19 August 2014), the BACTcontrol 01 was connected to the SAMP-FIL, whereas BACTcontrol 02 remained connected to the proprietary sample-pretreatment. In “Phase 3” (13 September 2014 to 13 October 2014), both BACTcontrol devices were connected to the SAMP-FIL.

To test the technical capability of the SAMP-FIL for long-term on-site operation, it was continuously operated since the installation in July 2014 for 16 months until November 2015.

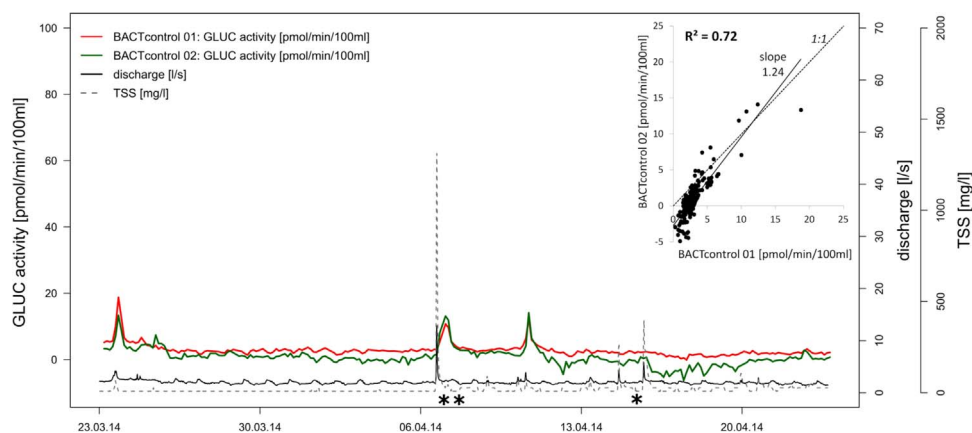
## 2.7. Reference analyses

Several studies described the attachment of faecal indicator bacteria to suspended particulate matter in aquatic habitats [4,6,7,11,25]. Enzymatic activity has been reported to be linked to fractions of suspended particulates in stream water [8,9]. As sample pre-filtration is an unavoidable step for automated GLUC measurements in sediment-laden waters, the authors tested the impact of a 100 µm filter on GLUC measurements and performed culture-based *E. coli* analyses. Grab samples were taken from the stream during different catchment conditions, i.e., regarding the hydrologic state and microbiological impact. One portion of each sample was filtered through a 100 µm filter (as mounted in the SAMP-FIL), whereas the other portion remained unaltered. Both portions were analyzed for GLUC activity and *E. coli*. GLUC activity measurements were performed with a ColiMinder laboratory measurement device. Although the units of the on-site and laboratory GLUC measurements are different, both ColiMinder (laboratory) and BACTcontrol (on-site) provide the same target-parameter, namely, the determination of beta-D-glucuronidase activity in waters [27]. Both constructions for GLUC determination yield results with an average one-to-one ratio between mMFU/100 ml (ColiMinder) and pmol/min/100 ml (BACTcontrol) [27]. The *E. coli* analyses were conducted using the Colilert18 method (ISO 9308-2:2012, MPN/100 ml).

## 3. Results

### 3.1. Field test

*Phase 1:* Measurements recorded by BACTcontrol devices connected to the proprietary pretreatment set-up showed significant damping and delay of the GLUC signals (Fig. 4). These effects were



**Fig. 4.** GLUC signals of BACTcontrol 01 (red) and BACTcontrol 02 (green) (both connected to proprietary sample pretreatment), discharge (black line) and TSS (dashed line) at the stream monitoring location during test phase 1. The linear regression analysis of BACTcontrol data for the same time period is shown on the right. The occurrences of GLUC signal damping (\*) and delay (\*\*) are highlighted with asterisks. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)

particularly pronounced during runoff events, when stream parameters, such as discharge and suspended sediment, showed a rapid response to changes in hydrologic conditions, whereas the response of GLUC signals was either delayed (for several hours) from that of parallel monitored stream parameters or appeared considerably damped (Fig. 4). Linear regression analysis of measurement data showed consistency between both BACTcontrol devices, with an  $R^2$  of 0.72 and a slope of 1.24. Measurements with negative GLUC values (Fig. 4) indicate malfunction of the BACTcontrol 02 device due to clogged reagent dosing or a contaminated fluorescence measurement window, presumably conditioned by insufficient sample pretreatment.

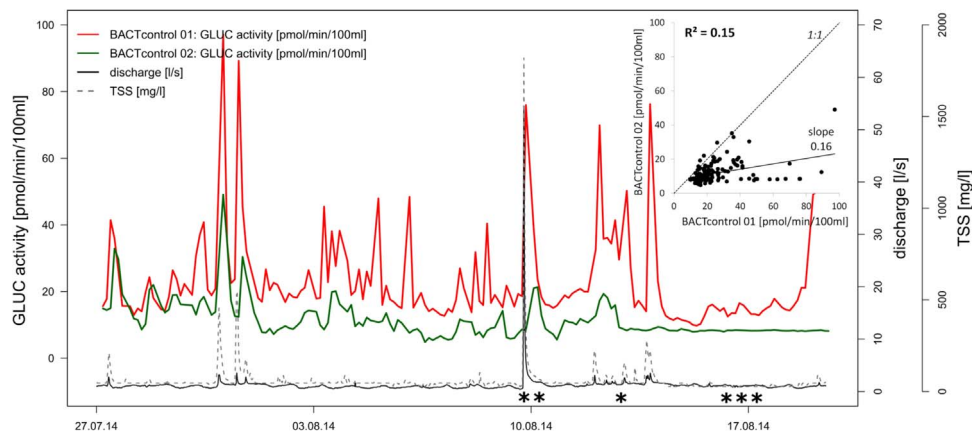
**Phase 2:** Damping and delay of GLUC signals due to the proprietary pretreatment set-up indicated during “Phase 1” were disclosed in “Phase 2”, in which BACTcontrol 01 was connected to the SAMP-FIL and BACTcontrol 02 retrieved sample water from the proprietary set-up (Fig. 5). Several precipitation events occurred during the test period, causing a potential input of faecal-associated contamination into the stream. BACTcontrol 01 recorded a significant peak of GLUC activity for each of these events, whereas BACTcontrol 02 showed a damped delayed response or even no response (Fig. 5). A linear regression coefficient  $R^2$  of 0.15 was found between the measurement results of BACTcontrol 01 and BACTcontrol 02. The regression slope of 0.16 (Fig. 5) demonstrates the higher sensitivity of the device connected to SAMP-FIL (BACTcontrol 01).

**Phase 3:** Connection of both the BACTcontrol apparatuses to SAMP-FIL resulted in highly consistent measurements between

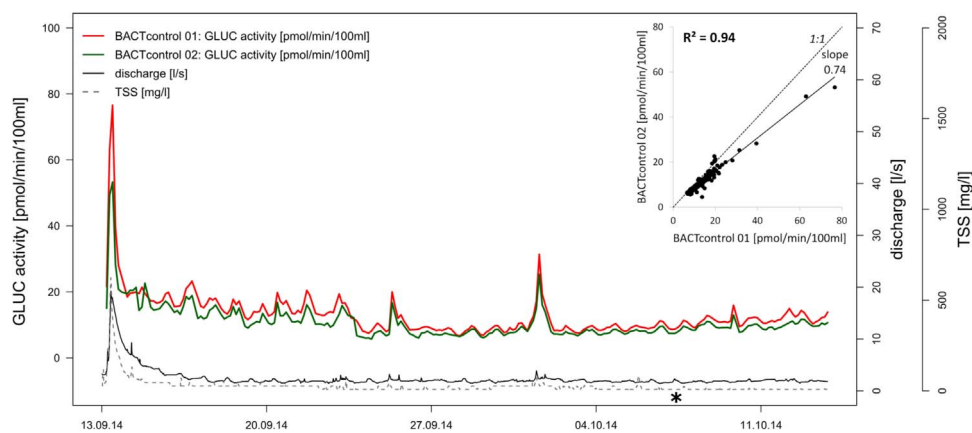
BACTcontrol 01 and BACTcontrol 02 and prompted responses of GLUC signals to changes in hydrologic conditions (Fig. 6). A linear correlation coefficient  $R^2$  of 0.94 was found between GLUC values gathered with BACTcontrol 01 and BACTcontrol 02. Both sets of GLUC signals are highly comparable regarding timing and range (Fig. 6). A regression slope of 0.74 indicates an offset between both devices (on average, BACTcontrol 01 yielded higher results than BACTcontrol 02 by 2.2 pmol/min/100 ml). For the first time since the installation of BACTcontrol devices at the monitoring location in 2012, diurnal fluctuations of GLUC activity (Fig. 6) in stream water during dry weather periods were captured by the BACTcontrol devices [27].

### 3.1.1. Continuous long-term operation

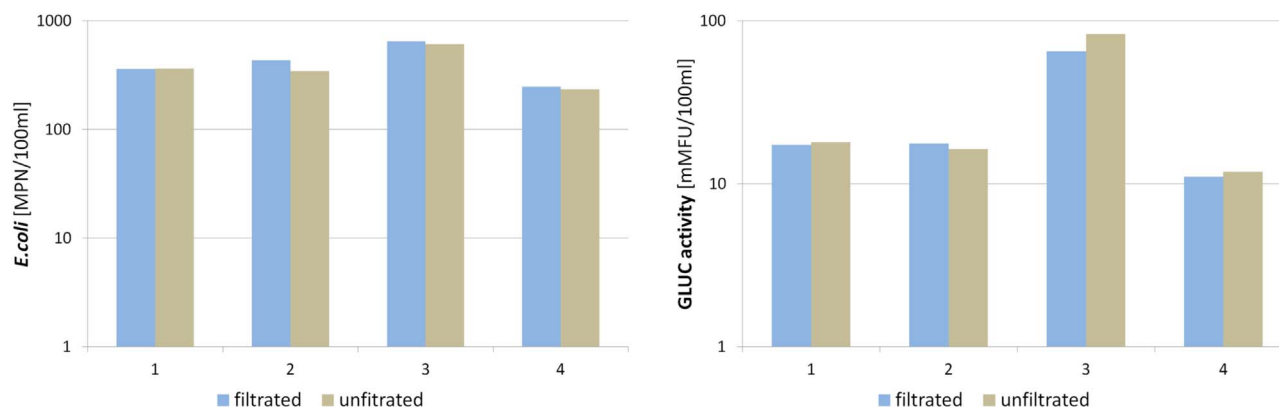
The SAMP-FIL was continuously operated from July 2014 to November 2015 (16 months). No technical failures of mechanical and electronic components occurred during this period. The coded script was autonomously continued after sporadic power outages (e.g., due to thunder storms). The error of the installed RTC of  $\pm 1$  min per month was maintained and corrected approximately every 3 months on-site by accessing the RaspberryPi via remote desktop. The filter cartridge remained unclogged but was preventively changed after 6 months of operation. No crucial sediment or bio-film accumulation within the tubing and sample vessel could be detected by visual inspection after the test period. The SAMP-FIL was preventively flushed with decalcifying and disinfecting cleaning solution after the test period before on-going operation continued.



**Fig. 5.** GLUC signals of BACTcontrol 01 (red, connected to the SAMP-FIL) and BACTcontrol 02 (green, connected to the proprietary sample pretreatment), discharge (black line) and TSS (dashed line) at the stream monitoring location during test phase 2. Linear regression analysis of BACTcontrol data for the same time period is shown on the right and indicates the higher sensitivity of BACTcontrol 01 connected to SAMP-FIL. The occurrences of GLUC signal (BACTcontrol 02) damping (\*), delay (\*\*), and no response (\*\*\*) are highlighted with asterisks. (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)



**Fig. 6.** GLUC signals of BACTcontrol 01 (red) and BACTcontrol 02 (green) (both connected to the SAMP-FIL), discharge (black line) and TSS (dashed line) at the stream monitoring location during test phase 3. The linear regression analysis of BACTcontrol data for the same time period is shown on the right. A high correlation ( $R^2=0.94$ ,  $p$ -value < 0.001) of GLUC measurements was achieved during this test period. The diurnal fluctuations in the GLUC activity in stream water were captured (marked with an asterisk). (For interpretation of the references to color in this figure legend, the reader is referred to the web version of this article.)



**Fig. 7.** Graphs of results from GLUC (left) and *E. coli* (right) analyses of four stream water samples, each portioned into a filtrated (through 100  $\mu$ m) and infiltrated part. No systematic impact of the tested filter on GLUC activity or *E. coli* concentration was found.

After the installation of SAMP-FIL, the GLUC measurements of the connected BACTcontrol devices achieved their highest quality in terms of consistency and sensitivity. The capability for long-term operation of BACTcontrol apparatuses in this technically challenging habitat increased as error-free running time and service intervals of up to 6 months were achieved.

### 3.2. Impact of sample pretreatment on GLUC activity and *E. coli* concentrations

Laboratory measurements of GLUC activity and culture-based *E. coli* analyses did not indicate a systematic influence of the 100  $\mu$ m filter on the measured signals (Fig. 7). Consequently, it is assumed that the chosen pore size does not adulterate on-site GLUC measurements.

### 3.3. Discussion and perspective

Low-cost equipment that allows the assembly of affordable equipment to be used as data loggers, sensors or accessory units will likely become of increased importance within various fields of today's environmental research, such as hydro-meteorological monitoring. Available inexpensive equipment has the potential to promote research and science into new fields and scales: To overcome the dearth of meteorological monitoring stations in the sub-Saharan Africa, Van de Giesen et al. [30] described the extensive installation of multitudinous monitoring stations based on new cost-effective technologies. The use of open-source technologies for the construction of low-cost laboratory equipment was delineated and discussed by [19].

The authors are aware that the capability of the used single-board computer was not fully exploited with the task described in this study. Nevertheless, the RaspberryPi proved to be the ideal choice for the intended purpose due to its robustness, usability, and low price (RaspberryPi 2 B: < 35€). This allowed resources to be used for the purchase of professional high-performance components (e.g., pumps, valves, casing, and tubing), resulting in failure-free on-site operation in a technically challenging habitat for 16 months (and on-going).

This work demonstrated that researchers can develop an effective device, controlled by a low-cost single-board computer, with a basic working knowledge in electronics but no background in electrical engineering or information technology (IT). The planning and construction of SAMP-FIL was exclusively conducted within the Vienna Doctoral Programme on Water Resource Systems (TU Wien).

The experimental results obtained during the various test phases demonstrated the relevance of best possible sample pretreatment with regard to minimal sample adulteration and the prevention of device fouling. This study quantitatively demonstrates how inadequate pretreatment procedures result in falsified and biased on-site measurements.

The SAMP-FIL version described in this paper is a prototype. Further endeavours will focus on the on-line connection of the SAMP-FIL, in particular, linking the SAMP-FIL to BACTcontrol to trigger SAMP-FIL activities by the connected measurement device.

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## References

- [1] M. Ali, J.H.A. Vlackamp, N.N. Eddin, B. Falconer, C. Oram, Technical development and socioeconomic implications of the raspberry pi as a learning tool in developing countries, *Comput. Sci. Electron. Eng. Conf. (CEEC)* (2013) 103–108. <http://dx.doi.org/10.1109/CEEC.2013.6659454>.
- [2] G. Blöschl, A.P. Blaschke, M. Broer, C. Bucher, G. Carr, X. Chen, A. Eder, M. Exner-Kittridge, A. Farnleitner, A. Flores-Orozco, P. Haas, P. Hogan, A. Kazemi Amiri, M. Oismüller, J. Parajka, R. Silasari, P. Stadler, P. Strauß, M. Vreugdenhil, W. Wagner, M. Zessner, The Hydrological Open Air Laboratory (HOAL) in Petzenkirchen: a hypotheses driven observatory, *Hydrol. Earth Syst. Sci. Discuss.* 12 (7) (2015) 6683–6753. <http://dx.doi.org/10.5194/hessd-12-6683-2015>.
- [3] G. Blöschl, G. Carr, C. Bucher, A.H. Farnleitner, H. Rechberger, W. Wagner, M. Zessner, Promoting interdisciplinary education – the Vienna Doctoral Programme on water resource systems, *Hydrol. Earth Syst. Sci. Discuss.* 8 (6) (2011) 9843–9887. <http://dx.doi.org/10.5194/hessd-8-9843-2011>.
- [4] I. Brettar, M.G. Höfle, Influence of ecosystemic factors on survival of *Escherichia coli* after large-scale release into lake water mesocosms, *Appl. Environ. Microbiol.* 58 (7) (1992) 2201–2210.
- [5] J.P. Cabral, Water microbiology. Bacterial pathogens and water, *Int. J. Environ. Res. Public Health* 7 (10) (2010) 3657–3703. <http://dx.doi.org/10.3390/ijerph7103657>.
- [6] B.C. Crump, E.V. Armbrust, J.A. Baross, Phylogenetic analysis of particle-attached and free-living bacterial communities in the Columbia river, its estuary, and the adjacent coastal ocean, *Appl. Environ. Microbiol.* 65 (7) (1999) 3192–3204.
- [7] S.C. Edberg, E.W. Rice, R.J. Karlin, M.J. Allen, *Escherichia coli*: the best biological drinking water indicator for public health protection, *Symp. Ser. (Soc. Appl. Microbiol.)* 88 (29) (2000) 106S–116S. <http://dx.doi.org/10.1111/j.1365-2672.2000.tb05338.x>.
- [34] A. Eder, M. Exner-Kittridge, P. Strauss, G. Blöschl, Re-suspension of bed sediment in a small stream - results from two flushing experiments, *Hydrol. Earth Syst. Sci.* 18 (2014) 1043–1052. <http://dx.doi.org/10.5194/hess-18-1043-2014>.
- [35] A. Eder, P. Strauss, T. Krueger, J.N. Quinton, Comparative calculation of suspended sediment loads with respect to hysteresis effects (in the Petzenkirchen catchment, Austria), *J. Hydrol.* 389 (2010) 168–176.
- [33] M. Exner-Kittridge, R. Niederreiter, A. Eder, M. Zessner, A simple and flexible field-tested device for housing water monitoring sensors at point discharges, *Water Sci. Technol.* 67 (2013) 1026. <http://dx.doi.org/10.2166/wst.2013.655>.
- [8] A.H. Farnleitner, L. Hocke, C. Beiwil, G.C. Kavka, T. Zechmeister, A.K. Kirschner, R.L. Mach, Rapid enzymatic detection of *Escherichia coli* contamination in polluted river water, *Lett. Appl. Microbiol.* 33 (3) (2001) 246–250. <http://dx.doi.org/10.1046/j.1472-765x.2001.00990.x>.
- [9] A.H. Farnleitner, L. Hocke, C. Beiwil, G.G. Kavka, R.L. Mach, Hydrolysis of 4-methylumbelliferyl- $\beta$ -D-glucuronide in differing sample fractions of river waters and its implication for the detection of fecal pollution, *Water Res.* 36 (4) (2002) 975–981. [http://dx.doi.org/10.1016/S0043-1354\(01\)00288-3](http://dx.doi.org/10.1016/S0043-1354(01)00288-3).
- [10] L. Fiksdal, M. Pommepuy, M.P. Caprais, I. Midttun, Monitoring of fecal pollution in coastal waters by use of rapid enzymatic techniques, *Appl. Environ. Microbiol.* 60 (5) (1994) 1581–1584.
- [11] T. Garcia-Armisen, P. Servais, Partitioning and fate of particle-associated *E. coli* in river waters, *Water Environ. Res.* 81 (1) (2009) 21–28. <http://dx.doi.org/10.2175/106143008x304613>.
- [12] I. George, M. Petit, P. Servais, Use of enzymatic methods for rapid enumeration of coliforms in freshwaters, *J. Appl. Microbiol.* 88 (3) (2000) 404–413. <http://dx.doi.org/10.1046/j.1365-2672.2000.00977.x>.
- [13] B.Horan, *Practical Raspberry Pi*, Apress, New York, NY, USA, 2013.
- [14] R.W. Hut, N.C. Van de Giesen, Delft University of Technology, *Civil Engineering and Geosciences: Water Management*, Delft University of Technology, Delft, Netherlands, 2013.
- [15] Installing Raspbian with NOOBS | Raspberry Pi Learning Resources. (<https://www.raspberrypi.org/learning/noobs-install/>) (accessed 04.02.16).
- [16] J. Koschelnic, W. Vogl, M. Epp, M. Lackner, Rapid analysis of  $\beta$ -D-glucuronidase activity in water using fully automated technology, *Water Resour. Manag.* 196 (2015) 471–481. <http://dx.doi.org/10.2495/WRM150401>.
- [17] M. Marriott, *Civil Engineering Hydraulics*, John Wiley & Sons, New York, NY, USA, 2009.
- [18] S. Monk, *Raspberry Pi Cookbook*, O'Reilly Media, Inc, Newton, MA, USA, 2013.
- [19] J.M. Pearce, *Open-Source Lab: How to Build Your Own Hardware and Reduce Research Costs*, Elsevier, Waltham, MA, 2013.
- [20] R. Pi, Index page. (<https://www.raspberrypi.org/forums/>) (accessed 10.12.15).
- [21] Raspberry Pi, View topic - The Correct way to add a RTC, (n.d.). (<https://www.raspberrypi.org/forums/viewtopic.php?T=85683>) (accessed 25.09.16).
- [22] M. Richardson, S. Wallace, *Getting Started with Raspberry Pi*, O'Reilly Media, Inc, Newton, MA, USA, 2012.
- [23] G. Ryzinska-Paier, T. Lendenfeld, K. Correa, P. Stadler, A.P. Blaschke, R.L. Mach, H. Stadler, A.K. Kirschner, A.H. Farnleitner, A sensitive and robust method for automated on-line monitoring of enzymatic activities in water and water resources, *Water Sci. Technol.* 69 (8) (2014) 1349–1358. <http://dx.doi.org/10.2166/wst.2014.032>.
- [24] A.E.U. Salam, T. Muh, M. Selintung, F. Maricar, Web based real time water pressure monitoring system, *Proc. Electr. Eng., Comput. Sci. Inform.* 1 (2014) 223–227. <http://dx.doi.org/10.11591/eecs1.1406>.
- [25] D. Savio, L. Sinclair, U.Z. Ijaz, J. Parajka, G.H. Reischer, P. Stadler, A.P. Blaschke, G. Blöschl, R.L. Mach, A.K.T. Kirschner, A.H. Farnleitner, A. Eiler, Bacterial diversity along a 2600 km river continuum, *Environ. Microbiol.* 17 (12) (2015) 4994–5007. <http://dx.doi.org/10.1111/1462-2920.12886>.
- [26] H. Stadler, P. Skritek, R. Sommer, R.L. Mach, W. Zerobin, A.H. Farnleitner, Microbiological monitoring and automated event sampling at karst springs using LEO-satellites, *Water Sci. Technol.* 58 (4) (2008) 899. <http://dx.doi.org/10.2166/wst.2008.442>.
- [27] P. Stadler, G. Blöschl, W. Vogl, J. Koschelnic, M. Epp, M. Lackner, M. Oismüller, M. Kumpan, L. Nemeth, P. Strauss, R. Sommer, G. Ryzinska-Paier, A.H. Farnleitner, M. Zessner, Real-time monitoring of beta-D-glucuronidase activity in sediment laden streams: a comparison of prototypes, *Water Res.* 101 (2016) 252–261. <http://dx.doi.org/10.1016/j.watres.2016.05.072>.
- [28] S. Sukaridhoto, D. Pramadihanto Taufiqurrahman, M. Alif, A. Yuwono, N. Funabiki, A design of radio-controlled submarine modification for river water quality monitoring, in: *Proceedings of the 2015 International Seminar on Intelligent Technology and Its Applications (ISITIA)*, IEEE, Piscataway, NJ, USA, 2015, pp. 75–80. (<http://dx.doi.org/10.1109/ISITIA.2015.7219956>)
- [29] E. Upton, G. Halfacree, *Meet the Raspberry Pi*, John Wiley & Sons, New York, NY, USA, 2012.
- [30] N. van de Giesen, R. Hut, J. Selker, The trans-African hydro-meteorological observatory (TAHMO), *Wires Water* 1 (2014) 341–348. <http://dx.doi.org/10.1002/wat2.1034>.
- [31] V. Vujovic, M. Maksimovic, Raspberry pi as a wireless sensor node: performances and constraints, in: *Proceedings of the 37th International Convention on Information and Communication Technology, Electronics and Microelectronics (MIPRO)*, IEEE, Piscataway, NJ, USA, 2014, pp. 1013–1018. (<http://dx.doi.org/10.1109/MIPRO.2014.6859717>)
- [32] F. Zibuschka, T. Lendenfeld, G. Lindner, Near real time monitoring von *E. coli* in Wasser, *Österreichische Wasser- und Abfallwirtsch.* 62 (11–12) (2010) 215–219. <http://dx.doi.org/10.1007/s00506-010-0240-z>.