

# Environmental Enrichment of Young Adult Rats (*Rattus norvegicus*) in Different Sensory Modalities Has Long-Lasting Effects on Their Ability to Learn via Specific Sensory Channels

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Sensory modalities individuals use to obtain information from the environment differ among conspecifics. The relative contributions of genetic divergence and environmental plasticity to this variance remain yet unclear. Numerous studies have shown that specific sensory enrichments or impoverishments at the postnatal stage can shape neural development, with potential lifelong effects. For species capable of adjusting to novel environments, specific sensory stimulation at a later life stage could also induce specific long-lasting behavioral effects. To test this possibility, we enriched young adult Norway rats with either visual, auditory, or olfactory cues. Four to 8 months after the enrichment period we tested each rat for their learning ability in 3 two-choice discrimination tasks, involving either visual, auditory, or olfactory stimulus discrimination, in a full factorial design. No sensory modality was more relevant than others for the proposed task per se, but rats performed better when tested in the modality for which they had been enriched. This shows that specific environmental conditions encountered during early adulthood have specific long-lasting effects on the learning abilities of rats. Furthermore, we disentangled the relative contributions of genetic and environmental causes of the response. The reaction norms of learning abilities in relation to the stimulus modality did not differ between families, so interindividual divergence was mainly driven by environmental rather than genetic factors.

**Keywords:** genetic versus environmental effects, phenotypic plasticity, learning, enrichment, Norway rats

**Supplemental materials:** <http://dx.doi.org/10.1037/com0000063.supp>

What may be the case with objects in themselves and abstracted from all this receptivity of our sensibility remains entirely unknown to us. We are acquainted with nothing except our way of perceiving them, which is peculiar to us, and which therefore does not necessarily pertain to every being . . . —Immanuel Kant

Sensory modalities constitute the link between the outside world and the information acquired about it. The sensory modalities used to obtain information about the environment vary enormously between different taxa, which mostly reflects the requirements of their specific environment and “way of life.” The fossorial and solitary blind mole-rat, for instance, apparently relies on the Earth’s magnetic field for spatial learning (Kimchi & Terkel, 2001), whereas visual followed by olfactory cues seem to be used preferentially in the same context by the terrestrial and highly social Norway rat (Maaswinkel & Whishaw, 1999). Likewise, for

food selection highly frugivorous spider monkeys use significantly more often olfactory cues than more carnivorous squirrel monkeys, who use primarily tactile cues (Laska et al., 2007).

Within species, the most efficient sensory channel may vary according to context and the task to be accomplished. In Norway rats, for instance, visual cues seem to be essential in the context of spatial navigation (Rossier et al., 2000), but they do not seem to be required in the social context, where olfactory and auditory cues are apparently more important (Gheusi et al., 1997).

The sensory modality by which individuals best obtain new information may diverge also between individuals of a species (Braithwaite & Guilford, 1995; Mahdjoubi & Akplotsyi, 2012; Salvanes et al., 2013). The relative contribution of genetic and environmental causes of such interindividual variance remains unclear (Schellenberg, 2015). Genotypic variability may allow adaptation to a particular environment across generations (Dukas, 2004; Jones et al., 1992; Nepoux et al., 2015), whereas environmental plasticity enables individual adjustment to changing conditions during a life span (Braithwaite & Guilford, 1995; Dukas, 2004; Piersma & Drent, 2003; Salvanes et al., 2013; Searle et al., 2015). Exposure to environmental enrichment can induce changes in the neural system and improve learning performance (Cai et al., 2009; Fischer, 2016; Girbovan & Plamondon, 2013; Hirase & Shinohara, 2014; Hullinger et al., 2015; Kotschal & Taborsky, 2010; Landers et al., 2011; Rampon et al., 2000; Rochefort et al., 2002; Sampedro-Piquero et al., 2013; Simpson & Kelly, 2011; Veyrac et al., 2009; Xu et al., 2009). Environments may be enriched in various ways, including enhancement in social inter-

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We thank Leif Engqvist for help with data analyses, Akos Dobay for creating the audio files, Evi Zwygart for care of the rats, Françoise Schenk for useful comments, and Markus Wymann for technical support. We acknowledge financial support of the Schweizerischer Nationalfonds (project Grant 310030B\_138660 to Michael Taborsky) and of the Société Académique Vaudoise (grant to Vassilissa Dolivo).

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actions, motor challenges, the presence of stimulating objects, and sensory stimulation. Even if the life stage during which environmental conditions are commonly assumed to play an important role for the plasticity of the brain is mostly the early postnatal phase (Braithwaite & Guilford, 1995; Cai et al., 2009; Cai et al., 2010; Kotrschal & Taborsky, 2010; Nichols et al., 2007; Prusky et al., 2000a; Salvanes et al., 2013; Xu et al., 2009; Zhu et al., 2014), effects of enrichment occurring at a later stage are also reported (Alwis & Rajan, 2013; Landers et al., 2011; Percaccio et al., 2007; Rampon et al., 2000; Rochefort et al., 2002; Veyrac et al., 2009). Nevertheless, they are sometimes considered to be weaker (Hirase & Shinohara, 2014), and with regard to the effects of sensory enrichments, they remain controversial (Xu et al., 2009; Zhang et al., 2009). For instance, regarding the auditory system, Zhang et al. (2002) determined a critical period for the formation of tonotopic maps in rats that ends at approximately 30 days of age, and Xu et al. (2009) found no significant differences in the NR2B protein expression in the auditory cortex between rats exposed to music and rats not exposed to music when the exposure was initiated later than during this critical period (Xu et al., 2009; NB: according to Xu et al., a high level of NR2B leads to a high plasticity of the auditory cortex). Nevertheless, Percaccio, et al. (2007) found that the auditory cortex of rats was affected by their auditory experience during adulthood. Concerning the visual system, to our knowledge no study reports any effect of specific visual enrichment or impoverishment when it occurs at an adult stage. This is different in the olfactory system, as the olfactory bulb is characterized by high plasticity throughout life (Martoncikova et al., 2011; Veyrac et al., 2009; Whitman & Greer, 2009), and odor enrichment of adults has thus revealed significant effects on neurogenesis and memory (Rochefort et al., 2002; Veyrac et al., 2009).

In species that are able to adapt to novel environments throughout life, as for instance Norway rats (Jones et al., 2008), sensory enrichments should induce long-lasting specific behavioral changes also after the early postnatal stage. Rats are able to use visual (Prusky et al., 2000b; Schneeberger et al., 2012), olfactory (Gheusi et al., 1997) and auditory cues (Burn, 2008; Rossier et al., 2000), and they adjust to diverse habitat conditions (Klemann & Pelz, 2006). This may be especially important when individuals disperse, which seems to mainly happen at an adult stage (McGuire et al., 2006). Climatic factors (seasons), food availability, and breeding periods may influence the movement of rats and cause changes in the selection of their habitats (Feng & Himsforth, 2014). We thus expect rats to be particularly sensitive to environmental conditions encountered during the “young adult” stage (i.e., adolescence), that is, when reaching sexual maturity, and we expect sensory enrichment to induce neurophysiological and behavioral changes especially during this period. The central nervous system should then still be sufficiently plastic to show long-term effects of sensory experience, as revealed for instance by effects of auditory experience during adulthood on the auditory cortex (Percaccio et al., 2007). Sensory enrichment may enhance selective attention, which has been shown to reduce response variability for instance in the auditory, visual and somatosensory systems (Strait et al., 2015). Furthermore, it has been argued that the use of a particular sensory signal during a learning task may induce physiological plasticity in connection with this particular signal in the cerebral cortex (Bieszczad & Weinberger, 2010). We

hypothesize, therefore, that more intensive exposure to a particular sensory modality (visual, auditory, or olfactory) during the ‘young adult’ stage will enhance the learning abilities specifically in response to stimuli encountered in the same modality later in life. Indeed, whereas general effects of enrichment have been determined in several species, the effects of specific sensory enrichment in situations involving specific sensory challenges are currently unclear.

In previous studies testing for effects of sensory enrichments on learning abilities, behavioral tests were usually performed briefly after (Rochefort et al., 2002; Zhu et al., 2014) or during the enrichment period (Xu et al., 2009). However, neurophysiological effects of early postnatal environmental enrichment have been observed in the auditory cortex of rats at least 2 months after enrichment had ended (Cai et al., 2010; see also Rochefort et al., 2002 for a note on comparable long-term enrichment effects on the olfactory memory of mice). To clarify whether specific environmental conditions encountered during the young adult stage can have long-lasting behavioral effects, we enriched female Norway rats in various sensory modalities (visual, auditory, or olfactory) and subsequently tested their learning abilities regarding the similarity or dissimilarity between the eventual specific enrichment they had encountered and the type of stimulus (visual, auditory, or olfactory) used for the test. In a learning test for which no sensory channel was per se more relevant than any other, we predicted that test rats would perform better with cues provided in the same sensory modality in which their earlier environment had been enriched.

In addition, the random assignment of sisters to divergent enrichment conditions allowed comparisons both between-families (genetic divergence effects) and between-enrichment conditions (plasticity effects; cf. Schellenberg, 2015). We were thus able to disentangle the relative contributions of genetic divergence and environmental plasticity.

## Method

### Ethics Statement

The procedure described in this article conforms to the legal requirements of Switzerland and the guidelines of the University of Bern, where the work was carried out (License Number BE98/11).

### Subjects

We intended to use 30 female wild-type Norway rats (source: Animal Physiology Department, University of Groningen, Netherlands) housed during all their lives in six different cages (P, R, S, T, U, X) containing five sisters each (except in three cages, where one of the five littermates was not a sister (T3 in the cage T, U4 in the cage U, and X5 in the cage X). One individual died before the end of the study (X1), and another one was not motivated to perform the learning task (S1); these two individuals are thus not considered further in this study, which is thus based on the results obtained with the 28 remaining rats. Housing cages (80 cm × 50 cm × 40 cm) were enriched with wood and paper toys, a tunnel, and a wooden shelter. Water and rat-pellets were pro-

vided ad libitum. In addition, the rats received special treats (seeds, fruits, vegetables, pasta or rice) on each afternoon.

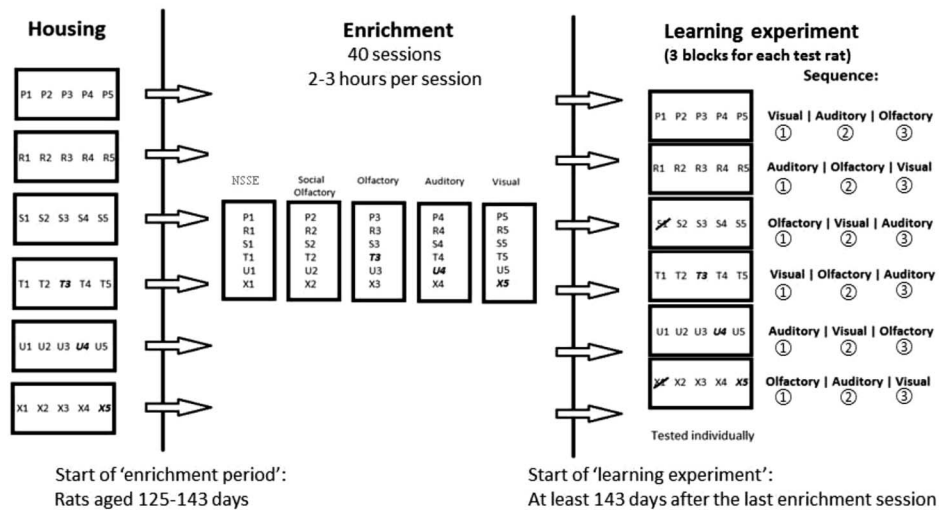
A sensory enrichment period of 40 sessions of 2 to 3 hr each spread over 60 days began when rats were aged 125 to 143 days (young adults). It was followed by a learning test (split in three blocks) for which no sensory channel was per se more relevant than any other, by using a nonsocial context in which no complex spatial navigation was required. During this test, each subject was exposed to three blocks of tests, each block involving a different sensory modality, in a design with balanced sequence (which means that each sensory modality was used in the first block for one third of the rats, in the second block for another third of the rats, and in the last block for the rest of the rats, thus avoiding any sequential bias; see Figure 1 and Table S1 in the online supplemental material). Between the last enrichment session and the first block of tests we maintained an interval of at least 143 days.

Moreover, as we had randomly assigned sisters sharing the same cage for all their lives, except during the enrichment sessions, to divergent enrichment conditions, it was possible to balance the potential influence of other factors than sensory enrichments. It also allowed us to disentangle the relative contributions of genetic divergence and environmental plasticity to the variability in reaction norms of learning abilities in relation to the stimuli (visual, auditory, or olfactory) provided during the learning test.

As rats are predominantly nocturnal, an inverted 12:12 light–dark cycle with lights on at 20:00 hours allowed us to work during the rats' natural activity phase (i.e., in the day hours under artificial red light). Rats are able to use visual information presented under red light conditions (Schneeberger et al., 2012).

## Enrichment Protocols

Rats were split into five treatment groups for the enrichment sessions: a group without specific sensory enrichment (1), and four groups with specific sensory enrichments involving social olfactory cues (2), nonsocial olfactory cues (3), auditory cues (4), and visual cues (5). Each treatment group contained exactly one rat from each housing cage (see Figure 1). For each enrichment session, rats belonging to a specific group were transferred into an empty cage (80 cm × 50 cm × 40 cm; located in a separate room) and kept together for 2 to 3 hrs, during which they were exposed to stimuli belonging to one specific sensory modality or no specific sensory modality in the case of group (1). At the end of each enrichment session, rats were returned to their respective housing cages. Each rat was exposed to 40 enrichment sessions spread over 60 days, with no more than one session per day. Knowing that novelty is crucial for enrichments to be successful (Cai et al., 2009; Veyrac et al., 2009), stimuli provided during the sessions were selected to be as broad as possible, as described in the following paragraphs.



## Learning experiment

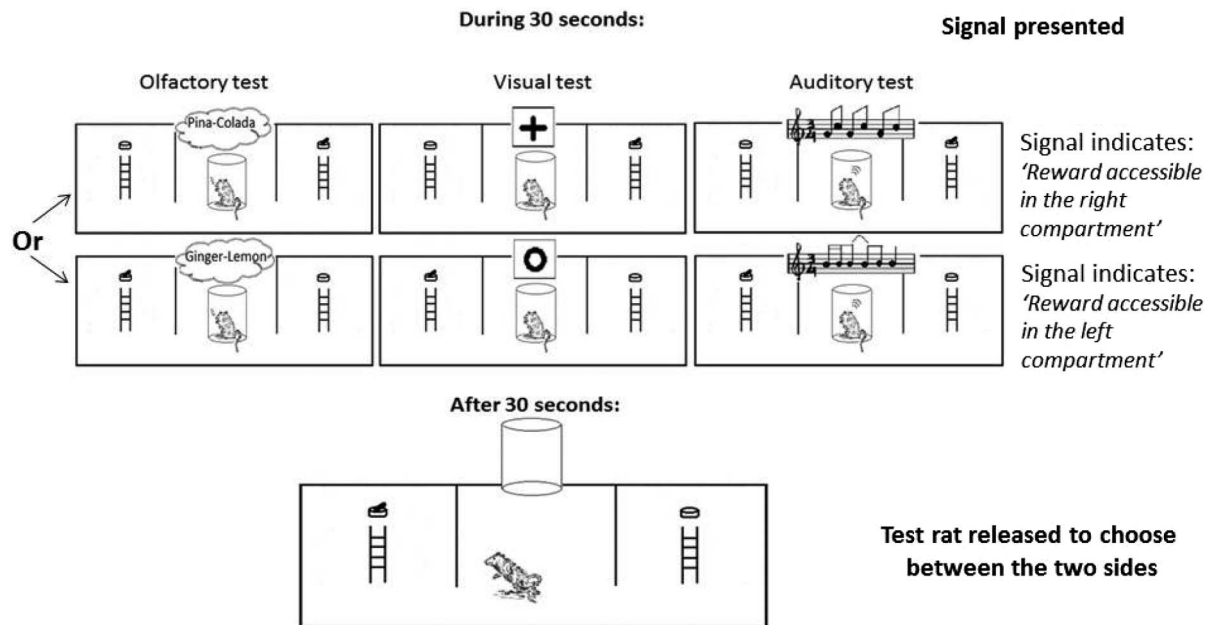
<b>1 st block:</b>	<b>Training:</b> day 1 – day 2 – day 3	<b>Test:</b> day 3 – day 6 – day 9	<b>At least 19 days between subsequent blocks</b>
<b>2 nd block:</b>	<b>Training:</b> day 1	<b>Test:</b> day 2 – day 3 – day 7	
<b>3 rd block:</b>	<b>Training:</b> day 1	<b>Test:</b> day 2 – day 3 – day 7	

*Figure 1.* Experimental schedule. Letters (P, R, S, T, U, X) refer to the housing cages. Numbers (1 through 5) refer to the kind of enrichment provided. Encircled numbers refer to the chronological order of the blocks. Each block consisted of a training phase followed by a test phase, which involved one specific sensory modality. Individuals printed in bold were not housed with siblings. S1 and X1 were excluded from the study (X1 died before the end of the experiment and S1 was not motivated by the reward).

1. No specific sensory enrichment (NSSE) group (4 subjects). Subjects of the NSSE group were transferred to a cage located in a separate room such as the rats from the four enrichment conditions, but they did not receive any specific sensory enrichment.
2. Social olfactory cues group (6 subjects). Subjects were exposed to a wide range of social odors during the enrichment sessions. During each session, two small cloth bags containing litter of unfamiliar conspecifics were attached to the sides of the cage. Odors of 40 different cages were used and exchanged before each session.
3. Nonsocial olfactory cues group (6 subjects). Subjects were exposed to a wide range of nonsocial odors during the enrichment sessions. During each session, two small cloth bags containing spices (e.g., basil, garlic, and cinnamon) and flavored teas were hanging on the sides of the cage. Odors were renewed and changed before and once during each session. A total of 80 different odors were used for enrichment.
4. Auditory group (6 subjects). Subjects were exposed to a wide range of nonsocial sounds during the enrichment sessions. Sounds consisted of different types of music, for instance piano pieces (Chopin, Tchaikovsky . . .), symphonic music (Addinsell, Ravel, Rachmaninov . . .), folk music (Russian, Brazilian . . .), or singing (Carmina Burana, Perry Como, Yves Duteil . . .), with more than 400 pieces in total.
5. Visual group (6 subjects). Subjects were exposed to a wide range of black-and-white pictures during the enrichment sessions. During each session, two new black-and-white pictures were fixed on the sides of the cage. Pictures were either figurative (e.g., tree, cat, bird) or nonfigurative (e.g., lines, curves, letters). A total of 80 different pictures were used for enrichment.

### Learning Experiment

**Nonsocial two-choice learning task.** For the learning experiment, we used cages ( $150 \times 68 \times 50$  cm) divided into three compartments by two cardboard partitions, with an adjustable door in each partition either allowing or blocking access between compartments (see Figure 2). A bowl containing a piece of pasta was presented in each of the two edge compartments. Using the principle of “false bottom,” the piece of pasta was either accessible or not: the bowl contained two superimposed bottoms with little holes in the upper one, allowing to reach the pasta only when it was located on the upper bottom and not between both bottoms. The rats could always smell the treat, so that the olfactory cues were similar between both conditions, regardless whether the food was actually accessible or not. To reach the bowls, rats had to climb either a rope, a ladder, or the grid-walls of the cage.



*Figure 2.* Symbolic depiction of the experimental set-up used for training and test during the learning experiment. The cage was separated into three compartments. At the beginning of a training or test trial, the rat was kept in a cylinder in the middle compartment for 30 s, during which she was presented with a signal (olfactory, auditory or visual). Two signals were used with each sensory modality, one indicating a reward in the left compartment and the other one indicating a reward in the right compartment. After 30 s, the cylinder was removed and the rat was free to access the reward. During the training phase, only the door offering access to the correct compartment associated with the provided signal was open. During the test phase, both doors were open and the rat could choose between the left and right compartments according to the presented signal.

An experimental block involving one specific sensory modality began with a training phase that was followed by a test phase. Experimental blocks were separated by intervals lasting at least 19 days. During the training phase of the experiment at the beginning of a trial, the subject was kept in a plastic cylinder surrounded by white paper in the middle compartment of the cage for 30 s. During these 30 s, she was presented with a signal (olfactory, auditory, or visual). Two signals were used for each sensory modality, one indicating that the reward was accessible in the left compartment and the other one indicating that the reward was accessible in the right compartment. After 30 s, the cylinder was removed and the door offering access to the compartment associated with the provided signal was opened. Each trial lasted until the rat had found the reward. The training phase lasted 3 days for the first block in which rats were exposed for the first time to the task and the set-up. For the second and third blocks, in which rats already knew the task and the set-up from their experience in the first block, but not the signals which were presented in a new sensory modality, the training phase was reduced to one day. Four trials per day were provided to each rat during the training phase (see Figure 1 and Table S1 of the online supplemental materials).

The test phase began on the last day of the training phase for the first experimental block, and one day after the training phase for the second and third experimental blocks. First blocks consisted of 10 trials each; second and third blocks consisted of 12 trials each. The fact that first blocks, for practical reasons, involved different trial numbers has neither conceptual nor analytical consequences because the way in which the analyses were performed allowed to interpret the results unequivocally. The test phase lasted 3 days, with four trials per day (except for the first day of the first block, which only consisted of two trials). Subsequent test days were separated by an interval of 1 to 4 days (see Figure 1 and Table S1 in the online supplemental materials). The beginning of a trial during the test phase was identical to the training phase. The only difference was that in the test phase, after the removal of the cylinder, both doors were opened and the rat could choose between the left and right compartments, according to the presented signal. The reward was located in the left compartment for one half of the trials, and in the right one for the other half (for more details, see Table S1 in the online supplemental materials). The response was assessed as right or wrong as soon as the rat had reached the correct or wrong bowl and checked for the reward. We noted the time taken to reach the bowl for each trial as a measure of decision speed.

We chose to standardize the procedure according to the criteria of the number of obtained rewards, thus avoiding our rats to lose their motivation if making consecutive mistakes during the first choice. Indeed, considering that our rats neither had been over-trained nor food deprived, the repetition of the trial after a wrong choice guaranteed that no rat would get less rewards than others, which might have caused a loss of motivation and of participation in the task. Thus, after each trial resulting in a wrong choice, the test rat was returned to the cylinder for 30 seconds and exposed to the same signal as presented before. Hence the rat received a second chance to choose the correct side and get the reward. If the choice was again wrong, this procedure was repeated once again. If the correct side was not chosen by the end of the third attempt, the correct bowl containing the reward was pointed out to the subject with a finger. Independently of the number of opportunities

provided to the rat to obtain the reward during one trial, only the result of the first choice (right or wrong) was used for the analyses. This treatment was identical for all test situations.

For the learning experiment involving the visual modality, a black cross fixed to the cylinder indicated a reward in the right hand compartment, and a black circle indicated a reward in the left hand compartment (see Figure 2). For the learning experiment involving the olfactory modality, a cloth bag containing "pina colada" tea was fixed to the top of the cylinder to indicate a reward in the right hand compartment, whereas a cloth bag containing "ginger-lemon" rooibos tea indicated a reward in the left hand compartment (see Figure 2). For the learning experiment involving the auditory modality, two short signals differing in the melody and rhythm were played four times while the subject was in the cylinder. One of these signals was associated with the right hand compartment, and the other one with the left hand compartment (see Figure 2; the auditory signals used in this experiment are provided in the online supplemental materials). All the stimuli used for the learning experiment (visual, olfactory, and auditory cues) had never been encountered previously by any of the rats.

## Data Analysis and Statistics

All analyses made use of the RStudio statistical software (Version 0.98.50) with the "lme4" and "multcomp" packages. For all analyses using generalized linear mixed models (GLMM) except those concerning the decision speed, a binomial distribution of the number of correct and wrong choices per test rat and block was assumed, which will further be referred to as *learning scores* (10 choices for the first block, 12 choices each for the second and third blocks). In the analyses concerning the decision speed, each trial was considered separately. Rat identity and cage were included as random factors in each GLMM, except in the cases where we tested for a family effect (in which only rat identity was included as random factor). To check for a family by-stimulus interaction, we used a likelihood ratio test (LRT). To test whether rats perform better with the sensory modality for which they had been enriched, a one-tailed test was used, as it has been widely established in previous studies that cognition, memory and learning are improved and not hampered by enrichment (Cai et al., 2009; Kotschal & Taborsky, 2010; Landers et al., 2011; Rampon et al., 2000; Rochefort et al., 2002; Veyrac et al., 2009; Xu et al., 2009). Model estimates are reported as  $\beta \pm SE$ .

## Results

### Did Test Rats Learn to Perform the Two-Choice Task Successfully?

Overall, in the two-choice tasks the test rats chose the correct side significantly more often than expected by chance (56.3% correct choices;  $\beta = -0.25 \pm 0.07$ ,  $p = .0001$ ; GLMM).

### Did Test Rats Learn Better if the Training Stimulus Matched the Sensory Modality Used for Enrichment?

We predicted specific sensory modality enrichment to improve the ability of individuals to learn a task if stimuli of the same sensory modality were used. Thus, we compared learning scores of

the 28 subjects with each of the three sensory modalities (84 learning scores, defined as a proportion of correct to wrong choices; cf. the Method section) between the cases in which the test sensory modality matched the enrichment modality with the cases in which the modalities differed. We assumed the olfactory modality in the learning test to match both the social and nonsocial olfactory enrichment conditions. For the NSSE group, we assumed that the test sensory modality never matched the enrichment modality. Learning scores were significantly higher if the test stimulus and the enrichment involved the same sensory modality ( $\beta = -0.27 \pm 0.15$ ,  $\chi^2 = 3.48$ ,  $df = 1$ ,  $p = .031$ ; Figure 3; GLMM).

### Did Genetic Divergence Influence Global or Modality-Related Learning Performance?

We tested whether global scores (cumulative ability of individuals to learn a task with each of the three sensory modalities used in this study) differed between families, and whether reaction norms of learning abilities differed between families in relation to the sensory modality of the stimulus (visual, auditory, or olfactory). There were no significant differences between families in the global scores ( $\chi^2 = 1.75$ ,  $df = 7$ ,  $p = .972$ ; GLMM including rat identity as random factor) and the reaction norms of learning abilities in relation to the sensory modality of the stimulus (LRT;  $\chi^2 = 0.14$ ,  $df = 5$ ,  $p = .999$  for the family-by-stimulus interaction).

### Did the Learning Performance Differ Between the Different Treatment Groups?

We did not detect differences in the proportion of correct decisions between the five experimental groups (all  $ps > 0.629$ ; multiple comparisons of means: Tukey contrasts, including rats' identity and cage as random factors). This does not indicate an influence of the sensory modalities used for enrichment on the rats' overall learning scores. This also does not indicate a difference between social and nonsocial olfactory enrichment on the rats' overall learning scores.

### Did the Learning Performance Differ Between Learning Tests Using Stimuli in Different Sensory Modalities?

There was no overall effect on learning performance of the sensory modality in which the indicator stimulus was presented in the learning test (Friedman rank sum test,  $N = 28$ ,  $\kappa = 3$ ,  $p = .137$ ). This remained true also when we excluded the cases in which the sensory modalities matched between enrichment phase and learning test ( $\chi^2 = 2.9$ ,  $df = 2$ ,  $p = .234$ ; GLMM). Thus no particular sensory modality in which the learning stimulus was presented allowed the population of test rats to learn significantly better than any other sensory modality.

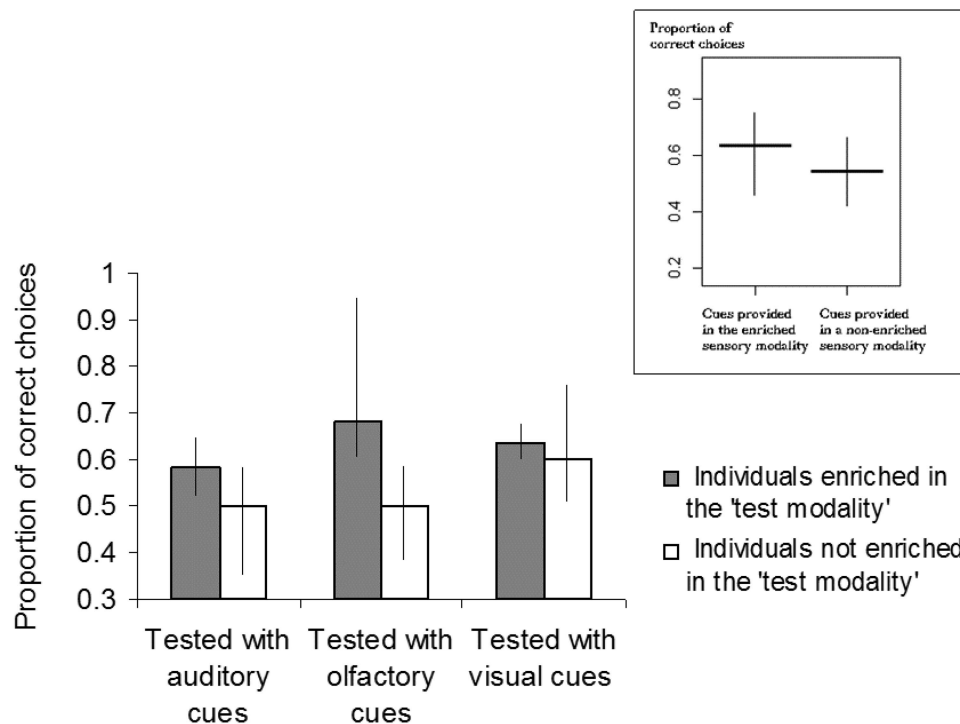


Figure 3. Proportion of correct choices during the learning task. Medians and quartile ranges are shown of proportion of correct choices with each type of sensory cue (auditory, olfactory, and visual) in dependence of whether or not the cue was provided in the enriched sensory modality. On the top right, median and quartile ranges are shown of overall proportion of correct choices in dependence of whether or not the cues were provided in the enriched sensory modality.

### Did Previous Experience With the Design and the Task Affect the Performance of Test Rats?

Test rats were submitted to three blocks of training and test for the discrimination task, with three different sensory modalities involved across the blocks. A new block of training and test was conducted between 19 and 55 days after the achievement of the previous one. The rank of the experimental block did not influence the proportion of correct choices ( $\beta = -0.02 \pm 0.08$ ,  $\chi^2 = 0.08$ ,  $df = 1$ ,  $p = .777$ ; GLMM).

### Did the Effect of Specific Sensory Enrichment Decrease With Increasing Delay Between Enrichment and Learning Test?

The time interval between enrichment experience and learning test did not affect the learning performance. This was tested for learning trials using stimuli that were matching the enrichment conditions ( $\beta = -0.003 \pm 0.005$ ,  $\chi^2 = 0.43$ ,  $df = 1$ ,  $p = .51$ ; GLMM).

### Did Test Rats Perform Quicker When Taking a Correct Decision, When Having Been Enriched in a Specific Sensory Modality, or When Tested With the Same Sensory Modality in Which They Had Been Enriched?

The decision speed did not differ between right and wrong choices ( $\chi^2 = 1.16$ ,  $df = 1$ ,  $p = .282$ ), and it was not affected by the modality of enrichment ( $\chi^2 = 2.84$ ,  $df = 4$ ,  $p = .585$ ) and the fact whether stimuli were used in the learning test that matched the sensory modality of the enrichment experience or not ( $\chi^2 = 1.88$ ,  $df = 1$ ,  $p = .17$ ). For these analyses, the decision speed was considered separately for each trial (GLMM with decision speed values normalized using a Box-Cox-transformation ( $\lambda = -.57$ )).

## Discussion

Our test subjects performed the learning task better when a signal was provided in the same sensory modality than previously used for their environmental enrichment, while decision speed remained unaffected by the experimental treatment. This reveals long-lasting specific behavioral effects of enrichment in a particular sensory modality, which conforms to results from neurophysiological studies suggesting organizational effects in the brain from specific sensory experience (Martoncikova et al., 2011; Percaccio et al., 2007; Rochefort et al., 2002; Veyrac et al., 2009; Xu et al., 2009). However, the neurophysiological and behavioral effects of sensory enrichment have been typically studied for exposure to a particular sensory environment occurring early in life, when plasticity of the central nervous system is considered to be maximal (Hirase & Shinohara, 2014; Salvanes et al., 2013; Xu et al., 2009). Effects of sensory enrichment presented later than at the early postnatal stage have received little attention and remain controversial (Xu et al., 2009; Zhang et al., 2009). Moreover, to the best of our knowledge no comparisons have yet been done between enrichment effects involving a range of different sensory modalities.

In this study, sensory enrichments were provided to wild-type Norway rats at a young adult stage, corresponding to the period in which rats likely encounter new environmental conditions, for instance due to dispersal, the start of reproduction, or seasonal changes in their natural environment. This period seems particularly suited for acquiring new skills to deal with both the physical and social environments. As expected, long-lasting effects of specific enrichments on learning abilities emerged already after less than 120 hr of enrichment. Moreover, these effects seem to remain stable over time. The learning performance of rats tested with stimuli matching their enrichment modality did not depend on the interval length between the end of enrichment and the start of the learning test. We can thus expect that the effects of a specific enrichment do not decrease with time per se. As a further perspective, it would be interesting to explore whether an additional subsequent sensory enrichment in another sensory modality would act in addition or in substitution to the first enrichment.

The test subjects performed significantly better than by chance, showing that they learnt to discriminate the provided signals. The proportion of correct answers was relatively low, which may be due to a rather weak motivation to avoid wrong choices. Our rats were neither food-deprived before the learning task, nor punished for wrong choices. Even if in absolute terms the learning score was not very high, it was nevertheless consistently higher than if individuals had acted randomly. Globally, no sensory modality in which the learning stimulus was presented allowed the population of test rats to perform significantly better than any other in our two-choice learning task. In a neutral context, in which no sensory modality seems a priori more valuable than any other to gain information, rats perform apparently as efficiently with visual cues as with auditory or olfactory ones. Previous studies have suggested, however, that there may be a context-dependent sensory hierarchy in Norway rats (Dolivo & Taborsky, 2015; Maaswinkel & Whishaw, 1999). It seems reasonable to expect selection to favor the use of particular sensory systems that would provide individuals with the best capability to adjust their behavior to the specific conditions of their environment. Thus, rats living in complex spatial environments might be favored if preferentially using vision, whereas those living in very large colonies might be favored if preferentially using olfaction. Our study suggests that preferential sensory modalities may be indeed selected via a mechanism of plastic adaptability to specific environments. Because of the limited sample size of each specific group in our study we cannot determine which sensory enrichments (olfactory, auditory, or visual) might have larger or smaller effects than others, and thus we do not know whether the strength of the effect of sensory enrichment depends on the kind of sensory modality involved. This would be an interesting aspect for future studies.

Cognitive and learning abilities are partially genetically determined (Koppik et al., 2015) and heritable (Smith et al., 2015), and learning responds to selection (Nepoux et al., 2015). Nevertheless, we found no difference between families in global learning scores and in reaction norms of learning abilities in relation to stimulus in our study. Apparently, the observed benefits in learning performance associated with specific sensory enrichments did not result from genetic differences between groups of individuals (cf. Schellenberg, 2015).

For *Homo sapiens*, it is commonly accepted that individuals differ in the sensory modality from which they best absorb infor-

mation, which might be an important aspect for school education (Mahdjoubi & Akplotsyi, 2012). Nevertheless, when considering music training, for instance, the role of the environment remains controversial (Schellenberg, 2015; Strait et al., 2015). It has been argued that musical practice may promote auditory attention and that the development of cortical mechanisms underlying selective auditory attention is associated with young-stage music practice (Strait et al., 2015). Alternatively, preexisting genetic differences between individuals could be involved, with good, genetically determined 'listening abilities' inducing children to take music lessons, thus leading to a biased interpretation of the hypothetical role of environmental enrichment (Schellenberg, 2015). Our study with rats as model organism suggests that at least partly the sensory modality from which the information is best absorbed may be determined by environmental aspects.

Decision speed may also reflect the perceived difficulty of a task. For instance, while assessing the visual acuity of rats, Prusky et al. found that more time is required to make a choice when difficulty is increased (Prusky et al., 2000b). In our case, one might expect the task to appear more difficult to subjects when the stimuli were not matching their enrichment modality. Nevertheless, neither the correctness of the choice nor the similarity between the test sensory modality and the enrichment sensory modality related to the decision speed.

No specific sensory enrichment seemed to improve the overall learning score more than others. Sensory enrichment enhances selective attention (Strait et al., 2015). This effect, plus the fact that all rats were living in generally enriched conditions, might explain why in our experiment the influence of specific sensory enrichments was restricted to learning processes involving these respective sensory modalities. Along these lines, the absence of discrimination in a discrimination task may not necessarily mean a lack of cognitive abilities, but merely a lack of attention to certain stimuli, where animals fail to discover upon which stimuli they should base their discrimination (Jeffery, 2007). On the other hand, it has been suggested (Bratch et al., 2016) that rats may process information via multiple working memory systems, independently of one other. This independency would also point toward specific rather than general effects of specific sensory enrichments. Before neurophysiological effects of our enrichment conditions have been investigated, we cannot determine whether specific sensory enrichments improve the recognition or memory system of rats, or whether they exclusively enhance their attention toward cues provided in the respective enriched modality (cf. Alwis & Rajan, 2013; Arisi et al., 2012; Percaccio et al., 2007; Rochefort et al., 2002; Strait et al., 2015).

Novelty is generally assumed to be the most important causal factor of enrichment effects (Cai et al., 2009; Veyrac et al., 2009). For this reason, we aimed to maximize diversity of the sensory enrichment conditions. It has been argued that for sensory inputs to be efficient, they must be salient for the enriched subjects (Percaccio et al., 2007). We expected social sensory cues to be more salient for rats than nonsocial cues, because Norway rats are highly social animals exchanging helpful behavior reciprocally among one another (Dolivo et al., 2016; Dolivo & Taborsky, 2015; Rutte & Taborsky, 2007, 2008). Therefore, we generated two groups enriched with olfactory cues, one with social and one with nonsocial cues. We could not detect any difference in the learning scores between these social and nonsocial olfactory enrichment

conditions. It is important to note that the enrichment sessions also provided a form of social enrichment by letting rats that were kept in separate home cages interact with each other. This was the same for all treatment groups, but it might have reduced the effects of specific sensory enrichments, thus reducing the 'between group' differences. In future studies, sensory enrichment of rats in isolation might be a means to accentuate the effects of specific sensory enrichments.

In conclusion, our results show that specific sensory enrichments provided at a young adult stage have long-lasting effects on the learning abilities of Norway rats when the learning task relies on a stimulus of the same sensory modality that was used for enrichment. In addition, phenotypic plasticity is apparently more important than genetic predispositions to explain the interindividual variation in the ability to use particular sensory channels when solving a discrimination task. Specific sensory stimulation, even if provided during a relatively short period at adulthood, seems to be an important determinant of the ability of rats to acquire and process information via a specific sensory channel.

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Received November 15, 2016

Revision received January 11, 2017

Accepted January 18, 2017 ■