

Journal of Visualized Experiments

Construction of a low-cost mobile incubator for field and laboratory use

--Manuscript Draft--

Article Type:	Methods Article - JoVE Produced Video
Manuscript Number:	JoVE58443R2
Full Title:	Construction of a low-cost mobile incubator for field and laboratory use
Keywords:	Incubator; field laboratory; microbial testing; field methods; Escherichia coli; drinking water; water quality testing
Corresponding Author:	Ariane Schertenleib Eawag Dübendorf, Zürich SWITZERLAND
Corresponding Author's Institution:	Eawag
Corresponding Author E-Mail:	ariane.schertenleib@eawag.ch
Order of Authors:	Ariane Schertenleib Jürg Sigrist Max ND Friedrich Christian Ebi Frederik Hammes Sara Jane Marks
Additional Information:	
Question	Response
Please indicate whether this article will be Standard Access or Open Access.	Open Access (US\$4,200)
Please indicate the city, state/province, and country where this article will be filmed . Please do not use abbreviations.	Überlandstrasse 133, 8600 Dübendorf, Zürich, Switzerland

Eawag | Ariane Schertenleib
Department Sandec | Project Officer
Überlandstrasse 133 | +41 58 765 53 23
CH-8600 Dübendorf | ariane.schertenleib@eawag.ch
Switzerland | www.sandec.ch

October 1st, 2018

Dear Dr Philip Steindel:

Thank you for the feedback on our Manuscript (JoVE58443R1, "Construction of a low-cost mobile incubator for field and laboratory use"), and for the opportunity to revise it.

We thank you for the fast and constructive feedback. We have detailed these edits within the point-by-point responses to editor comments, with edits indicated by underlined text. All edits are also visible in "track changes" within the revised manuscript uploaded on your platform.

Please do not hesitate to contact me if you have questions about the files submitted through the online system or require additional information. Thank you in advance for your consideration, and we look forward to hearing from you.

Yours sincerely,

A handwritten signature in black ink, consisting of stylized initials 'AS' followed by a long horizontal flourish.

Ariane Schertenleib

TITLE:**Construction of a Low-cost Mobile Incubator for Field and Laboratory Use****AUTHORS AND AFFILIATIONS:**

Ariane Schertenleib¹, Jürg Sigrist², Max ND Friedrich³, Christian Ebi⁴, Frederik Hammes², Sara J Marks¹

¹Department of Sanitation, Water & Solid Waste for Development, Swiss Federal Institute of Aquatic Science and Technology (Eawag), Dübendorf, Zürich, Switzerland

²Department of Environmental Microbiology, Eawag, Dübendorf, Zürich, Switzerland

³Department of Environmental Social Sciences, Eawag, Dübendorf, Zürich, Switzerland

⁴Department of Urban Water Management, Eawag, Dübendorf, Zürich, Switzerland

Corresponding Author:

Ariane Schertenleib

ariane.schertenleib@eawag.ch

Tel: +41 (0)58 765 53 23

Email Addresses of Co-authors:

Ariane Schertenleib (ariane.schertenleib@eawag.ch)

Jürg Sigrist (juerg.sigrist@eawag.ch)

Max ND Friedrich (max.friedrich@eawag.ch)

Christian Ebi (christian.ebi@eawag.ch)

Frederik Hammes (frederik.hammes@eawag.ch)

Sara J Marks (sara.marks@eawag.ch)

KEYWORDS:

Incubator, field laboratory, microbial testing, field methods, *Escherichia coli*, drinking water, water quality testing.

SUMMARY:

This paper describes a method for building an adaptable, low-cost and transportable incubator for microbial testing of drinking water. Our design is based on widely available materials and can operate under a range of field conditions, while still offering the advantages of higher-end laboratory-based models.

ABSTRACT:

Incubators are essential for a range of culture-based microbial methods, such as membrane filtration followed by cultivation for assessing drinking water quality. However, commercially available incubators are often costly, difficult to transport, not flexible in terms of volume, and/or poorly adapted to local field conditions where access to electricity is unreliable. The purpose of this study was to develop an adaptable, low-cost and transportable incubator that can be constructed using readily available components. The electronic core of the incubator was first developed. These components were then tested under a range of ambient temperature conditions (3.5 °C – 39 °C) using three types of incubator shells (polystyrene foam box, hard

cooler box, and cardboard box covered with a survival blanket). The electronic core showed comparable performance to a standard laboratory incubator in terms of the time required to reach the set temperature, inner temperature stability and spatial dispersion, power consumption, and microbial growth. The incubator set-ups were also effective at moderate and low ambient temperatures (between 3.5 °C and 27 °C), and at high temperatures (39 °C) when the incubator set temperature was higher. This incubator prototype is low-cost (< 300 USD) and adaptable to a variety of materials and volumes. Its demountable structure makes it easy to transport. It can be used in both established laboratories with grid power or in remote settings powered by solar energy or a car battery. It is particularly useful as an equipment option for field laboratories in areas with limited access to resources for water quality monitoring.

INTRODUCTION:

Culture-based methods for the detection of microbial contaminants are the state-of-the-art for water quality analysis in both industrialized and developing countries^{1,2}. Microorganisms exist in many environments and require different temperature conditions for optimal growth. Therefore, creating a temperature-stable incubation environment is a precondition for the reliable detection of microbial contaminants of concern in drinking water. According to the World Health Organization, *Escherichia coli* (*E. coli*) (or alternatively, thermotolerant coliforms (TTC)) are the most suitable indicators of fecal contamination in drinking water³. Detection of these organisms consists of, for example, filtering a 100 mL water sample through a membrane followed by incubation of the membrane on selective media at 35 – 37 °C (*E. coli*) or 44 – 45 °C (TTC)³.

Field-based applications of culture-based methods have become increasingly relevant in recent years. Under Sustainable Development Goal 6, Target 6.1, governments have committed to regularly report bacteriological quality of drinking water at the national level⁴. In addition to such public health surveillance efforts, operational monitoring of water infrastructure is regularly undertaken at the local or regional level⁵. These surveillance and monitoring campaigns are often in remote locations where the required laboratory infrastructure is inadequate or unavailable.⁶ Similarly, culture-based methods are widely used in medical diagnosis and microbiological research where local clinics and research institutions may be challenged by limited resources and insecure power supplies⁷.

In the above contexts, conventional incubators are often inadequate or unavailable. As an alternative, field incubators have been specifically developed for use outside the laboratory, *e.g.*, the Aquatest project⁸, University of Bristol, United Kingdom; DelAgua⁹, Marlborough, United Kingdom; or Aquagenx¹⁰, University of North Carolina, United States. However, these devices are relatively small in volume, thus limiting the number of samples that can be simultaneously processed. Field incubators on the market are also not designed to operate under very low (< 20 °C) or very high (> 40 °C) ambient temperature conditions, making their use in desert or alpine environments difficult. Further alternative solutions include yogurt-making appliances¹¹, body belts, and phase-change incubators¹². However, such unconventional incubators may function unreliably or be burdensome to operate¹¹.

There is thus a need for an incubator that offers the advantages of laboratory-based models (ease

of use, larger volume, and temperature precision) while remaining suitable for field applications (low-cost, easily transported and maintained, robustness to a range of ambient temperatures, energy efficient, and resilient to intermittent power supplies) (**Table 1**). The purpose of this protocol is to detail the fabrication process of a low-cost incubator designed to optimize the advantages of both conventional and field-based models using widely available material.

[Place **Table 1** here]

The following assembly protocol specifies the required materials and steps for building the incubator. It is structured in four steps: first, assembly of the heating unit; second, assembly of the control unit; third, assembly of the incubator electrical core; and fourth, assembly of the incubator. This protocol explains the construction of the electronic core of the incubator, that can work with a variety of incubator shells. See the **Table of Materials** for a full list of all components used in the Protocol and their technical specifications. The protocol below presents a functional example of the field incubator, but flexible use of different components is possible as long as they fulfill the electrical requirements. Using different components might influence the performances of the incubator. It is advised that the construction and wiring of electrical components be done by a person skilled in the electrical field.

PROTOCOL:

1. Heating Unit

1.1. Gather the following components (Figure 1**):**

- ① Support plate (280 x 250 mm) with required anchorage holes
- ② Axial fan (60 x 60 x 25 mm); 2x
- ③ Spacer (length 20 mm, internal diameter 4.25 mm (M4)); 4x
- ④ Luster terminal with three pins
- ⑤ Screw nut (M4); 4x and (M3); 4x
- ⑥ Washer (M4); 4x and (M3); 4x
- ⑦ Screw (M4); 4x and (M3); 4x

[Place **Figure 1** here]

1.2. Drill the required holes (Figure 2**) into the support plate ① to secure the axial fans ② as well as the luster terminal ④ (**Figure 1**).**

[Place **Figure 2** here]

1.3. Anchor the axial fans ② in the center on the support plate ① as shown in **Figure 3 with two M4 screws, screw nuts, and washers ⑤, ⑥, ⑦ per fan. Use the spacers ③ to leave a distance between the fans and the support plate (**Figure 3**).**

1.4. Anchor the luster terminal ④ to the support plate ① using M3 screws, screw nuts, and washers (**Figure 3**).

1.5. Connect the fan cables with the luster terminal. Connect the positive cables of each fan together and the negative cables of each fan together (**Figure 3**). The speed sensor is not required.

[Place **Figure 3** here]

NOTE: The cable colors mentioned correspond to the ones used in the figures. The cable colors might change depending on the material used.

2. Control Unit (Power Supply)

2.1. Gather the following components:

- ⑧ Universal enclosure (here 200 x 120 x 60 mm, but dimensions will depend on the size of the DC/DC converter and the PID temperature controller)
- ⑨ On/off-Switch
- ⑩ DC/DC converter, input voltage range 9 - 36V, output voltage 12V
- ⑪ PID temperature controller, 12 - 35 V/DC operating voltage
- ⑫ Cable gland, M12 x 15 mm, clamping range 2 - 7.5 mm (or according to the cable used)
- ⑬ Temperature sensor Pt100
- ⑭ AC power supply

NOTE: The incubator can be connected to the mains power supply or to a battery. In the case of the mains operation, the AC power supply is required and if the unit is exclusively connected to mains, the DC/DC converter is not mandatory. In the case of battery operation, the DC/DC converter is highly recommended, and a two-wire cable is required instead of the AC power supply. This protocol presents the version with the DC/DC converter and the AC power supply. An electrical diagram of the incubator electrical core is detailed in the supplementary material (**Figure S1**).

2.2. Mill the openings for the PID temperature controller, on/off switch, and cable glands into the enclosure with a drill and jigsaw, or an equivalent tool (**Figure 4**).

[Place **Figure 4** here]

2.3. Connect the DC/DC Converter to the on/off switch: connect the positive cable of the AC power adapter to the on/off switch and the negative cable of the AC power adapter to the "- Vin" of the DC/DC converter (**Figure 5**).

2.4. Use a cable to connect the on/off switch to the "+ Vin" of the DC/DC converter (**Figure 5**).

[Place **Figure 5** here]

2.5. Connect the cables from the heating unit to the PID temperature controller as follows (**Figure 6**):

2.5.1. Connect the terminal "1" of the PID temperature controller to the "DC –" wire from the heating unit connection and to the "- Vout" terminal of the DC/DC converter.

2.5.2. Connect the terminal "2" of the PID temperature controller to the terminal "4" of the PID temperature controller as well as to the "DC +" wire going to the heating unit (see point 3.2).

2.5.3. Connect the terminal "4" of the PID temperature controller to the "+ Vout" terminal of the DC/DC converter.

2.5.4. Connect the terminal "5" of the PID temperature controller to the "command" wire going to the heating unit. (see point 3.2).

2.5.5. Connect the temperature sensor to the terminals "10", "11" and "12".

NOTE: The red cable of the temperature sensor must be connected to terminal "11" of the PID temperature controller.

2.6. Anchor the DC/DC converter with Velcro tape at the bottom of the enclosure, and close the universal enclosure.

[Place **Figure 6** here]

NOTE: The functions of the PID temperature controller terminals used are given in **Table 2**.

[Place **Table 2** here]

3. Assembly of the Incubator Electrical Core

3.1. Gather the following components:

Heating unit from section 1

Control unit from section 2

⑮ Heating foils, self-adhesive, 100 x 200 mm, 12 V/20 W, 2x

3.2. Link the connection cables from the control unit to the heating unit as follows (**Figure 7**):

3.2.1. Connect the "DC-" wire from the control unit with one conductor of each of the heating foils and the negative wire of each fan.

3.2.2. Connect the "DC+" wire coming from the control unit with the positive cable of each fan.

3.2.3. Connect the “**command**” wire from the control unit to the remaining two conductors of the heating foils.

[Place **Figure 7** here]

NOTE: The completed field incubator electrical core of the incubator is shown in **Figure 8**.

[Place **Figure 8** here]

4. Assembly of the Incubator

4.1. Gather the following components:

- Incubator electrical core

- Incubator shell (here a polystyrene foam box, but can be any type of box made of insulating material)

- Support rack (here a metal rack, but can be another material)

4.2. Place the incubator components together as follows (**Figure 9**):

4.2.1. Place the incubator shell on its side, so that the opening of the incubator (door) is located on a side.

4.2.2. Place the support plate with the heating unit at the bottom of the incubator shell.

4.2.3. Place the support rack on top of the heating unit, leaving a space of a minimum of 10 cm between the heating unit and the support rack.

4.2.4. Place the temperature probe on the support rack. Ensure that the sensor does not touch any material.

4.3. Drill holes in the door of the incubator to allow for the entry of the cables (**Figure 9**).

4.4. Connect the incubator to the power source.

4.5. Turn the incubator on and adjust the settings of the PID temperature controller (see **Table S1** in the supplementary material for detailed settings).

[Place **Figure 9** here]

Note: The shell of the incubator can be a box of any type of material. It is recommended to use an insulating material, and that the box closes tightly to avoid dissipation of the heat. The support rack should contain large holes to avoid the accumulation of heat in the rack, and the material can be metal or other (e.g. plastic).

REPRESENTATIVE RESULTS:

The reliability of a robust field incubator lies in its ability to reach and maintain a set temperature under various conditions. To monitor the performance of the various incubator set-ups, the following measurements were made: time needed to reach the set temperature, effect of opening the door for one minute, power consumption over 24 hours of operation, inner temperature stability over 24 hours of operation, and observation of *E. coli* growth. The temperature inside the incubator was measured every minute with 4 temperature logging devices placed in different positions in the structure (support rack, wall, top, inside a growth plate). The set temperature was considered to be attained when all measurements were within plus or minus 2 °C, which is the acceptable range for the incubation of *E. coli*.¹³

The electronic core was tested with three types of shells, using materials that are typically found in many countries: a polystyrene foam box (78 liters), a hard plastic cooler box (30 liters), and a cardboard box covered with a survival blanket (46 liters) (**Figure 10**). To cover a range of ambient conditions that can be experienced in the field, these incubator set-ups were tested at three ambient temperatures: ambient (about 27 °C), cold (about 3.5 °C and 7.5 °C) and hot (about 39 °C). Performance measures were tested setting the inner temperature at 37 °C and 44.5 °C.

The time to reach the set temperature in the incubators was influenced by the ambient temperature and the material of the incubator shell. At an ambient temperature of about 27 °C, the three incubators set-ups reached the set temperatures (37 °C and 44.5 °C) in a similar time (**Figure 12a** and **Figure 13a**) and comparable with the performance of a standard incubator (**Table 3**). In cold environments (3.5 °C and 7.5 °C), the incubators with thicker shells, *i.e.*, the polystyrene foam and cooler box, reached the target set temperatures (37 °C and 44.5 °C) in a similar time; about four times longer than under an ambient temperature of 27 °C. With its lower insulation, the cardboard box with survival blanket never fully reached the set temperatures under cold ambient temperature conditions (**Figure 11b** and **Figure 12b**). In a warm environment (39 °C), the three incubator set-ups reached the target temperature of 44.5 °C in under 10 minutes (**Figure 12c**). However, when the set temperature was of 37 °C, *i.e.*, lower than the ambient temperature, none of the incubators could lower the temperature, resulting in overheating for all three incubator set ups (**Figure 11c**).

The ambient temperature and the type of incubator shell influenced the impact of opening the door of the incubator for one minute. The heat loss was greater in the cold environment, and the time to recover the inner set temperatures was longer, with the exception of the cardboard box incubator where the set temperatures were never reached (**Figure 13b** and **Figure 14b**). In the warmer environments, the heat loss was limited and the set temperatures were recovered in under 10 minutes (**Figure 13a,c** and **Figure 14,c**). In an ambient temperature of 39 °C and set temperature of 37 °C, opening the door did not cause nor reduce overheating of the incubators (**Figure 13c**).

The power consumption increased with cold environments and with an increase in the set temperature. Better insulating incubator shells (polystyrene foam and cooler box) showed a reduced power consumption as compared to the cardboard box incubator. In similar

environments (ambient temperature of about 27 °C), the three incubator set-ups consumed 0.22 to 0.52 kWh/24h less energy than the standard incubators tested (**Table 3**).

The temperature in the incubator remained stable over 24 hours with all type of incubator shells and ambient temperature tested (**Figure 13** and **Figure 14**). Slight variations of the measured temperature compared to the set temperature were observed according to the position of the temperature logging device in the incubator. With the exception of the tests with the ambient temperature (39 °C) warmer than the set temperature (37 °C) (**Figure 13c**), the variations in temperature were all within the 2 °C acceptable range for *E. coli* incubation.

All tests were performed in the presence of *E. coli* and total coliform measurement materials (membrane filter placed on growth plate). Replicates of a sample were placed in each incubator set-up and in a standard incubator for comparison. In all set-ups and conditions, the growth of *E. coli* and total coliform was successful and comparable to the growth observed in the standard incubator. A summary of the incubator configurations and ambient temperature conditions tested with results are shown in **Table 3**.

[Place **Table 3** here]

Figure and Table Legends

Figure 1: Individual components of the heating unit. ① Support plate, ② axial fans, ③ spacers, ④ luster terminal, ⑤ screw nuts, ⑥ washers and ⑦ screws.

Figure 2: Schematic diagram of support plate. Indications to drill the anchorage holes into the support plate to fix the axial fans as well as the luster terminal. Distances are given in millimeters.

Figure 3: Axial fans fixed on support plate.

Figure 4: Schematic diagram of the universal enclosure. (a) Indications to place the temperature controller ④ on/off switch ⑤ and the cable glands ⑥ into the universal enclosure; distances are given in millimeters. **(b)** 3D view of the universal enclosure.

Figure 5: Mounted control unit. Universal enclosure ⑧ with DC/DC converter ⑩ connected to PID temperature controller ⑪ and on/off switch ⑨.

Figure 6: Cable connection of DC/DC converter with PID temperature controller. DC/DC converter ⑩, PID temperature controller ⑪, connection to Incubator (cable A) and connection to temperature sensor (cable B).

Figure 7: Cable connection of heating foils ⑮ with PID temperature controller.

Figure 8: Complete field incubator electrical core. Heating unit ④ control unit ⑥ and temperature probe ⑦

Figure 9: Complete field incubator. Open (left) and closed (right). Heating unit ④ support rack ⑤ temperature probe ⑥ control unit ⑦ incubator shell ⑧ and holes for cables in the incubator shell (circled area).

Figure 10: Incubator shells tested. Open (upper row) and closed (bottom row). Polystyrene foam box (left), thickness of 3.5 cm, outer dimensions 39 x 56 x 36 cm; hard plastic cooler box (middle), thickness of 2.5 cm, outer dimensions 32 x 41 x 47 cm; cardboard box (right) covered with a standard survival blanket of a thickness of 12 µm folded twice, outer dimensions 30 x 42 x 37 cm.

Figure 11: Time to reach set temperature (37 °C) of the incubator set-ups under different ambient temperature conditions. Performances of incubators with a shell made of a polystyrene foam box, a hard cooler box, and a cardboard box covered with a survival blanket. At room ambient temperature (a), cold ambient temperature (b), and warm ambient temperature (c). Temperatures recorded on the support rack of the incubators.

Figure 12: Time to reach set temperature (44.5 °C) of the incubator set-ups under different ambient temperature conditions. Performances of incubators with a shell made of a polystyrene foam box, a hard cooler box, and a cardboard box covered with a survival blanket. At room ambient temperature (a), cold ambient temperature (b), and warm ambient temperature (c). Temperatures recorded on the support rack of the incubators.

Figure 13: Temperature variations over 24-hour period and effect of door opening under different ambient temperature conditions. Set temperature of 37 °C. Performances of incubators with a shell made of a polystyrene foam box, a hard cooler box, and a cardboard box covered with a survival blanket. At room ambient temperature (a), cold ambient temperature (b), and warm ambient temperature (c). Circled areas show the temperature variations due to the door opening for one minute. Temperatures recorded on the support rack of the incubators.

Figure 14: Temperature variations over 24-hour period and effect of door opening under different ambient temperature conditions. Set temperature of 44.5 °C. Performances of incubators with a shell made of a polystyrene foam box, a hard cooler box, and a cardboard box covered with a survival blanket. At room ambient temperature (a), cold ambient temperature (b), and warm ambient temperature (c). Circled areas show the temperature variations due to the door opening for one minute. Temperatures recorded on the support rack of the incubators.

Table 1: Characteristics of commercially available incubators (laboratory-based and field) and the optimized approach.

Table 2: Functions corresponding to the PID temperature controller terminals.

Table 3: Results summary for the incubator configurations and ambient temperature conditions tested. *Test 4: Absolute maximal and minimal temperatures recorded during the

stable periods, *i.e.*, from 10 minutes after the end of a disruptive event (time to reach set temperature, opening the door). [†]ND: No data, test not run.

Figure S1: Electrical diagram of incubator electrical core wiring. Alternatives for mains operation and battery operation are indicated.

Table S1: PID temperature controller settings. Display of set values; other parameters not necessary to run the incubator were left to default values.

DISCUSSION:

Under Sustainable Development Goal 6.1, the demand for water quality sampling is increasing, especially in remote rural areas where monitoring practices are less established¹⁴. A major barrier to implementing regular water quality testing in these settings is poor access to laboratories capable of supporting microbial methods⁶. This paper presents a method for a reliable incubator constructed from materials that are relatively cheap and widely available. The electrical components are relatively easy to source and assemble, requiring only limited expertise. Moreover, the incubator shell design is flexible and therefore can be constructed from locally available materials. This is especially desirable for those traveling to remote locations, since luggage space is not needed for a heavy and bulky shell. Depending on the shell used, the volume of the incubator is also adaptable and can be sized to accommodate a specific sample size. The presented set-up can be used on- and off-grid, which makes it robust to power cuts or absence of reliable electrical supply. While certain design limitations were observed, this set-up up generally proved to be effective under a range of ambient temperature conditions (3.5 °C to 39 °C).

There are several steps in the protocol that are critical for achieving an incubator design suitable to one's needs. The first is the selection of the electrical components of the incubator. Alternative components can be chosen based on the price or the local availability. Depending on the material selected and their technical specifications, the incubator may have altered performances as compared to the results presented. Another critical step in the protocol is the choice of shell material, which should be made based on the expected range of ambient temperatures, local power supply, and availability of materials. At lower ambient temperatures (< 25 °C), a shell constructed of polystyrene foam or a hard cooler box is recommended to achieve a set temperature of 37 °C to 44.5 °C. Based on the experimental data presented, these set ups can be expected to reach the set temperature in 45 – 96 minutes and consume 0.78 – 1.05 kWh/24h in cold environments (3.5 to 7.5 °C). The cardboard box with survival blanket is not recommended for use at lower ambient temperatures since this set up never reached a stable set temperature during the experimental observation period. At moderate ambient temperature (27 °C) any of the shell types tested are acceptable, with similar to slightly greater power consumption observed for the cardboard box set up. At higher ambient temperatures (39 °C), the incubator designs presented here were prone to overheating if unless the set temperature was even higher (*i.e.*, 44.5 °C). Therefore, such conditions would require a cooling device or use in a climate controlled space.

The cost of constructing the incubator presented here was about 300 USD when materials were sourced in Switzerland. However, these costs may be considerably lower in different locations, especially if shipping fees for the electronic core components can be kept to a minimum. Modification of the various components described in the protocol can further reduce costs. The protocol presented here is limited in that it compares only three shell material types at two set temperatures, as well as verification of microbial growth for *E. coli* only. Future research should test the suitability of this incubator design under a greater range of temperature parameters and using additional microbial indicator species (e.g., *Enterococcus*) and pathogens (e.g., salmonella, *Vibrio cholerae*). Future research should also focus on development of effective cooling techniques within the incubator, which would allow for its use in extremely warm environments (> 40 °C).

To our knowledge, there is no other known field incubator that offers adaptable volume capacity and is easily dismountable, while remaining transportable and low-cost. This innovative alternative to commercially available incubators fulfills a need for governments and organizations with water quality and other culture-based testing objectives where few laboratory facilities are available. When paired with simple water quality testing equipment, this incubator can help practitioners with limited capacities to establish permanent or seasonal laboratories at a reasonable cost. By increasing the number of laboratories in remote areas, efforts to conduct regular water quality surveillance or achieve punctual monitoring of system operations will become increasingly feasible.

ACKNOWLEDGMENTS:

This research was supported by the Swiss Agency for Development Cooperation and the REACH programme funded by UK Aid from the UK Department for International Development (DFID) for the benefit of developing countries (Aries Code 201880). The views expressed and information contained in it are not necessarily those of or endorsed by these agencies, which can accept no responsibility for such views or information or for any reliance placed on them. The authors also thank Arnt Diener for his contributions to early iterations of the polystyrene foam incubator prototype.

DISCLOSURES:

The authors have nothing to disclose.

REFERENCES

1. Bain, R. *et al.* A summary catalogue of microbial drinking water tests for low and medium resource settings. *International Journal of Environmental Research and Public Health*. **9** (1609–1625) (2012).
2. Köster, W. *et al.* Analytical methods for microbiological water quality testing. *Assessing Microbial Safety of Drinking Water*. 237–277 (2003).
3. World Health Organization (WHO). *Guidelines for Drinking Water Quality, 4th Edition*. WHO Press. Geneva. (2011).
4. World Health Organization (WHO). *Safely Managed Drinking Water - Thematic Report on*

483 *Drinking Water*. WHO Press. Geneva. (2017).

- 484 5. Peletz, R., Kumpel, E., Bonham, M., Rahman, Z., Khush, R. To what extent is drinking water
485 tested in sub-Saharan Africa? A comparative analysis of regulated water quality
486 monitoring. *International Journal of Environmental Research and Public Health*. **13** (3), 275,
487 doi: 10.3390/ijerph13030275 (2016).
- 488 6. Diener, A. *et al.* Adaptable drinking-water laboratory unit for decentralised testing in
489 remote and alpine regions. *40th WEDC International Conference, Loughborough, UK*. 1–6
490 (2017).
- 491 7. Malkin, R.A. Design of health care technologies for the developing world. *Annual Review*
492 *of Biomedical Engineering*. **9** (1), 567–587, doi: 10.1146/annurev.bioeng.9.060906.151913
493 (2007).
- 494 8. Rahman, Z., Khush, R., Gundry, S. Aquatest: Expanding Microbial Water Quality Testing for
495 Drinking Water Management. *Drinking Water Safety International*. **1** (4), 15–17 (2010).
- 496 9. DelAgua Water Testing Ltd. *DelAgua Portable Water Testing Kit: User Manual Version 5.0*.
497 Marlborough, UK. (2015).
- 498 10. Aquagenx LLC *Portable Incubator Fabrication Instructions*. Chapel Hill, NC. (2015).
- 499 11. Nair, J., Mathew, K., Ho, G.E. Experiences with implementing the H2S method for testing
500 bacterial quality of drinking water in remote aboriginal communities in Australia. *Water*
501 *for all life: A decentralized infrastructure for a sustainable future* (2007).
- 502 12. Kandel, P., Kunwar, R., Lamichhane, P., Karki, S. Extent of fecal contamination of household
503 drinking water in Nepal: Further analysis of Nepal Multiple Indicator Cluster Survey 2014.
504 *American Journal of Tropical Medicine and Hygiene*. **96** (2), 446–448, doi:
505 10.4269/ajtmh.16-0513 (2017).
- 506 13. Edberg, S.C., Rice, E.W., Karlin, R.J., Allen, M.J. Escherichia coli: the best biological drinking
507 water indicator for public health protection. *Journal of Applied Microbiology*. **88** (51),
508 1065–1165, doi: 10.1111/j.1365-2672.2000.tb05338.x (2000).
- 509 14. Taylor, D.D.J., Khush, R., Peletz, R., Kumpel, E. Efficacy of microbial sampling
510 recommendations and practices in sub-Saharan Africa. *Water Research*. **134**, 115–125, doi:
511 10.1016/j.watres.2018.01.054 (2018).

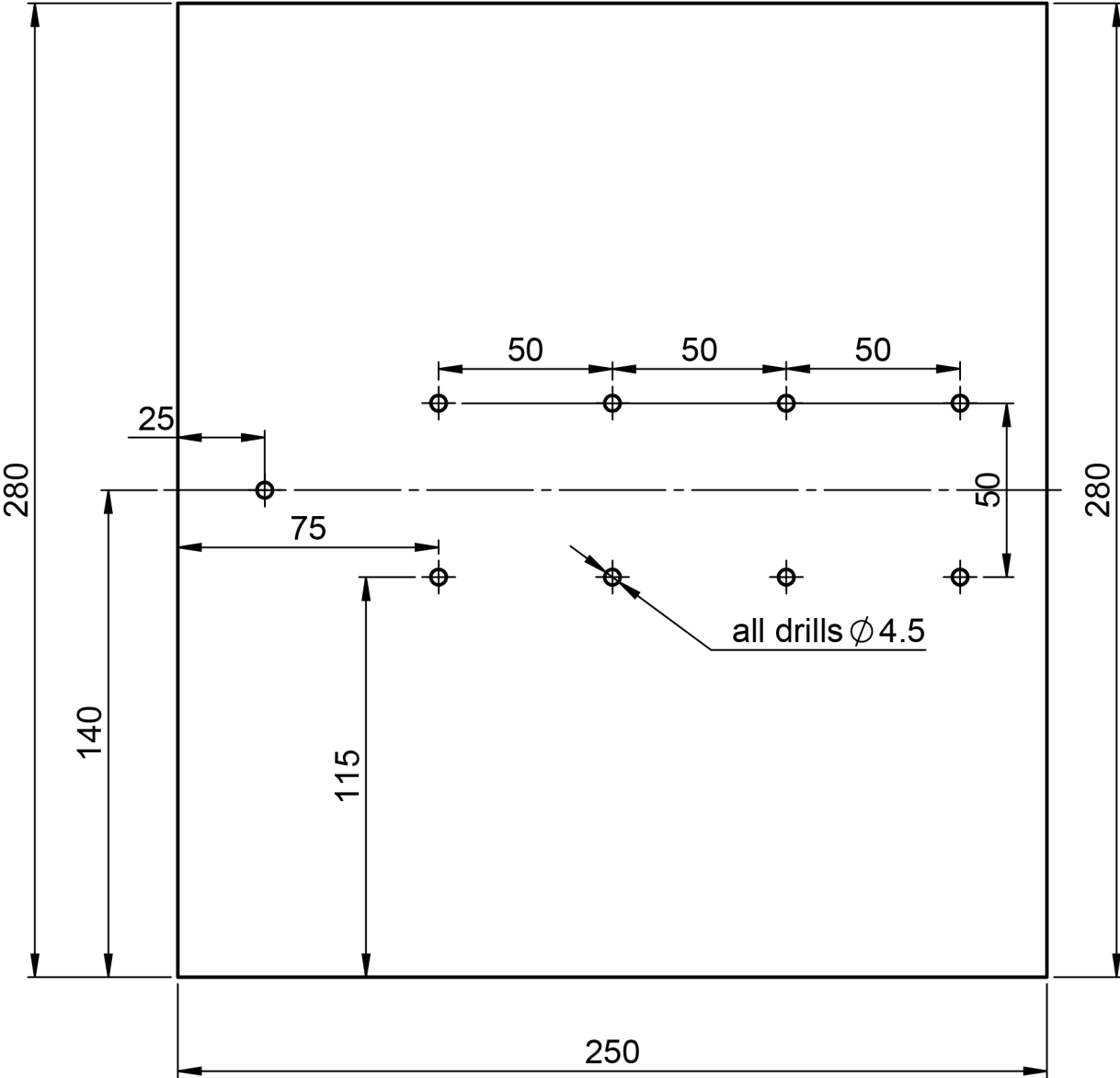
512

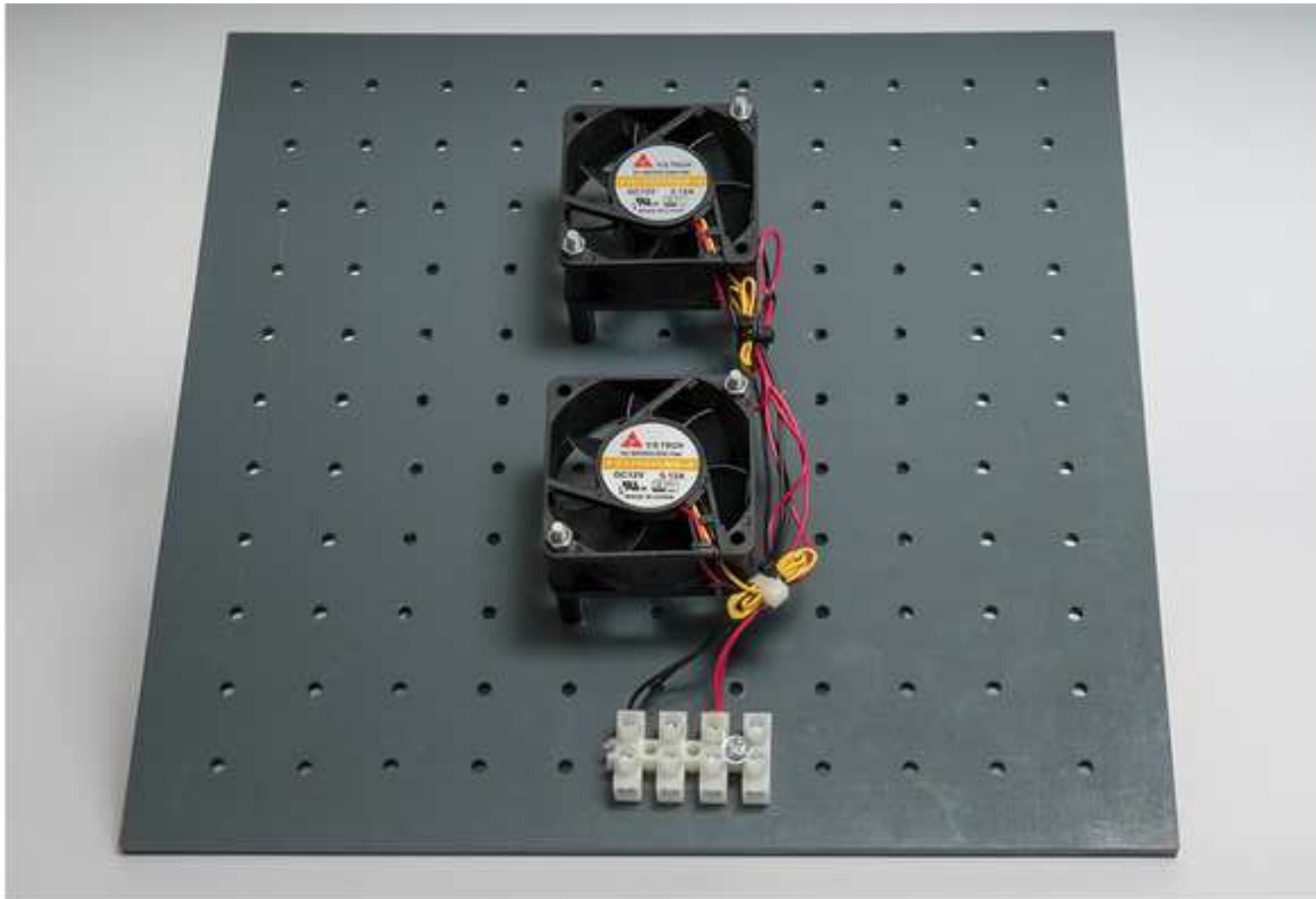
Figure 1

[Click here to access/download;Figure;Figure1.psd](#)



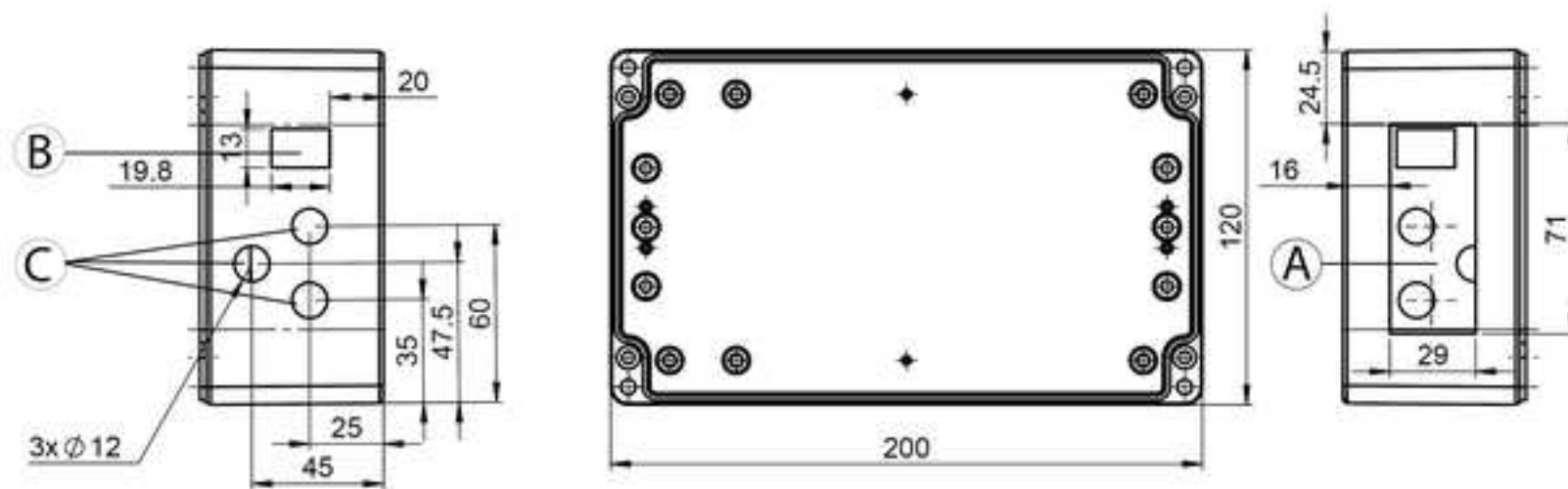
Figure 2





Cable color	Function
Black	Negative
Red	Positive
Yellow	Speed sensor

a.



b.

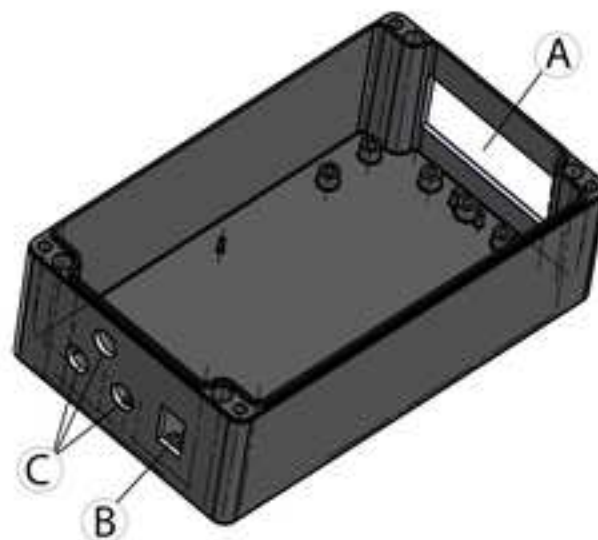
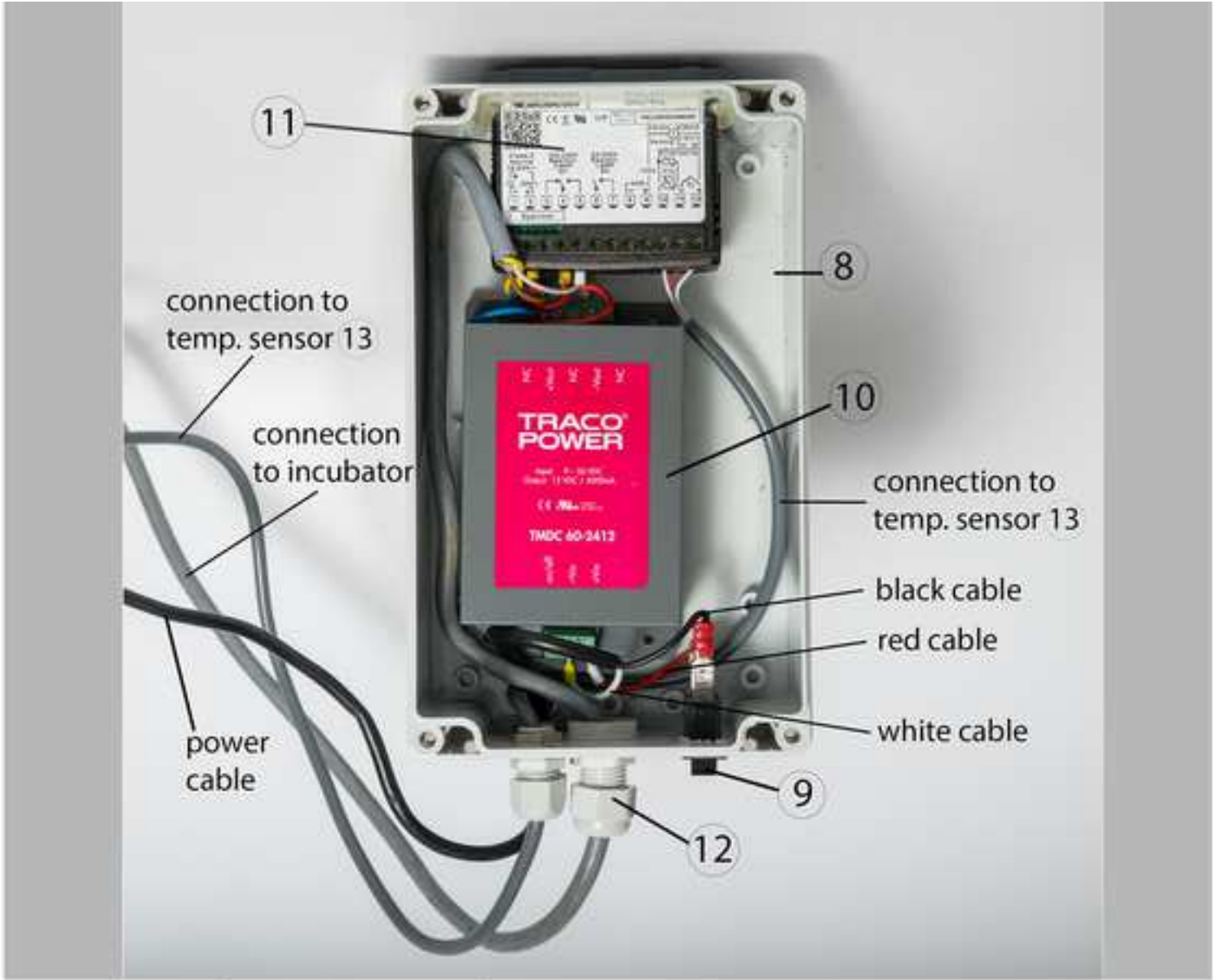
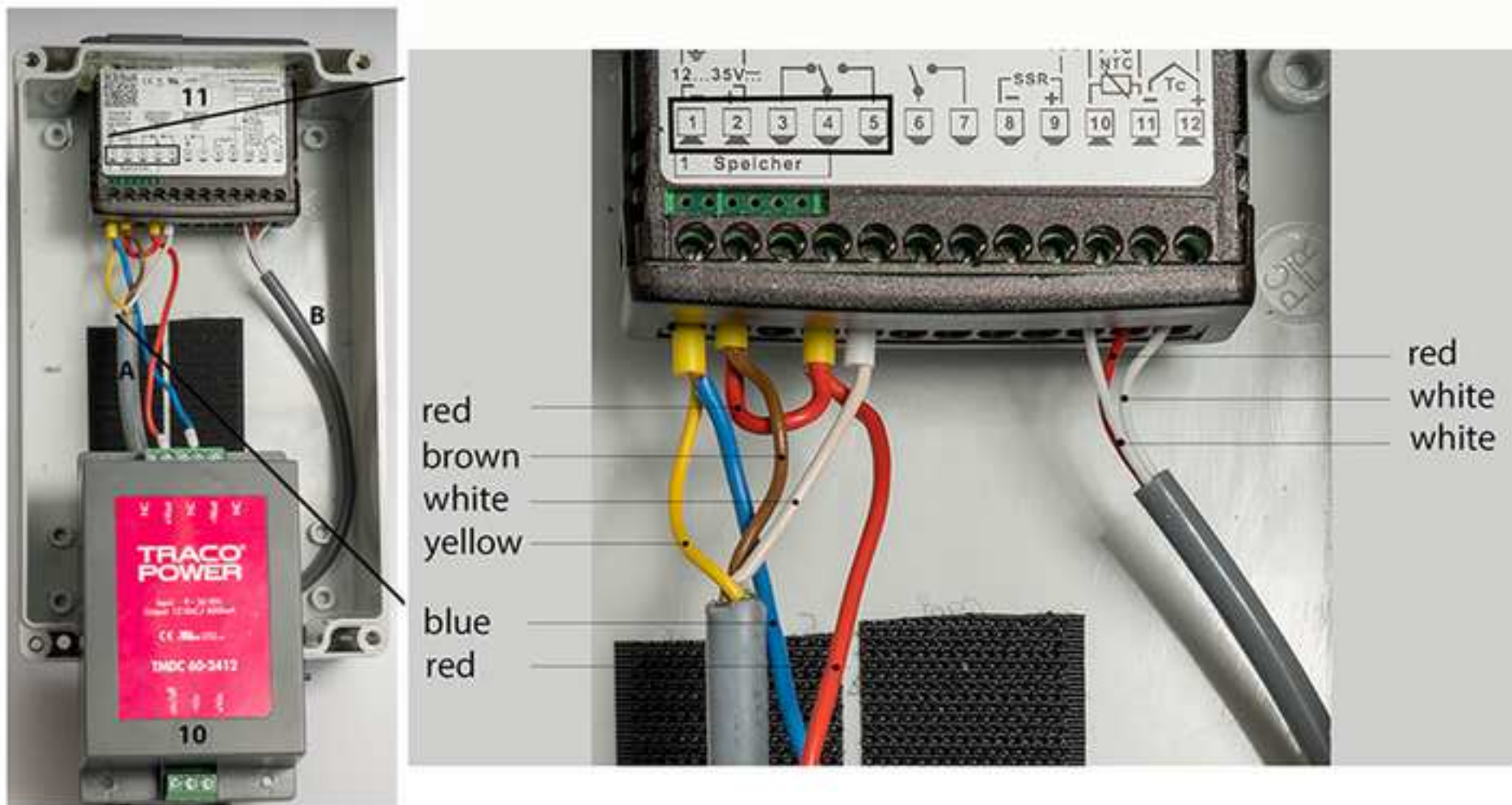


Figure 5



Cable color	Function	Connection
black	DC power +	AC power adapter to on/off switch
white	DC power -	AC power adapter to "-Vin" of DC/DC converter
red	DC power on/off	on/off switch to "+Vin" of DC/DC converter



Color	Function	Connection
Yellow	DC-	Terminal "1" of PID to heating unit of incubator
Blue	DC-	Terminal "1" of PID controller to "-Vout" of DC/DC converter
Red	DC+	Terminal "2" of PID controller to Terminal "4" of PID controller
Red	DC+	Terminal "4" of PID controller to "+Vout" of DC/DC converter
Brown	DC+	Terminal "2" of PID controller to heating unit of incubator
White	Command	Terminal "5" of PID to heating unit of incubator

Figure 7

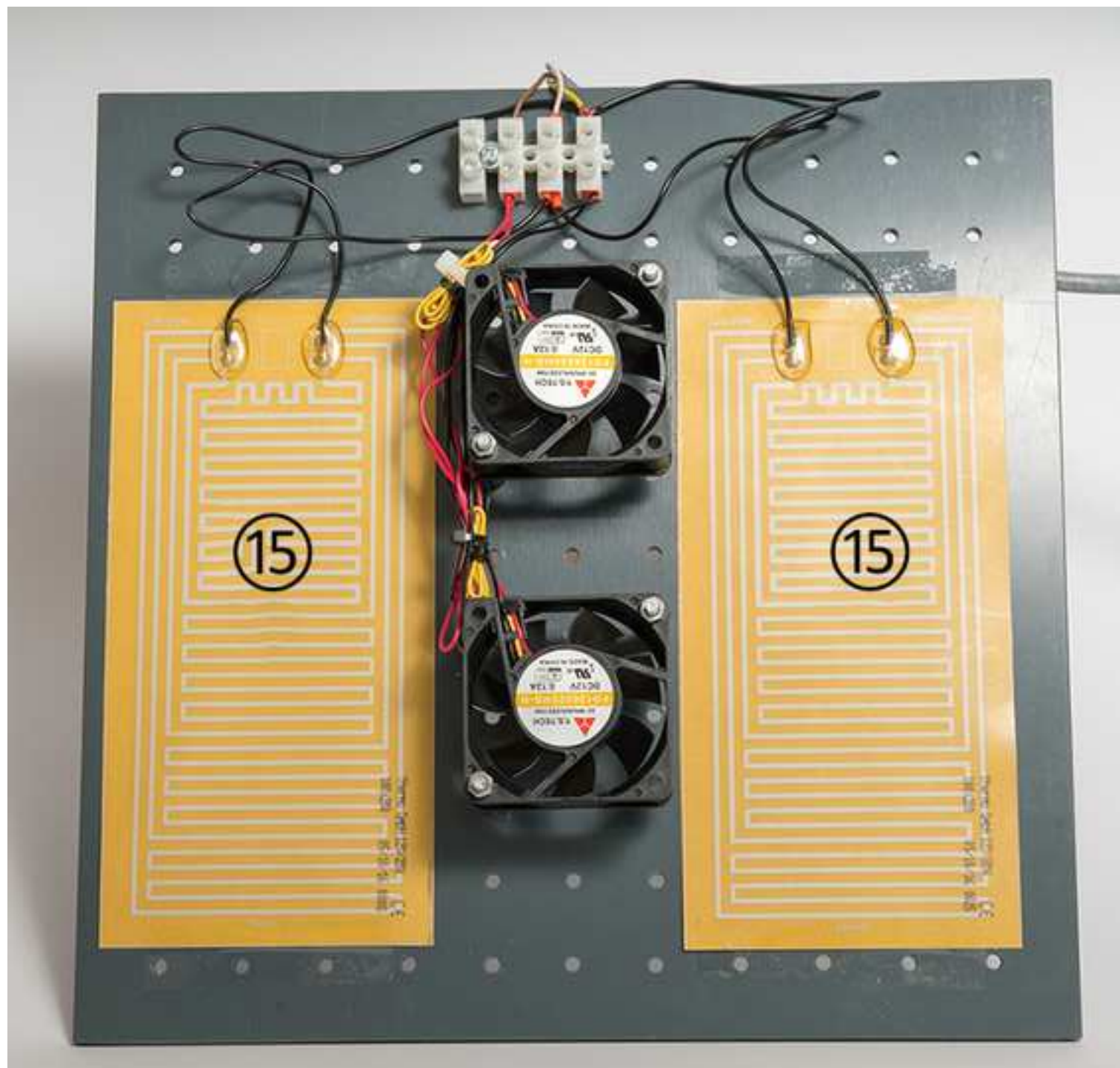


Figure 8



Figure 9

[Click here to access/download;Figure;Figure9.psd](#)

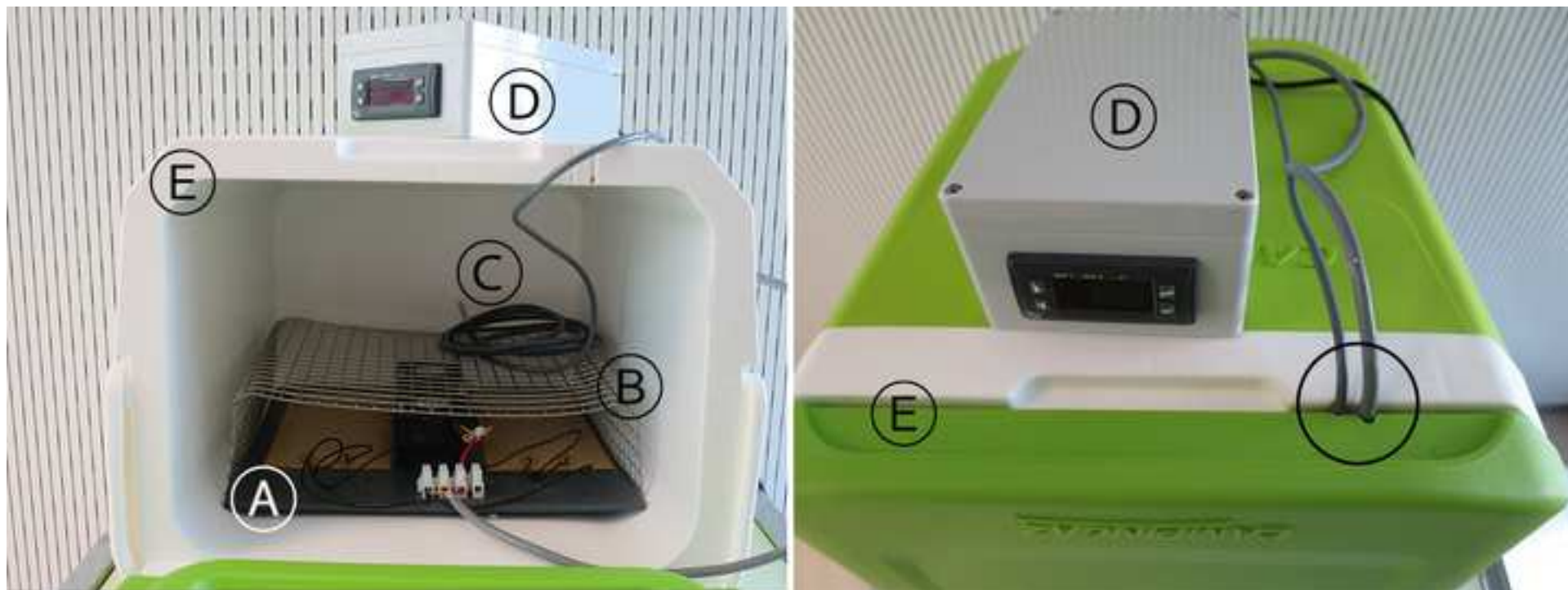
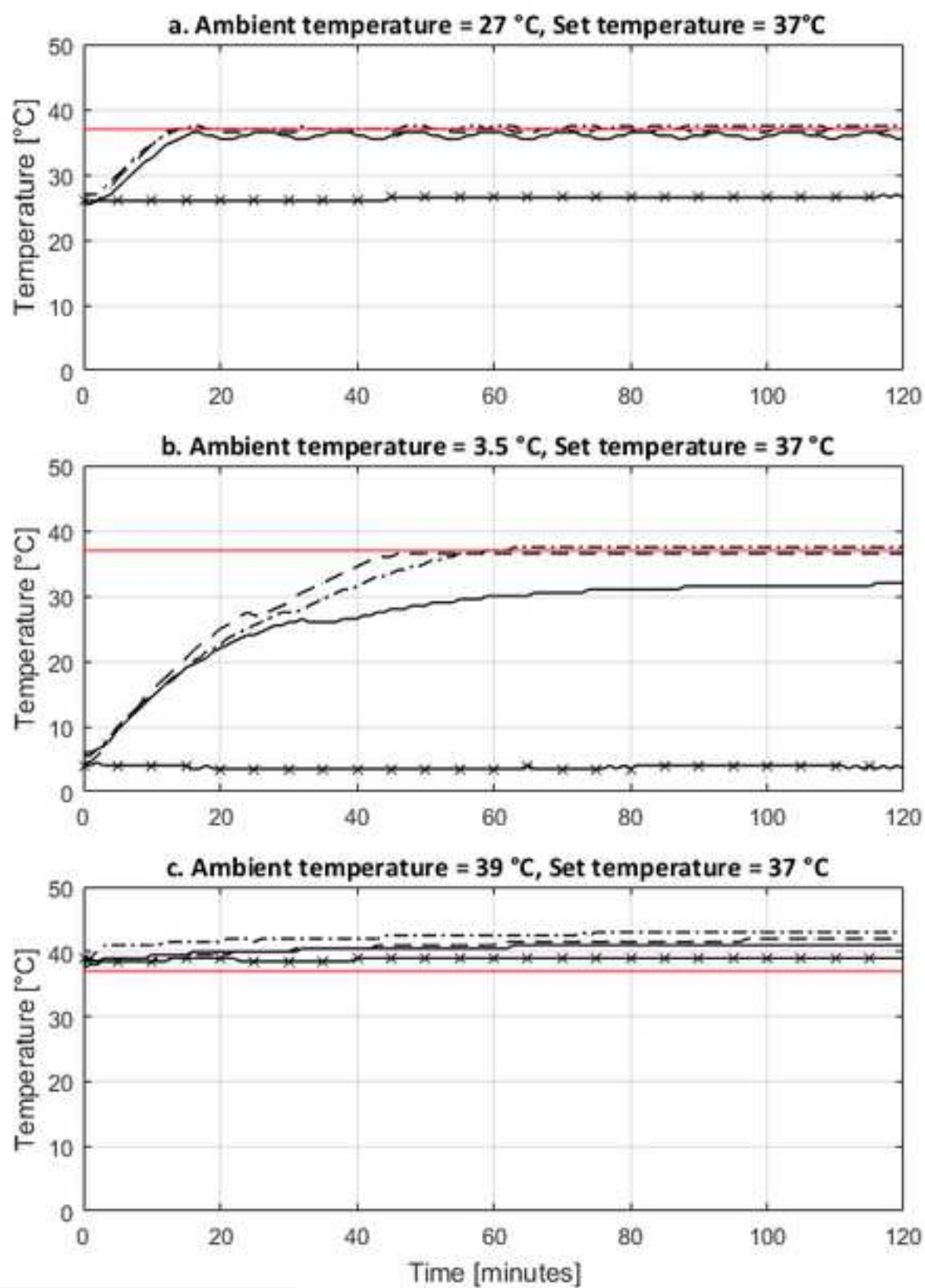


Figure 10

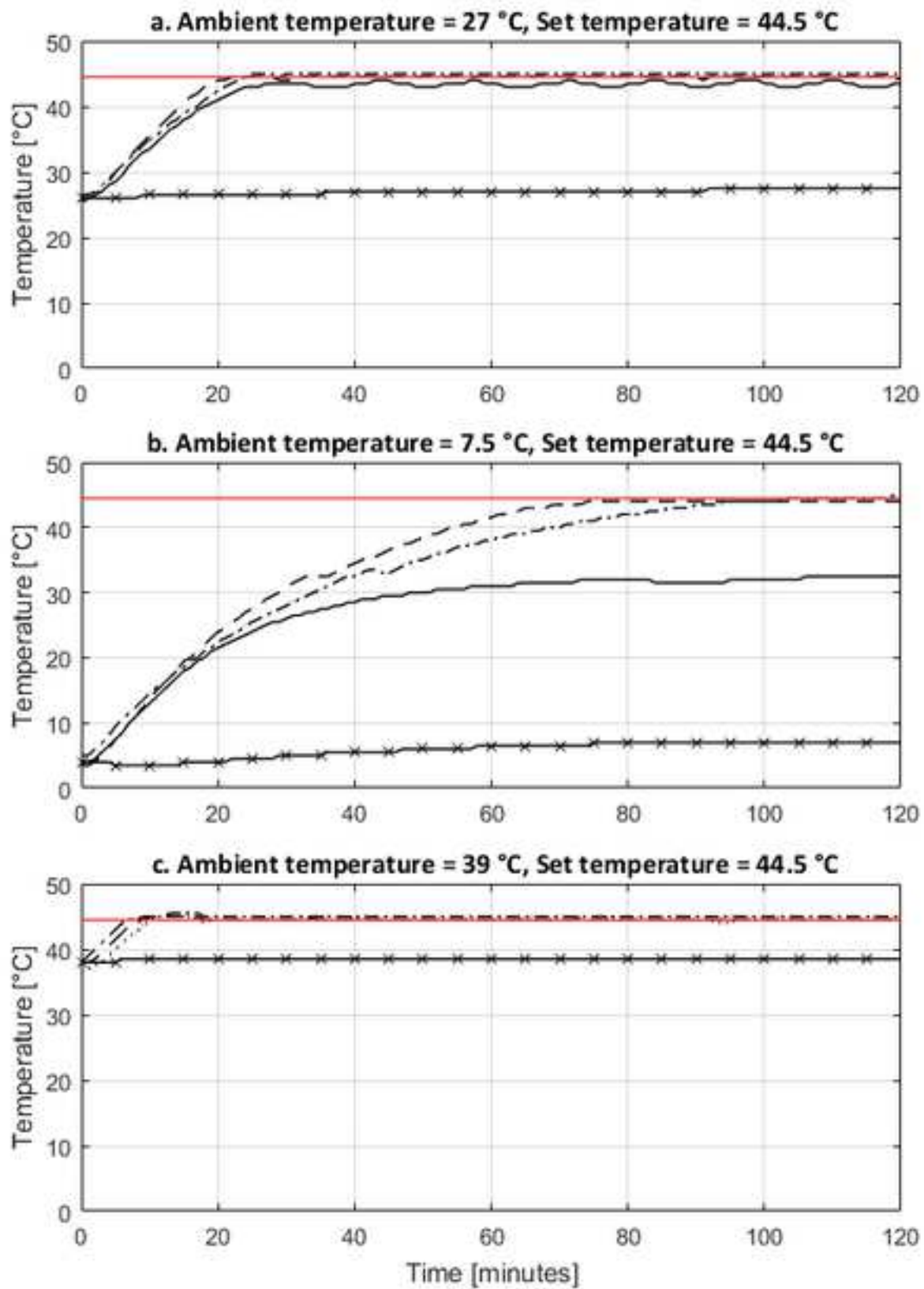
[Click here to access/download;Figure;Figure10.psd](#)



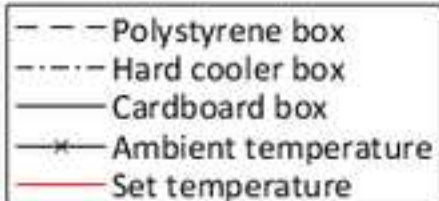
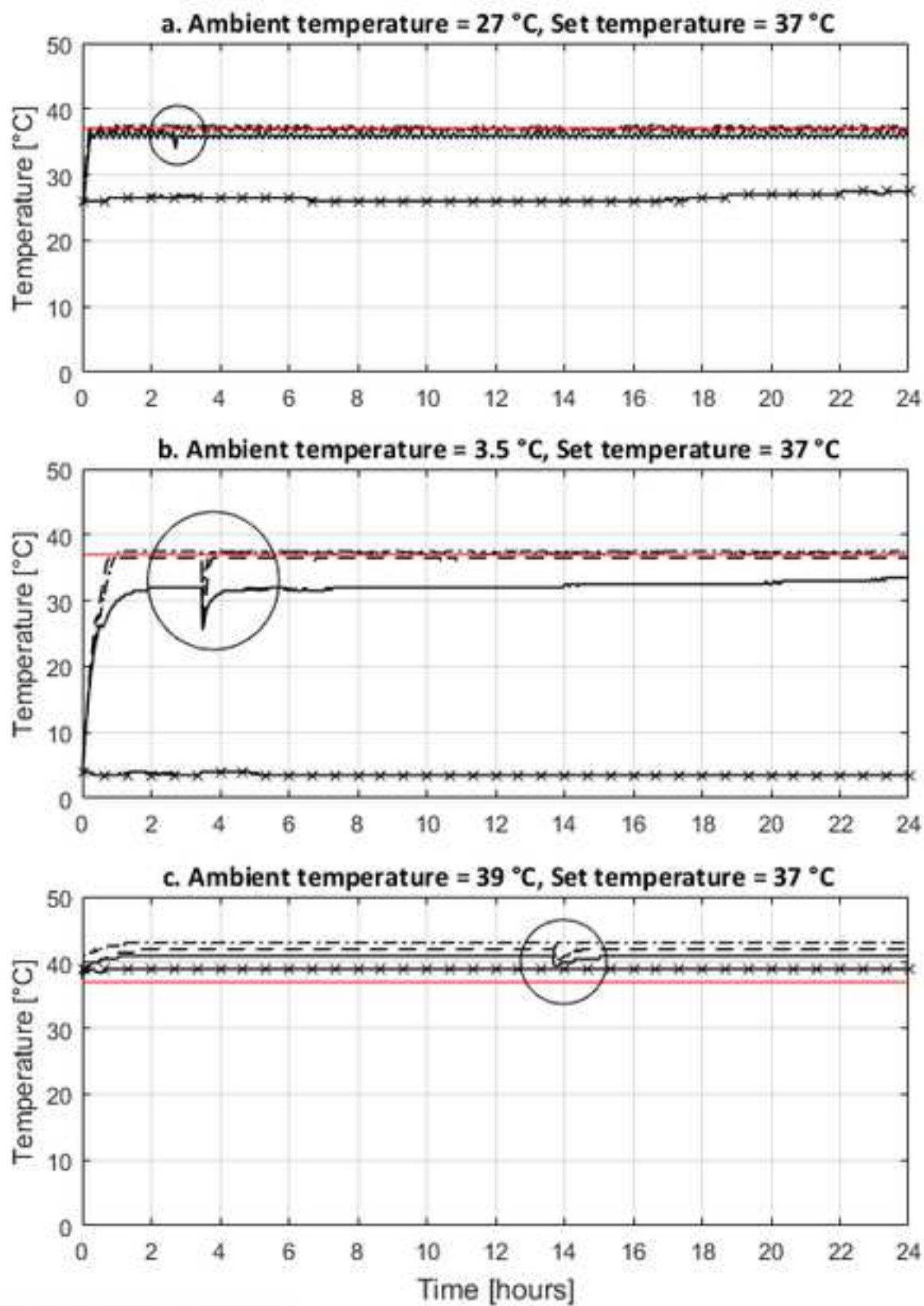
Time to set temperature



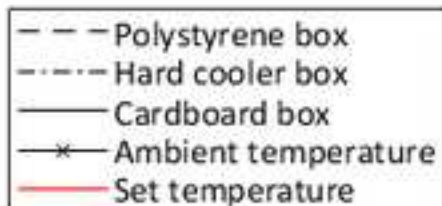
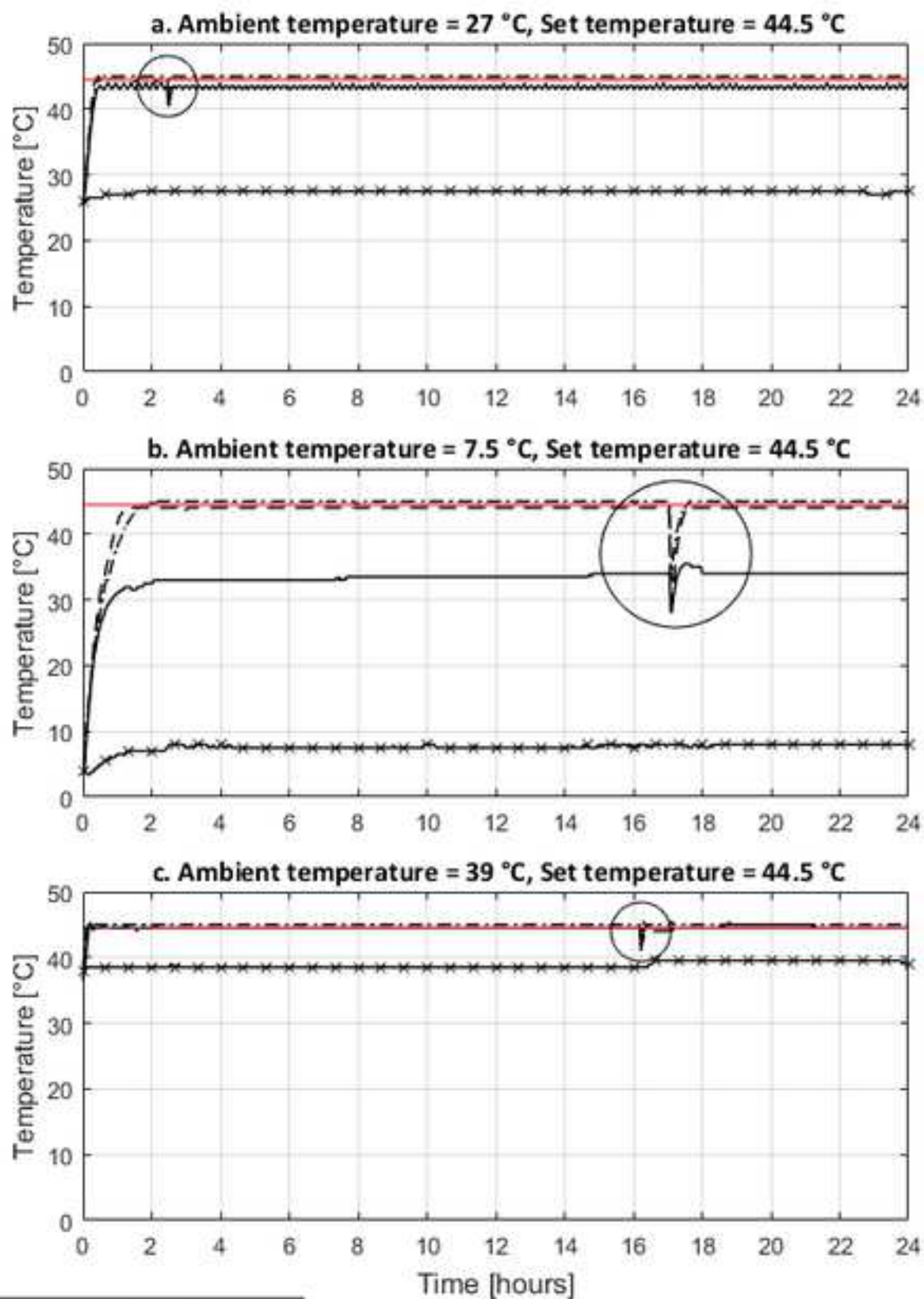
Time to set temperature



24 hours of operation



24 hours of operation



Characteristic	Laboratory-based	Field
User friendly design	✓	✗
Large capacity	✓	✗
Robust to wide range of ambient temperatures	✓	✗
Maintains constant temperature	✓	✗
Low cost	✗	✓
Easily transported	✗	✓
Energy efficient	✗	✓
Resilient to intermittent power supply	✗	✓

[illegible]

PID temperature controller terminal
Terminal "1"
Terminal "2"
Terminal "4"
Terminal "5"

Function
Supply input +
Supply input -
Control Output Common Contact
Control Output Normally Open Contact

			Test 1: Time to set temperature
	Ambient temperature	Set temperature	(min)
Polystyrene foam box	3.5 °C	37 °C	45
	7.5 °C	44.5 °C	74
	27 °C	37 °C	12
		44.5 °C	20
	39 °C	37 °C	0 (overheat)
		44.5 °C	7
Hard cooler box	3.5 °C	37 °C	54
	7.5 °C	44.5 °C	96
	27 °C	37 °C	13
		44.5 °C	25
	39 °C	37 °C	0 (overheat)
		44.5 °C	9
Cardboard box with survival blanket	3.5 °C	37 °C	Never reached (stable temperature after 109 min)
	7.5 °C	44.5 °C	Never reached (stable temperature after 120 min)
	27 °C	37 °C	15
		44.5 °C	24
	39 °C	37 °C	0 (overheat)
		44.5 °C	9
Standard incubator	27 °C	37 °C	18
		44.5 °C	23 (overheat)

Test 2: Side door opening one minute	Test 3: Power consumption over 24-hour period	Test 4: Temperature variation over 24- hour period	Test 5: <i>E. coli</i> growth observed
Maximal loss of temperature (°C); time to recover set temperature (min)	(kWh/24 h)	Absolute maximal temperature (°C); absolute minimal temperature (°C) *	(Yes / No)
10 °C; 17 min	0.78	37; 35.5	Yes
16.5 °C; 31 min	0.89	44.5; 42.5	ND [†]
2.5 °C; 3 min	0.28	37.5; 36.5	Yes
4.5 °C; 7 min	0.43	44.5; 43.5	ND [†]
2 °C; 0 min (overheat)	0.11	42.5; 42	Yes
3.5 °C; 5 min	0.17	45; 43.5	ND [†]
8 °C; 10 min	0.86	37.5; 36	Yes
12 °C; 30 min	1.05	45; 43	ND [†]
1.5 °C; 0 min	0.27	37.5; 36.5	Yes
2 °C; 4 min	0.50	45; 43.5	ND [†]
1 °C; 0 min (overheat)	0.11	43; 42.5	Yes
4 °C; 3 min	0.19	45.5; 44.5	ND [†]
6.5 °C; stable temperature after 30 min	1.24	33.5; 30.5	Yes
8 °C; stable temperature after 20 min	1.28	36.5; 32	ND [†]
2.5 °C; 6 min	0.42	36.5; 35.5	Yes
3 °C; 8 min	0.70	44.5; 42.5	ND [†]
1.5 °C; 0 min (overheat)	0.11	41.5; 40	Yes
2 °C; 0 min	0.20	45; 43.5	ND [†]
1 °C; 0 min (overheat)	0.64	38.5; 36	ND [†]
2.5 °C; 0 min	0.95	47.5; 43.5	ND [†]

Name of Material/ Equipment	Company	Catalog Number
Heating foil	Thermo	2115337
Axial fan	Yen Sun Technology Corp.	FD126025MB
PID Temperature Controller	Wachendorff Automation GmbH & Co. KG	UR3274S
Temperature sensor Pt100	Conrad	198466
Universal enclosure	OKW Gehäuse System	C2012201
ON/OFF Switch	SHIN CHIN INDUSTRIAL CO.	R13-70A-01
DC/DC converter	Traco Power	TMDC 60-2412
AC power adapter	Bicker Elektronik	BET-0612
Spacer	Schäfer Elektromechanik	20/4
Cable gland	WISKA	10066410
Luster terminal	Adels Contact	125312
Screw M4 x 50	Bossard	1579010
Screw nut M4	Bossard	1241478
Washer M4	Bossard	1887505
Screw M3 x 25	Bossard	1211099
Screw nut M3	Bossard	1241443
Washer M3	Bossard	1887483
Support plate	-	-

Comments/Description

Self-adhesive 10x20 cm; Operating voltage 12V; Power 20W

6x6x2.5 cm; Operating voltage 12VDC; Power 1.44W; Max. current consumption 60mA

PID controller 32x74 mm; Universal input for process signals, thermocouples, Pt100; Operating voltage 24 VDC; Outputs (thermostats) 10 A r

Temperature range -100°C to 200°C; Sensor Pt100, Type FS-400P

Dimensions 200 x 120 x 60 mm

Connection Type C CEE 7/16 plug 6.3 mm; Contact resistance Max 50 mΩ; Switching voltage 24 VDC; Switching current (mx.) 10A; Insulation r

Nominal voltage 24 VDC; Input voltage 9-36 VDC; Output voltage 12 VDC; Max. output current 5 A; Power 60W

Output voltage 12 VDC; Max. output current 5 A; Input voltage 115-230 VAC

Without thread; Thread size M4; Polystyrene; Distance 20 mm

M12 x 1.5 cm; clamping range 3 – 7 mm

Nominal current 25 A; Nominal Voltage 500V

M4 x 50 mm

M4

M4

M3 x 25 mm

M3

M3

Insulating material (plastic or other); 28 x 25 cm

elay, 5 A relay, SSR, RS 485

esistance Min 100 M Ω /500 Vdc



1 Alewife Center #200
Cambridge, MA 02140
tel. 617.945.9051
www.jove.com

ARTICLE AND VIDEO LICENSE AGREEMENT

Title of Article:

Construction of a low-cost field-robust incubator

Author(s):

Ariane Schertenleib, Jürg Sigrist, Max ND Friedrich, Christian Ebi, Frederik Hammes, Sara J Marks

Item 1 (check one box): The Author elects to have the Materials be made available (as described at

<http://www.jove.com/author>) via: ☐ Standard Access ☒ Open Access

Item 2 (check one box):

☒ The Author is NOT a United States government employee.

☐ The Author is a United States government employee and the Materials were prepared in the course of his or her duties as a United States government employee.

☐ The Author is a United States government employee but the Materials were NOT prepared in the course of his or her duties as a United States government employee.

ARTICLE AND VIDEO LICENSE AGREEMENT

1. Defined Terms. As used in this Article and Video License Agreement, the following terms shall have the following meanings: “**Agreement**” means this Article and Video License Agreement; “**Article**” means the article specified on the last page of this Agreement, including any associated materials such as texts, figures, tables, artwork, abstracts, or summaries contained therein; “**Author**” means the author who is a signatory to this Agreement; “**Collective Work**” means a work, such as a periodical issue, anthology or encyclopedia, in which the Materials in their entirety in unmodified form, along with a number of other contributions, constituting separate and independent works in themselves, are assembled into a collective whole; “**CRC License**” means the Creative Commons Attribution-Non Commercial-No Derivs 3.0 Unported Agreement, the terms and conditions of which can be found at: <http://creativecommons.org/licenses/by-nc-nd/3.0/legalcode>; “**Derivative Work**” means a work based upon the Materials or upon the Materials and other pre-existing works, such as a translation, musical arrangement, dramatization, fictionalization, motion picture version, sound recording, art reproduction, abridgment, condensation, or any other form in which the Materials may be recast, transformed, or adapted; “**Institution**” means the institution, listed on the last page of this Agreement, by which the Author was employed at the time of the creation of the Materials; “**JoVE**” means MyJoVE Corporation, a Massachusetts corporation and the publisher of *The Journal of Visualized Experiments*; “**Materials**” means the Article and / or the Video; “**Parties**” means the Author and JoVE; “**Video**” means any video(s) made by the Author, alone or in conjunction with any other parties, or by JoVE or its affiliates or agents, individually or in collaboration with the Author or any other parties, incorporating all or any portion of the Article, and in which the Author may or may not appear.

2. Background. The Author, who is the author of the Article, in order to ensure the dissemination and protection of the Article, desires to have the JoVE publish the Article and create and transmit videos based on the Article. In furtherance of such goals, the Parties desire to memorialize in this Agreement the respective rights of each Party in and to the Article and the Video.

3. Grant of Rights in Article. In consideration of JoVE agreeing to publish the Article, the Author hereby grants to JoVE, subject to **Sections 4** and **7** below, the exclusive, royalty-free, perpetual (for the full term of copyright in the Article, including any extensions thereto) license (a) to publish, reproduce, distribute, display and store the Article in all forms, formats and media whether now known or hereafter developed (including without limitation in print, digital and electronic form) throughout the world, (b) to translate the Article into other languages, create adaptations, summaries or extracts of the Article or other Derivative Works (including, without limitation, the Video) or Collective Works based on all or any portion of the Article and exercise all of the rights set forth in (a) above in such translations, adaptations, summaries, extracts, Derivative Works or Collective Works and (c) to license others to do any or all of the above. The foregoing rights may be exercised in all media and formats, whether now known or hereafter devised, and include the right to make such modifications as are technically necessary to exercise the rights in other media and formats. If the “Open Access” box has been checked in **Item 1** above, JoVE and the Author hereby grant to the public all such rights in the Article as provided in, but subject to all limitations and requirements set forth in, the CRC License.

ARTICLE AND VIDEO LICENSE AGREEMENT

4. Retention of Rights in Article. Notwithstanding the exclusive license granted to JoVE in **Section 3** above, the Author shall, with respect to the Article, retain the non-exclusive right to use all or part of the Article for the non-commercial purpose of giving lectures, presentations or teaching classes, and to post a copy of the Article on the Institution's website or the Author's personal website, in each case provided that a link to the Article on the JoVE website is provided and notice of JoVE's copyright in the Article is included. All non-copyright intellectual property rights in and to the Article, such as patent rights, shall remain with the Author.

5. Grant of Rights in Video – Standard Access. This **Section 5** applies if the "Standard Access" box has been checked in **Item 1** above or if no box has been checked in **Item 1** above. In consideration of JoVE agreeing to produce, display or otherwise assist with the Video, the Author hereby acknowledges and agrees that, Subject to **Section 7** below, JoVE is and shall be the sole and exclusive owner of all rights of any nature, including, without limitation, all copyrights, in and to the Video. To the extent that, by law, the Author is deemed, now or at any time in the future, to have any rights of any nature in or to the Video, the Author hereby disclaims all such rights and transfers all such rights to JoVE.

6. Grant of Rights in Video – Open Access. This **Section 6** applies only if the "Open Access" box has been checked in **Item 1** above. In consideration of JoVE agreeing to produce, display or otherwise assist with the Video, the Author hereby grants to JoVE, subject to **Section 7** below, the exclusive, royalty-free, perpetual (for the full term of copyright in the Article, including any extensions thereto) license (a) to publish, reproduce, distribute, display and store the Video in all forms, formats and media whether now known or hereafter developed (including without limitation in print, digital and electronic form) throughout the world, (b) to translate the Video into other languages, create adaptations, summaries or extracts of the Video or other Derivative Works or Collective Works based on all or any portion of the Video and exercise all of the rights set forth in (a) above in such translations, adaptations, summaries, extracts, Derivative Works or Collective Works and (c) to license others to do any or all of the above. The foregoing rights may be exercised in all media and formats, whether now known or hereafter devised, and include the right to make such modifications as are technically necessary to exercise the rights in other media and formats. For any Video to which this Section 6 is applicable, JoVE and the Author hereby grant to the public all such rights in the Video as provided in, but subject to all limitations and requirements set forth in, the CRC License.

7. Government Employees. If the Author is a United States government employee and the Article was prepared in the course of his or her duties as a United States government employee, as indicated in **Item 2** above, and any of the licenses or grants granted by the Author hereunder exceed the scope of the 17 U.S.C. 403, then the rights granted hereunder shall be limited to the maximum rights permitted under such

statute. In such case, all provisions contained herein that are not in conflict with such statute shall remain in full force and effect, and all provisions contained herein that do so conflict shall be deemed to be amended so as to provide to JoVE the maximum rights permissible within such statute.

8. Likeness, Privacy, Personality. The Author hereby grants JoVE the right to use the Author's name, voice, likeness, picture, photograph, image, biography and performance in any way, commercial or otherwise, in connection with the Materials and the sale, promotion and distribution thereof. The Author hereby waives any and all rights he or she may have, relating to his or her appearance in the Video or otherwise relating to the Materials, under all applicable privacy, likeness, personality or similar laws.

9. Author Warranties. The Author represents and warrants that the Article is original, that it has not been published, that the copyright interest is owned by the Author (or, if more than one author is listed at the beginning of this Agreement, by such authors collectively) and has not been assigned, licensed, or otherwise transferred to any other party. The Author represents and warrants that the author(s) listed at the top of this Agreement are the only authors of the Materials. If more than one author is listed at the top of this Agreement and if any such author has not entered into a separate Article and Video License Agreement with JoVE relating to the Materials, the Author represents and warrants that the Author has been authorized by each of the other such authors to execute this Agreement on his or her behalf and to bind him or her with respect to the terms of this Agreement as if each of them had been a party hereto as an Author. The Author warrants that the use, reproduction, distribution, public or private performance or display, and/or modification of all or any portion of the Materials does not and will not violate, infringe and/or misappropriate the patent, trademark, intellectual property or other rights of any third party. The Author represents and warrants that it has and will continue to comply with all government, institutional and other regulations, including, without limitation all institutional, laboratory, hospital, ethical, human and animal treatment, privacy, and all other rules, regulations, laws, procedures or guidelines, applicable to the Materials, and that all research involving human and animal subjects has been approved by the Author's relevant institutional review board.

10. JoVE Discretion. If the Author requests the assistance of JoVE in producing the Video in the Author's facility, the Author shall ensure that the presence of JoVE employees, agents or independent contractors is in accordance with the relevant regulations of the Author's institution. If more than one author is listed at the beginning of this Agreement, JoVE may, in its sole discretion, elect not take any action with respect to the Article until such time as it has received complete, executed Article and Video License Agreements from each such author. JoVE reserves the right, in its absolute and sole discretion and without giving any reason therefore, to accept or decline any work submitted to JoVE. JoVE and its employees, agents and independent contractors shall have

ARTICLE AND VIDEO LICENSE AGREEMENT

full, unfettered access to the facilities of the Author or of the Author's institution as necessary to make the Video, whether actually published or not. JoVE has sole discretion as to the method of making and publishing the Materials, including, without limitation, to all decisions regarding editing, lighting, filming, timing of publication, if any, length, quality, content and the like.

11. **Indemnification.** The Author agrees to indemnify JoVE and/or its successors and assigns from and against any and all claims, costs, and expenses, including attorney's fees, arising out of any breach of any warranty or other representations contained herein. The Author further agrees to indemnify and hold harmless JoVE from and against any and all claims, costs, and expenses, including attorney's fees, resulting from the breach by the Author of any representation or warranty contained herein or from allegations or instances of violation of intellectual property rights, damage to the Author's or the Author's institution's facilities, fraud, libel, defamation, research, equipment, experiments, property damage, personal injury, violations of institutional, laboratory, hospital, ethical, human and animal treatment, privacy or other rules, regulations, laws, procedures or guidelines, liabilities and other losses or damages related in any way to the submission of work to JoVE, making of videos by JoVE, or publication in JoVE or elsewhere by JoVE. The Author shall be responsible for, and shall hold JoVE harmless from, damages caused by lack of sterilization, lack of cleanliness or by contamination due to the making of a video by JoVE its employees, agents or independent contractors. All sterilization, cleanliness or decontamination procedures shall be solely the responsibility of the Author and shall be undertaken at the Author's


expense. All indemnifications provided herein shall include JoVE's attorney's fees and costs related to said losses or damages. Such indemnification and holding harmless shall include such losses or damages incurred by, or in connection with, acts or omissions of JoVE, its employees, agents or independent contractors.

12. **Fees.** To cover the cost incurred for publication, JoVE must receive payment before production and publication the Materials. Payment is due in 21 days of invoice. Should the Materials not be published due to an editorial or production decision, these funds will be returned to the Author. Withdrawal by the Author of any submitted Materials after final peer review approval will result in a US\$1,200 fee to cover pre-production expenses incurred by JoVE. If payment is not received by the completion of filming, production and publication of the Materials will be suspended until payment is received.

13. **Transfer, Governing Law.** This Agreement may be assigned by JoVE and shall inure to the benefits of any of JoVE's successors and assignees. This Agreement shall be governed and construed by the internal laws of the Commonwealth of Massachusetts without giving effect to any conflict of law provision thereunder. This Agreement may be executed in counterparts, each of which shall be deemed an original, but all of which together shall be deemed to be one and the same agreement. A signed copy of this Agreement delivered by facsimile, e-mail or other means of electronic transmission shall be deemed to have the same legal effect as delivery of an original signed copy of this Agreement.

A signed copy of this document must be sent with all new submissions. Only one Agreement required per submission.

CORRESPONDING AUTHOR:

Name:	Ariane Schertenleib	
Department:	Sandec	
Institution:	Eawag	
Article Title:	Construction of a low-cost field-robust incubator	
Signature:		Date: 08.05.2018

Please submit a signed and dated copy of this license by one of the following three methods:

- 1) Upload a scanned copy of the document as a pdf on the JoVE submission site;
- 2) Fax the document to +1.866.381.2236;
- 3) Mail the document to JoVE / Attn: JoVE Editorial / 1 Alewife Center #200 / Cambridge, MA 02139

For questions, please email submissions@jove.com or call +1.617.945.9051

JoVE submission JoVE58443R1 Construction of a low-cost mobile field-robust incubator for field and laboratory use
Author responses to comments

Dear Dr. Philip Steindel,

Thank you for the detailed and constructive feedback on our Manuscript (JoVE58443R1, “Construction of a low-cost mobile field-robust incubator for field and laboratory use”). Below please find our point-by-point responses to the editor comments, with edits indicated by underlined text. All edits are also visible in “track changes” within the revised manuscript.

Sincerely,

Ariane Schertenleib

Editorial comments:

1. 2.2: How exactly is the milling done here? We will need this information to be in the manuscript so that we can properly write the shooting script.
 - The text was modified to: “Mill the openings for the PID temperature controller, on/off switch and cable glands into the enclosure with a drill and jigsaw, or an equivalent tool (Figure 4)”
2. Please combine Figures 4a and 4b into one image file, with panels labeled.
 - Done. Figures 4a and 4b have been combined into one figure. The figure legend has also been modified to:
“**Figure 4a: Schematic diagram of the universal enclosure.** (a) Indications to place the temperature controller (A), on/off switch (B) and the cable glands (C) into the universal enclosure, distances are given in millimeter; (b) 3D view of the universal enclosure.”
~~“**Figure 4b: 3D view of the universal enclosure.** Openings for temperature controller (A), on/off switch (B), cable glands (C). ”~~
3. Figures 11-14: Why are the graphs labeled as A1, A2, etc.? It’s a little confusing as is. Are they intended to be panels (see above)?
 - Thank you for pointing out this confusing labeling. Figures 11-14 have been re-labeled a, b, c instead of A1, A2, etc.
References to these figures in the main text and in the figure legends have been modified as well.

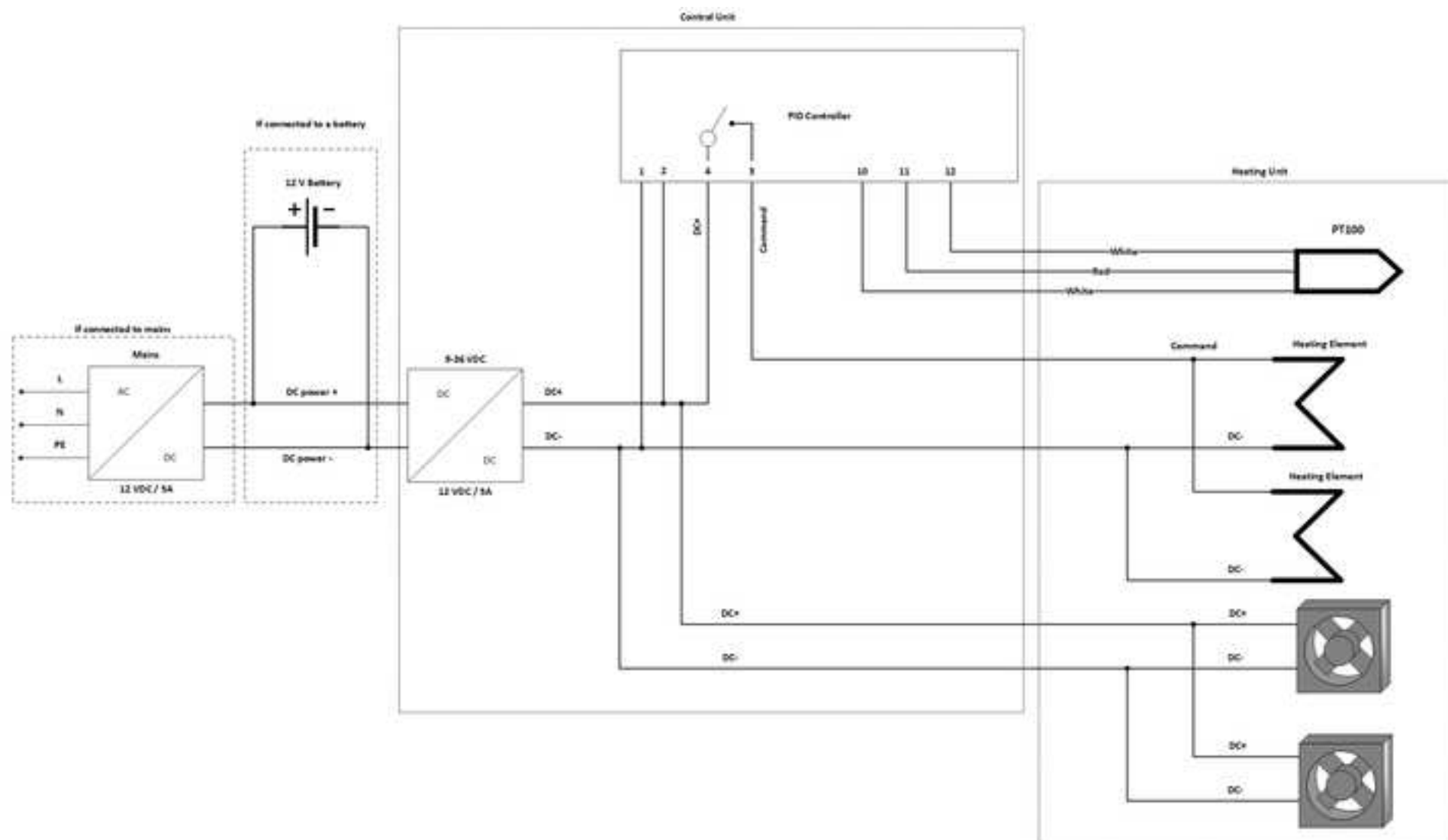
4. Table 3: What does ‘/’ mean in the last column? Also, what exactly is ‘successful’ *E. coli* growth?

- The paragraph at line 309 explains that replicates of water samples plated for *E. coli* were placed in each incubator and in a standard incubator to compare the growth. In Table 3, the column “Test 5” reports if the growth observed, and similar in the incubator and in the standard incubator. Tests were run only at 37 °C, as only *E. coli* were tested. Therefore, the rows in Table 3 corresponding to tests at 44.5°C do not report any results.

Table 3 has been modified as follows for more clarity:

- The header of the column was changed to “Test 5: ~~Successful~~ *E. coli* growth observed”
- The “/” in the last columns were replaced by “ND[†]” with a footnote in the legend:
“Table 3: Results summary for the incubator configurations and ambient temperature conditions tested. *Test 4: Absolute maximal and minimal temperatures recorded during the stable periods, i.e. from 10 minutes after the end of a disruptive event (time to reach set temperature, opening the door). [†]ND: No data, test not run.”
- Line 259: the reference to Test 5 has been changed to :” ... and ~~successful~~ observation of *E. coli* growth.”

Figure S1



	Parameter	Selected value
1	Type of control output	Control Q1 / Alarm Q2
2	Type of connected sensor	Pt100 (-200 to 140°C)
3	Lower limit selectable for setpoint value	0
4	Upper limit selectable for setpoint value	50
5	Type of control	Heating
6	ON/OFF hysteresis or dead band for P.I.D. control	0
7	Proportional bandwidth of the process expressed as units (°C if temperature)	1
8	Integral time. Inertia of the process expressed as seconds	80.0
9	Derivative time for P.I.D.	20.0
10	Cycle time for time-proportioning output	10
11	Allow/deny modification of setpoint values by frontal keyboard	Allow modification of all setpoints
12	Software filter. Number of readings to calculate the comparison value PV-SPV	10
13	Type of degree	°C
14	Type of cooling liquid	Air