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MICRO REPORT

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Open source code for behavior analysis in rodents

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Abstract

Aim: We have conducted a series of behavioral tests, which cover a broad range of behavioral domains, on various strains of genetically engineered mice. For the behavioral screening, we have been using Image J plugins that we developed for most of the tests in the battery. Our behavioral analysis system with the plugins enables systematic and automated image analysis of behavior. The plugins are freely available on the "Mouse Phenotype Database" website (http://www.mouse-phenotype.org/software.html). Here, we release the source code of the plugins in a Git repository with the aim of promoting their use and expanding their functionality.

Methods: We published the source code of the Image J plugins for behavioral analysis at Git repository (https://github.com/neuroinformatics). The source code for light/dark transition, elevated plus maze, open filed, T-maze, and fear conditioning tests was made publicly available in the repository.

Conclusions: The source code of the plugins for the behavioral tests as well as the pre-compiled binaries can be freely obtained. The open source code could promote the development and modification of the plugins for additional behavioral indices in these tests and for other behavioral tests.

KEYWORDS

comprehensive behavioral test battery, Image J, open source code

We have assessed behavioral phenotypes of more than 190 strains of mutant mice using a comprehensive test battery, which includes sensori-motor functions, emotion, learning and memory.[1,2] We have been using Image J plugins to analyze the behavioral data that were obtained in those studies. The plugins are based on the public domain Image J program (https://imagej.nih.gov/ij/index.html),[3] which is designed with an open architecture. Data acquisition, analysis, and processing can be customized by Java plugins. We developed the Image J plugins, and the pre-compiled plugins for light/dark transition test[4,5] (Image LD), elevated plus maze[6,7] (Image EP), open field test (Image OF), fear conditioning test[8,9] (Image FZ), and T-maze[10,11] (Image TM) are freely available on the website of "Mouse Phenotype Database" (http://www.mouse-phenotype.org/software.html). The automated data acquisition and analysis provide high-throughput quantification of animal behaviors and are useful in minimizing artifactual interpretations of behavioral data and reducing inter-observer variations and inter-subject variability. We have published video demonstrations, protocols, and results for the behavioral tests with the Image J plugins. The purposes and functionalities of each plugin, and references are provided in Table 1.

Here, we released the source code of the Image J plugins that we developed for the five behavioral tests, as mentioned above, under GNU

TABLE 1 Reference lists for behavior analysis with Image J plugins

Behavioral test	Characteristic behavior	Video demonstrations and detailed protocols	References
Light/dark transition test	Anxiety-like behavior (Crawley and Foodwin[4])	Takao and Miyakawa[1]	Sakai et al[20]
Elevated plus maze	Anxiety-like behavior (Handley and Mithani[6])	Komada et al[7]	Ohashi et al[19]
T-maze	Working and reference memory (Hepler et al[10])	Shoji et al[11]	Imai et al[18]
Fear conditioning test	Associative fear learning and memory (Fanselow[8])	Shoji et al[9]	Fujita et al[16]
			Hayashi et al[15]
			Umemura et al[14]
			Okuda et al[13]
			Maeta et al[12]
			Kanetake et al[17]

GPLv3 in the Git repository (https://github.com/neuroinformatics). The Image J plugins are written in Java, and are easily extendable. After installing the Java SE Development Kit (URL) and software package management system, the JAR file can be created by Apache Maven (https://maven.apache.org). Detailed instructions for compiling the plugins are described in a "readme.md" file available in the Git repository.

The hardware components of the behavioral analysis with the Image J plugins include a camera with a USB video capture device (compatible with JMF) or a USB web camera, and a Windows 7 or 10 (32-bit) computer. The camera should be mounted above the apparatus to monitor mouse behavior so that images of the apparatus and the mouse are captured by the computer. The usage instructions for each test are described in the "readme.text" files, which are downloadable with the pre-compiled plugins from the "Mouse Phenotype Database" website.

Our behavioral analysis system with Image J plugins consists of several simple components. The plugins convert recorded images into binary images using a user-selected brightness threshold and detect a mouse as a particle by background subtraction. The image processing is also applicable to behavioral analysis of rats and the other animals. The free availability of the plugins encourages the improvement and customization of the function of the plugins, thus potentially addressing new challenges in behavioral analysis. We have also developed the plugins for other behavioral tests, including home cage monitoring, social interaction, three-chamber social approach, Porsolt forced swim, tail suspension, and Barnes maze tests. Additional behavioral indices, such as the number of instances of circling and stretching, will be available as pre-compiled plugins and as open source code in the near future. These sets of open source code could further contribute to the development of novel methodology for behavioral analyses.

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CONFLICT OF INTEREST

The authors declare no conflict of interest.

AUTHOR CONTRIBUTIONS

Software development: KT and TM; Technical supports and discussions for open source code: SH, YO, KT, and YY; Manuscript preparation: SH and TM.

DATA REPOSITORY

We released the source code of the Image J plugins in the Git repository (https://github.com/neuroinformatics).

APPROVAL OF THE RESEARCH PROTOCOL BY AN INSTITUTIONAL REVIEWER BOARD

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INFORMED CONSENT

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REGISTRY AND THE REGISTRATION NO. OF THE STUDY/TRIAL

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