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BRIEF COMMUNICATION

Simple Device for Quantifying Drug Effects on the Righting Reflex

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REEVE, B., B. DINGWALL, C. L. DARLINGTON, S. J. SCOTT, A. J. SANSOM AND P. F. SMITH. *Simple device for quantifying drug effects on the righting reflex.* PHARMACOL BIOCHEM BEHAV 42(1) 183–185, 1992.—A simple, inexpensive device is described that allows quantification of the effects of drugs on the righting reflex. This device consists of a modified set of kitchen scales connected to a digital timer. Two moveable Hall effect switches are positioned around the pointer, which registers the weight of the animal on the scales; when the animal is placed on the scales in the supine position, the initiation of a righting reflex causes the pointer to cross one of the switches, stopping the digital timer and providing a measure of righting reflex latency (RRL). We describe an efficient protocol for using this device that provides quantification of drug effects on the RRL, which can then be subjected to analysis using parametric statistics such as analysis of variance.

Righting reflex Guinea pig Drug effects NMDA antagonists

IN psycho- and neuropharmacological studies using animals, it is often desirable to quantify the effects of a sedative or stimulant drug on motor behavior, which can then be compared with results from electrophysiological or binding studies using the same drug. Various procedures have been used for this purpose, including performance on rotarods (1) and treadmills (3). Although performance on these tasks can be quantified, they involve complex motor activity under voluntary control and variability between animals can arise from the drug having effects at different levels in the generation of the complex response. By contrast, the righting reflex is sometimes used as a measure of sedation because it is easily identified and results from the activation of a well-defined reflex pathway (4). However, the effects of drugs on the righting reflex are usually described qualitatively rather than by obtaining a quantitative measure of righting reflex performance. We describe here a simple, inexpensive device and an efficient protocol for obtaining a quantitative measure of the effects of drugs on the righting reflex latency (RRL) that we used to study the effects of sedative drugs on the righting reflex in guinea pigs.

APPARATUS

In lower mammals, the righting reflex is an axial rotation of the head and body (i.e., in the roll plane about the horizontal axis) into the prone position that is generated when an animal is placed in the supine position (i.e., on its back) (2). In drug-free guinea pigs with normal vestibular apparatus, this response usually occurs within a few seconds (see Fig. 1). We developed a device to quantify this response that consists of a modified set of kitchen scales (Hanson Dietetic Scales, Northbrook, IL, Model 1440) and a digital electronic timer (Lafayette Instrument Co., Lafayette, IN, Model 54417-A) (see Fig. 2). The scales are modified in two ways. First, the weighing tray is replaced by a semicylindical platform covered with felt so that the animal can comfortably lie on it in the supine position. Second, the pointer that registers the weight of the animal on the weight display has a magnet attached to it; two moveable Hall effect switches (I. C. Switch and Magnet, Stock No. 307-446, Radio Spares Components Ltd.) are positioned so that they are equidistant from the pointer and form an angle of approximately 20° (see below; see Fig. 2). When the animal is placed on its back on the weighing plat-

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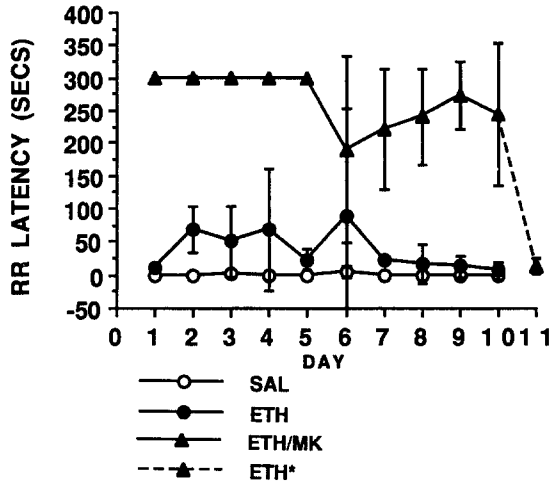

FIG. 1. Example of data obtained using the method of measuring RRL described in this article. Data points show average RRLs. SAL, 2 ml/kg IP saline (n = 3); ETH, 0.64 g/kg IP ethanol (n = 4); ETH/MK, 0.64 g/kg ethanol + 0.5 mg/kg MK-801 IP (n = 4); ETH*, animals in the ETH/MK group tested again on the eleventh day with ethanol (0.64 g/kg, IP) alone (n = 4). Bars represent ± 1 SD of the mean.

form, the pointer registers the animals's weight and the switches are positioned as described above; when the animal initiates a righting reflex movement, which causes displacement of the pointer, the pointer activates one of the switches by moving across it. The switches are connected to the digital timer, which is started manually when the animal is placed on the scales but stopped automatically when the pointer activates one of the switches. The digital timer therefore provides a latency (in seconds) for the initiation of a righting reflex (see Fig. 2). The position of the Hall effect switches with respect to the pointer on the scales has to be altered for animals of different weight; therefore, animals are weighed prior to measurement and the positions of the switches are adjusted appropriately. The angle the two switches form around the pointer is critical to determining the sensitivity of the device to the animal's movements. If the angle is too small, then very small movements that are not part of a righting reflex will activate the switches and stop the timer; on the other hand, if the angle is too large then even a righting reflex movement may not activate the switches. Determining the appropriate angle that will detect righting reflex movements but not other

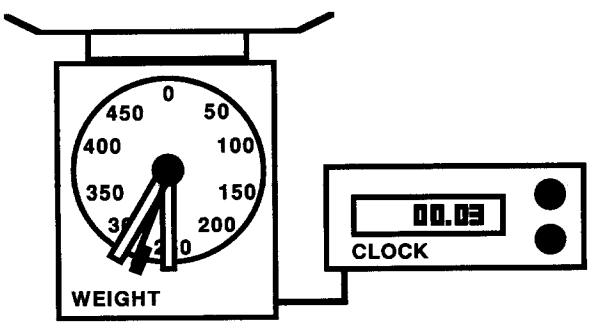

FIG. 2. Schematic diagram of apparatus.

movements requires a preliminary study. In our studies with guinea pigs, an angle of 20° fulfills these criteria. As an additional measure to ensure that movements other than righting reflex movements do not activate the switches, we videotape trials using a videocamera (Panasonic NV-M7) with a zoom lens, then replay the animals's movements in slow motion using a videorecorder (Mitsubishi E7 Black Diamond) and a color monitor (Sony Trinitron).

PROCEDURE

We developed a protocol that we use in conjunction with the apparatus described above. The most significant obstacle in obtaining accurate measurements of RRL using this device is the initial positioning of the animal on the weighing platform. This often requires several attempts and it is usually not possible for one person to both position the animal and start the timer (although this would be possible using a foot pedal to start the timer). We found that guinea pigs are easier to position on the platform if they have been handled regularly for a week or so before the experiment begins and are accustomed to the experimenter. Similarly, animals should be tested in exactly the same environment for each trial. The best way of placing the guinea pig on the platform in the supine position is to support it with both hands under the neck and sacrum.

One possible source of measurement error is a delay between the animal being placed on the weighing platform and the activation of the timer by a second person. However, with practice, any delay in activating the timer can be reduced to normal human reaction time for a simple button-press task (approx. 200 ms) and therefore would have negligible effect on the measurement of larger differences in RRL (see Fig. 1).

In drug trials, we test RRL twice: before and after injection of the drug or vehicle (the exact time following the injection depending upon the half-life of the drug). The change in RRL is calculated by subtracting preinjection latency from postinjection latency. We recently tested the device described in a study of the effects of ethanol and the *N*-methyl-D-aspartate (NMDA) channel antagonist (+)-5-methyl-10,11-dihydro-5*H*-dibenzo [*a*,*d*] cyclohepten-5,10-imine maleate (MK-801) on RRL in guinea pigs. Four conditions were used in which RRL was tested daily for 10 days: a) no injection (n = 4); b) saline (vehicle) injection (2 ml/kg, IP) (n = 3); c) ethanol injection (0.64 g/kg dissolved in 2 ml/kg saline, IP) (n = 4); d) ethanol + MK-801 injection (0.64 g/kg ethanol + 0.5 mg/kg MK-801, dissolved in 2 ml/kg saline, IP) (n = 4). In the latter three conditions, RRL was calculated as postinjection latency (obtained approximately 0.5 h following drug or vehicle injection) minus a preinjection latency (obtained approximately 0.5 h before drug or vehicle injection). An upper limit of 300 s was set for measurement of RRL. Average RRL was calculated for animals in each condition on each day and a mean RRL for each group was calculated. Two-way analysis of variance (ANOVA) with repeated measures was performed on the group means; the α rate was set at 0.05. At the dose used, ethanol had no significant effect on the RRL; however, ethanol + MK-801 significantly increased RRL relative to the ethanol condition, $F(1, 5) = 83.16$, $p < 0.0005$; there was no evidence of tolerance in any condition (see Fig. 1). In the group of animals that received the ethanol + MK-801 injections for 10 days, a single injection of ethanol alone (same dose) on the eleventh day resulted in a significantly shorter mean RRL compared to day 10 [paired two-tailed student's t-test, $t(3) = 4.36$, $p < 0.05$] (see Fig. 1). These results indi-

cate that the injection of ethanol and MK-801 together can increase RRL more than ethanol alone and that the device described above is useful in quantifying differences of this magnitude.

SUMMARY

We described a simple, inexpensive device and a measurement protocol that are useful for quantifying the effects of drugs on the RRL as an index of their effects on motor behavior. This method of quantification of drug effects on the righting reflex may be particularly useful in studies of behavioral drug tolerance and withdrawal, where it is often desirable to quantify gradual changes in a drug effect over a number of days or weeks and then subject the data to analysis with parametric statistics such as ANOVA.

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REFERENCES

1. Dolin, S. J.; Little, H. J. Are changes in neuronal calcium channels involved in ethanol tolerance? J. Pharmacol. Exp. Ther. 250:985–991; 1989.
2. Pellis, S. M.; Pellis, V. C.; Chen, Y -C.; Barzci, S.; Teitelbaum, P. Recovery from axial apraxia in the lateral hypothalamic labyrinthectomized rat reveals three elements of contact-righting: Cephalocaudal dominance, axial rotation, and distal limb action. Behav. Brain Res. 35:241–251; 1989.
3. Wenger, J. R.; Tiffany, T. M.; Bombardier, C.; Nicholls, K.; Woods, S. C. Ethanol tolerance in the rat is learned. Science 213: 575–577; 1981.
4. Wood, J. M.; Laverty, R. Metabolic and pharmacodynamic tolerance to ethanol in rats. Pharmacol. Biochem. Behav. 10:871–874; 1979.