



Research report

Arena geometry and path shape: When rats travel in straight or in circuitous paths?

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ABSTRACT

We show here that the global geometry of the environment affects the shape of the paths of travel in rats. To examine this, individual rats were introduced into an unfamiliar arena. One group of rats ($n=8$) was tested in a square arena ($2\text{ m} \times 2\text{ m}$), and the other group ($n=8$) in a round arena (2 m diameter). Testing was in a total darkness, since in the absence of visual information the geometry is not perceived immediately and the extraction of environment shape is slower. We found that while the level of the rats' activity did not seem to differ between both arenas, path shape differed significantly. When traveling along the perimeter, path shape basically followed the arena walls, with perimeter paths curving along the walls of the round arena, while being straight along the walls of the square arena. A similar impact of arena geometry was observed for travel away from the arena walls. Indeed, when the rats abandoned the arena walls to crosscut through the center of the arena, their center paths were circuitous in the round arena and relatively straight in the square arena. We suggest that the shapes of these paths are exploited for the same spatial task: returning back to a familiar location in the unsighted environment.

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1. Introduction

When humans and other animals travel from one place to another, or when traveling in a certain direction, the shortest path is not necessarily taken. Instead, travelers may veer toward salient landmarks [1], follow the boundaries of the area [2–7], or focus on security, on gathering information, or on visiting memorized locations [8]. For this, they rely on external and internal cues that are integrated into a spatial representation [9–14]. This environment representation enables path-planning, based on identifying origin, destination, directions, and distances [15]. The above processes have been revealed in various experimental environments, and one such environment is the open-field [16–18]. There are numerous versions of the open-field: for example, with or without walls, with or without objects inside it and, most commonly, a round or square open-field. This latter variation of a round opposed to a square open-field was employed in the present study, which was aimed at determining whether the different geometries of the apparatus influence the shape of the paths of spontaneous travel.

In a square open-field, rats and mice tend to spend extended periods at the corners [19–25], and therefore, the round open-field arena has been preferred by many experimenters due to the

absence of corners [26–31]. Previous studies that compared behavior in square versus round arenas have suggested that, in terms of the amount of activity, arena shape does not seem to be a crucial parameter, and that animals can be equally tested using either square or round arenas [32–34]. Similarly, in reviewing the literature on open-field behavior, it was suggested that there are no data available on the impact of open-field geometry on path shape [18]. However, spatial locomotor behavior is multi-dimensional, and thus needs to be studied using at least two independent dimensions: (i) the amount of activity; and (ii) the spatial patterns [24]. Accordingly, findings regarding a similar level of activity in round and square arenas do not contribute in determining spatial patterns, which may be similar or different in a square versus round arena.

The lack of information on the effect of arena shape on the trajectories of travel paths is surprising, considering that the global geometry of the environment has been suggested as comprising the key for orientation and navigation [35–38]; see however [39,40]. Moreover, the global geometry also has an impact on how the environment is encoded in the brain. Specifically, there are special neurons, termed place cells, which fire when the animal is in a specific area of the environment that is termed 'place field' [41–44]. It was found that for rats tested in a square arena, place fields were round, whereas for rats tested in a round arena, place fields had a crescent shape [45]. Accordingly, it was suggested that place cells are sensitive to the surface orientation of the arena walls [46,47]. It was also suggested that there are 'border cells', neurons that are

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sensitive to the periphery of the arena, and that these cells could be instrumental in planning trajectories and anchoring place fields to a geometric reference frame [48]. In light of the above studies, the aim of the present study was to determine whether round and square arenas differentially shape the paths of spontaneous travel in the arena.

2. Materials and methods

2.1. Animals

Male Wistar rats ($n=16$; age 3 months; weight 300–400 g) were housed in groups of 3–4 individuals in metal cages (60 cm \times 40 cm \times 25 cm) with sawdust bedding, in a temperature-controlled room (21 °C) under 12/12 dark/light cycle (lights on: 7 p.m.). Water and standard rodent chow were provided ad libitum. Rats were acclimated to handling for 10 min/day during the 5 days before testing.

2.2. Apparatus

Testing was carried out in the two following open-field arenas: (1) a square arena (200 cm \times 200 cm), enclosed with 50 cm-high opaque Plexiglas walls; and (2) a round arena (200 cm diameter), enclosed with a 50 cm high tin wall. The arenas were placed in a temperature-controlled room (21 °C). During testing, the room was in complete darkness, illuminated only with an infra-red light source, with an 830 nm filter that emits light not visible to the rats (Tracksys, UK). A video camera (Ikegami B/W ICD-47E, Japan) was placed 2.5 m above the center of the arena, providing a top view that was recorded onto a DVD (Sony 169GB).

2.3. Procedure

Rats were randomly assigned to one of two groups ($n=8$ each), and each group was tested in only one of the arenas. Testing took place during the dark period of the dark/light cycle since, as rats are nocturnal, their activity is higher and they tend to enter the arena center at this time. The test began when a rat was placed at a fixed start point near the arena wall (in the square arena at the same arena corner), facing the center of the arena. Each rat was tested only once, while its behavior was video-recorded for 20 min, after which it was returned to its cage. After each test, the arena was paper-wiped with detergent.

2.4. Data acquisition and analysis

Spatial behavior of the rats was analyzed using 'Ethovision' (Noldus Information Technologies, NL), which tracks progression in the arena, providing the time and location of the center of mass of the rat five times per second. For analysis, both arenas were divided into a perimeter area, which was a 15 cm wide strip along the arena wall, while the rest of the area was considered as the center. The following parameters were extracted from 'Ethovision' and exported to 'Microsoft Excel' for further analyses:

1. *Traveled distance*: the metric distance (m) traveled during the 20 min of observation.
2. *Velocity*: the speed of progression (cm/s) during the 20 min of observation.
3. *Duration at area*: the time spent at the perimeter or in the center areas.
4. *Short crosscuts*: departure from the perimeter area and travel into the center, followed by return to the perimeter at a distance no greater than a quarter of the perimeter length (200 cm in the square arena, 160 cm in the round arena).

5. *Long crosscuts*: departure from the perimeter area and travel into the center, followed by return to the perimeter at a distance greater than a quarter of the perimeter length (100 cm in the square arena, 80 cm in the round arena).
6. *Number of short and long crosscuts*: the frequency of long and short crosscuts during the 20 min of observation.

In addition, we measured for each crosscut the turn angle, which is the amplitude of the change in the direction of progression during the crosscut (Fig. 1). This was measured on-screen using the *Image-J* software (freeware by NIH).

2.5. Statistics

A *t*-test for independent samples and a two-way ANOVA with repeated measures were used to compare behavior between the square and round data. A chi-square test was used to compare the incidence of crosscuts between the square and round arena. Alpha level was set to 0.05, and a Bonferroni correction was applied for relatively dependent data.

3. Results

In terms of activity parameters, rats in the round and square arena did not differ in the total distance traveled and the mean velocity of travel. In the round arena, they traveled more along the arena wall and remained there for a longer duration compared with the square arena. However, as the various parameters of activity are not mutually independent, the level of significance was therefore set to 0.007 (after Bonferroni correction); and, accordingly, no significant difference was found between the amount of activity of the rats in the round compared with the square arena (Table 1).

Paths of travel in the round compared with the square arena are provided for four exemplary rats in Fig. 2. It is apparent that when the rats abandoned the arena walls and traveled through the center, the paths were relatively straight in the square arena and circuitous in the round arena.

To quantify the difference, as shown in Fig. 2, we extracted for each rat all the paths along which it moved at a distance of 15 cm or more away from the arena walls. As detailed in Section 2, we classified these sections, termed crosscuts, into short crosscuts, which were confined to a quarter of the arena, and long crosscuts, which extended into remote quadrants. A two-way ANOVA with repeated measures revealed a significant difference between the square and the round arena in the number of crosscuts (group effect; $F_{1,14}=12.1$; $p=0.004$); there was also a significant difference between the number of long and short crosscuts ($F_{1,14}=26.2$; $p<0.0001$). However, the interaction between arena shape and crosscut length was not significant ($F_{1,14}=0.05$; $p=0.8$). A Tukey HSD test revealed that in both the round and the square arena, the number of short crosscuts was significantly higher than the number of long crosscuts (Fig. 3a).

As seen in Fig. 2, crosscuts in the square arena were along relatively straight trajectories, whereas in the round arena the trajec-

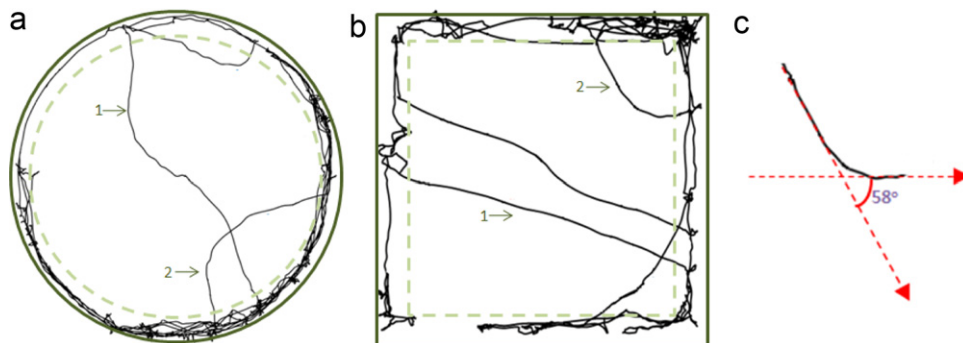


Fig. 1. Definition of crosscuts and turning angles. The paths of progression during a short bout of activity are depicted for the round arena (a) and the square arena (b). In each, the outer solid line represents the arena walls and the inner dashed line represents the border between the perimeter and center zones. Crosscuts are the paths within the center, and they could be long (marked by 1→) or short (marked by 2→). The turn angle was measured between the imaginary straight lines attached along the beginning and the end of the crosscut path (c).

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