Constructing an inexpensive and versatile homemade rodent treadmill

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Recording aerobic exercise in small animals is common in various fields as it provides an opportunity to investigate behavior and analyze tissues not normally accessible in human experiments. Pertinent to these studies is having access to equipment which allows manipulation of the exercise stimulus in order to reach overload. In such cases, small rodent treadmills are excellent tools which can control the exercise stimulus and aid in acquiring relevant data. They allow researchers to specify exercise duration, frequency, resistance, and intensity and can be used to quantify work, power and energy expenditure.

The main alternative to rodent treadmills is voluntary exercise equipment, such as running wheels. However, such methods do not allow for control of exercise duration, frequency or intensity as wheels measure the number of revolutions per 24 hours. In addition, voluntary wheel running requires specialized, individual housing, which increases the need for space and can thus become very expensive.

Though they can provide added control during aerobic exercise studies, the cost of commercially available treadmills can be quite high; some can cost upwards of several thousand dollars with any species-specific requirements only increasing the price. As an alternative to commercial models, we built a functional rodent treadmill for only $300, which solves the cost problem and has provided us with valuable aerobic exercise data (**Fig. 1**).

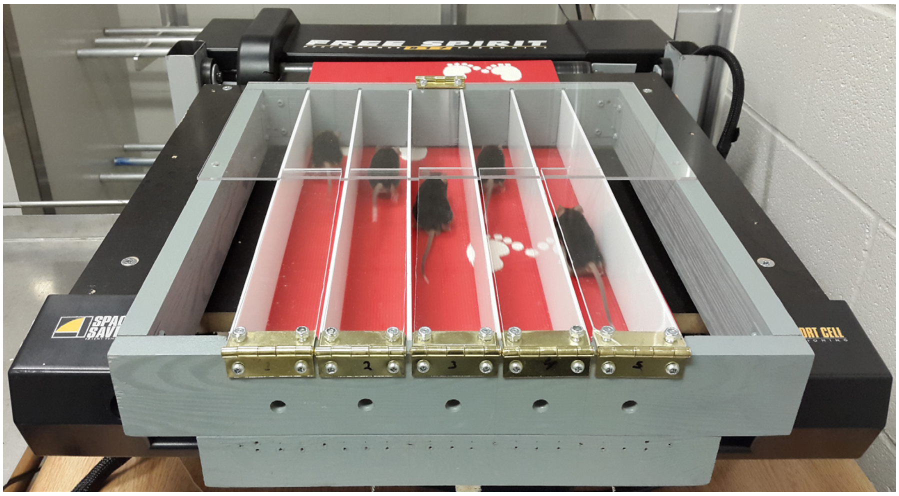

FIGURE 1 | Mice running on custom treadmill

Materials

Treadmill base and walking belt: We purchased a secondhand human treadmill from an online classified advertising website. This was used for the motor, overall base and control panel. Shorter walking belts can be customized, bought online, or found in specialty exercise equipment stores. We used the belt from an un-motorized child treadmill.

Animal-specific frame with lanes: To form the frame and running lanes, we used carpenter's pine wood coated with a plastic paint to allow for sterilization/cleaning using wipes. Lane dividers were made from white acrylic panels (46 cm x 92 cm x 0.3 cm). The whole frame was covered by a clear acrylic panel (61 cm x 92 cm x 0.3 cm).

Tools: The list of tools used includes: a drill, Allen keys, hammer, staple gun, wood saw, metal saw, circular saw, construction gloves, protective eyewear, scissors, camera, staple gun, hinges, finishing nails, screws, nuts and bolts, and plastic paint.

Methods

Resizing the treadmill: As we were working with a human-sized treadmill, the treadmill needed to be resized to provide a more convenient piece of equipment, both in terms of size (for storage and data collection) as well as weight (for easier portability). Furthermore, the stride length of a rodent is small enough that the full length of the treadmill was not needed to create appropriate experimental conditions.

Our first step was disassembly. Prior to disassembly, pictures were taken at every step for reference and guidance in the reassembly process. The treadmill was taken apart and the side moldings, rear caps, rear roller, treadmill belt, walking board and motor cover were removed (**Fig. 2**).

After disassembling the treadmill, we shortened the walking board and main frame to fit the child-sized belt. The extent that needed to be cut from the rear of the equipment was calculated as the difference between the long, adult sized belt and the child belt, divided by two. These were trimmed based on the calculated sizes using circular and metal hand saws.

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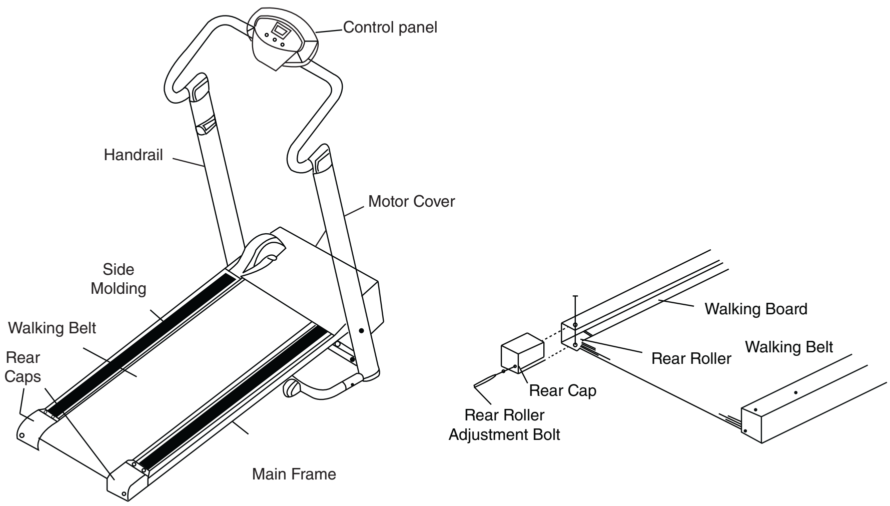

FIGURE 2 | Description and location of various treadmill parts

To decrease the treadmill's weight and increase its portability, we sought to remove as much metal as possible, which involved shortening the handrails. However, the control panel's wiring ran through the handrail to the motor. Therefore, we first had to remove the wiring. We started by disconnecting the control panel from the treadmill's circuit board (located in the motor cover) before the wiring was fished out. The handrails were subsequently cut using the metal hand saw to the height of the motor cover. The control panel then was reconnected by pulling the wiring back through the shortened hand rail. The excess wiring was kept, providing an extension to increase mobility when using the control panel.

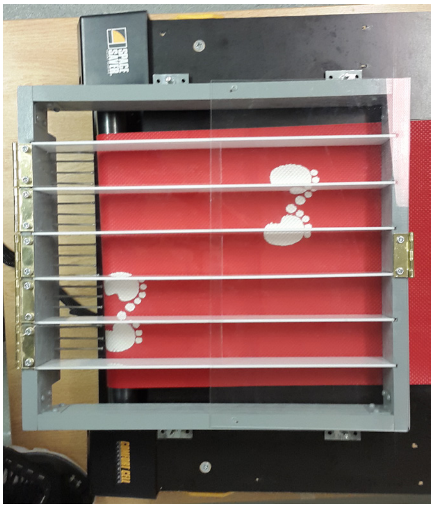

FIGURE 3 | Image showing the frame centered with the treadmill belt from a top view. The frame is wider than the belt but not as wide as the rear caps. The frame protrudes from the back of the treadmill to provide the exhaustion area.

Subsequently, the treadmill was reassembled using the shortened walking board and smaller treadmill belt. Once the belt was in place, adjustments were made to keep it centered and prevent shifting to either side. To accomplish this, we used an Allen key on the rear adjustment bolts located in the left and right rear caps. If the belt shifted to the left, the left adjustment bolt was tightened by half a turn while the right adjustment bolt was loosened by half a turn. Once the belt's large shifts had been minimized, the adjustments were fine-tuned until the belt remained centered. If the belt shifted to the right, this procedure would simply need to be inversed.

Preparing the frame with lanes: With a shortened treadmill ready, we then created a frame with running lanes to hold the rodents during trials. When designing the lanes within the frame, it is important that they meet the correct specifications for the target animal; several frames of different sizes can be prepared to accommodate various species. The maximum number of lanes depends on the width of the treadmill belt and the size of the animal. The design should allow for as many animals as possible to run during each trial. For mice, recommended lane dimensions are 40 cm long by 5.15 cm wide by 5 cm tall; for rats, 60 cm by 10.15 cm by 15 cm. The frame for the lanes must be slightly wider than the treadmill belt but cannot exceed the width of the rear caps (**Fig. 3**). The length and height of the frame should be the same as those of the chosen lane dimensions.

We used brackets to assemble the frame, though the method of assembly can vary as long as the dimensions of the frame are met. To insert lane dividers, we cut small slits (3 mm wide by 3 mm deep) for each panel across the height of the frame. These were marked with a pencil, etched out using a hand saw and smoothed using a file as shown in **Figure 4**. The lane dividers were measured, marked and cut from the white acrylic panel. The depth of both slits (6 mm total) must be added to the length of the dividers. The height of the dividers was the same as the lane's dimensional length. A variety of methods can be used to cut the acrylic panel—we used a ceramic tile saw. Once cut, the dividers should fit snugly into the slits. If not, a file may be used for minor adjustments. To keep the dividers from falling out of the frame, the front slits were blocked at the bottom using a staple gun while the slits at the back of the frame were blocked with a piece of pine wood. The back of the frame also serves as the animals' exhaustion area. This piece of wood should have a height 5 cm and a width 4 cm wider than the combined lanes, as shown in **Figure 5**. At this point the frame should be painted to block the pores of the wood and thus allow for easy cleaning and disinfection. While we used a plastic paint, this can be done using any preferred method.

To make the frame's cover, we used a clear acrylic panel attached to the frame with a hinge. In our method, the front half of the frame was covered with a single panel while each lane was covered separately in the back, as shown in **Figure 1**, to facilitate animal placement at the start of the trial. With each lane covered separately, the width of the lane lid should be the same as that of the lane and the length should exceed the length of the front half cover by one centimeter. This allows the front of the rear lane covers to rest on the back of the front frame cover.

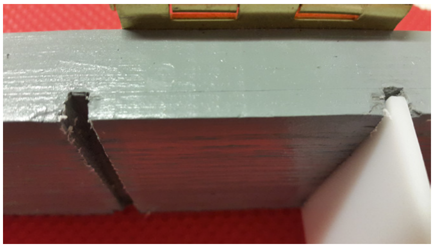

FIGURE 4 | Left side shows 3 mm wide slits to accommodate the lane divider. Right side shows the slits with the lane divider inserted.

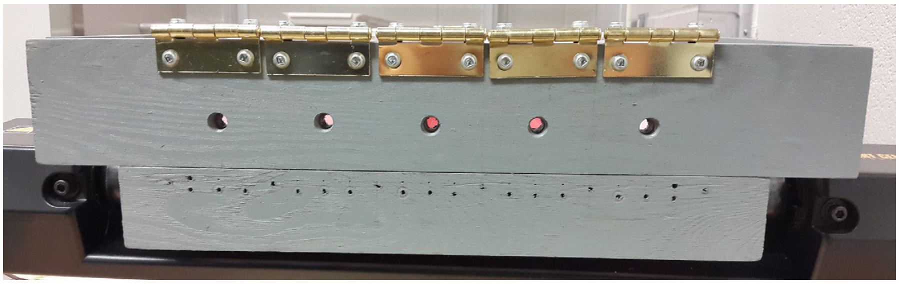

FIGURE 5 | The dimensions of the added piece of wood can be seen. Additionally, it blocks the slits formed the back of the frame and will hold the finishing nails for the exhaustion area.

Attaching the frame to the treadmill base: To place the frame on its base, we first screwed two "L" shaped brackets to the bottom left and right sides of the frame, 5 cm from the front and 15 cm from the back. Then, we placed the frame on the treadmill and centered it with the belt. The back of the frame should protrude from the rear of the treadmill one to two centimeters more than the width of the lane, to provide the exhaustion area shown in **Figure 3**. Using a pencil, we marked the placement of the holes on the walking board according to the four "L" brackets. Using a three tip drill bit, we drilled halfway through the top side of the walking board and placed four long bolts through these holes from the bottom out the top; the bolts were kept in place by a lock washer and a nut. This design allows for adjustments in height of all corners of the frame by placing a second nut on each bolt (**Fig. 6a**). The ideal height is slightly greater than the diameter of the animal's tail, as this will prevent the rodent from getting stuck between the frame and the walking belt. After placing the frame on the treadmill's bolts, we used a third set of nuts to secure the frame in place (**Fig. 6b**).

To finalize the exhaustion area in the back of the frame, we traced a line at the same height as the walking board onto the back of the frame. We then removed the frame and installed finishing nails at the marked height at a one centimeter distance with the exception of immediately below the lane dividers, where the finishing nails should be higher (**Fig. 6c**). The placement meant that the nails ended up within one centimeter of the belt when the frame was placed on the treadmill. This arrangement makes the back of the treadmill safe yet uncomfortable to the rodents. In addition, we used wire brushes fit between the nails to condition the animals to use the treadmill. Finally, we drilled a hole through the center of each lane through which a small air pump can be used to blast air at the animals and thus further motivate them to keep running (**Fig. 6d**).

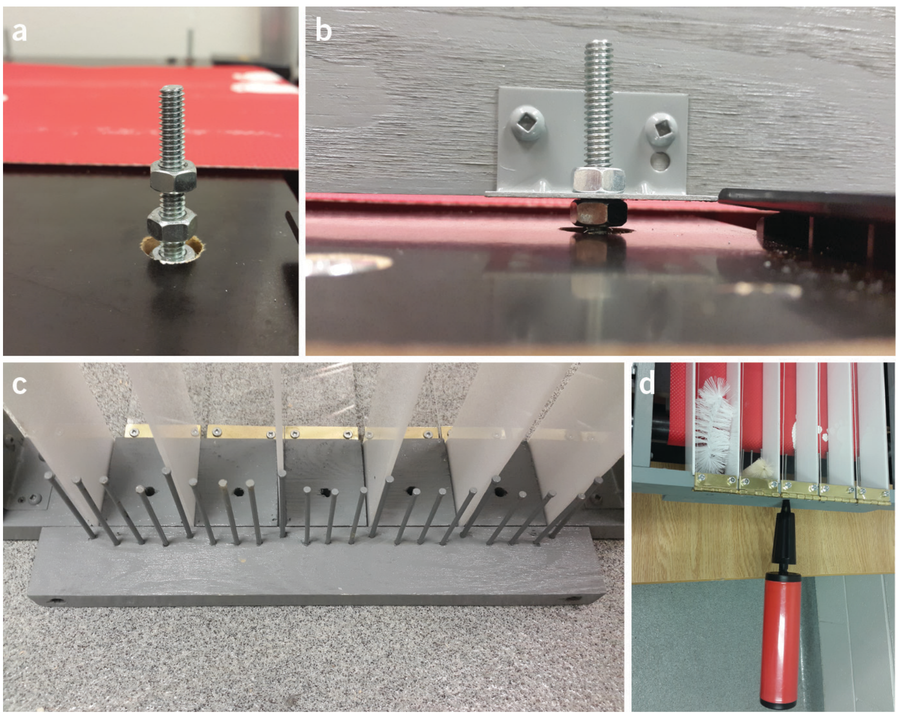

FIGURE 6 | Additional assembly details. (**a**) Lowest nut fastens the bolt in place to the base; the middle nut allows for adjustment of the frame's height; top nut fastens the frame in place. (**b**) Image of frame secured onto the treadmill base. (**c**) Arrangement of the finishing nails forming the exhaustion area. (**d**) Various methods to encourage the rodents to exercise. From the left, large wire brush in the first lane, smaller wire brush in second lane and air pump in the third lane.

Using our treadmill

Like any equipment used for forced exercise, the treadmill requires constant supervision and the animals must be trained how to use it. To test our treadmill, we acclimatized the rodents in the same room as the treadmill for a few hours before placing them in the lanes for 3 minutes. This was followed by turning the treadmill on for 3 minutes at a very low speed. Following this, we increased the speed to 3 m/min for 2 minutes then incrementally by an additional 3 m/min every two-minutes. If the animals went to the exhaustion area, a wire brush can be used to condition them to stay on the treadmill (**Fig. 6d**). Successful experiments have since been performed with our treadmill—for example, a six-week project recording ten mice on a low-carb/high-protein diet—which demonstrate its value and utility.

We consider our customized treadmill to be a versatile and economical solution to recording aerobic exercise in rodents, able to accommodate various species and costing only $300. The design is also more humane than some commercially available equipment—it relies on tail-tickling and air puffs rather than electric shocks. Researching, designing, and building our rodent treadmill took approximately one month, including a week of trial-and-error construction; we hope our documentation here can reduce that time to only a few days for others interested in building their own equipment.

COMPETING FINANCIAL INTERESTS
The authors declare no competing financial interests.