Behavioural Brain Research 191 (2008) 104–110

Contents lists available at ScienceDirect

Behavioural Brain Research

journal homepage: www.elsevier.com/locate/bbr

Research report

Effects of the genetic background on cognitive performances of TG2576 mice

Jean Michel Lassalle[a,b,*], Hélène Halley[a,b], Stéphanie Daumas[a,b,1], Laure Verret[a,b], Bernard Francés[a,b]

[a] Université de Toulouse, France
[b] CNRS, UMR 5169, Centre de Recherches sur la Cognition Animale, France

ARTICLE INFO

Article history:
Received 19 February 2008
Accepted 13 March 2008
Available online 21 March 2008

Keywords:
Tg2576
Transgenic mouse
Genetic background
Elevated plus maze
Morris navigation task
Contextual and cued fear conditioning

ABSTRACT

Animal models of genetic diseases obtained by transferring human mutated genes in the mouse are widely used in biomedical based research. They constitute efficient tools to study mechanisms underlying abnormal phenotypes. Unfortunately, the phenotype of the transgene is often obscured by the genetic background of the embryonic stem cells and that of the recipient strain used to create the transgenic line. It is also known, from the literature, that repeatedly backcrossing a transgenic strain to an inbred background may have unfavorable effects that can result in the loss of the transgenic line. In order to analyze the influences of the genetic background on the transgene expression, we studied the effects of the hAPPswe transgene involved in Alzheimer's Amyloid Pathology, in 3 genetic backgrounds differing by their genetic heterogeneity (homozygous vs heterozygous) and the strain of origin (C57BL6, CBA, B6SJL F1) after only one generation backcrossing. Three different behavioral paradigms were used to assess the psychological and cognitive phenotypic differences: elevated plus maze, morris navigation task and contextual fear conditioning. Our data indicate that the best solution to maintain the transgenic line is to backcross repeatedly the transgenic mice into the F1 hybrid cross that was used to create the transgenic strain, whereas phenotyping should be performed comparatively after only one generation backcrossing into various well chosen F1 or inbred backgrounds.

© 2008 Elsevier B.V. All rights reserved.

1. Introduction

In the last decade, animal models of genetic diseases were developed by transferring human mutated genes from families, in which the disease was inherited, to recipient mice [6,14,18,32]. One of the most successful transgenic murine models of Amyloid Pathology is the Tg(HuAPP695-SWE)2576 mouse developed by Hsiao et al. [16] and maintained by repeated backcrossing to B6 × SJL F1 hybrid. The behavioral, cognitive and neurophysiological phenotypes of this model have been extensively studied in this background [5,7,16,24,28,37]. However, the confusing influence of the genetic background on the expression of the transgene has been repeatedly pointed out in genetically modified mice [13,17,21,22,29–31] and has evolved into a widespread concern in mouse-based biomedical research [19,20,36].

In order to avoid confusing the phenotype of the transgene or the modified gene, and the phenotype of the lines they stem from [8,12,26,27], it has been proposed to backcross the transgenic line to an inbred strain [11,25]. Some authors [34,35] have proposed to follow the strategy that emerged as a consensus from the Branbury-Conference [2] that mutations should be maintained by 3 repeated backcrosses to at least 2 inbred strains while phenotypic characterization should be performed in F1 hybrids resulting from the cross between the 2 congenic lines. However, unexpectedly, repeated backcrosses of a transgene into an inbred background can have unfavorable effects such as inbreeding depression or increased sensitivity to aging, resulting in performance impairments that may preclude conclusive evidence of the deleterious effect of the transgene. It may also happen that backcrossing the transgene to a particular strain increases its toxicity, resulting in the loss of the transgenic line. That is what happened to the hAPP695 transgene which is lethal in inbred FVB/N [15] and B6 mice after only 3–4 backcrossing generations [4]. Thus, inserting the transgene in various genetic backgrounds would allow to study different modes of regulation of the transgene and its functions and help to reveal a phenotype that would have been obscured in a different background [4,23], improving the power of mutant models of human disorders [3,9]. Therefore, the aim of our study has been to analyze the effects of the genetic background on the expression of the hSWE

* Corresponding author at: Centre de Recherches sur la Cognition Animale, Université Paul Sabatier, Bât IVR3 b3, 118 route de Narbonne, 31062 Toulouse Cedex 4, France. Tel.: +33 561 556 170; fax: +33 561 556 154.
E-mail address: lassalle@cict.fr (J.M. Lassalle).
[1] Current address: Laboratory for Cognitive Neuroscience, Centre for Cognitive and Neural Systems, Division of Neuroscience, The University of Edinburgh, 1 George square, Edinburgh EH8 9JZ, UK.

0166-4328/$ – see front matter © 2008 Elsevier B.V. All rights reserved.
doi:10.1016/j.bbr.2008.03.017

APP695 transgene after only one generation backcrossing, in order to avoid increasing the lethality of the transgene to either the CBA/J or the C57BL/6J inbred backgrounds, or to the B6 × SJL F1 isogenic hybrid background.

Phenotypic differences between the backgrounds have been checked by comparing inbred C57BL/6, CBA and B6SJL F1 hybrid mice. Behavioral and cognitive performances of these mice have been assessed by comparing Tg+ and Tg− from the same litter, using three experimental paradigms. The elevated plus maze allowed to evaluate locomotor activity, anxiety reactions and disinhibition. The Morris navigation task was used to compare the acquisition, as well as short-term retention of a hippocampo-dependant spatial memory with distributed learning whereas the contextual fear conditioning measured the long-term retention of an episodic-like memory acquired during a single training session. We choose the B6SJL F1 and C5BL/6 backgrounds because of their ability to allow repeated backcrossing without deleterious consequences (B6SJL F1) and to increase the lethality of the APP transgene since the second or third backcross generation (C57BL/6), respectively. We also studied the CBA background because it is a non-albino strain that had never been used so far as a background strain for the APP transgene.

Our expertise in Tg2576 mouse breeding and the results of the present study confirm, as already established by Hsiao et al. [16], that the most suitable method to maintain the Tg2576 strain is to repeatedly backcross transgenic mice to F1 hybrids from the inbred strains that were used to create the transgenic construct. Meanwhile, the main outcome of this study is that backcrossing transgenic mice to various isogenic F1 hybrids and inbred strains for a single generation would constitute a cost effective and most likely an optimal strategy to detect most of the cognitive effects of a transgene in the mouse.

2. Methods

2.1. Animals

Two hemizigous Tg2576 (HuAPP695swe in a C57BL6/SJL genetic background) male mice created by K. Hsiao (1995) and generously gifted by Mayo Foundation for Medical Education and Research to J.M. Lassalle, were obtained from Charles River Laboratories. The Tg strain has been maintained in the CRCA mouse breeding facility by backcrossing Tg males to B6SJL F1 females.

Seventy female mice, 17 ± 1-month-old were used in this study. This time point, where mice are older than those generally used in this kind of studies, was chosen to allow us to consider the effects of senescence. We used female mice because they were still living in social groups in good conditions, whereas most 17-month-old males could hardly be kept in groups because of their aggressiveness. Mice were issued from B6SJL Tg+ progenitor males of the 4th generation, crossed to (i) B6SJL F1/j F1 females, (ii) C57BL6/j (B6) inbred females and (iii) CBA/j (CBA) inbred females. Mice were genotyped using PCR. C57BL/6 and CBA inbred mice as well as B6SJL F1 hybrids were also included in the experimental design. It has been shown in our laboratory that by 17 months of age, Tg2576 mice of both sexes display widely spread β-amyloid deposits associated to cognitive impairments (unpublished data). Some albino mice issued from the backcross to B6SJL F1 have been excluded from the study.

All mice used in this study were bred and reared in the same conditions in our institute breeding facility. They were housed in groups of three to five per cage and kept on at 12-h light–dark cycle, with lights on at 8 a.m., with constant ambient temperature (21 ± 1 °C) and humidity (50 ± 10%). Food and water were available *ad libitum* throughout the experiments. The breeding facility for transgenic mice was authorized by the committee for genetic engineering of the French ministry for research (#4161, July 8, 2004). All procedures followed the guidelines of the European Communities Council Directive of November 24, 1986 (86/609/EEC). Animal samples sizes and their characteristics in the experimental design were as indicated in Table 1.

2.2. Apparatuses and training

In order to avoid interferences between the 3 behavioral measurements, testing sessions were distributed over several weeks.

2.2.1. Elevated plus maze (EPM)

The experimental device was made of yellow polyvinyl chloride (PVC). The four arms were 30 cm long and 10 cm wide, the two opposite closed arms being equipped with 20 cm high walls. The EPM was elevated 100 cm above the floor. It was surrounded by a white curtain without any conspicuous cue, at a distance of 80 cm. The first week of behavioral testing the mouse was dropped on the maze in the 10 cm^2 central zone, facing an open-arm and videotaped for a 10 min period. Mouse behavior was analyzed using an ethological keyboard that allowed recording the number of visits and the time spent in each arm and the central zone.

2.2.2. Morris navigation task (MNT)

This test was performed on the second week of experiments. The experimental conditions replicated those routinely and successfully used in our laboratory [1]. The circular swimming pool (120 cm in diameter and 30 cm in height) was made of yellow PVC, filled with water (24 ± 0.5 °C), made opaque with the Opacifier 631® to 9 cm below the edge of the wall. A circular goal platform (8 cm in diameter) was laid 0.5 cm under the surface of the water and 16 cm from the wall. The device was placed in a regular room with a temperature of 22 °C. Dropped into the water from a different quadrant on each trial, mice had to learn to navigate to the invisible platform using the spatial cues available on a white curtain surrounding the pool at about 1.5 meter distance. After a three trial pre-training session to find out the procedural components of the task, mice were given three consecutive trials a day for 4 days. Shortly after the third trial of the last session, mice were submitted to a probe test for short-term spatial memory. The platform was removed and the mouse, starting from the opposite quadrant, was allowed a 1-min search for the platform. The path was videotaped and a spatial bias index was computed as the difference between the number of times a 12-cm-diameter annulus surrounding the former location of the platform was crossed and the mean number of crossings of three annuli, symmetrically laid out in the quadrants where the platform had never been, divided by the total number of annulus crossings.

2.2.3. Contextual fear conditioning (CFC)

We performed this test two weeks after the end of MNT. Conditioning took place in a conditioning chamber that consisted of a rectangular PVC box (length 35 cm, width 20 cm, and height 25 cm) with three light-brown sides and a Plexiglas front wall. The floor was made of a grid with stainless-steel rods connected to a generator (Campden Instruments) delivering shocks of defined duration (2 s) and intensity (0.7 mA) through a shock-scrambler unit. A loudspeaker producing the tone (85 dB, 30 s) was fixed on the top of the conditioning chamber. Experiments were videotaped. Contextual memory was checked in the same experimental conditions as conditioning, whereas tone memory was assessed in a modified context as already described by Daumas et al. [10].

Conditioning consisted of a single training session with two trials. Mice were dropped individually into the conditioning chamber via the ceiling. After a 2-min exploration period, a sound (CS) was emitted for 30 s, and a foot-shock (US) was superposed to the tone during the last 2 s. After an inter-trial interval of 2 min, the paired CS–US was repeated, and 30 s after the second foot-shock, mice were gently removed from the chamber and returned to their home cage. Twenty-four hours

Table 1
Experimental design

Experimental group	Number	Genotype	Background	
• B6SJL F1	7	Isogenic control	B6SJL	Heterozygous F1
• B6SJL Tg+	11	Tg+	B6SJL	Heterozygous BC (1/2 B6, 1/2 SJL)
• B6SJL Tg−	11	Tg−	B6SJL	Heterozygous BC (1/2 B6, 1/2 SJL)
• B6	7	Isogenic control	B6	Homozygous
• B6 Tg+	7	Tg+	B6	Heterozygous BC (3/4 B6, 1/4 SJL)
• B6 Tg−	8	Tg−	B6	Heterozygous BC (3/4 B6, 1/4 SJL)
• CBA	6	Isogenic control	CBA	Homozygous
• CBA Tg+	7	Tg+	CBA	Heterozygous BC (1/2 CBA, 1/4 SJL, 1/4 B6)
• CBA Tg−	6	Tg−	CBA	Heterozygous BC (1/2 CBA, 1/4 SJL, 1/4 B6)
Total	70			

Download English Version:

https://daneshyari.com/en/article/4315176

Download Persian Version:

https://daneshyari.com/article/4315176

Daneshyari.com