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MicroMI: A portable microbiological mobile incubator that uses inexpensive lithium power banks for field microbiology



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ABSTRACT

Incubation at controlled temperature is a key step in culture based microbiological tests. Access to culture-based microbiological testing requires access to conventional incubators in a laboratory. Portable incubators allow microbiological testing in the field and in resource-limited settings, and can eliminate the challenge of sample transportation, minimising the chance of sample degradation. Recent studies have reported low-cost portable incubator designs suitable for field or off-grid use, but these either need an external power supply (e.g. mains AC or 12 V DC), or rely on passive heating without thermostatic control. Here we report that small inexpensive uninterruptible power supply (UPS) products manufactured for consumer electronics and powered by lithium-ion battery packs allowing thermostatic temperature control in small portable incubators that can maintain precise temperatures with or without external power. We present an open-source design for a Microbiological Mobile Incubator (MicroMI) in two sizes for field use. The MicroMI is built from simple and widely available components and is easy to set up. The open source design can be customised for different numbers of samples. The smallest and most efficient design uses a vacuum insulated food flask that allows longer operation with smaller, lower capacity UPS. The larger flight case design has space for more samples, but depletes the battery faster. The UPS maintains a typical microbiology incubation temperature for up to 24 h without external power- ideal for typical incubation needed for culture methods. The battery capacity, incubator design, and external ambient temperature all affected duration of operation without requiring external power. We validated the MicroMI by conducting classical microbiological tests using agar petri dishes, slant cultures and dip slides, and biochemical tests. We conclude the MicroMI design allows inexpensive lithium battery products to be used to simplify field microbiology and increase access to vital analytical microbiology testing.

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Introduction

Specifications table

Hardware name	MicroMI (<u>Micro</u> biological <u>mobile incubator</u>)
Subject area	<ul style="list-style-type: none"> • Microbiology • Environmental surveillance • Training Tool
Hardware type	Microbiological testing
Cost of Hardware	£185 – 8300 mAH capacity 12v UPS large MicroMI£120 – small MicroMI
Source File Repository	Available with the article

Hardware in context

Infectious disease including bacterial infection remains a major global public health challenge. Incubation at a controlled temperature is required for culture-based analytical microbiology, and in spite of the emergence of important new molecular methods (e.g. sequencing, PCR and mass spectroscopy), culture methods are still widely used including for sterility testing, for microbial isolation and bacterial identification through growth on solid media, often combined with biochemical and phenotype measurements, and to measure pathogen growth on differing substrates. Culture-based methods alongside functional antibiotic drug susceptibility tests remain critical for public health [7]) [11]. Some methods, such as broth microdilution antibiotic susceptibility testing, still requires culture in an incubator, and biochemical phenotyping also requires controlled temperature [10]. The first temperature-controlled incubation devices for microbiological applications were described in 1958 [4]; since then, thousands of laboratory incubator designs have been introduced and sold on the market, becoming ubiquitous inside laboratories.

Currently analytical microbiology is conducted in laboratories equipped with bespoke incubators. A major limitation of laboratory services, for example in outbreak response or global health, is slow turnaround time. Sample testing turnaround time is a key performance indicator of laboratory activities and fast results are vital for example to support hospital treatment [2,9]. In remote areas or those with limited resources, laboratory access can limit the practical use of culture-based test methods. Alongside limited access to microbiological testing labs, the transportation of specimens from local sampling sites to the reference laboratory can be a major challenge, with concerns about sample degradation during transportation if not tested soon after the sample is collected, especially for culture and phenotypic methods. Transport to laboratories can also increase cost of testing. To help address those issues, several recent studies have reported low cost mobile incubators based on widely available materials and construction methods, convenience for use in low resources environments, and for integration into automated novel technology [3,5,6,8,11].

Current innovative incubator designs have made use of heated foam ice boxes [3] or food chiller boxes [1] powered by a 12 V car battery or similar flexible power supplies. Such designs have made use of widely available and inexpensive temperature control modules, coupled to resistive heater panels and fans [11]. These are compatible with a wide range of external power sources including 12v DC and 120–240v AC, and can often be operated off-grid using existing portable power supplies. Where laboratory power supplies are interrupted, i.e. during power outages, temperature may be maintained depending on the size/insulation of the incubator, but to avoid this laboratories must be supplied by expensive backup power supplies (generator, battery). To our knowledge however, all the electrically powered incubator designs require an external power supply, whether through mains infrastructure or off-grid via battery combined with field generation. The incorporation of lithium battery power supplies into these designs has not previously been reported. Alternatives to electrically heated and thermostatically controlled incubator have been proposed, including passive heating using hot water in bottles within an insulated box [3]. Whilst effective for many applications, the temperature maintained may vary and accuracy is dependent on both ambient temperature and hot water temperature, and many microbiological testing protocols specify stricter temperature control.

The continued fall in cost and increased power:weight ratio of rechargeable batteries for example using lithium-ion cells largely driven by consumer electronics (e.g. smartphones) now provides the potential for a fully portable incubator incorporating consumer rechargeable battery packs in the design. Even more convenience is possible if an external power supply can be combined with internal batteries without user input. Here, we explored whether the latest lithium battery packs in the form of inexpensive consumer Uninterruptible Power Supply (UPS) units have sufficient power and energy capacity to maintain temperature within portable microbiological incubators in various sizes and form factors, for applications where a continuous external power supply is not available or convenient. UPS products offer internal control circuits that switch seamlessly between external mains power (e.g. 120–240v AC) vs internal battery power. Recent demand in portable consumer electronics (smartphones) has led to lightweight highly portable UPS products based on lithium battery packs with 12v output and 5000–10000 mAh capacity (at 12v) becoming widely available and costing <£100. Even larger capacity



Fig. 1. The concept of MicroMI design (*) When unplugging from external electrical supply, the MicroMI with 12 V UPS maintains temperature from 9 to 12 h, depending on incubator size, external ambient temperature, and UPS battery capacity.

(>20000mAh) products are becoming more widely available. We developed MicroMI (Microbiological mobile incubator): an inexpensive mobile incubator concept, designed to solve barriers arising from transportation of samples to microbiology laboratories, to improve turnaround time for public health microbiological testing, and to help move analytical microbiology testing into the field. We combined low cost (under £100), portable consumer UPS providing dual 12v and 5v USB outputs, with other readily available components, to create an incubator design which can be unplugged from external power and transported (Fig. 1). We found these small portable incubators maintain temperature reliably for up to 24 h without external power, long enough to complete a typical microbiology incubation between charging.

We provide here designs to build two different sized inexpensive (£125 to £185) portable incubators powered by small and lightweight consumer UPS products containing modest capacity lithium-ion batteries (manufacturer specified capacity ~ 6-8Ah at 12 V corresponding to ~<100 Wh or ~<360 kJ). The large MicroMI design has space to incubate agar plates and can be constructed entirely from off-the-shelf parts. The smaller vacuum flask design was designed to be contained within a small backpack (<15L capacity) to aid transportation and field use, yet has sufficient space to incubate 4 dip-slides or slant agar cultures; this design requires two 3D printed components in addition to off-the-shelf parts. We provide validation data indicating the power required for these two different configurations with the smaller vacuum flask requiring significantly lower power allowing it to maintain temperature for longer using the same UPS capacity.

The key design features of these mobile incubators include:

Small size (W380 × H145 × D400 mm at 2.64 kg for suitcase, and 180 × 122 × 118 mm for vacuum flask at 650 g)

3. Makes use of small portable UPS to maintain suitable microbiological culture temperatures (typically 37 °C) without external power, for example outside the laboratory or during power outages.

Simple customisable designs to make use of locally available and inexpensive (<£100) components.

This paper provides design details for both size variants and outlines the critical design parameters to facilitate customisation and use of locally-available components. We include validation data showing temperature profiles, estimated power consumption, and successful analytical microbiology testing of reference stains, real microbiological samples and different test formats including petri dish, slant tube, and dip slides. These microbiological assays are required in a range of situations where laboratory access is limited – environmental testing, point of care testing, food safety monitoring and outbreak management especially in low resources settings.

Hardware description

Overall system design

Both configurations of the MicroMI were built up from widely available off-the-shelf components, with availability from both online and local hardware suppliers

The main parameters to consider before selecting the components are: 1) Size, type and number of samples or items to incubate; and 2) Length of time required between plugging into external power supply (e.g. mains). When required, external power (12 V or AC mains) can both re-charge the internal UPS, and power the incubator to extend incubation. The ambient operating temperature will also significantly affect the battery capacity required; higher ambient temperatures will require significantly less power (and thus deplete the battery slower) to maintain 37 °C. These parameters are highlighted alongside consideration of the location and distance from field site to the local or reference laboratory, that combines with the type of

testing and location. Additionally, the laboratory space is another aspect to consider; the small scale of these incubators permits them to be set up and used in temporary testing sites or alongside other facilities (e.g., farm, health clinic). This small capacity of incubator is a good choice for laboratories with small sample numbers each day, with no need to invest in a large volume incubator for large scale testing.

The most important components are:
Uninterruptible Power Supply:

- Portable size, 12 V output, with sufficient capacity for chosen application (we evaluated products specifying 5000 and 8000 mAh at 12 V) and capable of providing sufficient current (up to 1A). The UPS battery packs we evaluated weighed <0.5 Kg. Whilst these products had a nominal 12 V output, in use they delivered between 10.6 and 12.1 V.

Incubation chamber one of two sizes:

- A flight case or briefcase with dimensions: W380 × H145 × D400 mm
- or
- Vacuum flask- consumer hot food container, we selected a mid-sized 800 ml product, but larger and smaller versions are widely available.

Heating system:

- Temperature controller STC – 1000 rated at 12 V for switching 12 V heater.
- V resistive heater element; capable of providing up to 10 W heat.
- Small fan 20 × 20 mm or 40 × 40 mm either 12 V or 5 V (widely sold for 3D printers or computer cooling)

The power output of the UPS should be carefully matched to the heater element to avoid drawing higher currents than the battery is rated for.

Large MicroMI

For the large incubator version based on an easy to carry handheld flight case or briefcase, we constructed a simple rectangular frame from widely available aluminium extrusion (20 × 20 mm) plus a glass/acrylic sheet to build the main heated chamber which was fixed permanently to the bottom of the case with double-sided adhesive tape, providing enough space for incubating multiple standard samples. We found the flight case was quite heavy (2.6 kg for case alone), so lighter boxes of similar size might be better suited to some applications where portability is more important than strength. A transparent plastic sheet was connected to the extrusion using adhesive tape hinges on top of the chamber as a lid. The case was filled with foam insulation to reduce heat loss. The chamber was big enough to accommodate petri dishes, dip slides, cell culture bottles and microwell plates. A resistive heater element was fitted on the glass floor of the chamber and a small fan used to achieve homogenous distribution of heat throughout the chamber (Fig. 2). To facilitate consistent photo imaging of samples

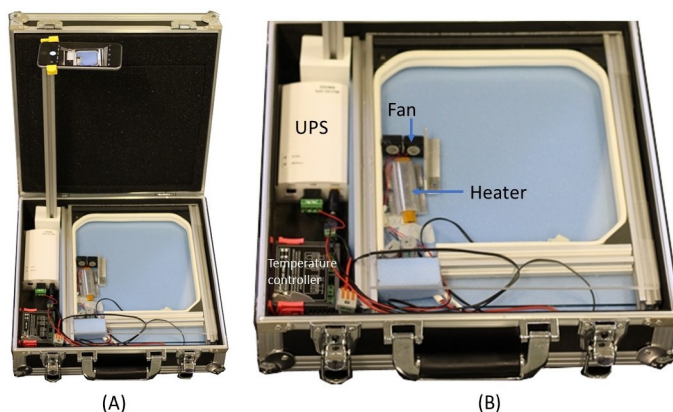


Fig. 2. Layout of large MicroMI based on flight case (A): Large MicroMI with cell phone mounted to take digital photographs and image microbiology samples during and after incubation without removing from incubator chamber. (B): Key components identified inside case including UPS power bank, temperature controller, and heated chamber layout including location of fans and resistive heating element.

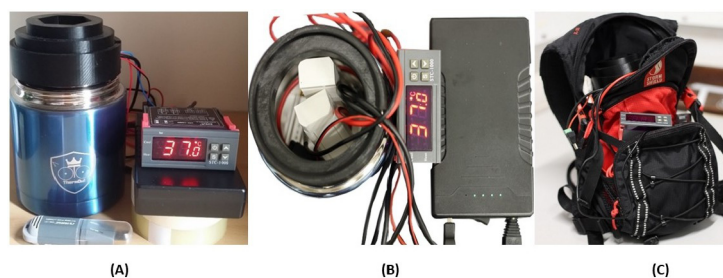


Fig. 3. Small MicroMI based on food jar stainless steel vacuum flask (A); layout of MicroMI with temperature control and UPS power pack alongside (B); samples visible inside small MicroMI with upper lid removed (C); Small MicroMI in a small backpack for carrying and operation in the field.

during or after incubation the design included a cell phone dock, this was produced by 3D printing a bar holder and clip which fixed to the corner of the case above the transparent top of the incubator. In this way, samples can be imaged consistently without removing them from the incubator or even opening the incubator.

Small MicroMI

To avoid the need of carrying our larger version of MicroMI for field testing of a smaller number of samples, and to see if a better insulated incubator chamber could reduce the battery capacity requirement, we made a smaller version of MicroMI using a small vacuum flask in the form of a consumer stainless steel food flask intended to keep a small meal warm (800 ml). We 3D-printed a double-lid designed with a permanent outer lid ring that incorporates wiring to the heater, fan, and thermocouple that were all included inside the flask (Fig. 3), and a removable inner lid to close the chamber after samples were added. The power supply and thermostatic control module were kept externally, and the components transported together in a small backpack for portability.

Design files

Most components are freely available consumer products (listed below in bill of materials; BOM), however a small number of custom bespoke designs were used. A customised lid was designed and 3D printed for the vacuum flask. The smartphone camera mount requires two 3D printed parts. We provide here CAD designs (OpenSCAD) for modification to fit different vacuum flasks and/or smartphone sizes, plus STL mesh for direct 3D printing the exact parts used in building our own MicroMI. The full set of design files is also available at <https://doi.org/10.5281/zenodo.5550785>. All the design information including all essential information for procuring and making parts, and for assembly of the MicroMI is included in the tables below.

Design part name	File type	Location (all files are also available at https://doi.org/10.5281/zenodo.5550785)
Camera extrusion holder	STL	http://dx.https://doi.org/10.17632/mgkzyj8m9t.1
Camera extrusion holder	OpenSCAD	http://dx.https://doi.org/10.17632/t4dr6nnbn7.1
Phone holder to bolt on extrusion (Adapted from open source design: https://www.thingiverse.com/thing:11435)	STL	http://dx.https://doi.org/10.17632/pb9r7d2dgh.1 ; this STL has holder sized for Xperia L1 phone
Phone holder CAD design (Adapted from open source design: https://www.thingiverse.com/thing:11435)	OpenSCAD	http://dx.https://doi.org/10.17632/k7nt6g9c64.1
Lid for vacuum flask for small MicroMI	STL	http://dx.https://doi.org/10.17632/kr47gmm3m3.1
Lid for vacuum flask for small MicroMI	OpenSCAD	http://dx.https://doi.org/10.17632/dc7m6n9mm2.1

These four components were designed in OpenSCAD (or in the case of the phone holder, adapted from a previous open source design), and STL files were sliced using Cura software and transferred to the 3D printer to print the components using standard parameters:

- 3D Printer: Creality Ender 3 or Prusa I3 MK3
- PLA (Polylactic Acid): 1.75 mm
- Printing condition: 0.2 mm layer height, infill: 20%

Bill of materials

Component	Qty/unit	Cost per unit (GBP; purchase price in May 2020)		Source of material
		Large	Small	
Uninterruptable Power Supply (UPS)				
NOTE: different capacity battery packs were compared; only 1 UPS needed per incubator				
GM322 Mini UPS 7800MAH 12 V 2A – KTC5336FBA, 15.4 × 13.6 × 4.6 cm; 340 Grams)	1	26.99	26.99	https://www.amazon.co.uk/Docooler-Protection-Charger-Portable-Applications-White/dp/B07BF4SR6S
TalentCell Rechargeable 72 W 100WH 12 V/8300mAh 12 V/9V/5V DC Output Lithium-ion Battery Pack, YB1208300-USB – 13.7 × 3.9 × 7.9 cm; 500 Grams	1	56.99	56.99	https://www.amazon.co.uk/TalentCell-Rechargeable-11000mAh-20000mAh-Portable/dp/B06Y5G3C8Z
TalentCell Rechargeable 36 W 12 V/6000mAh 5 V/12000mAh DC Output Lithium-Ion Battery Pack for LED Strip, Tape Light, CCTV Camera and More, Black, YB1206000-USB, 12.95 × 2.54 × 7.62 cm; 350 Grams	1	49.99	49.99	https://www.amazon.co.uk/TalentCell-Rechargeable-6000mAh-12000mAh-Lithium/dp/B0713T4XT9
Controller				
Aideepen STC-1000 DC 12 V-72 V LED Digital Temperature Controller Thermoregulator Thermostat with Heater And Cooler For Incubator, A7X13068,Accuracy: +/- 1° C (-50° C ~ 70° C).Temperature measuring range: -50° C ~ 99° C)	1	8.69	8.69	https://www.amazon.co.uk/Aideepen-Temperature-Controller-Thermoregulator-Thermostat/dp/B08DFPHZ3M
Heater				
Enclosure heating element 30 W, 80 °C, 12 → 24 V, 60x8.5x 35 mm Mfr. Part No.: FG14745.4	1	14.98		https://uk.rs-online.com/web/p/heating-elements/2995770/
Enclosure heating element 90 °C 12–30 V, 40x8.5x35mmMfr. Part No.:HPG-1/09-40X35-12–30	1		7.63	https://uk.rs-online.com/web/p/heating-elements/7256474/
Container				
Universal Flight Case – Medium - ACC-CASE-M, Dimensions: Inside (W × H × D): 350 × 115 × 350 mm, Outside (W × H × D): 380 × 145 × 400 mm, Weight: 2.66 kg	1	£39	–	https://cpc.farnell.com/pulse/acc-case-m/flightcase-universal-medium/dp/DP31699
Vacuum soup container Jar Lunch Box Food flask with handle 800 ml (18 × 12.2 × 11.8 cm; 650 Grams)	1	–	22	https://www.amazon.co.uk/ThermOwl-Stainless-Insulated-Leakproof-Container/dp/B07MDNJ6W4
Fan				
Axial Fan, Brushless Motor, Tubeaxial, Vapo, 5 V, DC, 20 mm, 10 mm, 1.5 cu.ft/min, 0.042 m ³ /min	1	8.66	–	https://uk.farnell.com/multicomp/mc33873/dc-fan-axial-20 mm-5vdc-0-179a/dp/2395867

(continued)

Component	Qty/unit	Cost per unit (GBP; purchase price in May 2020)		Source of material
		Large	Small	
RS PRO, 5 V dc, DC Axial Fan, 40 × 40 × 10 mm, 11.9 m ³ /h, 1.92 W	1	–	6.85	https://uk.rs-online.com/web/p/axial-fans/7897858/
Others				
V 2020 Black Aluminium Extrusion VSlot 6 Profile 20x20mm (2 × 1 m)	1	10	–	https://ooznest.co.uk/product/v-slot-linear-rail-20x20mm-cut-to-size/
50CM X 50CM X 5CM High Density Upholstery Firm Foam Rubber Sheet Cushion Replacement	1	10.25	–	https://www.amazon.co.uk/Density-Upholstery-Cushion-Replacement-Mattress/dp/B016ADSKJS
Double-side tape	<1m	Under £1	–	
90°Cast Corner	4	1.50	–	https://ooznest.co.uk/product/90-degree-cast-corner/
Silicone insulated wire 18 AWG Black and Red (approx. 1 m required)	1	6.54	6.54	https://www.amazon.co.uk/BNTECHGO-Silicone-Flexible-Strands-Stranded/dp/B01708AYYQ
WAGO Lever connectors 2-way (Part 222–412) and 3-way (Part 222–413)	1	2.70 plus 2.40 (10 per pack)	2.70 plus 2.40 (10 per pack)	https://uk.rs-online.com/web/p/standard-terminal-blocks/7581650/ https://uk.rs-online.com/web/p/standard-terminal-blocks/4751437/
PLA filament for 3D printing (white)	15 m 1.75 mm filament	under £5	–	https://ooznest.co.uk/product/pla-3d-printer-filament-1-75 mm/
		Total		
With GM322 Mini UPS		£170		
With TalentCell 12 V/8300mAh		£187		
With TalentCell 12 V/6000mAh			£125	

The total cost for each version (large vs small) of MicroMI depends on the capacity of UPS selected, as this was the most expensive single component. Two different types of UPS manufacturers were tested in the larger version of MicroMI and therefore, the total cost of building large MicroMI ranged from £170 to £185. For the smaller version of MicroMI only the 6000mAh TalentCell UPS was tested, so the total cost was £120. However, the price, availability and performance of these products varies considerably, and selection of different UPS should be made based on local availability and budget, and will be influenced by the duration the incubator will need to be used without any external power supply. As the power consumption varies depending on the ambient external temperature, careful consideration of battery capacity is required depending on the application and environment.

Build instructions

Large MicroMI

The following steps provide a step-by-step guide for building and assembling the larger version of MicroMI (Fig. 4):

1. Cut a hole (3.6 × 7.6 cm to fit dimensions of temperature controller) in the sidewall of the flight case and mount the temperature controller.
2. Cut the Aluminium Extrusion Slot Profiles into four pieces with the appropriate size to fit flight case interior.
3. Connect them together with nut and screw bolts using angle brackets.
4. Fix onto glass side and adjoin to the underneath of suitcase with double-sided adhesive tape.
5. Follow the instructions supplied with digital temperature controller product, connect positive and negative charge to 12 V power supply from the UPS, and to the heater, fan, and temperature sensor. Position these elements on the glass bottom plate to ensure fan distributes heat from the resistive heating element, and to ensure the temperature sensor is positioned near to the sample location.

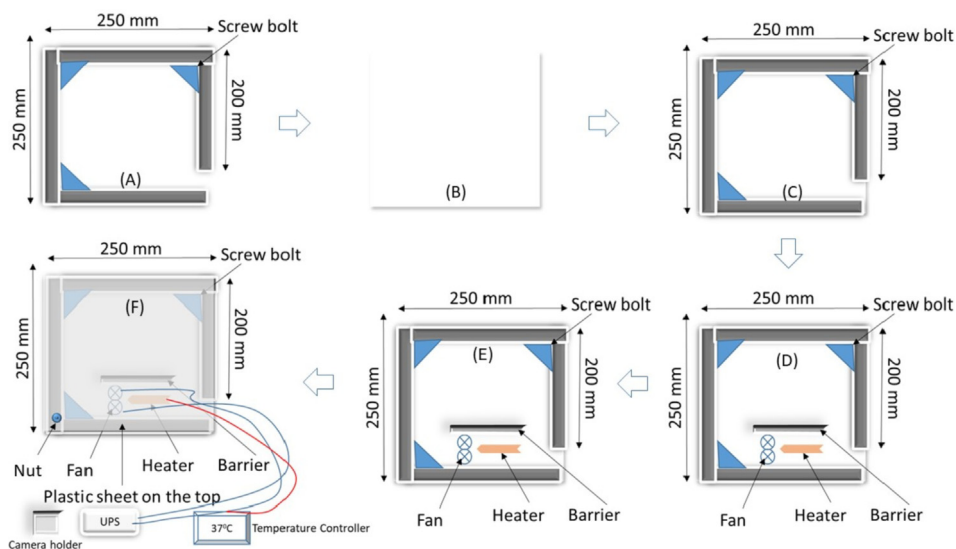


Fig. 4. Essential step to construction Heat – chamber for large MicroMI (A): Create the frame for the chamber using angle brackets bolted onto v-slot extrusion. (B): Glass plate fixed in the bottom of the flight case. (C) Fix chamber onto the glass plate using double-sided tape. (D): Fix fan and heater onto glass bottom. (E): Put the plastic sheet on the top hinged in place with tape. (F): Connect to battery and temperature controller. Then, fix UPS and camera holder into the case.

6. Put the plastic sheet ($W308 \times H145\text{mm}$) on the top of frame, as a lid and fix with two layers of adhesive tape to hinge one edge. To secure the lid when closed, adhesive magnets can be fixed to the opposite corners to the hinge attached to the extrusion frame.
7. Use insulation foam ($W308 \times H145\text{mm} \times D50\text{mm}$ or as appropriate to fit in chosen case) fixed into the bottom of the suitcase with double tape. This helps to maintain temperature inside and reduces heat loss, extending the time without external power.
8. Program the temperature controller to 37°C as instructed by the manufacturer.
9. Put the UPS inside the flight case positioned in a convenient space to allow easy connection to an external power supply when available. We cut a rectangular hole in the case so the temperature screen and controls were visible outside, but this is optional.
10. Put the camera holder in the corner of suitcase at a location suitable for holding digital smartphone camera to image the samples within the incubator. The bar holding the camera clip is removable and can be stored inside the suitcase when not in use.
11. When capturing the photo, the phone holder is fixed on the Aluminium Extrusion using T-slot nuts. The bar is placed in the holder inside the case, then place smartphone into the holder clip and take the photo. The incubator lid is transparent. After finishing, cell phone and camera holder must be removed to close the lid of the case for transportation and to maintain temperature.

Small MicroMI

The follow step will help to summarize all essential steps (Fig. 5):

1. Put the vacuum custom 3D printed lid on the top of flask.
2. Fix the heater on the bottom side edge inside the flask.
3. Fix fan at the edge of the wall towards the bottom of the flask.
4. Follow the manufacturer instructions for the temperature controller to connect power, temperature probe, and heater.
5. Leave the temperature sensor inside the flask.

Operation instructions

These large MicroMI and small MicroMI are easy to use in many microbiological applications for agar petri dish, 96 well – plate, and dip-slides. This mobile incubator can set up range temperatures from below 37°C to 42°C for microbiota detec-

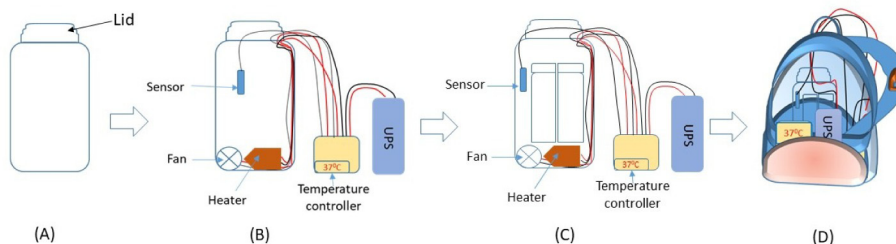


Fig. 5. Essential step to construct small MicroMI – vacuum flask (A): 3D-print a two-parts lid for the flask , (B): Add heater and fan to the bottom of flask which is connected to the temperature and UPS as introduction of temperature controller, with wiring routed through channel in lower lid. (C): Put samples such as frame dip slide, slant cultures, petri dish inside and add upper lid. (D): Small MicroMI can put inside the backpack to carry without interrupting operation.

tion, depending on the culture requirements. The device has no active cooling so cannot maintain temperatures below ambient without further modification. User instruction for operation is simple:

1. Carry this mobile incubator to the field location, temporary testing site, or anywhere required to perform the test.
2. Turn the MicroMI on at least 20–30 min before placing the samples inside the chamber so that target incubation temperature (37 °C) is achieved, based on large or small MicroMI respectively. Open the suitcase and lid of chamber to add samples.
3. As long as UPS battery is charged, incubator will operate without external power. However, when available, UPS unit should be plugged in allowing direct power by external source alongside battery charging.

This mobile device can be applied for biochemical testing of strains or used to culture and observe the bacteria present from samples collected from field sites, environmental or agricultural testing or clinical patient samples. In case of dip-slides which are simple, ready-to-use solid media for isolation and identification of many pathogens, just directly dip the dip-slides into samples and place into large or small MicroMI.

4. Appropriate safety equipment such as gloves, and good hand hygiene with detergent or antimicrobial such as alcohol is recommended prior to operation to maintain sterility and avoid contamination, and after handling samples. If handling clinical or potentially pathogenic samples, all essential microbiological safety procedures must be followed.

5. Close the large or small MicroMI and bring it back to local laboratory or reference laboratory.

6. The operating time without external power will depend on the ambient temperature outside; we evaluated these with an ambient temperature of approximately 18–20 °C (during UK winter) and 22–27 °C (during UK summer) and found temperature could be maintained from between 3 and > 24 h depending on the size and UPS capacity (Table 1). The smallest capacity UPS in the large MicroMI with under 20 °C ambient temperature only lasted ~ 3 h, in contrast the mid-sized 6000 mAh TalentCell UPS pack maintained the Small MicroMI for 24 h.

The time maintained at 37 °C under battery power depended on the capacity of the UPS, with temperature stability and duration discussed in more detail below in section 7.

To validate the MicroMI we compared the results of a wide range of example microbiological assays including biochemical identification tests, agar petri dish colony culture, and smaller samples including slant tube and dip-slides (Figs. 6–9). We cultured three lab reference strains representing major gram-negative and gram-positive pathogens, *E. coli* ATCC 25922 and *Staphylococcus aureus* ATCC 12600, plus *Klebsiella pneumoniae* ATCC 13883. In addition, 3D printed dip slides were used to

Table 1
Comparison of large and small MicroMI incubators.

	Large MicroMI	Small MicroMI
Time to reach 37 °C	35–40 mins	15–25 mins
Duration of power supply by UPS without any external power supply		
Power Bank Portable Power for 12 V (GM322 Mini UPS)	3–3.5 h with 15–20 °C ambient temperature	Not tested
TalentCell Rechargeable 36 W 12 V/6000mAh	Not tested	Over 24 h with 20–25 °C ambient temperature
TalentCell Rechargeable 72 W 100WH 12 V/ 8300mAh	8–9 h with 15–20 °C ambient temperature	Not tested
Time maintaining 37 °C after empty battery	1 h	2 h
Amplitude of temperature fluctuation	±0.5–1 °C	±0.5–1 °C
Application	Biochemical test, normal size petri dish, slant tube, frame dip slides.	Mini petri dish, slant tube, frame dip slides.



Fig. 6. Biochemical test performed in MicroMI alongside replicate in laboratory incubator room.



Fig. 7. Bacterial identification by colony growth on MacConkey's agar in small petri dishes performed using the MicroMI. The three reference strains were streaked onto three segments, and a fourth segment was negative control.

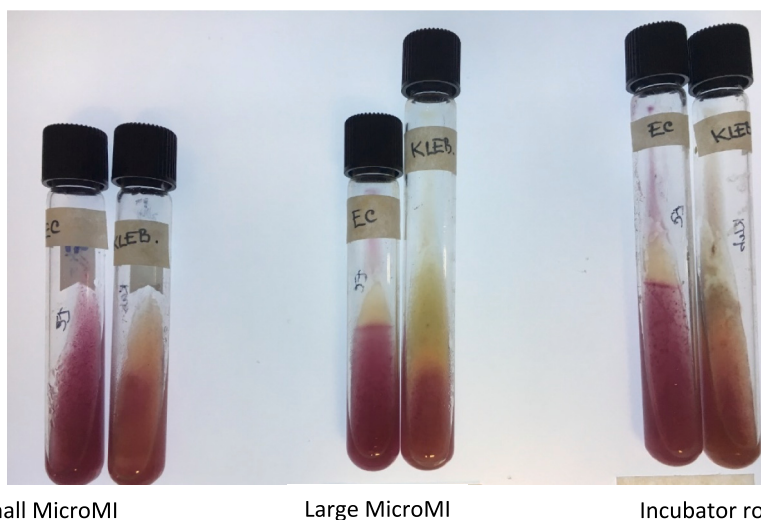


Fig. 8. Comparing bacterial growth in the MicroMI with conventional incubator- *E. coli* and *Klebsiella* on agar slant tubes of MacConkey agar.

detect bacteria in milk samples collected from cows with mastitis. All the results showed equivalent findings in terms of bacterial growth or biochemical test results observed using large and small MicroMI (both flight case and vacuum flask) to identical samples incubated in parallel in a conventional laboratory incubator room at the same culture temperature of 37 °C (Figs. 6-9) (Table 2).

The Microbact (Thermo Fisher Scientific Oxoid, Basingstoke UK) biochemical test panel for identification of different staphylococcus species was performed according to manufacturer's instruction, and with duplicate samples incubated either in the large MicroMI or a conventional microbiological incubator room (Fig. 6). This comparison did not include the small

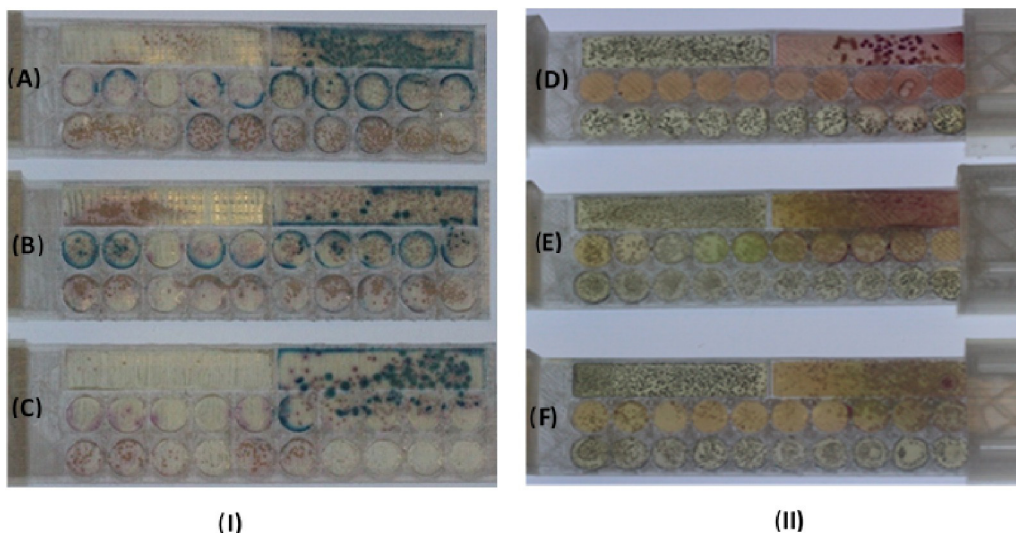


Fig. 9. Dip-slides constructed from 3D printed frames tested with mastitis milk samples (I), Frame dip slide on *E. coli* and *Staphylococcus aureus* ATCC strains (II). (A) and (D) were incubated at large MicroMI, (B) and (E) were incubated in the small MicroMI, (C) and (F) were incubated in a normal microbiological laboratory incubator.

MicroMI because these tests are liquid and the small MicroMI does not have suitable sample space for liquid microwell strips.

Biological safety requires careful containment of samples, and a range of measures are likely to be needed- as with use of any microbiological incubator and associated work practices. The large MicroMI has a securely closed lid which ensure the incubator chamber cannot open during transportation, but within the chamber careful planning is needed to ensure liquids or condensation cannot leak and microbially contaminate the inside of the incubator. We found hook-and-loop straps were ideal for securing the additional 3D printed lid onto the small MicroMI but sample containment is still required to prevent spillage that could contaminate the internal incubation chamber. The screw-topped slant tubes are enclosed after sample addition. Petri dishes require further containment such as zip-lock bags or parafilm. We 3D printed individual cases for the custom dip-slide frames (manuscript submitted). Sufficient air must be included to supply oxygen for any tests where aerobic growth is expected, which can be challenging when trying to seal samples against leakage. Biochemical tests in microwells cannot be transported as they will spill, so the MicroMI needs to be stationary during these tests.

In addition to biological safety, electrical safety should be considered. We used substantial spring connectors for rapid build and modification. We found the relatively low power drawn during heating of a peak of 1A (larger capacity battery in large MicroMI) and significantly lower peak of 0.5A (smaller TalentCell battery in small MicroMI) did not ever lead to any battery heating, even when the incubator heater chamber was heating up from ambient (i.e. heating element constantly on). As lithium batteries can potentially overheat and even catch fire if not used as directed, it remains important to use

Table 2
Comparison with the specified temperature control for commercially available microbiological incubator.

Manufacturer	Model	Temperature deviation	Preheat times (device empty, to 98% of working temperature)	Recovery times (device empty, door 30 s open, return to 98% of working temperature)	References
Manufacturers specification					
Thermo Scientific	IMC18	±1 °C at 37 °C	37 °C–15 min	37 °C–5 min	Thermo Scientific https://www.coleparmer.co.uk/
Coleparmer	H2220-HE	±1.5 °C at 37 °C	Not specified	Not specified	
Cultura ^R		±1 °C	Not specified	Not specified	https://echamicrobiology.com/
MicroMI comparison					
Our open hardware design - MicroMI		±0.5 °C at 37 °C	37 °C–30 to 45 min	37 °C–3 min	This paper- Fig. 10

these products safely and to ensure the UPS battery pack specification and any external 12v power supply used can provide the expected maximum current.

Validation and characterization

Alongside validation in use to perform microbiological tests, we performed technical performance, evaluating the stability and uniformity of temperature and the duration of operation.

We measured temperature at 5 points in the chamber (4 point at corner – 2 cm distance from the edge of chamber and 1 in the middle – 125 mm in the centre of the chamber), this temperature check was replicated at 3 different timepoints to assess range of temperature (Fig. 10). For small MicroMI, we measured two points: bottom and top – near the lid of flask, 30 mm long distance from the top. Similar temperature measurements were obtained with two different USB data loggers and the inbuilt temperature probe from the temperature controller, confirming the temperature controller was able to measure temperature accurately.

At each point, we fixed the sensor for 5 min, then measuring the temperature. The fluctuation of temperature was around $37\text{ °C} \pm 0.3\text{--}0.5$. All corner position varied around 37 °C with range of temperature of $0.01\text{--}0.03\text{ °C}$. In the centre of the chamber, the temperature did not change substantially, the temperature stayed within $\pm 0.1\text{ °C}$ of the target temperatures and there was no significant difference in temperature between the sensor and data logger. For the small MicroMI, the temperature of bottom position was always higher than the top of flask. (Fig. 10)

Both the size of incubator and the ambient temperature outside are important parameters that influence the time to reach temperature for bacterial growth. For the large MicroMI, it took nearly 35 min to get 37 °C when the temperature outside was cooler (around $12\text{--}19\text{ °C}$). The temperature only fluctuated between 0.3 and 0.5 °C around 37 °C , (Fig. 10), an acceptable variation in temperature that won't affect bacterial growth, and similar performance to commercial incubators (Table 2). The UPS maintained this temperature for 9 h in the large MicroMI before the largest capacity battery tested ran out and temperature fell, in contrast to the small MicroMI that maintained temperature for over 24 h without external power. The time depended on the battery capacity of the UPS, and the ambient temperature. For the small MicroMI, it took less time to reach 37 °C , only 16–23 min, with the fan ensuring rapid warm up and even temperature distribution (Fig. 10). We conclude the small MicroMI has superior insulation, as it stayed warm for far longer with the same capacity UPS and was faster to heat up, but it also had a smaller volume (thus fewer samples) that could also account for the lower power consumption and faster warmup. For the small MicroMI to achieve optimal incubation temperature for samples, the controller was set to 35 °C which ensured the homogenous heat inside did not exceed 37 °C , as we found with the data logger that the sample temperature was consistently 2 °C higher than the probe temperature. We conclude that minor calibration of the controller may be needed for each device.

To check the accuracy of the thermal controller temperature sensor probe, we used an additional data logger (Omega OM – EL USB – 2 – LCD). As the temperature logger recorded the same temperature as displayed on the temperature controller, we concluded the probe was calibrated accurately. We also used these USB data loggers to measure any difference in temperature at different points inside the incubator, and to see the rate of temperature drop after the UPS battery was depleted and the temperature controller switched off.

The large MicroMI lost around 5° after 1 h once the UPS battery was depleted, having stayed at target temperature for 4 h, using a smaller capacity UPS product. For this reason, we added a larger capacity UPS battery product, which lasted significantly longer. Because the larger size, larger heater element, and less effective insulation leads to far faster battery consumption, the large design is better suited to situations where there is intermittent access to external power. For example, during the day alongside travel away from external power, samples can be added to the incubator and culture experiment started. As long as external power – or a second fully charged UPS battery – is available after 4–9 h powered by battery, a full 24 h culture incubation period can be maintained. For example, the MicroMI can be plugged in overnight. Fig. 10C illustrates practical use of UPS to operate the incubator from 11:30am to 8:30 pm, at which point it was plugged in. By the following morning, mains power had maintained temperature and at the same time fully charged the battery. Larger capacity batteries are also readily available for additional cost if longer running is needed without external power.

We observed in the UK's temperate climate some differences in duration of temperature maintained by the battery between winter and summer periods. Covid-19 pandemic restrictions limited lab access during parts of this study, and limited access to temperature-controlled laboratory meant we could only test duration of battery for fixed times. Longer runs were performed without samples at home, during the summer, where ambient temperatures varied from 20 to 27 °C . We expect that in warmer ambient temperatures closer to typical culture temperature of 37 °C , such as tropical areas including Vietnam, Thailand, and Malaysia the temperature inside will be more stable, the UPS battery will last longer and the chamber will take longer time to cool down. With the current design these incubators can only be used when the ambient temperature remains at or below 37 °C , as no active cooling is included. However, the simple inexpensive thermal controllers used are also available with additional switched cooling circuit, so that active battery-powered cooling could be added alongside heating, if required. For example, fans plus a water source could be used to drive evaporative cooling. This remains an important opportunity for future innovation. Alternatively, the incubator could be operated in an air-conditioned location to avoid overheating.

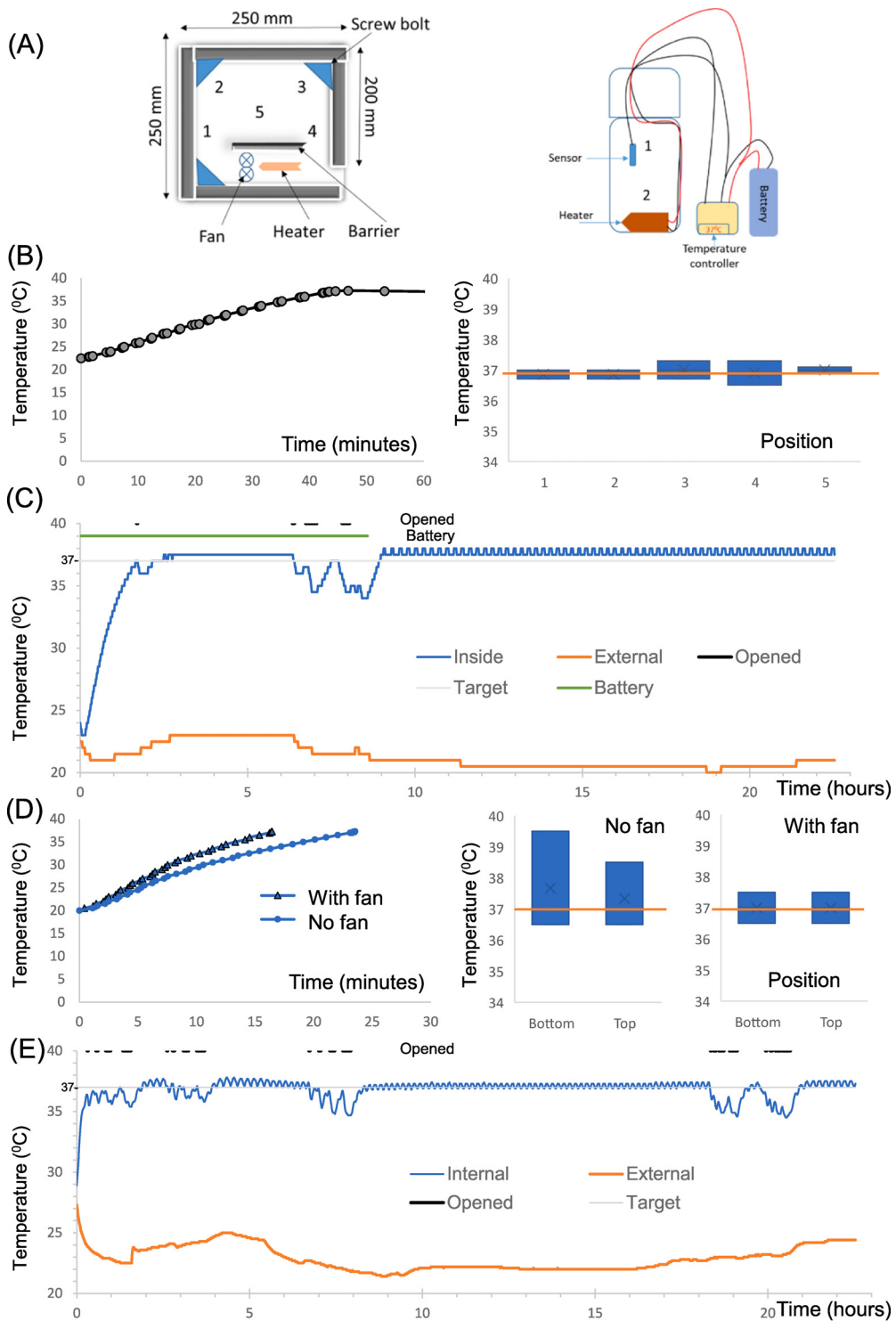


Fig. 10. The various temperature at different position. (A): Position of sensor in each MicroMI. (B): Time to reach 37 °C of large MicroMI and the homogenous temperature inside the chamber at different measure positions. (C): Datalogger temperature profile inside large MicroMI for 24 h operation, running for first 8.5 h until the UPS battery was depleted, and plugged into mains power thereafter (indicated by green line top). Black lines above indicate when incubator chamber was opened for increasing lengths of time, with respective dip and recovery in temperature. (D) Shows the increasing temperature to 37 °C from ambient, inside small MicroMI with or without fan (left), and shows how addition of fan leads to more uniform temperature than without (right). (E):Datalogger temperature profile inside small MicroMI for 24 h operation running on UPS battery alone. Black lines above indicate when incubator chamber was opened for increasing lengths of time to represent sample addition or removal, with respective dip and recovery in temperature. . (For interpretation of the references to colour in this figure legend, the reader is referred to the web version of this article.)

Declaration of Competing Interest

The authors declare that they have no known competing financial interests or personal relationships that could have appeared to influence the work reported in this paper.

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