Meta-analysis of strategy choice by 94 mutant mouse lines in a standardized place navigation task identifies behavioral flexibility as performance limiting factor

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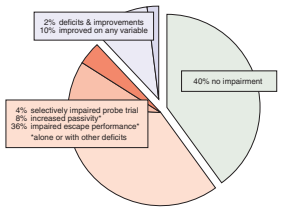
Summary

When tested in procedures that were originally developed for rats, mice may be affected by manipulations in different ways than rats. A meta-analysis comparing hippocampally lesioned mice with 94 different mutant lines shows that performance of mice in a watermaze place navigation task is limited by behavioral flexibility rather than by spatial memory.

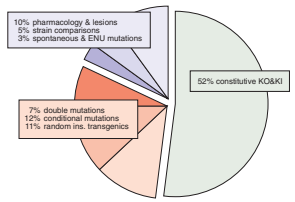
We categorized trials into 8 exclusive categories according to swimming strategy: circling, floating, wall hugging, random swimming, scanning, chaining, focal searching, direct swims. Analysis of strategy transitions between trials revealed that normal mice initially tend to perseverate on non-spatial strategies, giving up this tendency more or less rapidly in favor of spatially directed strategies. Hippocampal lesions mainly disrupt the flexibility to explore different strategies, limiting the choice to wall hugging or floating, and preventing progression to stages where processing of spatial information becomes relevant. Mutations that block plasticity-relevant signaling pathways or model human cognitive deficits produce a similar syndrome or a milder form. Selective spatial impairments occur rarely, e.g. in Arg3.1/Arc or nestin/GR knockout mice.

1 Differences are most often observed in variables of training performance

Study outcome



Mutations / models



5356 animals were grouped into 124 studies with one experimental and one control group each. As needed, number of groups was reduced by merging statistically indistinguishable groups or splitting into sub-studies with a shared control group. The median number of animals per study is 32.

In 124 studies conducted 1987-2003 in the same laboratory, along with strain comparisons, lesion and pharmacological experiments, 94 different mutant lines were analyzed, carrying modifications in 80 different target genes that are all expressed in the CNS.

Methods

2 watermaze place navigation protocols in 1.5m pool. Standard protocol (109 studies, 4922 animals): 3 days x 6 trials acquisition & 2 days x 6 trials reversal, max. trial duration 120s, ITI 30-60 min, probe trial = first of reversal. Difficult protocol (15 studies, 434 animals): 14 days x 2 trials, max trial duration 90s, ITI 60s, probe trial 24h after training. Video tracking at 256x256 pixel and 4.2Hz. Data analysis in Wintrack 2.4, www.dpwolfer.ch/wintrack. Statistical comparisons: ANOVA with transformations as necessary. Multiple comparisons filtered using false discovery control procedure.

Mutations: APLP1+/- on APLP2-/- background, A1AR-/-, APLP1-/-, bAPP-/- on APLP1-/- background, bAPP$/$, bAPP-/- , Arg3.1/Arc-/-, ARHGEF6-/-, brl/brl (barrelless mutation of adenylyl cyclase type I gene), calretinin-/-, CHL1-/-, Nes/Cre;c-Junlx/lx, CREBa/aλ , CREBaλ/-, CaMKII/Cre;CREBlx/lx on CREM-/- background, CaMKII/Cre;CREBlx/lx, Nes/Cre;CREBlx/lx, CaMKII/CREB(A133), Cx32-/-, EphB2extdrezd, EphB2-/-, EphB3-/-, Nes/Cre;Ephrin B1lx/lx, CaMKII/Cre;Ephrin B2lx/lx, F52-/-, GABAAα5(H105R)/α5(H105R), NSE/GAP43(CAM), NSE/GAP43(G19), Gd1Y/-, GFAP/L1, GlyRa3-/-, GR(A458T)/(A458T), Ras-GRF1enu/enu, Ras-GRF1-/-, Nes/Cre;GRlx/lx, Hmgb2-/-, MAG-/-, Met(2C)^2G^, Metgrb2^grb2^, MR-/-, CaMKII/Cre;MRlx/lx, NSE/NAB2, mCD24-/-, Ncam-/-, Ncam-/- x NF-L/Ncam180, Nr1neo-/- (neo in intron 20), Ns-/-, Thy-1/Ns x Thy-1/PA, Thy-1/Ns, neurotrypsin+/-, Thy-1/neurotrypsin(trunc), CaMKII/Cre;N-WASPlx/lx, ERK1-/- MF1, CaMKII/Cre;PAC1lx/lx, PAC1-/-, Per1-/-, Per2-/-, Prp-/- Büeler x Sakagushi, Prp-/- Büeler x Sakagushi x Cozzio, Prp-/- Büeler, Pv-/-, Rab3a-/-, Rag1-/-, rb/rb (Robertsonian translocation), rd/rd, Ras-GRF1ma (maternal allele not inactivated), Rsk2-/-, RyR3-/-, S100A1-/-, Sgk3-/-, BAC/Sim-2, Ras-GRF1-/- x SOS+/-, Thy-1/bAPP(Swe) x Thy-1/tPA, Thy-1/APLP2, CaMKII/Cre;trkBlx/lx, CaMKII/Cre;trkBlx/-, tlx-/-, tPA-/-, uPA-/-, NSE/Vegf

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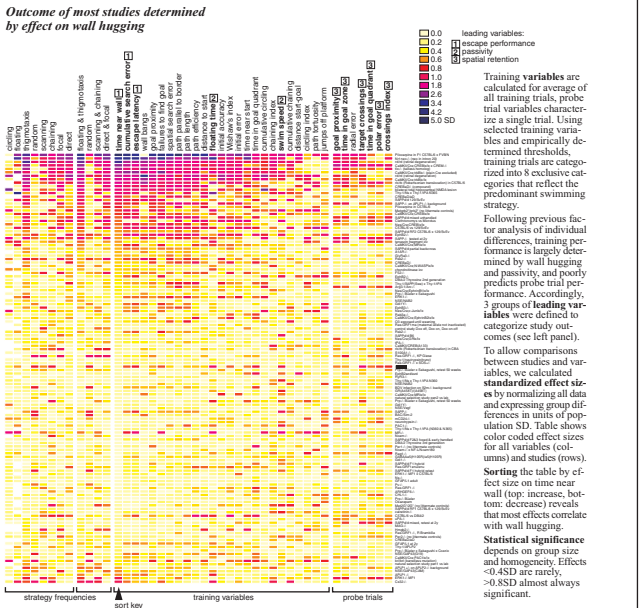
2 Mutation effects on training performance are larger than those on probe trial retention

Outcome of most studies determined by effect on wall hugging



Training **variables** are calculated for average of all training trials, probe trial variables characterize a single trial. Using selected training variables and empirically determined thresholds, training trials are categorized into 8 exclusive categories that reflect the predominant swimming strategy.

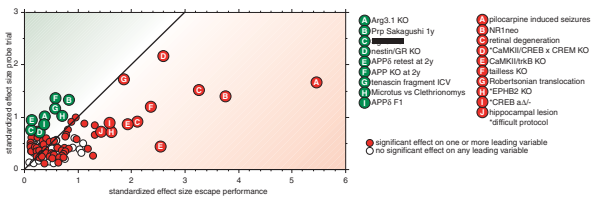
Following previous factor analysis of individual differences, training performance is largely determined by wall hugging and passivity, and poorly predicts probe trial performance. Accordingly, 3 groups of **leading variables** were defined to categorize study outcomes (see left panel).

To allow comparisons between studies and variables, we calculated **standardized effect sizes** by normalizing all data and expressing group differences in units of population SD. Table shows color coded effect sizes for all variables (columns) and studies (rows).

Sorting the table by effect size on time near wall (top: increase, bottom: decrease) reveals that most effects correlate with wall hugging.

Statistical significance depends on group size and homogeneity. Effects <0.4SD are rarely, >0.8SD almost always significant.

Hippocampal lesions and genetic disruption of synaptic plasticity strongly increase wall hugging



Strongest impairments are found in animals with genetic or excitotoxic ablation of the hippocampus, as well as after genetic inactivation of NMDA or trkB receptor function. These models show a strong increase of thigmotaxis. The impairment of blind mice is similar and can only be differentiated using a visible platform protocol. A selective impairment of probe trial performance was observed in a relatively small number of studies.

3 Behavioral flexibility rather than spatial memory limits training performance

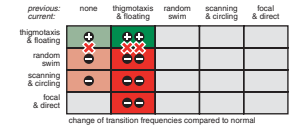
106254 strategy transitions in mice that learn normally

previous: / *current:*	none	thigmotaxis & floating	random swim	scanning & circling	focal & direct
thigmotaxis & floating	54%	46%	10%	5%	4%
random swim	26%	11%	20%	7%	5%
scanning & circling	12%	22%	34%	39%	27%
focal & direct	8%	21%	36%	49%	64%

transition frequencies

Using video tracks and an automatic algorithm, swim trials are categorized into 8 exclusive categories according to the predominant swimming strategy. To simplify analysis of strategy transitions between subsequent trials, categories are combined to reduce their number to 4: “thigmotaxis & floating” = passive and wall oriented behavior, “random swim” = lack of detectable spatial selectivity, “scanning & chaining” = partial spatial selectivity, “focal & direct” = strong spatial selectivity. Each table row shows relative frequencies of a particular category depending on the category of the preceding trial, as well as for the very first trial of training.
In normal animals, first trials are often thigmotactic (floating is rare in normal mice) and there is a tendency to perseverate on this behavior. Once this is overcome, animals tend to oscillate between weakly and strongly spatial trials. 70% of random swims are followed by a spatially directed trial, indicating that one random hit of the goal is in many cases sufficient to remember its location at least approximately.

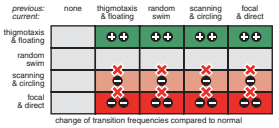
Predicted changes in mice trapped in thigmotaxis by behavioral inflexibility



change of transition frequencies compared to normal

If previous trial was thigmotactic probability of thigmotaxis increased, all other decreased. Similar but less pronounced pattern for very first trial. Transition pattern from non-thigmotactic trials should be same or similar as in control animals.

Predicted changes in mice “thigmotactic because they do not know where to go”



change of transition frequencies compared to normal

Independently of previous strategy reduction of spatial and increase of thigmotactic trials, but no change of random swim probability. Normal frequencies during very first trial when recall of spatial information is irrelevant.

Pattern of mildly to moderately impaired mice best explained by behavioral inflexibility

previous: / *current:*	none	thigmotaxis & floating	random swim	scanning & circling	focal & direct
thigmotaxis & floating	+4%	+18%	+1%	+1%	+1%
random swim	-2%	-18%	0%	+1%	+1%
scanning & circling	-13%	-22%	-10%	-3%	-5%
focal & direct	0%	-13%	+4%	-2%	0%

change of frequencies compared to normal (% of possible)

previous: / *current:*	none	thigmotaxis & floating	random swim	scanning & circling	focal & direct
thigmotaxis & floating	56%	56%	12%	7%	5%
random swim	25%	9%	20%	8%	5%
scanning & circling	11%	17%	31%	38%	26%
focal & direct	9%	18%	38%	48%	64%

transition frequencies (%)

Data from studies with significant impairment but standardized effect size on escape performance smaller than after hippocampal lesion. If previous trial was thigmotactic probability of thigmotaxis increased, all other decreased. Similar but less pronounced pattern for very first trial. No consistent decrease of frequency of spatial trials after non-thigmotactic trials.

Pattern of severely impaired mice reveals combination of inflexibility and spatial deficit

previous: / *current:*	none	thigmotaxis & floating	random swim	scanning & circling	focal & direct
thigmotaxis & floating	+34%	+65%	+24%	+20%	+35%
random swim	-33%	-66%	0%	+7%	+6%
scanning & circling	-75%	-72%	-37%	-20%	-19%
focal & direct	+2%	-56%	-25%	-36%	-53%

change of frequencies compared to normal (% of possible)

previous: / *current:*	none	thigmotaxis & floating	random swim	scanning & circling	focal & direct
thigmotaxis & floating	70%	81%	32%	24%	37%
random swim	17%	4%	20%	14%	11%
scanning & circling	3%	6%	22%	31%	22%
focal & direct	10%	9%	31%	31%	30%

transition frequencies (%)

Data from 7 studies with standardized effect size on escape performance equaling or exceeding hippocampal lesion. Strong changes as predicted by inflexibility hypothesis. In those animals that can overcome thigmotaxis, the pattern of strategy transitions is altered in a way that can best be explained by assuming a spatial orientation deficit.