

BRIEF COMMUNICATION

Improved Automated Apparatus for Recording Rotation (Circling Behavior) in Rats or Mice¹

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(Received 7 June 1974)

GREENSTEIN, S. AND S. D. GLICK. *Improved automated apparatus for recording rotation (circling behavior) in rats or mice.* PHARMAC. BIOCHEM. BEHAV. 3(3) 507-510, 1975. — An improved automated apparatus for measuring rotation (circling behavior) is described. The apparatus may be designed for either mice or rats. The logic circuit differentiates between complete 360° rotations and incomplete oscillatory turns; preliminary data indicate that this distinction has functional significance.

Rotation	Circling behavior	Caudate nucleus	Amphetamine
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RATS or mice with unilateral lesions of the nigro-striatal system rotate or turn in circles following the administration of drugs known to affect dopamine metabolism (e.g. [2,6]). Because a specific relationship between rotation and the nigro-striatal system has been established [3], rotation is now commonly employed as a behavioral assay of nigro-striatal function. Apparatuses, for rats, designed to record rotation automatically have been described previously [1,6]. However, such apparatuses have not adequately distinguished complete 360° rotations from incomplete oscillatory turns [6]; the latter may introduce considerable artifact into the recorded results. The present apparatus distinguishes between rotations and oscillations, differentiates between rotations and oscillations in different directions and may be designed, with minor differences, for either rats or mice.

METHOD

Apparatus and Logic Circuit

The apparatus, for mice, consists of two 8 in. (20.3 cm) (12 in. or 30.5 cm for rats) Plexiglas hemispheres (commercially available lamp shades) used as upper and lower pieces. The lower hemisphere is glued to a 3 in. (7.6 cm) length of 4 in. (10.2 cm) (8 in. or 20.3 cm for rats) diameter cylindrical Plexiglas which serves as a base. Three 1 in. (2.5 cm) by 1/2 in. (1/3 cm) by 1/8 in. (3.2 mm) Plexiglas strips are

evenly spaced and bolted to the top edge of the hemisphere acting as guides on which the upper hemisphere is placed. The position sensing device consisting of two Plexiglas discs is located on top of the upper hemisphere. The lower disc measuring 2 1/2 in. (6.4 cm) in diameter by 1/4 in. (6.4 mm) thickness is fastened to the upper hemisphere. Four photocells (Clairex No. C19031) are mounted 3/4 in. (1.9 cm) from the center of the disc and 90 degrees apart, face flush with the top of the disc. A ball bearing (bore = 0.375 in., O.D. = 0.875 in., PIC Design Corp. No. E 1-14) is mounted in the center of the disc. The upper disc measuring 2 1/4 in. (5.7 cm) in diameter by 1/8 in. (3.2 mm) thick is mounted 1/8 in. (3.2 mm) above the lower disc on a shaft fitted in the ball bearing. A single 1/8 in. (3.2 mm) diameter hole is drilled 3/4 in. (1.9 cm) from the center of the upper disc. A light source is mounted above the upper disc, such that, as the upper disc revolves 360 degrees, it sequentially illuminates the four photocells.

A mouse (or rat) harness is fabricated from 3-0 stainless steel wire sheathed in silicone tubing and formed into a slip-loop which can be tightened behind the animal's forelegs. The silicone tubing inhibits slippage. The other end of the wire is terminated with a button magnet. A ferrous washer is screwed to the bottom of the center shaft which projects 1 in. into the hemisphere. Once the animal is secured in the harness and placed in the lower hemisphere the magnet is placed on the ferrous washer on the center

¹ This research was supported by NIMH grant MH25644 and NIMH Research Scientist Development Award (Type 2) DA70082 to S. D. Glick.

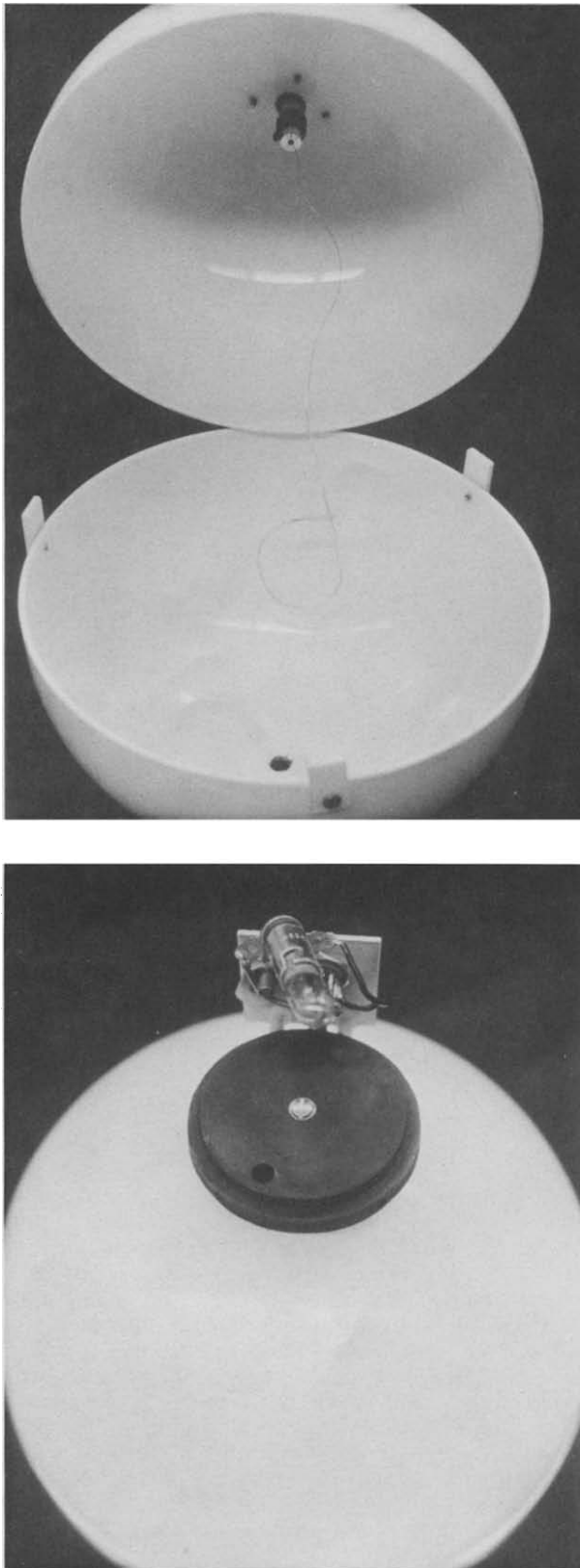


FIG. 1. Photographs of rotometer: (a) inside view with harness attached, (b) photocell sensing device on top.

shaft. As the animal rotates, the upper disc follows. The entire apparatus is shown in Fig. 1.

The four photocells effectively divide the hemisphere into 4 areas or quadrants. When the animal enters a quadrant (causing a photocell to be illuminated), a flip-flop representing that quadrant is set and, in turn, triggers a one shot (Tech Serv Digibit logic modules). If a flip-flop representing a quadrant to the immediate left or right was also set (i.e. the animal was just in that quadrant), a gating network would register a quarter turn count for the respective direction, pulse a 4 count shift register for the respective direction, and reset all quadrant flip-flops except the one just entered. This operation repeats every time the animal enters a new quadrant. If the animal enters 4 quadrants sequentially in the same direction (i.e. moves 360°), the 4 count shift register will enter a full turn count for the respective direction and reset to 0. If, however, an animal enters a quadrant from the opposite direction of the previous move, the shift register of the previous direction is reset to 0 and the shift register of the new direction counts 1. The resulting output is quarter turns left, quarter turns right, full turns left, and full turns right. The circuit is diagrammed in Fig. 2.

Application

A pilot experiment demonstrated that the distinction between full and quarter turns is, perhaps, more functionally important than was originally thought. Nine naive female CF1 mice received unilateral electrolytic lesions (2 mA for 5–10 sec) of the caudate nucleus using a previously described stereotaxic method [5]. One week after surgery, each mouse was administered 5.0 mg/kg of d-amphetamine sulphate 15 min before being tested in the apparatus for 15 min. After testing, all mice were killed and perfused with 10% Formalin, their brains were removed, immersed in Formalin and subsequently sectioned ($40\ \mu$) and stained (Luxol blue and cresyl violet) for histological study. All lesions were centrally located in the anterior caudate nucleus [4] but varied in size (1–2½ mm in diameter) as intended.

RESULTS AND DISCUSSION

Table 1 shows all data recorded for the nine mice. Also note that extra quarter turns were computed; that is, if all quarter turns were parts of full turns, then dividing quarter turns by 4 should produce the same numbers as the recorded full turns. This was never the case; extra quarter turns are therefore the result of subtracting 4 times the number of full turns from the number of quarter turns. It should be noted that most full turns were always made in the direction ipsilateral to the lesion, as should be the case [2,6]. Extra quarter turns, however, were generally distributed equally to the left and right. To possibly determine the significance of full turns vs. extra quarter turns, regression correlations of lesion size with turning behavior were performed (Table 2). Lesion size was correlated with net rotations (ipsilateral full turns minus contralateral full turns; this is the measure of rotation usually employed in other studies), total (ipsilateral plus contralateral) full turns, net quarter turns, total quarter turns and total extra quarter turns. As shown in Table 2, only the inverse correlation of extra quarter turns with lesion size was significant. In other words, the elimination of extra quarter turns by

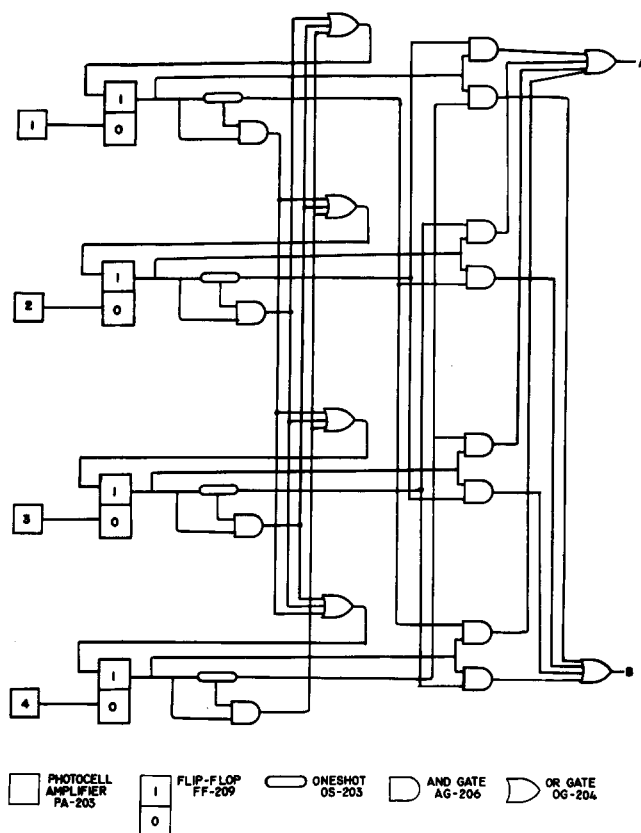


FIG. 2. Circuit diagram for distinguishing left and right turns. The four photocells on the sensor are numbered clockwise and are connected to the four photocell amplifiers, respectively. Output A generates "right $\frac{1}{4}$ turns" which go to a counter and a 4 count shift register. Output B is the same for "left $\frac{1}{4}$ turns." The shift registers count full turns.

TABLE 1
EFFECT OF UNILATERAL CAUDATE LESIONS ON TURNING BEHAVIOR

Mouse No.	Lesion Diameter (mm)	L $\frac{1}{4}$ Turns	R $\frac{1}{4}$ Turns	L Full	R Full	L Extra	R Extra
1-L*	2.0	563	62	139	13	11	10
2-L	2.2	542	103	129	22	26	31
3-R	2.0	82	268	15	61	22	24
4-R	2.2	19	564	3	140	7	4
5-L	1.8	486	145	115	27	26	37
6-R	1.2	282	450	56	100	58	50
7-L	2.5	551	126	135	29	11	10
8-R	1.5	184	610	30	136	64	66
9-L	1.0	490	232	104	40	74	72

*side of lesion: L = left, R = right

TABLE 2
CORRELATION OF TURNING BEHAVIOR AND LESION SIZE*

Lesion Diameter vs	
Net Full Turns (i.e. rotation)	$r = 0.59$
Total Full Turns	$r = -0.09$
Net ¼ Turns	$r = 0.60$
Total ¼ Turns	$r = -0.44$
Total Extra ¼ Turns	$r = -0.90^*$

* $p < 0.001$

the lesion seemed to be the most sensitive index of lesion size. With a larger lesion, the animal's behavior was less random. In subsequent work (Glick and Greenstein, in preparation), we found that extra quarter turns were positively correlated and net rotations were negatively correlated with spontaneous locomotor activity when mice tested for rotation were retested a week later in an open field apparatus (photocell box). Thus the present apparatus and logic circuit allow one to differentiate between two very different behaviors of possibly very different functional significance.

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