METHODS & DESIGNS

An infrared device for detecting locomotor activity

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An electronic device for detecting locomotor activity in the home cage of small animals is described. The system involves a dense array of infrared (IR) beams designed to function as switches and capable of penetrating plastic walls of standard rodent cages without operational decrement. The device is reasonably inexpensive, is simple to calibrate, can be used with groups of animals, may be used for extended periods without maintenance, and is reliable with different cages.

Infrared-actuated motion detectors can provide automatic monitoring of an animal's locomotor activity in its home cage without perturbing the pattern of its normal behavior or initiating the spurt of exploration occasioned by transfer to a novel environment. Although an activity monitor based on photocells is not novel, only the recent advent of inexpensive, high-output, narrow-beam infrared (IR) LEDs and of sensitive IR phototransistors makes dense arrays of these devices practical without focusing apparatus. Commercially built systems of this type are available, but are rather expensive.

Perhaps the most common form of activity detector is some variety of the so-called "jiggle platform." The principle of this device is the translation of the movements of an animal on a balance into electrical signals (e.g., Campbell, 1954; Cunningham, Crowell, Eaton, & Brown, 1973; Zeier & Tschannen, 1968). These "stabilimeters," because of the combined mass of the platform and animal, have a relatively large inertia, which tends to reduce their sensitivity. Stabilimeters suffer from a lack of interinstrument reliability and, as Tarpy and Murcek (1984) pointed out, are difficult to calibrate.

Inexpensive ultrasonic (Akaka & Houck, 1980) and doppler-radar (Martin & Unwin, 1980) motion detectors are now available. Existing ultrasonic motion detectors, however, operate at a frequency (40 kHz; see Akaka & Houck, 1980) to which rodents and other small animals

are known to be acoustically sensitive (Paranjape, Secord, & Voss, 1983). An unfortunate interaction exists between the physical dimensions of the target animal and the amplitude of the recorded activity. The latter defect makes it invalid to compare animals of the same age and strain but of differing body masses or to use more than a single animal in a given apparatus. The doppler-radar systems suffer from similar defects. The orientation of the animal (i.e., the cross-section presented to the radar detector) at the time of movement determines the magnitude of return. These problems can be compensated by autocorrelational analysis (see Buchan & Sattelle, 1979), but it is doubtful whether they can be resolved completely.

Some motion-detection devices employ electronic circuits that are designed to respond to changes of resistance across conducting plates or grids as the animal moves about (Morgret & Albee, 1974; Tarpy & Murcek, 1984). The disadvantage of these devices lies ultimately in their susceptibility to contamination (dry urine or feces on the plates or grids forms an insulative coating) and, because of this susceptibility, the difficulty in using them over extended periods or in the animal's home cage. The system proposed by Tarpy and Murcek (1984) minimizes this problem by the use of a sophisticated circuit that employs the resistance found in a previous interval as a standard against which to compare resistance in the present to determine whether movement has occurred (autocorrelation). Even these authors, however, recommend occasional flushing with a nonionic solvent to maintain trouble-free operation.

A novel approach to the problem of automatic monitoring of activity is found in the television/computer system for recording open-field activities outlined by Livesey and Leppard (1981). This approach, in addition to utilization

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CIRCUIT DESCRIPTION

Eight infrared LED/phototransistor pairs are arranged in an alternating array and are embedded in aligned holes drilled in blocks of opaque Plexiglas (Figure 1). In operation, the rodent cage is positioned between these blocks against a protruding stop. The two blocks are bonded to a similarly opaque bedplate of Plexiglas. The eight IR phototransistors (Q_1 – Q_8) are normally kept in a conducting state by the action of the eight IR LEDs (LED_1 – LED_8) in the holes opposite. As the animals move about, their bodies selectively occlude the IR beams passing through the cage. Each occlusion stops conduction in the pho-

The positive voltage pulse from the phototransistor is seen at the comparator's noninverting input, IC₁ (Figure 3). If no further movement occurs, the capacitors (C₁–C₈) block any additional voltage from IC₁. The negative pulse that occurs when an animal moves away from an occluded transistor is blocked by the diodes (CR₁–CR₈), which also form part of an eight-legged OR gate.

As long as the pulse on IC₁ exceeds the noise-rejection level, as established by rheostat R₁₉, an output is produced by the comparator. The minimal setting of this noise-rejection rheostat is determined by the ambient levels of electrical noise in the environment and by the length of the cable connecting the main chassis to the sensing unit (in this case, 2 m of 12-conductor ribbon cable).

The reliability and simplicity of the design derives largely from the characteristics of the IR LEDs and phototransistors. These circuit elements are capable of supplying usable signals to the OR gate at a distance far greater than the 17 cm used in the sensing unit. This causes the LED/phototransistor pairs to act against a ceiling effect (i.e., to behave essentially as though they were switches rather than analog devices).

Figure 1. Dimensions of the Plexiglas sensing unit are presented. This unit is designed to hold a standard 30.5 × 19.5 × 13-cm acrylic rodent cage. For clarity, the unit is shown without electronic components, terminal strips, wiring, or cables.

Figure 2. A circuit diagram of the main-chassis current supply and the electronic components on the sensing unit. (Note that if metal-shelled plugs are not available for the sensing-unit's ground return, one of the unused pins on P1/J1 may serve as a ground.)

Figure 3. Circuit diagram of the detection and interfacing components (IC₁, IC₂, & IC₃).

The one-shot multivibrator (IC₂) produces a 5- to 25-msec pulse (depending on the setting of R₂₃) each time it is triggered by the comparator (Figure 3). This feature makes it possible to use the device at the most advantageous pulse width in width-sensitive systems. Below a 15-msec pulse width, for example, many electromechanical counters are unable to complete their cycles. The digital

counter (Figure 4) is not pulse-width sensitive within the limits of R₂₃.

The voltage follower (IC₃) forms an interface between the one-shot multivibrator and the recording equipment. The zener diode (Z₁) reduces the output of the follower to TTL levels for the computer and digital counter. The counter has a five-digit, seven-segment LED display.

Figure 4. The optional five-digit LED readout circuit. Although not critical, this circuit is very useful for accumulating grand totals and for checking the operation of computer channels.

The MOS/FET electromechanical interface is designed to drive recorders with a low-active output to 8 A of near-instantaneous current at 60 V or less.

The loaded YES circuit (Figure 5) "cleans up" the TTL output for noise-sensitive computer-input channels. The use of this circuit is an option that depends on the chassis wiring, the noise characteristics of the TTL zener diode (Z_1), and the input characteristics of the computer-input channel.

CONSTRUCTION

The most critical phase of construction involves the alignment of the LED/phototransistor pairs (Figure 1). The utmost care must be taken to insure that the vertical Plexiglas blocks housing these pairs are of equal size. The blocks should be clamped together, and the holes that will house the pairs should be bored in-line by a drill-press. If rods are placed through these holes prior to bonding of the blocks to the bedplate, a more precise alignment results. After the phototransistors and LEDs are soldered to terminal strips at the rear of the blocks, the unit should be energized, and voltage readings should be taken between the collector of each transistor and chassis ground. The physical alignment of the pairs then should be changed by bending their leads so as to achieve a reading of -8 V or less on a voltmeter.

The dimensions of the sensing unit in Figure 1 were selected to accommodate a standard $30.5 \times 19.5 \times 13$ -cm acrylic rodent cage. It previously had been determined that these cages passed IR beams with little if any attenuation. The height of the plane of LED/phototransistor pairs above the bedplate is the sum of the height of the cage floor, 500 ml of evenly spread Sanicel litter, and the normal shoulder height of a 35-g mouse. Prior to wiring and installation of the electrical components, the entire unit should be sanded lightly and rendered opaque by the application of flat-black enamel. This simple expedient shields the photosensitive elements from external light and precludes interaction between elements. External sources

Figure 5. Diagram of the loaded YES circuit for noise suppression.

of light, depending on their spectral character and intensity, might otherwise present a problem.

The digital counter, consisting of IC₄-IC₁₃, R₃₀-R₆₅, and the five seven-segment LEDs, are mounted on a 9.5 × 14-cm grid board (Figure 4). This board is positioned on stand-off insulators against the front of a metal cabinet. The face of the cabinet has a Plexiglas-covered aperture through which the seven-segment LEDs are visible. The three regulators (7912, 7812, and 7805) are mounted against the rear of the cabinet on mica insulators (Figure 2). The 5-V regulator is mounted on a double heat sink (both inside and outside of the cabinet). The MOS/FET is mounted on a heat-sink assembly and a mica insulator and attached to the bottom of the cabinet. Other electrical components (diode bridge, capacitors, resistors, etc.) are mounted on terminal strips. The IR LED current-limiting resistor (R₁₇) should be mounted clear of other components to promote dissipation of thermal energy.

The circuit pictured in Figure 3 is mounted on a 9 × 4.5-cm PC board, placed on four stand-off insulators, and positioned against the rear of the cabinet.

All hardware and all electrical and electronic components were purchased from Radio Shack for approximately \$175. If electromechanical or TTL output alone is sufficient, the construction cost can be cut in half by eliminating the digital counter (Figure 4) with its associated voltage regulator (7805) and heat sinks.

QUALIFICATIONS

Photoelectric-cell activity monitors have always been limited in sensitivity. This limitation is a function of the number of sensing devices. The practical number of these devices depends on the dispersion angle of the radiant-energy transmitters and the acceptance angle of the receivers. The wider the angles, the greater the chance that a single transmitter will simultaneously excite several receivers. The IR LEDs used in this device have a dispersion-angle, half-power point of 24°. This makes possible a greater density of sensing devices than was practical heretofore without costly lens systems.

Our activity monitor is unidirectional, in that it is insensitive to motion parallel to the IR beams. The reasons for this design involve simplicity of construction and ease of calibration. If needed, as many as three additional LED/phototransistor pairs may be added at right angles to the eight existing pairs to correct this condition (the longer baseline of these pairs necessitates a decrease in their density because of the problem outlined in the preceding paragraph).

Calibration of the activity monitor is simple because of the binary nature of the circuit. Each time a pencil or another small, opaque object is run along a path orthogonal to the IR beams, a count of 8 (one for each IR LED/transistor pair) should appear on whatever readout device is being monitored. Inaccurate counts indicate that a comparator reference-level adjustment R₁₉ is required. If two or more activity monitors are being calibrated, it

is necessary to set their comparator reference voltage levels and pulse widths to the same value.

Sanicel litter is recommended because rodents tend to pile woodshavings against the cage walls, blocking one or more IR beams and thus reducing the sensitivity of the motion detector. Dry Sanicel, like dry sand, spreads out and normally cannot be piled high enough to block the beams if used in moderation and renewed periodically.

Although we anticipated that only new and pristine rodent cages would prove satisfactory in the IR sensing unit, it was later found that even cages so scratched and aged as to be virtually translucent presented no problems. The IR beams pass through them with no detectable spread and little, if any, attenuation.

TESTING THE IR-ACTUATED MOTION DETECTOR

After calibration of the motion detector, a test was conducted with live mice.

Method

Subjects. Four male BALB/CANN mice (Charles Rivers Labs, Inc., Stoneridge, NY), all 70 days of age, were used to test the movement detector. The mice were supplied ad lib with Teklad Lab chow (Harlan Sprague Dawley, Inc. Co., Winfield, IA) and water. They were maintained on a 12:12 light:dark (LD) schedule (lights on at 6:00 a.m.). Sanicel litter (Anderson Cob Division, Delphi, IN) covered the cage floor.

Apparatus. The TTL output of the IR-actuated detector was run through the YES circuit and connected to the counter input of an ISAAC-91A data-acquisition system (Cyborg Corp., Newton, MA). The ISAAC-91A system is connected to an external power source (schematic available from the senior author) to reduce the electrical noise inherent to the Apple IIe microcomputer (Apple Computer, Cupertino, CA) to which the ISAAC-91A is connected. The Apple computer is equipped with Apple Duo-disk disk drives and an Apple Monitor II.

Procedure. At 10:00 a.m., 2 h prior to initiation of data collection, the four mice were placed in a standard acrylic rodent cage, as described above. The cage was placed in the sensing unit, and the unit and cage were placed in a room adjoining the location of the data-collection-and-analysis equipment. (The requirement for this precaution is greatly reduced by the virtually silent operation of the programming and recording equipment once an experiment has begun; the electromechanical output option of the motion detector was not used in this test.) The only associated noise was generated by the disk drives when data were saved to disk at 5-h intervals. To obtain one complete circadian cycle, we chose arbitrarily to collect locomotor movement data at successive 1-min intervals for 24 h.

Two relatively simple programs (Appendices A and B) were written in Applesoft and Labsoft, a proprietary enhancement to Applesoft provided by the Cyborg Corporation for use with the ISAAC-91A.

The first program (Appendix A) was designed to receive the input from the motion detector into counter channel No. 4 of the ISAAC-91A at 1-min intervals (& Pause = 60) over a 24-h period. At the end of 300 min, the data stored in an array were saved to disk. Loss of data during file saves was minimized by the use of a binary file (up to 16 times faster than a simple sequential file). The decision regarding the frequency of saving data was made arbitrarily; it should be based on how many data an investigator is willing to lose in the event an experiment is inadvertently terminated. At the end of 24 h, the program was terminated. All data exist in a binary file, and they may be analyzed or manipulated at the convenience of the investigator. First, however, the binary file must be accessed and read from the disk into a suitable program.

The program in Appendix B was written to read the existing data file and to reduce the totals of 1,440 1-min counts to 144 10-min counts. The choice of data reduction was performed solely for illustrative purposes.

Results

The total accumulated counts recorded by the IR-actuated motion detector was 76,721 (on the digital counter). The program controlling the ISAAC-91A registered 76,642 counts. Over a period of 24 h, there was a discrepancy of 79 counts which yielded agreement of 99.9% between the two systems. The results of the total counts accumulated per hour and at 10-min intervals are presented in Figure 6. The counts per minute ranged from 0 to 359. The hourly and daily count totals were comparable to those that we had previously obtained for mice on a commercial (LKB Animex, Type DSE) activity meter.

Discussion

Locomotor activity is an arbitrarily defined unit of measurement that depends on the resolution of the detection system and the objectives to be attained in a given ex-

periment. If the experimenter wishes to define only gross body movements as signals, and maintenance behaviors as noise, then only very sophisticated and expensive activity monitors, such as those based upon television/computer systems or interlaced beam photoelectric devices, will be satisfactory. The number of beam intersection points or the resolution of the computer-generated electronic grid, however, is the limiting factor in both of these schemes. The IR-actuated activity monitor described in this paper can be triggered by an opaque object of only a few millimeters diameter. It must be kept in mind that a mouse grooming in an IR beam can produce many counts of something other than locomotor activity.

In this test, we chose arbitrarily to accumulate counts at 1-min intervals with the movement detector set at maximum pulse width (25 msec). As illustrated in Figure 6, the classic circadian pattern of activity is easily discerned and, depending on the interval chosen (e.g., hours or minutes), the resolution (length of interval) can be readily adjusted. The movement detector operating in conjunction with the ISAAC-91A data-acquisition system and the software yielded reliable counts at intervals measured in seconds. For example, based on 5-sec sampling intervals, 500 counts were artificially generated. The ISAAC system yielded 496 counts for accuracy of 99.2%. Concordance between detector output and the counts recorded does not diminish appreciably until measurement intervals of less than 1 sec are attempted.

The movement detector is not the limiting factor. Rather, it is the capability of the software that presents problems as the measurement interval is decreased. In this case, there are two periods when counts may be missed. The first is inherent in the finite amount of time that is required for an Applesoft Basic "for-next" loop to execute (Appendix A, Lines 120-300). The second case is when the subroutine (Lines 400-600) for saving data to disk is executed. During bench tests, we found that at intervals as short as 1 sec, no counts were missed within a given counting period [i.e., during the execution of "& PAUSE" and "COUNTERIN, (TV) = A(B)" statements (Appendix A, Lines 140, 150)]. The speed of the "for-next" loop could be increased by deleting the portion of the program that prints incoming data to the screen, a relatively time-consuming process that was inserted to yield a visual indication that the system is operating appropriately during the test. The time consumed by saving the data to disk at regular intervals is within the control of the experimenter. We chose to use a binary file format, as that is the least time-consuming method. Also, as stated above, the number of times a "file save" is executed is at the discretion of the experimenter.

In conclusion, we have found the IR-actuated movement detector to be a highly sensitive and reliable device in spite of relatively modest cost. It is appropriate to a wide range of experiment protocols and data-recording systems, and it avoids many of the problems inherent in previous devices.

Figure 6. Graph of the locomotor activity counts of four mice taken over a 24-h period and summed at hourly and at 10-min intervals.

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APPENDIX A

Applesoft and Labsoft Program to Collect and Save Movement Data

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100 TEXT: HOME: A = 2: B = 0: D = 0: E = 1440: DIM A (1500): A(0):
    B = 0: F = 0: A$ = CHR$(4): PRINT A$: "NOMON CIO": VTAB 11: PRINT
    "OUTPUT FILE NAME? ": PRINT: INPUT "": B$: REM CLEAR SCREEN,
    ESTABLISH VARIABLES, AND GET OUTPUT FILE NAME FROM KEYBOARD INPUT
110 & CNTFMT = 4: REM GET INPUT ONLY FROM ISAAC CHANNEL NO. 4
120 FOR G = 1 TO 5: FOR H = 1 TO 300: B = B + 1
130 & CLR COUNTER, (C#) = 4: REM SET CHANNEL NO. 4 TO ZERO COUNTS ON
    EACH LOOP.
140 & PAUSE = 60: REM ACCUMULATE COUNTS FOR ONE MINUTE
150 & COUNTERIN, (TV) = A(B): REM ASSIGN COUNTS TO ARRAY
160 F = F + A(B): PRINT B, A(B): IF B = 1440 THEN G = 5: H = 300: GOTO
    500: REM PRINT TOTAL COUNTS FOR EACH MINUTE TO THE MONITOR SCREEN AND

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UPON COMPLETION OF DATA COLLECTION LEAVE LOOP CLEANLY, SAVE THE DATA, AND END.

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200 NEXT H: IF G = 1 THEN GOSUB 400
300 GOSUB 500: NEXT G
400 J = (E + 1) * 5: REM ESTABLISH LENGTH OF ARRAY IN PREPARATION TO
    SAVE AS A BINARY FILE.
500 A(0) = PEEK(131) + PEEK(132) * 256: D = A(0): PRINT A$: "BSAVE ":
    B$: "A": D: ",L": J: ",D": A: IF B = 1440 THEN PRINT "THAT'S ALL
    POLKS:": PRINT F: END: REM SUBROUTINE TO SAVE ARRAY AS A BINARY
    FILE AND END PROGRAM WHEN ALL DATA HAS BEEN COLLECTED.
600 RETURN

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APPENDIX B

Applesoft Program to Read a Binary Array of the Movement Data from Disk and Convert to Counts Per 10-Min Interval

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10 TEXT: HOME: D = 2: D$ = CHR$(4): REM CLEAR SCREEN, SET TO TEXT,
    AND ESTABLISH DISK DRIVE 2 AS THE DATA DRIVE.
20 DIM MOVES(1440), X(145): REM CREATE SPACE IN MEMORY FOR TWO
    ARRAYS.
40 MOVES(0) = PEEK(131) + PEEK(132) * 256: REM FIND THE ACTUAL
    MEMORY LOCATION OF THE ARRAY TO STORE THE ORIGINAL MOVEMENT
    DATA AND ASSIGN THE VALUE TO MOVES(0).
50 PRINT D$: "BLOAD ": F$: "A": MOVES(0): ",D": D: REM READ THE
    ORIGINAL DATA FROM THE DISK FILE AND PUT IN THE MOVES() ARRAY.
80 FOR J = 1 TO 1440 STEP 10: REM STEP THROUGH THE MOVES ARRAY IN
    INCREMENTS OF 10.
90 M = J + 9: REM SET M EQUAL TO THE NEXT TENTH DATA POINT.
100 FOR JA = J TO M: REM LOOP THROUGH EACH SUCCESSIVE TEN DATA
    POINTS IN THE MOVES() ARRAY.
110 X(M/10) = X(M/10) + MOVES(JA): REM PLACE EACH 10 M TOTAL IN THE
    X() ARRAY.
120 NEXT JA: NEXT J
130 FOR I = 1 TO 144: PRINT I, X(I): NEXT: REM PRINT CONVERTED DATA
    TO THE MONITOR SCREEN. HERE IS A PLACE TO INSERT A FILE SAVE FOR
    YOUR FAVORITE ANALYSIS AND GRAHICS PROGRAM OR TO INSERT YOUR OWN
    ANALYSIS ROUTINES.
140 PRINT D$: END: REM CLOSE FILE, END THE PROGRAM.

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