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Research report

Enrichment-induced exercise to quantify the effect of different housing conditions: A tool to standardize enriched environment protocols

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HIGHLIGHTS

- A direct correlation exists between EE and EE-induced exercise.
- Enrichment-induced exercise positively correlates with the degree of enrichment.
- Enrichment-induced exercise could be used to standardize EE protocols.

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ABSTRACT

Enriched environments (EE) have been used for a long time to promote recovery in many neurological disorders, however, a growing body of inconsistent results strongly calls for a rigorous standardization of experimental EE paradigms. Although some core principles are well accepted as standards, a method to quantitatively assess the complexity of EE in various experimental designs is still lacking. In this study, we tracked and recorded the physical exercise of rats in four housing conditions, namely isolated condition, social condition, novel condition and EE. Then, we analyzed whether and to what extent, enrichment-induced exercise reflected the degree of enrichment. We next examined rat exercise in a conventional environment condition and under different light intensities, to explore whether environment-related exercise could be considered a parameter to quantify the degree of enrichment. The results obtained showed that (1) both inanimate and social stimulations enhanced the exercise level and (2) EE combined the effects of the two stimulations. Furthermore, exercise durability which correlated positively with degree of enrichment, was an objective measure of different housing conditions. Exercise-related parameters also sensitively reflected the impacts of light intensity even in the same enrichment arrangements. Our results indicate that there is a direct and measurable correlation between degree of environmental enrichment and enrichment-induced exercise, and therefore enrichment-induced exercise could be used as a helpful tool to evaluate the degree of housing conditions and to standardize the EE protocols.

1. Introduction

An enriched environment (EE) is classically defined as 'a combination of complex inanimate and social stimulations' [1]. It is supposed to facilitate enhanced sensory, cognitive, motor and social stimulations compared to standard laboratory housing conditions [2]. Many studies have demonstrated that environmental stimulation elicits several positive effects in the nervous system at molecular, anatomical and functional levels [2–4], such as increased hippocampal neurogenesis [3,5], enhancement in learning and memory [3,6,7] and induction of neural plasticity [8,9]. However, despite this largely positive body of evidence, non-significant or even negative effects are also reported [10]. For example, animals exposed to enriched environments seem to display increased anxiety compared to those exposed to standard conditions [11,12]. In addition EE have been reported (i) to either

Abbreviations: EE, enriched environment; IC, isolated condition; SC, social condition; NC, novel condition; SHC, standard housing condition; CNS, central nervous system.

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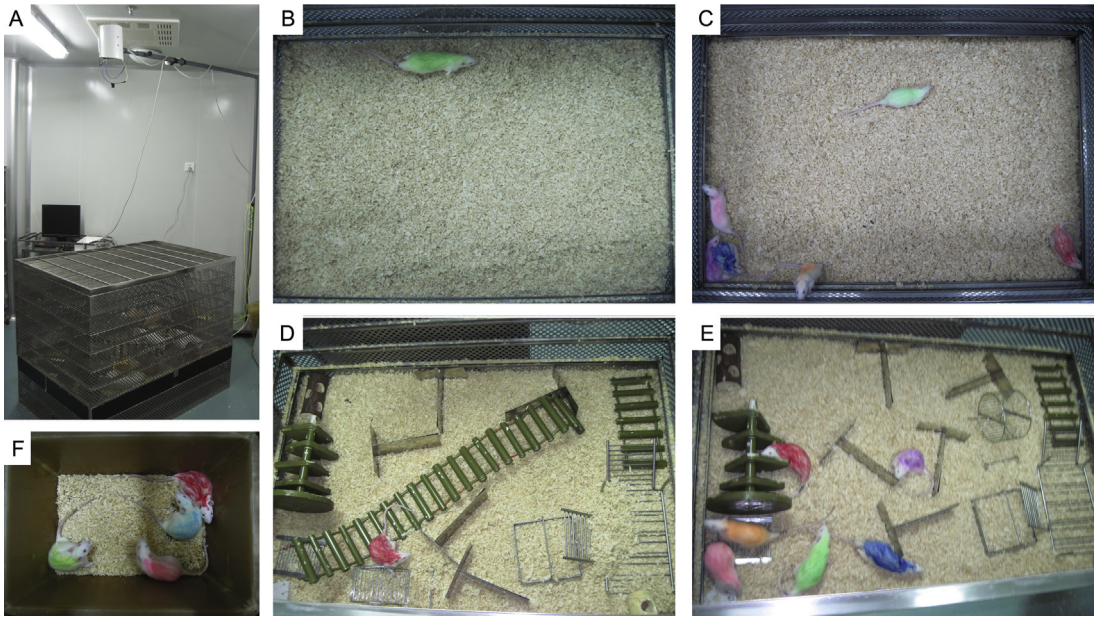

Fig. 1. Experimental set up and housing conditions used in the study. (A) Exercise testing and monitoring device. Typical rat/rats tested and tracked in: (B) isolated condition; (C) social condition; (D) novel condition; (E) enriched environment; (F) standard housing condition.

have no effect on corticosterone levels [13] or to increase them [11,14]; (ii) to enhance the development of amyloid plaques, in Alzheimer's models [2]; (iii) to induce stronger drug preference compared to control animals [15]; and (iv) to fail to reduce infarct volume of rats following cerebral ischemia [16–18]. Overall, the reason for such complex and sometimes inconsistent results could be the wide variety of EE protocols that may be difficult to compare each other [19]. Thus, increasing attention is being given to efforts aimed at standardizing enrichment protocols in order to obtain more consistent results. In this respect, a major obstacle is represented by the lack of a method to measure the extent of enrichment in various protocols.

Physical exercise has been proposed as a critical component of enrichment [20]. This is supported by the observation that exercise and enrichment share similar structural and behavioral effects [3]. For example, both can increase adult hippocampal neurogenesis and improve spatial learning ability [5,21]. However, evidence to prove a direct relation between enrichment-borne inanimate and social stimulations and enrichment-induced exercise is still lacking. Furthermore, whether enrichment-induced exercise could reflect or even accurately quantify the degree of enrichment is largely unknown.

In this study, we tested whether a correlation exists between enriched stimulation and enrichment-induced exercise, and whether the enrichment-induced exercise could be used as a tool to quantify the degree of enrichment in a variety of housing conditions.

2. Experimental procedures

2.1. Animals

All procedures involving animals were performed in accordance with the Animal Care and Use Committee of Fudan University and every effort was made to minimize the number of animals. Two month old male Sprague-Dawley rats were kept under 12 h light/12 h dark cycles (07:00 lights on) with free access to food and water. All experiments were conducted during the light period. The rats were initially placed in the test arena for a two week habituation phase prior to experiments. For exercise tracking experiments, the rats were marked up with non-toxic colored ink to allow for the computer imaging recognition from the videotapes. The color was applied to the back of the body and covered approximately 60% of the body (Fig. 1).

2.2. Experimental conditions and tracking system

The testing lab was illuminated with eight 36 W fluorescent lamps during the experiments to ensure all tests, except for the light intensity test, were implemented under the same light intensity. An infrared video camera was used to record the animal's physical exercise during the experiment (Fig. 1A). The level of voluntary exercise was observed and recorded using an automated tracking system (EthoVision XT 7.0, Noldus Information Technology, Fig. 2).

2.3. Rat selection

Rats were placed in standard cages (40 cm × 30 cm × 20 cm) and housed in groups of four per cage (Fig. 1F). We tracked their physical exercise 6 h per day for one week. We compared the exercise level of 80 rats and selected 48 of them, who did not show significant differences in this performance, to be included in the subsequent experiments.

2.4. Relation between exercise and enrichment test

For this test (Fig. 1B–E) colored rats were assigned by random number table to one of the following groups (n = 12/group): isolated condition (IC), social condition (SC), novel condition (NC) or EE. IC rats were housed one per cage; SC rats were housed in groups of six per cage; NC rats were housed one per cage in an experimental cage containing a variety of objects (e.g., boxes, chains, metal barrels, ladders, etc.) which were changed daily; EE rats were housed is groups of 6 per cage with a variety of objects which were also changed daily. The tracking manipulations were administered 2 h per day (09:00–11:00) for 28 days and they were carried out in a cage of the same size (120 cm × 80 cm × 100 cm) to avoid the influence of a spacial factor. Changing the wood shavings between tests ensured that any residual odor of alcohol was eliminated before these animals were introduced into the test apparatus again the next day. After each test, the color was washed out and the rat was dried and returned to its home cage.

2.5. Conventional EE paradigm test

Rats were assigned by random number table to either the enriched environment (EE, n = 12) group or the standard housing condition (SHC, n = 12) group. In the EE group, rats were housed in the environment described above (Fig. 1E). In the SHC group, they were housed in a standard cage devoid of objects (40 cm × 30 cm × 20 cm, four rats in each cage, Fig. 1F). Also this manipulation was administered 2 h per day, for 28 days.

2.6. Light intensity test

Rats were randomly assigned to one of the following four groups (n = 6/group): enriched environment plus strong light (EESL), enriched environment plus dim light (EEDL), isolated condition plus strong light (ICSL) and isolated condition plus dim