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HumidOSH: A self-contained environmental chamber with controls for relative humidity and fan speed

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**ABSTRACT**

Relative humidity can affect physical, biological, and chemical changes in biological samples through modification of water activity and is also known to be important in the fabrication of sensitive electronic devices. The HumidOSH is a free, open source, and self-contained system for creating a controlled relative humidity environment within the range of 3 to 97% with a 0.2% tolerance. Each HumidOSH unit also comes with a fan with adjustable fan rotational speed to improve moisture uniformity inside the chamber. The system includes many additional features such as glove ports for manipulating samples, a sample door for transferring objects in and out of the system, ceiling lights for illuminating the work area inside the chamber, and two-point calibration for the relative humidity sensor. While relative humidity and fan rotational speed readings are displayed in real-time on the built-in user-friendly interface, the readings can also be recorded through a USB connection to a laptop or computer and the optional computer program. The design files, source code, and build instructions of the HumidOSH can be accessed at https://dx.doi.org/10.17605/OSF.IO/WCKHM.

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**Specifications Table**

| Hardware name | HumidOSH |
|---------------|----------|
| **Subject Area** | Biological Sciences |
| | Educational Tools and Open Source Alternatives to Existing Infrastructure |
| **Hardware type** | Biological sample handling and preparation |
| **Open Source License** | GNU General Public License (GPL) 3.0 |
| **Cost of Hardware** | $495.44 for one HumidOSH system and approximately $10 for consumables used during construction and operation |
| **Source File Repository** | https://dx.doi.org/10.17605/OSF.IO/WCKHM |

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1. Hardware in context

Relative humidity (RH) is defined as the ratio of two quantities: the vapor pressure of water present in air and the saturation vapor pressure of water in air. In other words, it is a measure of how much moisture is present in air relative to the maximum amount of moisture that can be held by the air in vapor form. RH can affect samples in various manners, either directly or indirectly. Electrostatic discharge has been shown to be affected by RH and it is thus necessary to control RH when fabricating sensitive electronic devices [1]. When most biological samples are placed in an environment with a fixed RH for a sufficient time, the vapor pressure of water in the sample will eventually equilibrate to that of the air around the sample. This equilibrium RH of the sample is defined as water activity and is expressed as a decimal quantity.

Water activity, $a_w$, is a measure of the availability of water, and thus it affects the rate of any reactions that are directly or indirectly affected by the presence of water. As such, the $a_w$ in a biological sample such as food can affect changes in its physical, biological, and chemical qualities. The glass transition temperatures of food powders are affected by $a_w$ and the maintenance of low $a_w$ is vital for ensuring flowability and non-aggregation of food powders [2–4]. Water activity is a crucial parameter in controlling the proliferation of microorganisms in foods and even affects the thermal resistance of microorganisms in low-moisture foods [5, 6]. Chemical reactions in foods such as autoxidation of lipids, degradation of anthocyanins, Maillard browning reaction, and most enzymatic activities are also affected in varying degrees by $a_w$ [7]. As such, the control of $a_w$ and hence RH, is indispensable in research on biological samples.

The control of RH has traditionally been achieved with the use of saturated binary salt solutions made of pure water and non-volatile salts such as lithium chloride or sodium chloride. These solutions will either absorb or desorb water to maintain the RH of a closed environment to be equal to the equilibrium RH or $a_w$ of the saturated salt solution [8]. Ease of preparation and maintenance makes this method attractive for simple RH control, but the fixed equilibrium RH of the solutions means that multiple salt solutions must be prepared to achieve a range of RH. The advent of small, affordable, and reliable RH sensors has paved the way to programmable electronic RH-controlled chambers. These commercially available chambers control RH through a combination of electronic RH sensors, control loops, and a variety of methods to generate or remove humidity such as steam generators and condensers. Due to the high cost of these systems, a few custom RH control systems have been constructed such as a system that dries air with silica gel beads and bubbles air through water to add moisture [9], an open source humidity controller which mixes dry nitrogen gas with water-saturated nitrogen gas [10], Agenator: an open source humidity control system for dry aging of meat [11], and Polar Bear: an open source environmental chamber which controls temperature in addition to RH [12]. The construction of such custom RH control systems can be motivated by cost and the desire to customize the systems according to research needs.

The ability to condition samples in an RH-controlled environment is invaluable to researchers from various fields. However, the high costs of commercially available equipment can make it difficult to condition large amounts of samples. Although custom alternatives exist, complete build instructions are either unavailable or the systems are missing desirable features such as easy manipulation of samples and a self-contained design. These needs, along with the many advantages of open source scientific equipment [13, 14], eventually culminated in the HumidOSH: a self-contained environmental chamber with controls for RH and fan speed. This work describes the design, construction, operation, and performance of the HumidOSH along with a case study on food samples.

2. Hardware description

The HumidOSH (Fig. 1) is a large yet portable chamber with a user-friendly interface for adjusting the inside RH and fan rotational speed to create a controlled environment for samples. The RH sensor utilizes the SHT85 digital humidity sensor (Sensirion AG, Staefa ZH, Switzerland) which is specified to have an RH accuracy of ±1.5% and hysteresis of ±0.8% [15]. The system is capable of adjusting RH to within the range of 3 to 97% and maintaining it within 0.2% of the target. The specified range of achievable RH is a conservative estimate; in actual usage, most HumidOSH units were able to exceed the limits of the range without issues. Although the RH sensor also acquires temperature readings, these data is not displayed during operation but can be acquired through the optional computer program. The system also includes a fan inside the chamber for circulating air and improving moisture transfer with the sample. The rotational speed of the fan can be adjusted between 1200 to 7500 RPM and will be maintained within 100 RPM of the target. A higher fan rotational speed results in higher average air velocity in the chamber, thus accelerating the equilibration of the sample with the surrounding air. With a chamber capacity of approximately 125 L and an air pump capable of pumping 5 L/min under atmospheric pressure, a HumidOSH unit with its air pump operating at full duty cycle undergoes approximately 2.4 air exchanges per hour. The duty cycle of the pump is controlled by pulse-width modulation and is reduced as the RH in the chamber approaches the target RH. The air pump can be replaced with a more powerful pump if a faster air exchange rate is desired. This upgrade could be done by either direct replacement with a stronger 12 V air pump, or by
replacing the 12 V air pump with a 12 V relay that switches an externally powered air pump. Other features of the system include:

- Glove sleeves with replaceable hand gloves for handling samples inside the chamber.
- Sample door for adding/removing objects to/from the chamber during operation.
- Ceiling LED lights for illuminating the work area inside the chamber.
- Visual indicators for system operation status.
- Two-point calibration for the RH sensor.
- Refillable cartridges for humidifying or dehumidifying the air.
- Power extension cord for operating electronic devices inside the chamber.
- Self-contained system: every part of the system is either housed within or connected to the chamber, allowing for easy relocation of the system.
- Autoclavable aluminum trays and stainless steel tray rack for holding samples.
- HumidOSH units can be stacked on top of each other and are also appropriately sized for placement in commercially available shelves.
- Optional USB connection to a laptop or computer for recording real-time RH, temperature, and fan rotational speed readings.

A single HumidOSH unit costs about a tenth or less of leading commercial humidity-controlled chambers. Excluding the custom printed circuit boards, the HumidOSH is purposely designed to be built from commercially available components to reduce the time and expertise needed to build the system. The system utilizes disposables (e.g. gloves, silica gel beads) that can be replaced at affordable costs. Sanitation and cleaning of the system could be performed using paper towels soaked with cleaning or disinfecting solutions, or by utilizing deeper sanitation methods such as fumigation with chlorine dioxide. The system does not have a temperature control system and is thus incapable of directly controlling the temperature of samples. The HumidOSH can be used for various applications such as:

- Adjusting the $a_w$ or moisture content of samples.
- Performing accelerated shelf life studies in a high RH environment.
- Storage of moisture-sensitive samples.
The design of HumidOSH was inspired by and improves upon a system described by Smith et al. that utilized silica gel beads and a water column for controlling water activity in food products [9].

3. Design files

Table 1 lists the files needed for constructing or operating HumidOSH units and an optional computer program that can be used during operation of HumidOSH units. The printed circuit board (PCB) design files consist of both the control box PCB and the RH sensor PCB. There are many companies that can manufacture these PCBs when given the PCB design files. The laser cutting files are used with a laser cutter to cut holes on the control box for mounting various electronics on it, though the cutting can be done manually if no laser cutter is available. The Arduino code will be uploaded to the Arduino Nano microcontroller in the control box. The optional computer program is a Windows executable file for recording live readings from HumidOSH units. The design files are stored in online repositories (linked in Table 1) that contain “Wikis” explaining the use of the files and how to download them.

4. Bill of materials

The materials required for constructing one HumidOSH system are listed in Tables 2–4. Consumables used during the construction and operation of the HumidOSH are given in Table 5. In Table 6, specialized tools that are used in the construction process are listed. Although these tools are not absolutely necessary to construct HumidOSH systems, they will make the construction process more efficient. Some items are sold in bulk but only a few quantities are needed; these are denoted by decimal quantities in the tables.

5. Build instructions

Instructions on constructing the HumidOSH can be found at https://dx.doi.org/10.17504/protocols.io.6a5shag6. The designators used in the build instructions are defined in the bills of materials (Tables 2–6).

6. Operation instructions

6.1. Basic operation

Before operating the HumidOSH, the “wet” and “dry” columns must be filled sufficiently. These columns are located on the left side of the chamber. The wet column contains hydrated water beads made of water-absorbing polymers that slowly release moisture into the air to humidify the air. The dry column contains silica gel beads which absorb moisture from the air to dehumidify it. To fill the wet column, it is necessary to hydrate the wet beads (X5 from Table 5) with water at a mass ratio of 1:200 overnight. Approximately 5 g of water beads is sufficient to fill up one wet column. Excess water should be removed before transferring the water beads into the wet column. To add the water beads into the wet column, first unscrew and remove the wet column housing from the HumidOSH unit (Fig. 2A). Then, remove the cap and plastic filter piece from the cartridge. Pour the hydrated water beads into the cartridge until it is about 4/5 full (Fig. 2B). Reinstall the plastic filter piece and cap and insert the assembled cartridge back into the column housing. Ensure that the gasket on the cap of the inner plastic column is well-seated before screwing the wet column back onto the HumidOSH unit (Fig. 2C). The procedure for filling the dry column is similar to that of the wet column but uses silica gel beads (X3 and X4 from Table 5) instead of water beads. Although both X3 and X4 will dehumidify the air, X4 is able to change color from orange to blue as the beads become saturated with water which is a useful visual indicator as to when to change the beads. However, X4 is more expensive than X3, therefore it is recommended to mix X4 with X3 at a mass ratio of approximately 1:10 to reduce costs while preserving the visual indicator feature.

Operation of the HumidOSH begins by preparing the samples that will be placed into the chamber. Distribute the samples among a maximum of six aluminum trays (T2 from Table 4) and slide them into the tray rack. Position the tray rack at the center of the chamber and plug the cables of the RH sensor and fan into the appropriate ports on the wall of the chamber (Fig. 3A). Connect the LED lights on the lid to the power cable on the wall and close the lid over the chamber (Fig. 3B).

| Design file name          | File type | Open source license | Location of the file          |
|---------------------------|-----------|---------------------|-------------------------------|
| Printed circuit board design files | Electronics | GNU General Public License (GPL) 3.0 | https://dx.doi.org/10.17605/OSF.IO/579FQ |
| Laser cutting files for control box | CAD | GNU General Public License (GPL) 3.0 | https://dx.doi.org/10.17605/OSF.IO/QG5F6 |
| Arduino code              | Software  | GNU General Public License (GPL) 3.0 | https://dx.doi.org/10.17605/OSF.IO/M8WEK |
| Computer program          | Software  | GNU General Public License (GPL) 3.0 | https://dx.doi.org/10.17605/OSF.IO/DGMQS |
Table 2
Bill of materials for components of the control box printed circuit board.

| Designator | Component | Quantity | Cost per unit (USD) | Total cost (USD) | Source of materials |
|------------|-----------|----------|---------------------|------------------|---------------------|
| PCB        | Printed circuit boards (Pack of 10) | 0.1 | $5.00 | $0.50 | Send PCB design files from Table 1 to a PCB manufacturer (e.g. https://jlcpcb.com/) |
| C1 C2 C3 C6 | Unpolarized capacitor, 0.1 µF, 0603 | 4 | 0.0352 | $0.14 | https://www.arrow.com/en/products/cl10b104kb8nnnc/samsung-electro-mechanics |
| C4 C5 C7 | Unpolarized capacitor, 10 µF, 0603 | 3 | 0.456 | $1.37 | https://www.arrow.com/en/products/grm188rb1c106ma73d/murata-manufacturing |
| D1 D2 D3 | Schottky Diode, 30 V, 1A | 3 | 0.3 | $0.90 | https://www.arrow.com/en/products/pmej3010gwx/nexperia |
| J6 | RJ-45 jack, R/A | 1 | 0.7402 | $0.74 | https://www.arrow.com/en/products/rjhse5080/amphenol |
| J12 | Female header, 15 positions, 2.54 mm pitch | 2 | 1.38 | $2.76 | https://www.digikey.com/product-detail/en?keywords=SAM1213-15-ND |
| J7 | Mini-DIN 6 Receptacle, R/A | 1 | 1.73 | $1.73 | https://www.arrow.com/en/products/md-60sm/cui-inc |
| J11 | DC barrel jack, 2.1 × 5.5 mm | 1 | 0.5276 | $0.53 | https://www.arrow.com/en/products/pj-102a/cui-inc |
| J9 J10 | Header, R/A, 2 positions, white | 2 | 0.4654 | $0.93 | https://www.arrow.com/en/products/0039301020/molex |
| J8 | Header, R/A, 2 positions, black | 1 | 0.59 | $0.59 | https://www.digikey.com/product-detail/en?keywords=50-36-2457 |
| J4 | Shrouded header, straight, 4 positions | 1 | 0.8204 | $0.82 | https://www.digikey.com/product-detail/en?te-connectivity |
| J5 | Shrouded header, straight, 9 positions | 1 | 2.26 | $2.26 | https://www.digikey.com/product-detail/en?te-connectivity-amp-connectors |
| J1 J14 | Shrouded header, 2 positions | 2 | 0.45 | $0.90 | https://www.arrow.com/en/products/292207-4-te-connectivity |
| J2 J3 | Shrouded header, 4 positions | 2 | 1.1 | $2.20 | https://www.arrow.com/en/products/pj-007/cui-inc |
| J13 | DC barrel jack, 1.35 × 3.5 mm | 1 | 0.76 | $0.76 | https://www.arrow.com/en/products/pj-007/cui-inc |
| Q1 Q2 Q3 Q4 Q5 Q6 | MOSFET, N-channel, 30 V, 3.4 A | 6 | 0.399 | $2.39 | https://www.arrow.com/en/products/jf16346trpbj/infineon-technologies-ag |
| R5 R8 | Resistor, 120 Ω, 0.1%, 0603 | 2 | 0.3515 | $0.70 | https://www.arrow.com/en/products/rt0603br07120rl/yageo |
| R4 R6 R7 R9 R10 R15 R17 R19 | Resistor, 604 Ω, 0603 | 8 | 0.0017 | $0.01 | https://www.arrow.com/en/products/rc0603fr-07604rl/yageo |
| R11 R14 R16 R18 | Resistor, 1.5 kΩ, 0603 | 4 | 0.0013 | $0.01 | https://www.arrow.com/en/products/rc0603jr-071k5l/yageo |
| R20 R21 | Resistor, 2.2 kΩ, 0603 | 2 | 0.0017 | $0.00 | https://www.arrow.com/en/products/rc0603fr-132k2l/yageo |
| R11 R14 R16 R18 | Resistor, 5.1 kΩ, 0603 | 4 | 0.0017 | $0.01 | https://www.arrow.com/en/products/rc0603fr-075k1l/yageo |
| R12 R13 | Resistor, 10 kΩ, 0603 | 2 | 0.0018 | $0.00 | https://www.arrow.com/en/products/rc0603fr-101k0l/yageo |
| R3 | Resistor, 3.3 MΩ, 0603 | 1 | 0.0017 | $0.00 | https://www.arrow.com/en/products/rc0603fr-073m3l/yageo |
| U2 | PWM Fan Controller | 1 | 0.9195 | $0.92 | https://www.arrow.com/en/products/emc2301-1-acz1r-tr/microchip-technology |
| U3 | Linear Regulator, 5 V to 3.3 V, 1 A | 1 | 0.5197 | $0.52 | https://www.arrow.com/en/products/mc7805bdg/on-semiconductors |
| U1 | I2C Buffer | 1 | 2.802 | $2.80 | https://www.arrow.com/en/products/pca9615dpjnxp-semiconductors |

Table 3
Bill of materials for components of the relative humidity sensor printed circuit board.

| Designator | Component | Quantity | Cost per unit (USD) | Total cost (USD) | Source of materials |
|------------|-----------|----------|---------------------|------------------|---------------------|
| PCB        | Printed circuit boards (Pack of 10) | 0.1 | $5.00 | $0.50 | Send PCB design files from Table 1 to a PCB manufacturer (e.g. https://jlcpcb.com/) |
| C1 C2 | Unpolarized capacitor, 0.1 µF, 0603 | 4 | 0.0352 | $0.14 | https://www.arrow.com/en/products/cl10b104kb8nnnc/samsung-electro-mechanics |
| J1 J2 | RJ-45 jack, straight | 1 | 0.7517 | $0.75 | https://www.arrow.com/en/products/rjhse5080/amphenol |
| R1 | Resistor, 1.5 kohm, 0603 | 2 | 0.0013 | $0.00 | https://www.arrow.com/en/products/rc0603jr-071k5l/yageo |
| R2 R3 | Resistor, 2.2 kohm, 0603 | 2 | 0.0017 | $0.00 | https://www.arrow.com/en/products/rc0603fr-132k2l/yageo |
| R4 R5 | Resistor, 3.3 MΩ, 0603 | 2 | 0.3515 | $0.70 | https://www.arrow.com/en/products/rc0603fr-073m3l/yageo |
| R11 R14 R16 R18 | Resistor, 10 kΩ, 0603 | 2 | 0.0018 | $0.00 | https://www.arrow.com/en/products/rc0603fr-101k0l/yageo |
| R3 | Resistor, 1.5 kΩ, 0603 | 1 | 0.0017 | $0.00 | https://www.arrow.com/en/products/rc0603fr-075k1l/yageo |
| U1 | I2C Buffer | 1 | 2.802 | $2.80 | https://www.arrow.com/en/products/pca9615dpjnxp-semiconductors |
| U2 | PWM Fan Controller | 1 | 0.9195 | $0.92 | https://www.arrow.com/en/products/emc2301-1-acz1r-tr/microchip-technology |
| U3 | Linear Regulator, 5 V to 3.3 V, 1 A | 1 | 0.5197 | $0.52 | https://www.arrow.com/en/products/mc7805bdg/on-semiconductors |
| U1 | I2C Buffer | 1 | 2.802 | $2.80 | https://www.arrow.com/en/products/pca9615dpjnxp-semiconductors |

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| Designator | Component | Quantity | Cost per unit (USD) | Total cost (USD) | Source of materials |
|------------|-----------|----------|---------------------|------------------|-------------------|
| L1         | 12 V LED Strip Light, SMD 2835, 16.4 ft | 0.13 | $7.99 | $1.06 | https://www.amazon.com/gp/product/B08HFS65MC/ |
| L2         | LED Strip to DC Female Plug Connector (Pack of 10) | 0.1 | $9.99 | $1.00 | https://www.amazon.com/gp/product/B01D7M7F800/ |
| L1         | Clear epoxy resin mix, 1 Gal kit | 0.2 | $62.97 | $12.59 | https://www.amazon.com/gp/product/B01LYK2NAG/ |
| L2         | Silicone seal with adhesive backing, red, 10 ft | 0.8 | $20.50 | $16.40 | https://www.mcmaster.com/1129a994-1129a954 |
| L3         | Draw-latch, 2-5/16" Long × 15/16" Wide (pack of 10) | 1 | $10.57 | $10.57 | https://www.mcmaster.com/1390a13 |
| L4         | Truss Screws, #5–40, 1/4" long (pack of 100) | 0.2 | $6.12 | $1.22 | https://www.mcmaster.com/91770A124 |
| L5         | Locknuts, #5–40 (pack of 100) | 0.2 | $5.54 | $1.11 | https://www.mcmaster.com/90633A006 |
| L6         | Cable ties, 0.09" width, 0.04" thick, 3" long (pack of 100) | 0.01 | $5.52 | $0.06 | https://www.mcmaster.com/7130K101 |
| L7         | Cable tie mount (pack of 50) | 0.02 | $10.01 | $0.20 | https://www.mcmaster.com/7566k62 |
| C1         | Storage Box, 132 qt | 1 | $38.99 | $38.99 | https://www.irisusainc.com/clear-box-with-buckles-132-qt-cb-130-clear |
| C2         | Duct Flange, Galvanized Steel, Size 5 | 2 | $9.22 | $18.44 | https://www.mcmaster.com/1758K24 |
| C3         | Truss Screws, 1/4”–20, 5/8” long (pack of 100) | 0.14 | $8.17 | $1.14 | https://www.mcmaster.com/90271A539 |
| C4         | Flat washers, 1/4” (pack of 100) | 0.14 | $3.37 | $0.47 | https://www.mcmaster.com/92141A029 |
| C5         | Locknuts, 1/4”–20 (pack of 100) | 0.14 | $4.39 | $0.61 | https://www.mcmaster.com/95615A120 |
| C6         | Deck Plate Kit, 6” | 1 | $15.98 | $15.98 | https://www.mcmaster.com/gp/product/B011J5J60/ |
| C7, A4     | Truss Screws, #10–24, 1/2” long (pack of 100) | 0.1 | $5.22 | $0.52 | https://www.mcmaster.com/90272A242 |
| C8, A3     | Flat Washers, #10 (pack of 100) | 0.2 | $2.33 | $0.47 | https://www.mcmaster.com/92141A011 |
| C9, A7     | Locknuts, #10–24 (pack of 100) | 0.1 | $3.31 | $0.33 | https://www.mcmaster.com/90631A011 |
| C10        | Unthreaded spacers, 3/8” OD, 7/8” Long, for Number 6 Screw Size (pack of 100) | 0.2 | $12.59 | $2.52 | https://www.mcmaster.com/94639A410 |
| C11        | Locknut, #6–32 (pack of 100) | 0.2 | $2.91 | $0.58 | https://www.mcmaster.com/90633A007 |
| C12        | Truss Screws, #6–32, 1–1/4” long (pack of 100) | 0.2 | $7.56 | $1.51 | https://www.mcmaster.com/91770A155 |
| C13, T,F5  | Flat Washers, #6 (pack of 100) | 0.22 | $1.17 | $0.26 | https://www.mcmaster.com/92141A008 |
| C14        | Cable Gland - Waterproof Rj-45 | 1 | $9.94 | $9.94 | https://www.arrow.com/en/products/927/adatafruit-industries |
| C15        | RJ 45 cable, 1 ft long (pack of 10) | 0.1 | $15.99 | $1.60 | https://www.amazon.com/gp/product/B00K2E4X2U/ |
| C16        | Mini DIN-6 bulkhead connector, female-female | 1 | $5.23 | $5.23 | https://www.mccmarter.com/Cables/Calrad_35_498_BH_6_Bulk_Head_Chrome_plated_6_Pin_p/wal22-35-498-bh-6.htm |
| C17        | Mini DIN-6 cable, male-male, 2 m | 1 | $3.23 | $3.23 | https://www.arrow.com/en/products/ak678-2/assmann-wsw-components-inc |
| C18        | Hose Barb Thru-Panel Elbow Adapter, 1/4” × 1/4”, natural nylon | 3 | $2.45 | $7.35 | https://www.kempcospec.com/ProductDetails.asp?ProductCode=KITPLA4-4-4NN |
| C19        | Submersible Cord Grip, 0.18”–0.39” Cord OD, M16 Knockout Size | 1 | $4.03 | $4.03 | https://www.mcmaster.com/7310K32 |
| C20        | Extension cord, 10 ft, 0.38” OD | 1 | $16.94 | $16.94 | https://www.mcmaster.com/7576K24 |
| C21        | Plug, NEMA 5–15 | 1 | $8.80 | $8.80 | https://www.mcmaster.com/7216K51 |
| C22        | Submersible Cord Grip, 0.14”–0.32” Cord OD, PG-9 Knockout Size | 1 | $3.33 | $3.33 | https://www.mcmaster.com/7310K12 |
| C1L        | DC Power Pigtail Cable, 2.1 mm × 5.5 mm Barrel Plug, 50 cm long (pack of 10) | 0.1 | $7.99 | $0.80 | https://www.amazon.com/gp/product/B01CPLM8VG/ |
| C1L2       | Power Barrel Connector Plug 1.35 mm ID, 3.50 mm OD | 1 | $1.14 | $1.14 | https://www.arrow.com/en/products/pp3-002d/cusinc |
| A1         | Triple Bracket for Standard 10” Canisters (U Style) | 1 | $9.99 | $9.99 | https://www.bulkreefsupply.com/triple-bracket-for-standard-10-ro-canisters-u-style-bulk-ref-supply.html |
| A2         | Truss Screws, #10–24, 3/4” long (pack of 100) | 0.1 | $4.44 | $0.44 | https://www.amazon.com/gp/product/B0071A245 |
| A5         | 12 VDC vacuum diaphragm pump | 1 | $10.24 | $10.24 | https://www.ebay.com/itm/DC12V-65-120kpa-5L-min-Micro-Vacuum-Pump-Negative-Pressure-Suction-Pump-Holder/322354285216 |
| A6         | Rubber Washers, #10 (pack of 100) | 0.06 | $10.33 | $0.62 | https://www.mcmaster.com/1556a54 |
| A8         | Zinc-Plated Steel Corner Bracket, 2” × 2” × 5/8” | 2 | $0.92 | $1.84 | https://www.mcmaster.com/|
| A9         | Screws, #8–32, 3/8” long (pack of 100) | 0.06 | $3.44 | $0.21 | https://www.mcmaster.com/90275A192 |
| A10        | Flat Washers, #8 (pack of 100) | 0.06 | $2.00 | $0.12 | https://www.mcmaster.com/92141A009 |
| A11        | Locknuts, #8–32 (pack of 100) | 0.06 | $3.16 | $0.19 | https://www.mcmaster.com/90631A009 |
| A12        | Power Supply, 12 V DC 2A, 5.5 mm – 2.1 mm (Pack of 5) | 0.2 | $34.99 | $7.00 | https://www.amazon.com/dp/B07HN2R3K/ |
| A13        | Tube Clamps, 1/4” to 5/16” ID (pack of 20) | 0.1 | $9.68 | $0.97 | https://www.mcmaster.com/9579K62 |
| ALC1       | 10” Reverse Osmosis Canister 1/4” Ports | 2 | $16.99 | $33.98 | https://www.bulkreefsupply.com/reverse-osmosis-canisters.html |
| Designator | Component | Quantity | Cost per unit (USD) | Total cost (USD) | Source of materials |
|------------|-----------|----------|---------------------|------------------|---------------------|
| AC2        | 10" BRS Reactor Refillable Cartridge - Hard Shell | 2 | $9.99 | $19.98 | [Link](https://www.bulkreefsupply.com/10-brs-reactor-refillable-cartridge-hard-shell.html) |
| AC3, A.S2  | Elbow Adapter, 1/4" Tube ID x 1/4" NPT | 0.8 | $9.58 | $7.66 | [Link](https://www.mcmaster.com/5463k489) |
| AS1        | Solenoid Valve, 12 VDC, 1/4" NPT, N/C | 2 | $11.99 | $23.98 | [Link](https://www.ebay.com/itm/1-4-NPT-12V-DC-Electric-Solenoid-Valve-12-Volt-DC-NC-RO-Air-Water-BBF/290723310425?hash=item43b075ab59:g:bpcAAOSw5xV2gfdz) |
| AS3, A.P3  | Plug contacts, 22–28 AWG, crimp | 6 | $0.08 | $0.48 | [Link](https://www.mcmaster.com/5233k56) |
| AS4        | Plug, 2 positions | 2 | $0.31 | $0.62 | [Link](https://www.amazon.com/B01CSWPJRG/) |
| AP1        | 2 Conductor Wire, 50' Long | 0.01 | $9.95 | $0.10 | [Link](https://www.mcmaster.com/7060k15) |
| AP2        | Insulated Quick-Disconnect Terminals, Single Crimp Female, 22–18 Gauge, 0.187" Wide x 0.02" Thick Tab (Pack of 100) | 0.02 | $15.62 | $0.31 | [Link](https://www.mcmaster.com/94812a300) |
| AP4        | Plug, 2 positions, black | 1 | $0.33 | $0.33 | [Link](https://www.mcmaster.com/90272A081) |
| AT1        | PVC Clear Tubing, 1/4" ID, 3/8" OD (sold in ft) | 4 | $0.28 | $1.12 | [Link](https://www.mcmaster.com/5463k723) |
| AT2        | Wye, 1/4" Tube ID | 0.1 | $17.50 | $1.75 | [Link](https://www.mcmaster.com/5463k723) |
| AT3        | Check Valve, 1/4" ID, Polypropylene | 2 | $0.75 | $1.50 | [Link](https://www.usplastic.com/catalog/item.aspx?itemid=32233) |
| AT4        | HEPA Air Filter (1/4" In-line) | 2 | $3.99 | $7.98 | [Link](https://www.grainger.com/product/SHOWA-Chemical-Resistant-Glove-4JF22) |
| AB1        | Plastic case, 8.5" x 5.1" x 2" (Pack of 10) | 0.1 | $16.99 | $1.70 | [Link](https://www.amazon.com/WYPH-ATmega328P-Microcontroller-Development-Pre-soldered/dp/B07KC9C6H5/) |
| AB2        | Threaded Hex Standoff, Nylon, #6–32, 1/4" Hex Size, 1/4" Long | 8 | $0.28 | $2.24 | [Link](https://www.digikey.com/products/en?keywords=LCD-14074) |
| AB3        | Nylon Hex Nut, #6–32 (Pack of 100) | 0.04 | $6.37 | $0.25 | [Link](https://www.digikey.com/products/en?keywords=14662-sparkfun-electronics) |
| AB4        | Arduino Nano V3.0 with USB cable | 0.33 | $12.35 | $4.12 | [Link](https://www.digikey.com/products/en?keywords=G1) |
| AB5        | Push Button, Black, N/O, SPST, Momentary Contact, Panel Mount | 1 | $1.07 | $1.07 | [Link](https://www.mcmaster.com/7060k15) |
| AB6        | Switch, Rocker, SPST, 10A, 125 V | 1 | $1.02 | $1.02 | [Link](https://www.mcmaster.com/7060k15) |
| AB7        | Cable assembly, 2 positions | 2 | $0.53 | $1.05 | [Link](https://www.mcmaster.com/7060k15) |
| AB8        | Push Button, Green, Illuminated, N/O, SPST, Momentary Contact, Panel Mount | 1 | $1.72 | $1.72 | [Link](https://www.mcmaster.com/7060k15) |
| AB9        | Push Button, Blue, Illuminated, N/O, SPST, Momentary Contact, Panel Mount | 1 | $1.95 | $1.95 | [Link](https://www.mcmaster.com/7060k15) |
| AB10       | Cable assembly, 4 positions | 2 | $1.10 | $2.20 | [Link](https://www.mcmaster.com/7060k15) |
| AB11       | 20 x 4 LCD, Black on RGB, 3.3 V | 1 | $25.00 | $25.00 | [Link](https://www.mcmaster.com/7060k15) |
| AB12       | Shrouded header, straight, 4 positions | 1 | $0.82 | $0.82 | [Link](https://www.mcmaster.com/7060k15) |
| AB13       | Flat Flex Cable Assembly, 4 Position, 8.00" Long | 1 | $4.34 | $4.34 | [Link](https://www.mcmaster.com/7060k15) |
| AB14       | Keypad, 12 Button | 1 | $3.95 | $3.95 | [Link](https://www.mcmaster.com/7060k15) |
| AB15       | Shrouded header, straight, 9 positions | 1 | $1.92 | $1.92 | [Link](https://www.mcmaster.com/7060k15) |
| AB16       | Flat Flex Cable Assembly, 9 Position, 8.00" Long | 1 | $5.39 | $5.39 | [Link](https://www.mcmaster.com/7060k15) |
| AB17       | Screws, #2–56, 1/4" long (pack of 100) | 0.08 | $4.75 | $0.38 | [Link](https://www.mcmaster.com/7060k15) |
| AB18       | Flat Washers, #2 (pack of 100) | 0.16 | $1.40 | $0.22 | [Link](https://www.mcmaster.com/7060k15) |
| AB19       | Unthreaded spacers, 3/16" OD, 3/16" Long, for Number 2 Screw Size (pack of 100) | 0.04 | $9.24 | $0.37 | [Link](https://www.mcmaster.com/7060k15) |
| AB20       | Locknuts, #2–56 (pack of 100) | 0.08 | $3.51 | $0.28 | [Link](https://www.mcmaster.com/7060k15) |
| G1         | Nitrile gloves, large | 1 | $9.95 | $9.95 | [Link](https://www.mcmaster.com/7060k15) |

(continued on next page)
Fasten the latches along the walls of the chamber to the strike plates on the lid. If desired, add a small amount of talcum powder (X1 from Table 5) to the inside of the gloves to lubricate the insides.

With the samples in the chamber and the lid secured, all that is left is to set the target RH and fan rotational speed. First, ensure that the power supply adapter for the control box is plugged into an electrical outlet. Once the control box has performed its boot up sequence, it will display the readings screen (Fig. 4(A)) which shows the readings for RH and fan rotational speed, and the status of the control system for both. At this point, the statuses should be "IDLE," indicating that...
the control systems are not running. RH readings are obtained and displayed every second while the system is powered but the fan rotational speed readings will only appear if the fan control system is activated. Otherwise, “N/A” will be displayed for the fan rotational speed reading. Pressing the black button once will change the screen to the adjustment of target RH screen (Fig. 4(B)). Here, the current target for RH is displayed. At the bottom of the screen, the user is prompted for the new target RH. To set the new target, simply key in the desired target with the keypad, keeping in mind that the given value should have only one decimal place and be in the range of 0.0 to 100.0%, inclusive. If it is not desired to change the target RH, do not key in any value or clear any entered values using the backspace key. Press the black button to save the new target or, if the new target was left blank, keep the old target and move to the next screen. The next screen is for setting the target fan rotational speed (Fig. 4(C)) and is mostly similar to the one for RH. When keying in the new target, it should be an integer (i.e. no decimals) and be between 1200 to 7500 RPM, inclusive. Press the black button to change the screen to the RH sensor calibration screen (Fig. 4(D)) which is not required for typical operation and will be described in Section 6.3. Pressing the black button will return the screen back to the readings screen. At this point, pressing the green button will start the control system for RH, which is indicated by the green button lighting up and flashing arrows beside the RH reading on the screen (Fig. 4(F)). Pressing the blue button will initiate a similar sequence of events for the fan control system. At anytime during operation, the targets for RH and fan rotational speed can be changed without stopping the control systems by scrolling to the appropriate screens with the black button and keying in new targets. The control systems will automatically adjust to the new targets once the new targets have been saved. To stop any of the control systems, simply press the appropriate button.
(green or blue) and hold for four seconds. A message will be displayed on the screen to show the remaining time to hold the button before the control system is turned off.

At any time during operation of the HumidOSH, samples in the chamber can be manipulated with the gloves at the front of the chamber. Before manipulating samples, it is recommended to turn on the LED lights in the chamber by flipping the switch at the front of the control box (Fig. 4(A)). In addition, the sample door on the left side of the chamber can be opened to transfer objects/samples in and out of the chamber. Manipulation of samples with the gloves changes the pressure inside the chamber which causes external air to seep in through tiny leaks. The same happens when the sample door is opened. Experience with using the HumidOSH demonstrated that these activities can cause a temporary change in RH as much as 4% depending on the difference in RH between the inside and outside of the chamber. If electrical devices such as heat sealers, weighing scales, or vortexes are to be used inside the chamber, they can be plugged into the extension cord attached to the left wall of the chamber.

6.2. Maintenance

During long-term operation of the HumidOSH, a few maintenance activities are recommended, as described below. The interior of the chamber should be cleaned periodically to remove spilled samples and prevent contamination of future samples. Before any cleaning is done, it is extremely important to remove the RH sensor and place it away from the chamber to prevent contaminating the sensor with the cleaning chemicals. In addition, power to the control box and extension cord should be disconnected. The tray rack should be removed from the chamber to be cleaned separately. The insides of the
chamber can then be sprayed with a disinfectant such as 70% ethanol or an appropriate cleaning solution and then wiped down with paper towels. The tray rack and aluminum trays can be autoclaved if necessary or cleaned with the same cleaning solution. The fan on the tray rack should be removed before autoclaving is performed. The hand gloves attached to the glove sleeves can either be cleaned or replaced with a new pair. Once everything has been cleaned, place everything except the RH sensor back into the chamber, leave the lid open, and turn on the fan in the chamber to evaporate residual moisture in the chamber. Verify that the inside surfaces of the chamber are dry before reinstalling the RH sensor.

Over time, the contents of the wet and dry columns of the system will need to be replaced, especially if it is desirable to adjust the RH to extreme values. The wet beads will shrink in size after prolonged usage and can either be soaked in water to rehydrate them or be replaced with a new batch of wet beads. If the silica gel beads in the dry column have mostly turned orange to blue in color, they can either be regenerated by heating at 120 °C for about 2 h or be replaced with new silica gel beads.

The RH sensor is sensitive to contamination and will show some inaccuracies over long periods of usage. Although this can be addressed by Section 6.3, it is also possible to simply replace the sensor by following step 79 of the build instructions in Section 5.

6.3. Calibration of relative humidity sensor

Over time, the readings of the RH sensor may drift to inaccurate values. This drifting can be compensated with the two-point calibration included with HumidOSH. This calibration is a “soft” calibration; it merely applies scaling and offset to readings from the sensor. In addition, the calibration parameters are stored within the control box of HumidOSH and not the sensor, so the calibration values are not carried over when transferring the sensor to another HumidOSH system. To access the calibration protocol, press the black button on the control box until the calibration screen is shown (Fig. 4(D)). Here, the user can calibrate one of the two points or clear the saved calibrations. Based on the instructions on the screen, press either key “1” or “2” on the keypad to begin calibrating one of the points (Fig. 4(E)). Place the RH sensor in an airtight container that contains a reference standard for RH calibration. For example, saturated salt solutions with known equilibrium RH such as high-purity sodium chloride and lithium chloride can be used [8]. Preparation of these solutions involve dissolving as much of the high-purity salt as possible in hot pure water until no more salt can be dissolved, then letting the solution cool down. If an airtight container is not available, simply pour some saturated salt solution into a beaker, place the RH sensor in the container without touching the solution, and then seal the opening of the beaker to the RH sensor cable with Parafilm or a flexible plastic film. It is extremely important that the RH sensor does not come into direct contact with the salt solution as that may damage the sensor. Allow the air in the container to equilibrate with the salt solution for at least 10 min. The raw RH reading displayed on the screen (Fig. 4(E)) should also stabilize during this time. Once the raw RH reading is stable, key in the reference RH reading i.e. the known equilibrium RH of the saturated salt solution. Press the black button to save this calibration point. Repeat the calibration procedure for the second point with another saturated salt solution and the calibration procedure is complete. If a new RH sensor is installed into the HumidOSH system and the saved calibrations are no longer needed, erase the saved calibrations by scrolling to the calibration screen (Fig. 4(D)) and then press key “3” followed by “5,” as shown by the instructions on the screen.

6.4. Computer program

An optional computer program is available for recording readings from HumidOSH systems. In order to use the program, the microcontroller inside the control box must be connected to a computer or laptop with a USB cable. Multiple instances of the program can be opened to acquire readings from multiple HumidOSH systems. In this section, the colored labels in Fig. 5 will be used to refer to the various sections of the program.

11 is a dropdown list of all the open communication ports of the computer or laptop. The communication ports can be used by various devices such as USB devices. The port that is connected to the HumidOSH system needs to be selected here. Some guesswork may be required here, so select a port from the list and press 6 to attempt communication. If an error appears, select the next port and repeat. Sometimes, the first communication attempt to the HumidOSH system may fail, so it may be necessary to try twice. Once the communication attempt is successful, 1 displays the RH reading, 2 the temperature, 3 the fan rotational speed, 4 the target RH of the control system, and 5 the target fan rotational speed of the control system. Pressing 6 again while the readings are being acquired will end communications with the HumidOSH system. To record the readings, press 2 to open a dialog box for choosing the location and file name for storing the readings; the path to the file will appear in 1 when the selection is confirmed. Press 3 to begin recording the data or to stop recording. All recorded data are stored in Comma Separated Value (CSV) files which can be opened with spreadsheet software such as Microsoft Excel or text editors.

7. Validation and characterization

To test the ability of the HumidOSH to maintain a stable RH, samples of whole milk powder (28.5% milkfat, Land O’Lakes, Inc., St. Paul, MN) were placed in HumidOSH units set to target RH of either 5% or 80% and 5000 RPM for target fan rotational
speed. For the 80% target RH, the whole milk powder samples were used as is, with a native \( a_w \) of 0.2030 ± 0.0033 \( a_w \). As for the 5% target RH, the \( a_w \) of the samples were first adjusted to a higher \( a_w \) through the addition of deionized water and then mixed using a kitchen mixer (KSM8990OB, KitchenAid, Benton Harbor, MI) with a wire whip attachment (W10361360, KitchenAid, Benton Harbor, MI) for 15 min at speed 4. The hydrated milk powder was then left in sealed plastic bags at room temperature overnight to allow the mixture to equilibrate to 0.4291 ± 0.0044 \( a_w \). When inserting the whole milk powder samples into the HumidOSH units, 500 g of whole milk powder were distributed across two aluminum trays per HumidOSH system and placed at the third and sixth positions from the top of the tray rack. Five HumidOSH systems were used for each target RH. The computer program described in Section 6.4 was used to record readings from all the HumidOSH systems. Every day, two random samples were taken out from each HumidOSH system and measured for \( a_w \) with a water activity meter (4TE, METER Group, Pullman, WA). The validation study was performed continuously for 6 days.

The RH readings and \( a_w \) measurements of the validation study are shown in Fig. 6. When the HumidOSH units were set to 80% target RH, the RH rose rapidly from approximately 25% to 50% within the first few hours of operation and then slowly rose to 80% over the next two days. This behavior in RH increase is likely due to a large difference in vapor pressure between the air and the water beads in the wet column of the HumidOSH units at the beginning of the study which subsequently decreased as the RH of the air increased. The reverse of this trend was observed when drying the air to a target RH of 5%, though the initial decrease was not as rapid. In addition, there is a noticeable spike in RH readings for all the HumidOSH units that occurred every day around the same time; these RH disturbances were caused by extraction of samples from the HumidOSH units for \( a_w \) measurements. It should be noted that the time of RH disturbances and \( a_w \) readings do not coincide exactly in Fig. 6 because the RH readings are plotted in real-time format while the \( a_w \) measurements are plotted in daily format. Upon achieving the target RH, all the HumidOSH units were able to maintain the RH within a tight tolerance (within 0.2% of the target RH), as evident by the small standard deviations.

In general, the \( a_w \) of the whole milk powder samples lagged behind the RH during the first few days because of the time needed for vapor pressure equilibration between the sample and the air inside the HumidOSH units. The \( a_w \) readings stabilized after the third day and remained relatively constant for the remainder of the study. However, some of the stabilized \( a_w \) readings, especially when the target RH was 80%, deviated from the target RH. This deviation is likely due to inaccuracies of the RH sensor in some of the HumidOSH units which led to inaccurate control of the RH and subsequently inaccurate \( a_w \) in the samples after equilibration. The largest deviation was 0.06 \( a_w \), or, equivalently, 6% RH which is larger than the 1.5% accuracy tolerance given by the manufacturer of the RH sensors used in HumidOSH. The deterioration in accuracy of the sensors can be explained by prolonged use of the HumidOSH units; all the HumidOSH units used in the validation studies had been
used to condition various food samples for almost a year before the validation studies were performed. Therefore, it is recommended to either calibrate the sensors every few months (Section 6.3), replace the sensors periodically (Section 6.2), or simply apply an offset to the target RH to account for the sensor drift.

In this characterization study, it took approximately three days before the HumidOSH was able to achieve the target RH. While this is acceptable for certain applications such as inoculating low-moisture food samples, the long equilibration time could pose a problem for time-sensitive studies. There are several ways to overcome this issue such as reducing the amount of sample or upgrading the air pump. The former is the simplest method; by decreasing the amount of sample, the system has a smaller moisture load to deal with. On the other hand, upgrading the air pump will require finding a suitable drop-in replacement or a proper relay and externally powered pump (as described in Section 2). The importance of equilibration time and the choice of a suitable solution is dependent on the user’s application of HumidOSH and should be evaluated on a case-by-case basis.

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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Appendix A. Supplementary data

Supplementary data to this article can be found online at http://doi.org/10.1016/j.johx.2020.e00141.

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