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An inclined plane system with microcontroller to determine limb motor function of laboratory animals

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Abstract

This study describes a high-accuracy inclined plane test system for quantitative measurement of the limb motor function of laboratory rats. The system is built around a microcontroller and uses a stepping motor to drive a ball screw, which changes the angle of the inclined plane. Any of the seven inclination speeds can be selected by the user. Two infrared (IR) LED/detector pairs function as interrupt sensors for objective determination of the moment that the rat loses its grip on the textured flooring of the starting area and slips down the plane. Inclination angle at the moment of IR interrupt (i.e. rat slip) is recorded. A liquid crystal display module shows the inclination speed and the inclination angle. The system can function as a standalone device but a RS232 port allows connection to a personal computer (PC), so data can be sent directly to hard disk for storage and analysis. Experiments can be controlled by a local keypad or by the connected PC. Advantages of the presented system include easy operation, high accuracy, non-dependence on human observation for determination of slip angle, stand-alone capability, low cost and easy modification of the controlling software for different types of experiments. A fully functional prototype of the system is described. The prototype was used experimentally by a hospital group testing traumatic brain injury experiments, and some of their results are presented for system verification. It is found that the system is stable, accurate and easily used by investigators.

Keywords: Animal model; Laboratory animal; Inclined plane; Motor function; Neurologic examination

1. Introduction

Experimental studies of pharmacology, neuropathology and pathophysiology commonly measure the motor function of rats. It is generally believed that the central nervous system is designed to produce behaviour; behavioural assessment is the ultimate performance of brain or spinal cord function. The behavioural performance can be used as an index for neural disease process or therapy efficiency. According to the review of Whishaw et al. (1999), most of the behavioural methodology comes from research on rats. However, the ethograms of rodents are similar so that the methods developed for rats can be applied to other rodents. The following testing methods have been developed: (a) motor scores based on screen, balance and prehensile-traction tests (Combs and D'Alecy, 1987); (b) beam walking test (Feeney et al., 1982); (c) limb-placement test (Ryck et al., 1989); (d) neurological examination based on posture (Bederson et al., 1986); (e) open field score (Giulian and Silverman, 1975; Koob et al., 2006); (f) inclined plane test (Rivlin and Tator, 1977; Yonemori et al., 2000; Carter et al., 2002; Hallam et al., 2004; Spangenberg et al., 2005). Methods (a–d) contain many parameters, which require subjective evaluation by an individual human observer, from which overall motor function impairment is estimated. The results of such test methodology may vary for different observers.

On the other hand, the inclined plane test methods (f) is objective and quantitative because the only parameter tested is the angle at which the rat can no longer maintain its grip on the tilted plane and begins to fall. The inclined plane test was used to test muscle performance capacity of the rats. This test was able to measure motor functions after neural damage in rats, by assessing the rat's ability to prevent itself from falling over and also assessing the endurance strength in the upper and lower limbs. The lever of tilt is mechanically obtained so the angle can be measured easily and accurately. Since timing of the angular measurement is based on the rat falling bodily off the plane, the measurement does not vary for different operators. Finally, the test easily detects differences of motor function with respect to

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cerebral laterality and spatial orientation by readily allowing initial placement of the rat on the plane in right-headed, left-headed, up-headed or down-headed position.

The present study was performed to investigate the effects of traumatic brain injury (TBI) on motor function using an improved inclined plane testing system. The inclined plane testing system is driven by a stepping motor and equipped with infrared sensors for detecting the moment that the rat slips down the inclined plane. In addition, the firmware of the system is easily modified for use in different experimental requirements.

2. Materials and methods

2.1. Inclined plane system

The following describes a new microcontroller-based inclined plane system for measuring the motor function in small laboratory animals such as rats. The physical apparatus consists of two major physical components, i.e. an inclining plane and a support box. Within the box are the electrical and mechanical elements for elevating the plane, and the electronic components for controlling and sensing the system. A parallel port is provided for connection to a conventional PC, which is used to write and load the firmware used in the microcontroller. The PC can be used to directly store collected data via a RS232 port and also to link the inclined plane test system to a LAN or the Internet.

2.1.1. Apparatus

A photo of the lab-built apparatus is shown in Fig. 1. An inclined plane made from a 60 cm × 60 cm acrylic plane is coupled by a pair of hinges to a support box. Near the edge of the plane that achieves maximum elevation a three-walled enclosure is mounted so that the missing wall faces down the slope of the plane as it inclines. The floor of the enclosure is covered with hook-side Velcro fabric, providing a secure area on which the rat can grab and hold on. In fact, this enclosure is easily modified or exchanged for special use. For example, the enclosure seen in Fig. 1 was being used in real hospital experiments when photographed.

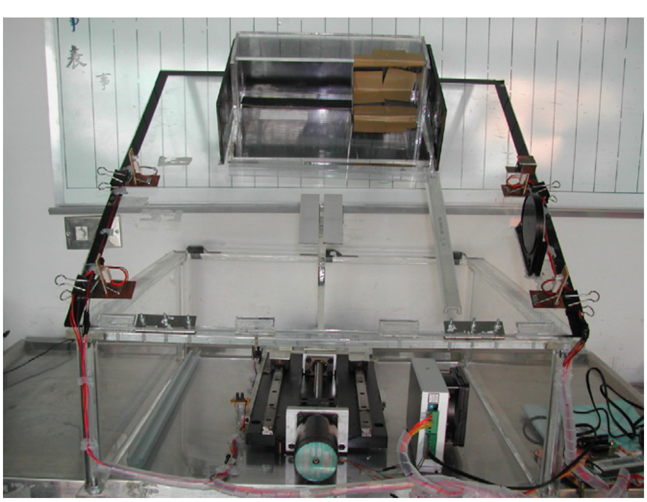

Fig. 1. Apparatus of the inclined plane test system.

The inclination of the plane is controlled by a ball screw turned by a stepping motor and its driver. Two mechanical switches are mounted by the slide rail of the ball screw so the plane's angular motion is limited to the range from 30° to 70°. The speed of inclination can be selected by the user from a local keypad or by command from a conventional PC that is coupled to the inclined plane system. In our laboratory prototype, seven pre-set inclinations speeds are available for push-button selection by a user. The number of speeds and their specific values are adjustable during initial design of the firmware for the microcontroller. Two infrared (IR) source/detector pairs are mounted on the plane such that two IR light beams are directed across the path through which a rat must fall if it loses its grip on the Velcro. The locations of these beams are adjustable on the board, allowing for experimental variation. The configuration seen in Fig. 1 has one source/detector pair close to the enclosure and one near the bottom of the board. This is so that a rat hanging onto the Velcro but dangling out of the enclosure can be detected as a distinct event. Then, when the rat fully loses its hold on the Velcro and falls, the second source/detector pair will capture this event clearly and distinctly. The black round object connected to the side of the plane between the upper and lower source/detector pairs in a goniometer, which is used for measuring the angle of incline during calibration.

2.1.2. Control unit

A block diagram of the control unit hardware is shown in Fig. 2. The control unit consists of a microcontroller, an LCD display, a touch-button keypad, an infrared carrier signal generator and multiplexer switch, a RS232 voltage level converter and a power supply. The following presents brief descriptions of these components.

2.1.2.1. Microcontroller. The system is based on an ATmega32L microcontroller (ATMEL Corp.), which has a RISC architecture and provides fast execution time. The ATmega-32L includes four programmable ports, three external interrupt sources, three timer/counters, one programmable serial USART and 32 K program memory and 2 K SRAM for data memory. Assignment of the various functions to the various ports is shown in Table 1.

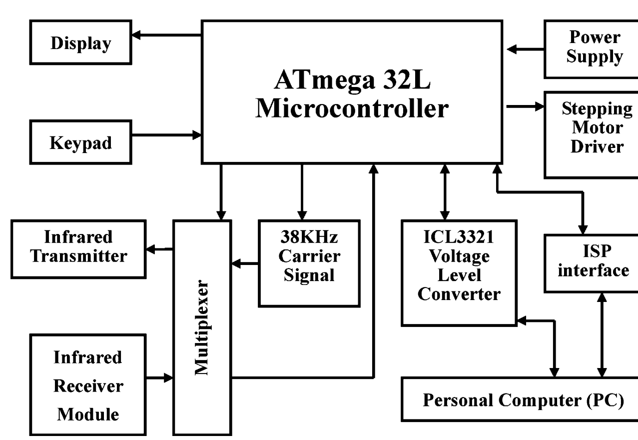

Fig. 2. Block diagram of the microcontroller-based control unit.