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# **Supplemental information**

# A behavioral paradigm for measuring

### perceptual distances in mice

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### **Supplemental Information**

#### Table S1. Odors used in the experiments, Related to Figures 1, 2, and 3

Set	Odor name	Abbreviation	Liquid	Headspace	Air dilution
<u> </u>	Cippomoldobydo	CinAld			Tange
1			0.0004	0.9 X 10	
	Ethyl butyrate	EB	0.0008	9.1 X 10 <sup>-7</sup>	
	2-Methylbutyric acid	2MBAcd	0.0004	8.9 x 10 <sup>-9</sup>	
	2,2-Dimethylbutyric acid	22DMBAcd	0.0004	2.3 x 10 <sup>-9</sup>	0.01 - 0.05
	Cyclopentanecarboxylic acid	CPAcd	0.0012	1.1 x 10 <sup>-8</sup>	0.01 0.00
	2-Heptanone	2Hep	0.0002	4.2 x 10 <sup>-8</sup>	
	Isobutyric acid	IBAcd	0.0004	7.6 x 10 <sup>-8</sup>	
	Isovaleric acid	IVAcd	0.0004	1.6 x 10 <sup>-8</sup>	
2	3-Heptanone	ЗНер	0.001	1.8 x 10 <sup>-7</sup>	
	Methylvalerate	MVT	0.0002	1.6 x 10 <sup>-7</sup>	
	Ethyl butyrate	EB	0.0018	2.0 x 10 <sup>-6</sup>	
	Propionic acid	PpAcd	0.0063	2.0 x 10 <sup>-7</sup>	0.02 - 0.1
	Butyric acid	ButAcd	0.001	4.5 x 10 <sup>-8</sup>	
	(+)-α-Pinene	Pinene	0.001	2.9 x 10 <sup>-7</sup>	
	Benzaldehyde	BzAld	0.0004	4.9 x 10 <sup>-8</sup>	
	5-Methyl-2-Hexanone	5M2H	0.0002	6.9 x 10 <sup>-8</sup>	
3	3-Heptanone	ЗНер	0.001	1.8 x 10 <sup>-7</sup>	
	Ethyl butyrate	EB	0.0018	2.0 x 10 <sup>-6</sup>	
	Valeric acid	ValAcd	0.0002	3.5 x 10 <sup>-9</sup>	
	3-Methylvaleric acid	3MVAcd	0.0002	2.3 x 10 <sup>-9</sup>	
	3,3-Dimethylbutyric acid	33DMBAcd	0.0002	4.6 x 10 <sup>-9</sup>	0.01 – 0.05
	(+)-alpha-Pinene	Pinene	0.001	2.9 x 10 <sup>-7</sup>	
	Benzaldehyde	BzAld	0.0004	4.9 x 10 <sup>-8</sup>	
	Isovaleric acid	IVAcd	0.0002	7.8 x 10 <sup>-9</sup>	
	4-Methylvaleric acid	4MVAcd	0.0002	2.1 x 10 <sup>-9</sup>	
	Hexanoic acid	HexAcd	0.0002	2.8 x 10 <sup>-9</sup>	

\* - Headspace concentration was approximately estimated based on dilution and saturated vapor pressure of a chemical

Set	# of mice	# of trials	# of sessions	Trial # / session
				(mean ± std)
1	10	75195	308	244.1 ± 73.2
2	6	24295	107	227.1 ± 75.0
3	12	58232	259	224.8 ± 80.3

Table S2. Trial statistics of different odor sets, Related to Figures 1, 2, and 3



**Figure S1. Experimental setup, Related to Method Details session.** *Left:* the odor delivery system. Odors are delivered using a two-cassette air dilution olfactometer. Each cassette has 8 odor vials and two Mass Flow Controllers (MFCs) for flow ranges of 0-1000 ml/min and 0-100 ml/min. To prepare an odor, a pair of valves for a single odor vial is opened and an odorized air flow (1000 ml/min) is first directed to the exhaust via the final valve. Clean air of the same flow is delivered to the odor port. After approximately 1 sec of the flow stabilization, the final valve redirects the odorized flow to the odor port and the clean air to the exhaust. The concentration is controlled by the ratio of the MFC flow rates. To deliver a binary mixture, two vials from different cassettes are opened simultaneously. The air is continuously pumped away from the odor port with the same air flow rate. *Right:* behavioral setup. A mouse is positioned on a freely rotating running wheel with its head fixed. The mouse snout is placed in the odor port. The water delivery spouts are positioned below the odor port opening.



**Figure S2.** Probability of a no-go choice by odor identity and concentrations, Related to Figure 1. Trials were assigned to a specific odor identity if that odor was presented either on first or second presentations. Circles correspond to individual mice, and bars are averages across mice (n = 10 mice, 75195 trials, odor set #1 (Table S1)).



Figure S3. Relationship between distance metric and logit of probability of nogo choice, Related to Figure 3. a. plot showing the dependence of  $logit(P_{nogo})$  (left-hand side of (Eq. 2)) on the distance metric between odor pairs. The x-axis represents percentile of distance metric across all odor pairs. b. same as in **a** for (Eq. 3).

**a:** 
$$D(A, B) = 1 - \frac{1 - P_{nogo}(A, B)}{\sqrt{(1 - P_{nogo}(A, A))(1 - P_{nogo}(B, B))}}$$
  
**b:**  $D(A, B) = P_{nogo}(A, B)$   
**c:**  $D(A, B) = \max\left(P_{nogo}(A, B) - P_{nogo}(A, A), P_{nogo}(A, B) - P_{nogo}(B, B)\right)$ 



**Figure S4. Comparison of regression analysis for different distance metrics, Related to Figure 3 a,b.** Absolute value of regression coefficients in the logistic regression model (**Eq.2**) for odor identity (odor), concentration of the first (conc 1) and the second (conc 2) presented odors, earlier vs later trials in a session (trials), and sequence of odor presentation A->B vs B->A (seq) (n = 10 mice, 75195 trials) for **a**, original distance metric **Eq.1** (the same as **Fig. 3a**); **b**, for distance metric;  $D(A, B) = P_{nogo}(A, B)$ ; **c**, for distance metric  $D(A, B) = \max \left( P_{nogo}(A, B) - P_{nogo}(A, A), P_{nogo}(A, B) - P_{nogo}(B, B) \right)$