Designing a chamber for studies involving manipulation of light:dark cycles

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The authors designed and built a device that can house mice or rats and allow researchers to control the light:dark cycles inside. They developed this chamber for neuroscientists who are studying the condition-dependent plasticity of the mouse visual cortex. The chamber, which (when closed) completely blocks outside light, consists of two units. Each unit can hold eight small mouse cages or six rat cages. Each unit contains an optical sensor that triggers an audible and visual alarm when light is detected. Researchers can monitor the environmental conditions inside each unit using a control panel located outside the unit. Researchers have reported that this chamber is ideal for use in their work involving manipulations of light:dark cycles.

Neuroscientists at Children's Hospital Boston, who are carrying out research on condition-dependent plasticity of the mouse visual cortex, were planning to study how synaptic function in the brain is influenced by visual experience. They asked for help controlling the length of light:dark cycles for C57/B6 mice and Black Swiss mice (Charles River Laboratories, Wilmington, MA). The research team wanted to test the effects of modifying the visual experiences of adult mice, pregnant female mice and female mice with litters of various ages.

Because the researchers wanted to study only a small number of animals in a facility where cage density is high and space is limited, altering the light:dark cycle of an entire animal room was not feasible. Their study required that animals be kept in dark conditions for the entire length of the study, which lasted between 2 d and 32 d. We determined that the best solution would be to design a self-contained chamber (Fig. 1) that could be placed in an existing animal room. Our challenge was to design and build a portable chamber that was 'lighttight' and that allowed for internal temperature and humidity to be monitored. The chamber also needed to be easy to sanitize and needed to accommodate multiple groups of animals with distinct light cycle requirements. This project was a collaboration between staff members in animal resources management at a research facility and at a plastic manufacturing firm; we worked together to design and build the portable chamber.

We designed a two-unit, wheeled chamber made of black polypropylene. The chamber is 38 in wide, 25 in long and 73.75 in high (**Fig. 2**). These dimensions allow the full chamber to fit easily through standard facility doors. We used polypropylene because it can be easily cleaned using liquid sanitizer. Each unit in the chamber can accommodate eight small mouse cages or six rat cages. Mice and rats are not housed together in the chamber at the same time.

To ensure that there would be complete darkness inside each enclosed unit, we designed doors with an overlapping gasket seal. We planned to install an optical sensor in each unit that could verify light-tight conditions and could alert users if light was detected, but we could not find a light sensor that was capable of detecting extremely low light levels. Therefore, engineers at Plastic Concepts, Inc. (North Billerica, MA) designed an upgraded version of an existing sensor that could detect low light levels (4 lumens). They used an existing lowlumens light detection device (Plastic Concepts, Inc.) to confirm that the upgraded sensors had enhanced light-detection capability.

To determine where to install the light sensor inside each unit, we carried out extensive testing using a light fixture wired to a dimmer switch. We varied the position of the light source and the intensity of the light inside and outside the chamber until we found the location where the presence of any interior light could be

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FIGURE 1 | The completed light:dark chamber, which consists of two separate units.

detected as quickly as possible. We determined that the best location for each sensor was on the floor, towards the front of the unit. We placed a light sensor inside a small electrical box inside each unit. When the sensor detects light, it sends an electronic signal to the control panel mounted outside each unit. This signal trips a relay switch that activates an audible alarm (85 decibels) and a flashing light (Idec, Sunnyvale, CA) to indicate that light is present inside the chamber. To silence the alarm, users can press a button on the control unit, which connects to a latching relay.

We installed one 20-watt fluorescent light bulb (30 in long), fully encapsulated in lexan tubing (K&H Industries, Inc., Newberry, MI), in each unit of the chamber. Each chamber unit contains tubular shelving that allows us to light two rows of cages. We installed one KT-V series digital timer (Koyo Electronics Industries Co., Tokyo, Japan) into the control panel for each unit; each timer can be programmed to turn the light bulb on and off in accordance with the desired light:dark cycle. We placed a double wall around all electrical connections and around the air intake and exhaust outlets (**Fig. 3**) to prevent any reflected light from entering the chamber.

We installed a temperature probe and a humidity probe inside each unit. The probes are connected to digital readout displays in the control panel (**Fig. 4**), which report the environmental conditions inside each unit. Personnel monitor these environmental conditions with a Series THC temperature and humidity switch (Dwyer Instruments, Inc., Michigan City, IN).

A small, filtered-air fan installed in the side of the chamber supplies ambient air to each unit inside; an exhaust outlet for each unit is located at the top of the chamber. We placed internal baffles in the air intake and exhaust outlets to prevent light from entering the chamber through these outlets.

We placed a fan (4-in diameter; capable of moving air at 28 ft³ per min; McMaster-Carr, Atlanta, GA) in the side of each unit to circulate fresh air from the room (filtered through high-efficiency particulate air filters) throughout the unit, thereby achieving a uniform environment inside each unit. We selected the fans based on the manufacturer's specifications for air movement. We used a velocity meter to confirm these specifications after the fans had been placed in the completed chamber. The air inside each unit is cycled at a minimum rate of 15 air changes per h to ensure that it matches room conditions. The Guide for the Care and Use of Laboratory Animals states that for secondary enclosures, an air exchange rate of 10-15 free air changes per h is an acceptable general standard¹. We chose the exchange rate of 15 air changes per h to prevent heat build-up and to eliminate the excess humidity generated by the animals themselves in an enclosed environment. We purchased the fans for use in the design to try to attain the recommended air exchange rates cited in the Guide¹.

A control panel (Fig. 4) located on the right side of each unit is used to monitor the environmental conditions of each unit. The control panels are also



FIGURE 2 A sketch of the chamber, which is 38 in wide × 25 in long × 73.75 in high, showing key components: exhaust vents, upper unit, lower unit, two control panels and push handle. Each unit can house either eight mouse cages or six rat cages. Mice and rats are not housed in the same chamber at the same time.



FIGURE 3 | Locations of the light towers, air supply units and exhaust vents.

used to control the output settings of the light detection alarm for each unit. Users can turn off the light or the sound of the alarms. The panels also contain connections that relay information about temperature, humidity, air flow, door position and the presence of light to the investigators' computers that are directly connected to the chamber.

To date, we have built three chambers. Researchers at Children's Hospital Boston started using the first chamber in August 2008. Investigators using the chambers are primarily responsible for observing the animals and for changing the cages. Animals are maintained in standard mouse cages (7.5 in wide × 11.5 in long \times 5 in high), which are changed on a weekly basis. Alternatively, rat cages (9 in wide \times 17 in long \times 8.5 in high) could be placed in the chamber. The chambers are cleaned with Clidox (base:water:activator; 1:5:1; Pharmacal Research Laboratories, Inc., Naugatuck, CT) before cages are placed inside and after each cage changing. Animal facility technicians also observe the health of the animals on a daily basis. All work is carried out in the dark, so military night goggles (Lab Safety Supply Inc., Janesville, WI) must be worn during cage changes and husbandry procedures.

The neuroscientists have reported that the chambers are ideal for their research involving manipulation



FIGURE 4 | Components of the control panel. 1, main power switch and cord; 2, temperature and humidity control; 3, exhaust recirculation fan switch; 4, light switch; 5, timer switch; 6, timer; 7, alarm silence switch; and 8, audio and visual alarm.

of light:dark cycles. We continue to observe that mice placed in these chambers seem to remain healthy and show no signs of stress. Researchers in Singapore have used our design to build their own light:dark chamber and are currently using it at a research facility.

Neuroscientists at Children's Hospital Boston have used these chambers to carry out research that has been published in multiple papers^{2,3}. Studies using the chamber were approved by the IACUC of Children's Hospital Boston and were carried out in accordance with the *Guide for the Care and Use of Laboratory Animals*¹.

COMPETING FINANCIAL INTERESTS

The authors declare no competing financial interests.

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