

## *Supplementary Information*

# **Aversive gustatory learning and perception in honey bees**

---

**Marie Guiraud<sup>1,‡</sup>, Lucie Hotier<sup>1</sup>, Martin Giurfa<sup>1,\*</sup> and María Gabriela de Brito Sanchez<sup>1,\*</sup>**

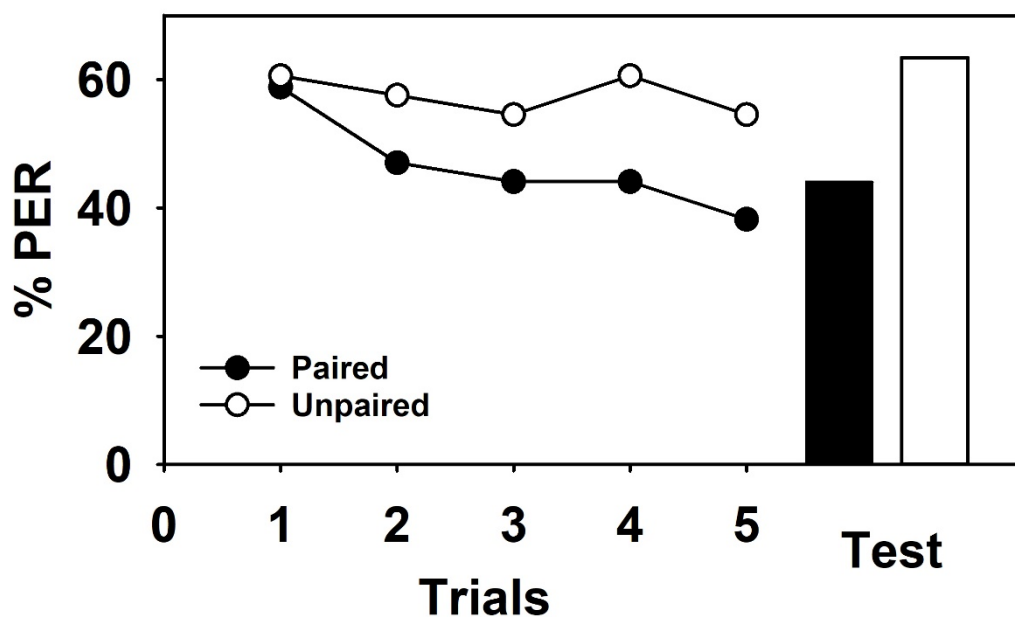
---

\*senior authorship shared

<sup>1</sup>: *Centre de Recherches sur la Cognition Animale (CRCA), Centre de Biologie Intégrative (CBI), Université de Toulouse; CNRS, UPS, 31062 Toulouse cedex 9, France*

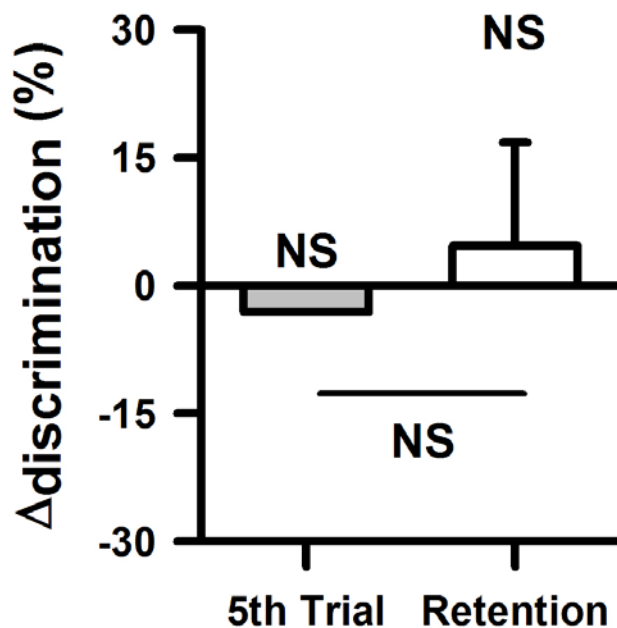
<sup>‡</sup>: *Current address: Queen Mary University of London, School of Biological and Chemical Sciences, Biological and Experimental Psychology, Mile End Road, London E1 4NS, United Kingdom*

Figure S1



**Figure S1: Appetitive responses (PER) to sucrose of bees from Fig. 2a.** The responses of a paired group trained with sucrose solution contingent with electric shock (black circles;  $n = 34$ ) and of an unpaired group presented with sucrose non-contingent with electric shock (white circles;  $n = 33$ ) during 5 trials. PER to sucrose never reached typical levels observed in foragers trained appetitively (90-100%); it remained high (between 55% and 60%) and constant along trials in the unpaired group ( $F_{4,128} = 0.15$ ;  $P = 0.96$ ). In the paired group, levels of PER (between 40% and 60%) were also lower than typical response levels and did not vary along trials ( $F_{4,132} = 1.12$ ;  $P = 0.35$ ). No differences were observed between both groups ( $F_{1,65} = 2.03$ ;  $P = 0.16$ ). One hour after conditioning, PER to sucrose was lower in the paired group (44%) compared to that of the unpaired group (64%) but the difference was again not significant ( $F_{1,65} = 2.59$ ;  $P = 0.11$ ).

Figure S2



**Figure S2: Control experiment showing that antennal side is not learned *per se* in differential gustatory SER conditioning.** Bees ( $n = 64$ ) were presented with a single tastant, sucrose solution 1.0 M, delivered alternately to the left and the right antenna, with only one antennal side being associated with shock. The  $\Delta_{\text{discrimination}}$  indexes obtained for the last conditioning trial and for the retention test were not different from zero ( $t_{63} = 0.50$ ;  $P = 0.62$  and  $t_{63} = 0.77$ ;  $P = 0.44$ , respectively) and did not differ from each other ( $t_{63} = 0.96$ ;  $P = 0.34$ ). Thus, bees were unable to learn the discrimination between sucrose punished and sucrose non-punished based on antennal side, showing that this variable was not learned during gustatory SER conditioning.