

Post-Weaning Housing Conditions Influence Freezing during Contextual Fear Conditioning in Adult Rats

Natalie Schroyens, Christian Luis Bender, Joaquín Matias Alfei, Victor Alejandro Molina, Laura Luyten, Tom Beckers

-- Appendix A: Statistical Analyses performed in JASP (Version 0.8.5.1) --

This overview contains results of all preregistered statistical analyses (<https://osf.io/q92v8>):

1. The amnestic effect of MDZ in the standard housing (SH) group
→ *One-sided T-tests*
2. The influence of enriched housing (EH) on the amnestic effect of MDZ
→ *ANOVAs with factors Housing (EH vs. SH) and Treatment (SAL vs. MDZ)*
3. The influence of impoverished housing (IH) on the amnestic effect of MDZ
→ *ANOVAs with factors Housing (IH vs. SH) and Treatment (SAL vs. MDZ)*
4. Group differences in the temporal pattern of % freezing during the test session
→ *ANOVAs with BS factors Housing and Treatment and WS factor Time*
5. The influence of housing conditions on % freezing during pre- and post-shock period and reactivation.
→ *ANOVAs with factor Housing (EH, IH, SH)*
6. The influence of housing conditions on body weight
→ *ANOVA with factor Housing (EH, IH, SH) and ANOVA with WS factor Age and BS factor Housing (EH, IH, SH)*

1. The amnesic effect of MDZ in the standard housing (SH) group

A one-sided T-test (SAL > MDZ) was used to compare % freezing during the test session (first 5 min and complete 10-min session) between rats that received post-reactivation SAL vs. MDZ.

A. For the complete 10-min test session

Group Descriptives

	Group	N	Mean	SD	SE
T_10min	MDZ	8	30.83	7.828	2.767
	SAL	8	31.65	8.849	3.129

Independent Samples T-Test

	Test	Statistic	df	p	Effect Size	95% CI for Effect Size	
						Lower	Upper
T_10min	Student	-0.195	14.00	0.424	-0.097	-∞	0.727
	Mann-Whitney	28.000		0.360	-0.125	-∞	0.349

Note. For the Student t-test, effect size is given by Cohen's *d* ; for the Mann-Whitney test, effect size is given by the rank biserial correlation.

Note. For all tests, the alternative hypothesis specifies that group MDZ is less than group SAL .

Test of Normality (Shapiro-Wilk)

		W	p
T_10min	MDZ	0.956	0.769
	SAL	0.811	0.037

Note. Significant results suggest a deviation from normality.

Test of Equality of Variances (Levene's)

	F	df	p
T_10min	0.075	1	0.789

B. For the first 5 min of the test session

Group Descriptives

	Group	N	Mean	SD	SE
T_5min	MDZ	8	23.88	5.589	1.976
	SAL	8	29.46	9.020	3.189

Independent Samples T-Test

	Test	Statistic	df	p	Effect Size	95% CI for Effect Size	
						Lower	Upper
T_5min	Student	-1.488	14.00	0.079	-0.744	-∞	0.122
	Mann-Whitney	15.000		0.041	-0.531	-∞	-0.102

Note. For the Student t-test, effect size is given by Cohen's *d* ; for the Mann-Whitney test, effect size is given by the rank biserial correlation.

Note. For all tests, the alternative hypothesis specifies that group MDZ is less than group SAL .

Test of Normality (Shapiro-Wilk)

		W	p
T_5min	MDZ	0.902	0.301
	SAL	0.759	0.010

Note. Significant results suggest a deviation from normality.

Test of Equality of Variances (Levene's)

	F	df	p
T_5min	1.819	1	0.199

As a control, a two-sided T-test was used to assess whether there was a difference in % freezing during reactivation between rats of the SAL vs. MDZ group. In case of a between-group difference in % freezing during reactivation, we planned to perform a mixed ANOVA with within-subjects factor Session (reactivation vs. test) and between-subjects factor Treatment (SAL vs. MDZ).

Group Descriptives					
	Group	N	Mean	SD	SE
R_5min	MDZ	8	45.46	12.88	4.555
	SAL	8	49.46	12.17	4.303

Independent Samples T-Test							
	Test	Statistic	df	p	Effect Size	95% CI for Effect Size	
						Lower	Upper
R_5min	Student	-0.638	14.00	0.534	-0.319	-1.301	0.673
	Mann-Whitney	27.000		0.636	-0.156	-0.630	0.402

Note. For the Student t-test, effect size is given by Cohen's *d* ; for the Mann-Whitney test, effect size is given by the rank biserial correlation.

Test of Normality (Shapiro-Wilk)			
		W	p
R_5min	MDZ	0.798	0.027
	SAL	0.952	0.727

Note. Significant results suggest a deviation from normality.

Test of Equality of Variances (Levene's)			
	F	df	p
R_5min	0.888	1	0.362

2. The influence of enriched housing (EH) on the amnestic effect of MDZ

A two-way ANOVA with factors Housing (EH vs. SH) and Treatment (SAL vs. MDZ) was performed to assess the influence of enriched housing on the amnestic effect of MDZ on % freezing during test (first 5 min and complete 10-min session).

A. For the complete 10-min test session

Descriptives - T_10min				
Housing	Treatment	Mean	SD	N
EH	MDZ	35.75	8.822	8
	SAL	32.81	14.187	8
SH	MDZ	30.83	7.828	8
	SAL	31.65	8.849	8

ANOVA - T_10min							
Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	74.014	1	74.014	0.707	0.408	0.024	0.025
Treatment	9.031	1	9.031	0.086	0.771	0.003	0.003
Housing * Treatment	28.125	1	28.125	0.269	0.608	0.009	0.010
Residual	2930.715	28	104.668				

Note. Type III Sum of Squares

B. For the first 5 min of the test session

Descriptives - T_5min				
Housing	Treatment	Mean	SD	N
EH	MDZ	30.71	9.447	8
	SAL	28.08	12.414	8
SH	MDZ	23.88	5.589	8
	SAL	29.46	9.020	8

ANOVA - T_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	59.59	1	59.59	0.670	0.420	0.022	0.023
Treatment	17.50	1	17.50	0.197	0.661	0.006	0.007
Housing * Treatment	134.75	1	134.75	1.514	0.229	0.050	0.051
Residual	2491.79	28	88.99				

Note. Type III Sum of Squares

As a control, a one-way ANOVA was performed to assess whether there was a difference in % freezing during reactivation between the four groups (EH-SAL, EH-MDZ, SH-SAL, SH-MDZ).

Descriptives - R_5min

Group	Mean	SD	N
EH-MDZ	48.83	10.10	8
EH-SAL	49.25	12.15	8
SH-MDZ	45.46	12.88	8
SH-SAL	49.46	12.17	8

ANOVA - R_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Group	84.75	3	28.25	0.200	0.895	0.021	0.021
Residual	3946.81	28	140.96				

Note. Type III Sum of Squares

3. The influence of impoverished housing (IH) on the amnestic effect of MDZ

A two-way ANOVA with factors Housing (IH vs. SH) and Treatment (SAL vs. MDZ) was performed to assess the influence of impoverished housing on the amnestic effect of MDZ on % freezing during test (first 5 min and complete 10-min session).

According to a predefined exclusion criterium, 8 rats (3 IH-SAL, 5 IH-MDZ) were excluded since they showed freezing levels of <25% during the reactivation session. Statistical analyses were performed without these 8 rats (3.1) and with all rats included (3.2).

3.1 Analyses in which 8 rats were excluded

A. For the complete 10-min test session

Descriptives - T_10min

Housing	Treatment	Mean	SD	N
IH	MDZ	22.56	15.953	3
	SAL	18.37	2.777	5
SH	MDZ	30.83	7.828	8
	SAL	31.65	8.849	8

ANOVA - T_10min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	593.24	1	593.24	7.821	0.011	0.275	0.281
Treatment	14.55	1	14.55	0.192	0.666	0.007	0.010
Housing * Treatment	31.93	1	31.93	0.421	0.524	0.015	0.021
Residual	1516.95	20	75.85				

Note. Type III Sum of Squares

B. For the first 5 min of the test session

Descriptives - T_5min

Housing	Treatment	Mean	SD	N
IH	MDZ	19.78	18.446	3
	SAL	18.67	5.798	5
SH	MDZ	23.88	5.589	8
	SAL	29.46	9.020	8

ANOVA - T_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	282.99	1	282.99	3.530	0.075	0.144	0.150
Treatment	25.53	1	25.53	0.319	0.579	0.013	0.016
Housing * Treatment	57.21	1	57.21	0.714	0.408	0.029	0.034
Residual	1603.16	20	80.16				

Note. Type III Sum of Squares

As a control, a one-way ANOVA was performed to assess whether there was a difference in % freezing during reactivation between the four groups (IH-SAL, IH-MDZ, SH-SAL, SH-MDZ).

Descriptives - R_5min

Group	Mean	SD	N
IH-MDZ	40.44	9.335	3
IH-SAL	34.20	6.265	5
SH-MDZ	45.46	12.884	8
SH-SAL	49.46	12.171	8

ANOVA - R_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Group	772.7	3	257.6	2.036	0.141	0.234	0.234
Residual	2530.2	20	126.5				

Note. Type III Sum of Squares

3.2 Analyses in which all rats were included

A. For the complete 10-min test session

Descriptives - T_10min

Housing	Treatment	Mean	SD	N
IH	MDZ	16.50	10.729	8
	SAL	18.73	7.735	8
SH	MDZ	30.83	7.828	8
	SAL	31.65	8.849	8

ANOVA - T_10min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	1485.125	1	1485.125	18.888	< .001	0.400	0.403
Treatment	18.503	1	18.503	0.235	0.631	0.005	0.008
Housing * Treatment	4.014	1	4.014	0.051	0.823	0.001	0.002

ANOVA - T_10min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Residual	2201.632	28	78.630				

Note. Type III Sum of Squares

B. For the first 5 min of the test session

Descriptives - T_5min

Housing	Treatment	Mean	SD	N
IH	MDZ	12.83	12.553	8
	SAL	16.75	8.923	8
SH	MDZ	23.88	5.589	8
	SAL	29.46	9.020	8

ANOVA - T_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	1128.125	1	1128.125	12.900	0.001	0.300	0.315
Treatment	180.500	1	180.500	2.064	0.162	0.048	0.069
Housing * Treatment	5.556	1	5.556	0.064	0.803	0.001	0.002
Residual	2448.583	28	87.449				

Note. Type III Sum of Squares

As a control, a one-way ANOVA was performed to assess whether there was a difference in % freezing during reactivation between the four groups (IH-SAL, IH-MDZ, SH-SAL, SH-MDZ).

Descriptives - R_5min

Group	Mean	SD	N
IH-MDZ	22.29	16.53	8
IH-SAL	28.04	10.06	8
SH-MDZ	45.46	12.88	8
SH-SAL	49.46	12.17	8

ANOVA - R_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Group	4172	3	1390.5	8.078	<.001	0.464	0.464
Residual	4820	28	172.1				

Note. Type III Sum of Squares

Since there was a between-group difference in % freezing during reactivation, a mixed ANOVA with within-subjects factor Session (Reactivation vs. Test) and between-subjects factors Housing (IH vs. SH) and Treatment (SAL vs. MDZ) was conducted.

A. For the complete 10-min test session

Descriptives

Session	Housing	Treatment	Mean	SD	N
React	IH	MDZ	22.29	16.531	8
		SAL	28.04	10.058	8
	SH	MDZ	45.46	12.884	8
		SAL	49.46	12.171	8
Test 10 min	IH	MDZ	16.50	10.729	8
		SAL	18.73	7.735	8
	SH	MDZ	30.83	7.828	8
		SAL	31.65	8.849	8

Within Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Session	2260.210	1	2260.210	29.242	< .001	0.474	0.511
Session * Housing	300.444	1	300.444	3.887	0.059	0.063	0.122
Session * Treatment	45.002	1	45.002	0.582	0.452	0.009	0.020
Session * Housing * Treatment	0.111	1	0.111	0.001	0.970	0.000	0.000
Residual	2164.177	28	77.292				

Note. Type III Sum of Squares

Between Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	5160.03	1	5160.03	29.744	< .001	0.506	0.515
Treatment	163.63	1	163.63	0.943	0.340	0.016	0.033
Housing * Treatment	10.03	1	10.03	0.058	0.812	0.001	0.002
Residual	4857.40	28	173.48				

Note. Type III Sum of Squares

B. For the first 5 min of the test session**Descriptives**

Session	Housing	Treatment	Mean	SD	N
React	IH	MDZ	22.29	16.531	8
		SAL	28.04	10.058	8
	SH	MDZ	45.46	12.884	8
		SAL	49.46	12.171	8
Test 5 min	IH	MDZ	12.83	12.553	8
		SAL	16.75	8.923	8
	SH	MDZ	23.88	5.589	8
		SAL	29.46	9.020	8

Within Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Session	3885.444	1	3885.444	66.675	< .001	0.652	0.704
Session * Housing	434.028	1	434.028	7.448	0.011	0.073	0.210
Session * Treatment	0.063	1	0.063	0.001	0.974	0.000	0.000
Session * Housing * Treatment	11.674	1	11.674	0.200	0.658	0.002	0.007
Residual	1631.681	28	58.274				

Note. Type III Sum of Squares

Between Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	4669.444	1	4669.444	23.195	< .001	0.437	0.453
Treatment	370.562	1	370.562	1.841	0.186	0.035	0.062
Housing * Treatment	0.007	1	0.007	3.450e-5	0.995	0.000	0.000
Residual	5636.847	28	201.316				

Note. Type III Sum of Squares

4. Group differences in the temporal pattern of % freezing during the test session

In addition to the analyses described above, and based upon pilot data from two animals showing a substantial increase in freezing during the first minutes of testing, we also assessed **temporal changes throughout the 10-min test session**, using mixed repeated-measures ANOVAs including within-subjects factor Time (freezing per minute), and between-subjects factors Treatment and Housing. A graphical presentation of these data can be found in [Appendix C](#).

4.1 Analyses in which 8 rats were excluded

Within Subjects Effects

	Sphericity Correction	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Test min	Greenhouse-Geisser	20632 ^a	5.890 ^a	3502.7 ^a	10.242 ^a	< .001 ^a	0.204	0.231
Test min * Housing	Greenhouse-Geisser	6364 ^a	11.780 ^a	540.2 ^a	1.579 ^a	0.101 ^a	0.063	0.085
Test min * Treatment	Greenhouse-Geisser	1541 ^a	5.890 ^a	261.7 ^a	0.765 ^a	0.596 ^a	0.015	0.022
Test min * Housing * Treatment	Greenhouse-Geisser	4243 ^a	11.780 ^a	360.2 ^a	1.053 ^a	0.402 ^a	0.042	0.058
Residual	Greenhouse-Geisser	68492	200.264	342.0				

Note. Type III Sum of Squares

^a Mauchly's test of sphericity indicates that the assumption of sphericity is violated ($p < .05$).

Between Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	9931.4	2	4965.7	4.865	0.014	0.218	0.222
Treatment	385.8	1	385.8	0.378	0.543	0.008	0.011
Housing * Treatment	429.7	2	214.9	0.210	0.811	0.009	0.012
Residual	34705.9	34	1020.8				

Note. Type III Sum of Squares

4.2 Analyses in which all rats were included

Within Subjects Effects

	Sphericity Correction	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Test min	Greenhouse-Geisser	27674	^a 6.115	^a 4525.4	^a 15.188	^a < .001	^a 0.238	^a 0.266
Test min * Housing	Greenhouse-Geisser	7400	^a 12.231	^a 605.0	^a 2.031	^a 0.021	^a 0.064	^a 0.088
Test min * Treatment	Greenhouse-Geisser	1624	^a 6.115	^a 265.6	^a 0.891	^a 0.503	^a 0.014	^a 0.021
Test min * Housing * Treatment	Greenhouse-Geisser	3293	^a 12.231	^a 269.2	^a 0.904	^a 0.545	^a 0.028	^a 0.041
Residual	Greenhouse-Geisser	76528	256.843	298.0				

Note. Type III Sum of Squares

^a Mauchly's test of sphericity indicates that the assumption of sphericity is violated ($p < .05$).

Between Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	25209.074	2	12604.537	12.740	< .001	0.374	0.378
Treatment	0.145	1	0.145	1.462e -4	0.990	0.000	0.000
Housing * Treatment	570.185	2	285.093	0.288	0.751	0.008	0.014
Residual	41552.674	42	989.349				

Note. Type III Sum of Squares

5. The influence of housing conditions on % freezing during pre- and post-shock period and reactivation.

ANOVAs with factor Housing (EH vs. SH vs. IH) were performed to compare % freezing during training (pre- and post-shock period) and reactivation between the three housing conditions.

5.1 Analyses in which all rats were included

A. Effect of Housing on baseline freezing (Day 1)

Descriptives - pre_3 min

Housing	Mean	SD	N
EH	21.562	7.537	16
IH	4.861	5.265	16
SH	18.299	7.159	16

ANOVA - pre_3 min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	2507	2	1253.75	27.70	< .001	0.552	0.552
Residual	2037	45	45.26				

Note. Type III Sum of Squares

Post Hoc Comparisons - Housing

	Mean Difference	SE	t	Cohen's d	p _{tukey}
EH IH	16.701	2.378	7.022	1.014	< .001
SH	3.264	2.378	1.372	0.198	0.364
IH SH	-13.438	2.378	-5.650	-0.815	< .001

Note. Cohen's d does not correct for multiple comparisons.

pre_3min

Student-Newman-Keuls^{a,b}

Housing	N	Subset	
		1	2
IH	16	4,861111111	
SH	16		18,29861111
EH	16		21,5625000
Sig.		1.000	.177

Means for groups in homogeneous subsets are displayed.
Based on observed means.
The error term is Mean Square(Error) = 45.256.
a. Uses Harmonic Mean Sample Size = 16.000.
b. Alpha = 0.05.

Student Newman-Keuls post-hoc tests (SPSS Statistics) suggest that IH rats freeze significantly less during baseline freezing, compared to SH and EH rats.

B. Effect of Housing on post-shock freezing (Day 1)

Descriptives - post3

Housing	Mean	SD	N
EH	57.50	18.31	16
IH	35.50	19.41	16
SH	65.13	19.45	16

ANOVA - post3

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	7572	2	3786.1	10.42	< .001	0.317	0.317
Residual	16352	45	363.4				

Note. Type III Sum of Squares

Post Hoc Comparisons - Housing

	Mean Difference	SE	t	Cohen's d	p _{tukey}
EH IH	22.000	6.740	3.264	0.471	0.006
SH	-7.625	6.740	-1.131	-0.163	0.500
IH SH	-29.625	6.740	-4.396	-0.634	< .001

Note. Cohen's d does not correct for multiple comparisons.

post3

Student-Newman-Keuls^{a,b}

Housing	N	Subset	
		1	2
IH	16	35,50	
EH	16		57,50
SH	16		65,13
Sig.		1.000	.264

Means for groups in homogeneous subsets are displayed.
Based on observed means.
The error term is Mean Square(Error) = 363.372.
a. Uses Harmonic Mean Sample Size = 16.000.
b. Alpha = 0.05.

Student Newman-Keuls post-hoc tests suggest that IH rats freeze significantly less after the third shock, compared to SH and EH rats.

C. Effect of Housing on freezing during reactivation (Day 2)

Descriptives - R_5min

Housing	Mean	SD	N
EH	49.04	10.80	16
IH	25.17	13.55	16
SH	47.46	12.28	16

ANOVA - R_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	5704	2	2851.8	18.97	<.001	0.457	0.457
Residual	6765	45	150.3				

Note. Type III Sum of Squares

Post Hoc Comparisons - Housing

	Mean Difference	SE	t	Cohen's d	p _{tukey}
EH IH	23.875	4.335	5.508	0.795	<.001
SH	1.583	4.335	0.365	0.053	0.929
IH SH	-22.292	4.335	-5.142	-0.742	<.001

Note. Cohen's d does not correct for multiple comparisons.

R_5min

Student-Newman-Keuls^{a,b}

Housing	N	Subset	
		1	2
IH	16	25,1666667	
SH	16		47,4583333
EH	16		49,0416667
Sig.		1.000	.717

Means for groups in homogeneous subsets are displayed.

Based on observed means.

The error term is Mean Square(Error) = 150.330.

a. Uses Harmonic Mean Sample Size = 16.000.

b. Alpha = 0.05.

Student Newman-Keuls post-hoc tests suggest that IH rats freeze significantly less during reactivation, compared to SH and EH rats.

5.2 Analyses in which 8 rats were excluded

The preregistration of the current study (<https://osf.io/8ezmq/register/565fb3678c5e4a66b5582f67>) contained the following exclusion criterion: 'Rats that freeze less than 25% during the reactivation session will be excluded from the analysis. Explorative analyses including all subjects will also be performed.' The aim of this predefined criterion was to exclude rats that did not sufficiently acquire the context-shock association because this could have hampered the investigation of memory interference.

On the other hand, in order to study the effect of housing conditions on % freezing during training and reactivation (preregistered under 'exploratory analyses'), it is not necessarily relevant to exclude rats that show low freezing during reactivation. Nevertheless, since the exclusion criterion was included in the preregistration and applied for analyzing other freezing data, we present the results of these analyses based on this subset of the data as well. The impairing influence of IH on baseline freezing is significant in this subset of rats as well.

A. Effect of Housing on baseline freezing (Day 1)

Descriptives - pre_3 min

Housing	Mean	SD	N
EH	21.562	7.537	16
IH	3.472	3.892	8
SH	18.299	7.159	16

ANOVA - pre_3 min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	1819	2	909.42	19.49	<.001	0.513	0.513
Residual	1727	37	46.67				

Note. Type III Sum of Squares

Post Hoc Comparisons - Housing

	Mean Difference	SE	t	Cohen's d	p _{tukey}
EH IH	18.090	2.958	6.115	0.967	<.001
SH	3.264	2.415	1.351	0.214	0.375
IH SH	-14.826	2.958	-5.012	-0.792	<.001

Note. Cohen's d does not correct for multiple comparisons.

pre_3min

Student-Newman-Keuls^{a,b,c}

Housing	N	Subset	
		1	2
IH	8	3,4722222	
SH	16		18,2986111
EH	16		21,5625000
Sig.		1.000	.249

Means for groups in homogeneous subsets are displayed.

Based on observed means.

The error term is Mean Square(Error) = 46.671.

a. Uses Harmonic Mean Sample Size = 12.000.

b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

c. Alpha = 0.05.

Student Newman-Keuls post-hoc tests suggest that IH rats freeze significantly less during baseline, compared to SH and EH rats.

B. Effect of Housing on post-shock freezing (Day 1)

Descriptives - post3

Housing	Mean	SD	N
EH	57.50	18.31	16
IH	45.50	18.32	8
SH	65.13	19.45	16

ANOVA - post3

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	2065	2	1032.7	2.928	0.066	0.137	0.137
Residual	13050	37	352.7				

Note. Type III Sum of Squares

C. Effect of Housing on freezing during reactivation (Day 2)

Descriptives - R_5min

Housing	Mean	SD	N
EH	49.04	10.797	16
IH	36.54	7.601	8
SH	47.46	12.282	16

ANOVA - R_5min

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	897.4	2	448.7	3.760	0.033	0.169	0.169
Residual	4415.9	37	119.3				

Note. Type III Sum of Squares

Post Hoc Comparisons - Housing

	Mean Difference	SE	t	Cohen's d	p _{tukey}
EH IH	12.500	4.731	2.642	0.418	0.031
SH	1.583	3.862	0.410	0.065	0.911
IH SH	-10.917	4.731	-2.308	-0.365	0.067

Note. Cohen's d does not correct for multiple comparisons.

R_5min			
Student-Newman-Keuls ^{a,b,c}			
Housing	N	Subset	
		1	2
IH	8	36,5416667	
SH	16		47,4583333
EH	16		49,0416667
Sig.		1.000	.725

Means for groups in homogeneous subsets are displayed.

Based on observed means.

The error term is Mean Square(Error) = 119.349.

a. Uses Harmonic Mean Sample Size = 12.000.

b. The group sizes are unequal. The harmonic mean of the group sizes is used. Type I error levels are not guaranteed.

c. Alpha = 0.05.

Student Newman-Keuls post-hoc tests suggest that IH rats freeze significantly less during reactivation, compared to SH and EH rats.

6. The influence of housing conditions on body weight

An ANOVA with factor Housing (EH vs. SH vs. IH) was performed to assess whether housing conditions influenced body weight as measured before the start of the fear-conditioning protocol (PND66).

Descriptives - Weight PND66

Housing	Mean	SD	N
EH	286.4	19.12	16
IH	215.8	18.57	16
SH	270.8	12.16	16

ANOVA - Weight PND66

Cases	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	44011	2	22005.6	76.90	<.001	0.774	0.774
Residual	12877	45	286.2				

Note. Type III Sum of Squares

PND66					
Student-Newman-Keuls ^{a,b}	Group	N	Subset		
			1	2	3
.b	IH	16	215.81		
	SH	16		270.75	
	EH	16			286.44
	Sig.		1.000	1.000	1.000

Post Hoc Comparisons - Housing

	Mean Difference	SE	t	Cohen's d	p _{tukey}
EH IH	70.62	5.981	11.809	1.704	<.001
SH	15.69	5.981	2.623	0.379	0.031
IH SH	-54.94	5.981	-9.186	-1.326	<.001

Note. Cohen's d does not correct for multiple comparisons.

Student Newman-Keuls post-hoc tests suggest that all housing conditions differ significantly in body weight at PND 66.

A mixed ANOVA with within-subjects factor Age and between-subject factor Housing (EH vs. SH vs. IH) was performed to assess whether housing conditions influenced the increase in body weight throughout development.

Within Subjects Effects

	Sphericity Correction	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Age	Greenhouse-Geisser	3.100e +6 ^a	1.934 ^a	1.602e +6 ^a	4664.16 ^a	< .001 ^a	0.962	0.990
Age * Housing	Greenhouse-Geisser	90982 ^a	3.869 ^a	23516.70 ^a	68.46 ^a	< .001 ^a	0.028	0.753
Residual	Greenhouse-Geisser	29904	87.049	343.53				

Note. Type III Sum of Squares

^a Mauchly's test of sphericity indicates that the assumption of sphericity is violated (p < .05).

Between Subjects Effects

	Sum of Squares	df	Mean Square	F	p	η^2	η^2_p
Housing	150280	2	75140	56.80	< .001	0.716	0.716
Residual	59533	45	1323				

Note. Type III Sum of Squares