SUPPLEMENTAL INFORMATION

Molecular Annotation of Integrative Feeding Neural Circuits

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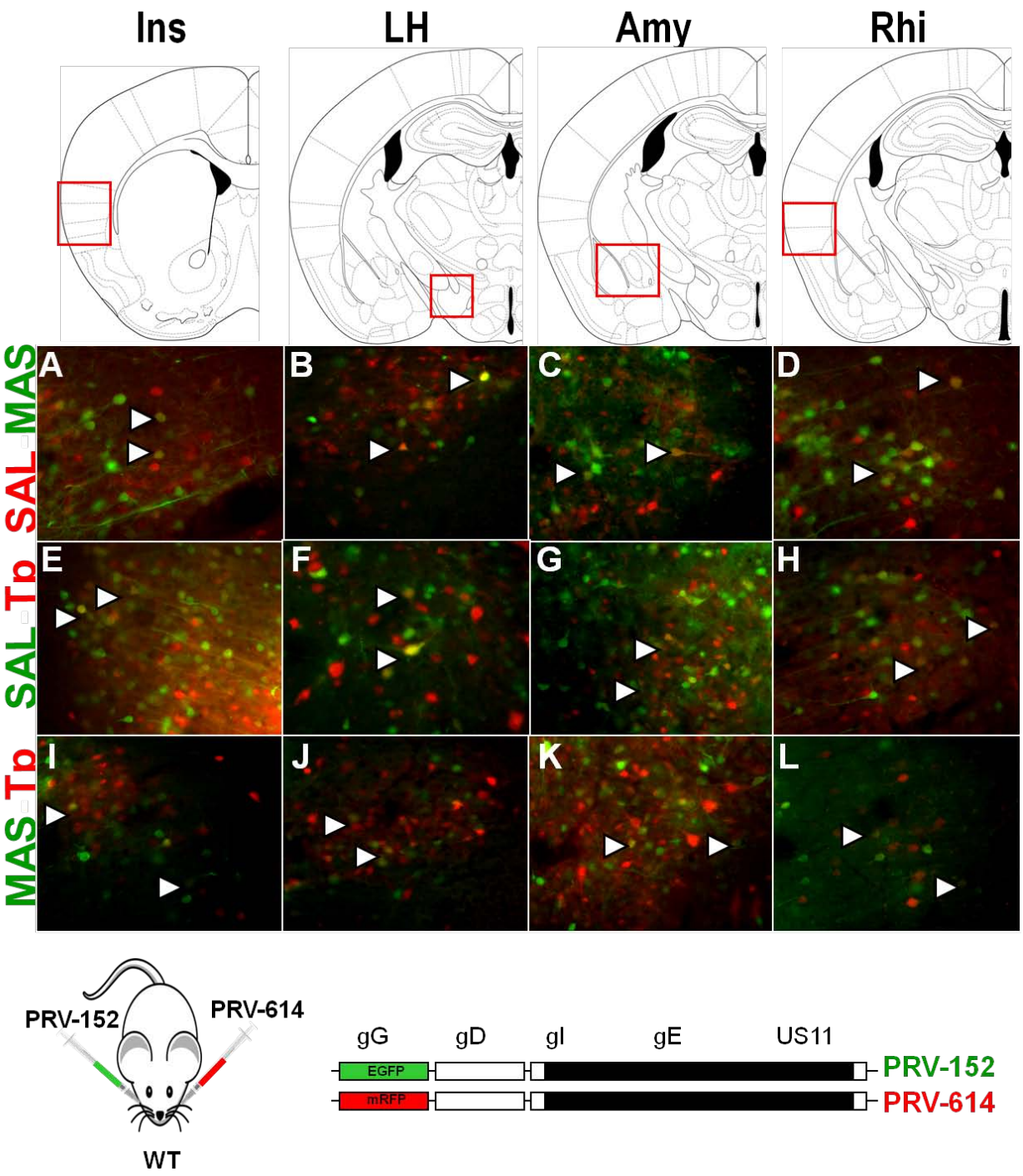

Figure S1 (related to Fig. 2)

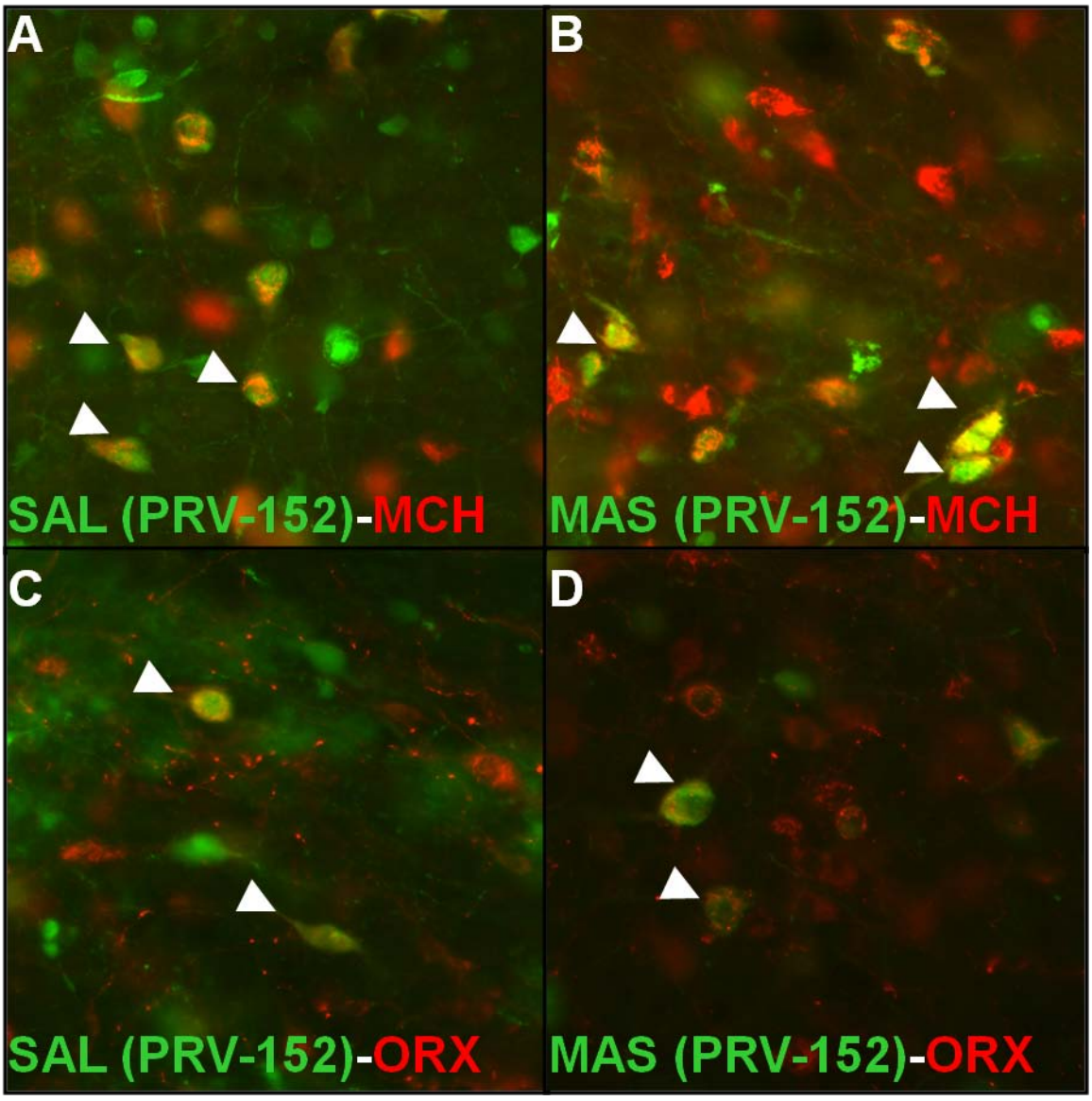

Figure S2 (related to Fig. 3)

Figure S1 (related to Fig. 2): Dual PRV Labeling Reveals Neurons Integrated Into Multiple Neuronal Circuits

Two isogenic PRV strains encoding different reporters (see scheme at the bottom), PRV-152 (encoding EGFP) and PRV-614 (encoding mRFP) were simultaneously injected in pair-wise combinations of SAL, MAS or T_P tissues. Double injections for each of the three pair-wise tissue combinations, SAL-MAS (**3A-D**), SAL-T_P (**3E-H**) and MAS-T_P (**3I-L**), were performed and the distribution of EGFP and mRFP signals was analyzed 96-120h post injection in the Ins (**3A, E, I**), LH (**3B, F, J**), Amy (**3C, G, K**) and Rhi (**3D, H, L**) by determination of EGFP immunofluorescence (in green) and mRFP intrinsic red fluorescence, respectively. Double labeled neurons were evident in all the regions studied (see arrowheads). There were also neurons that showed only EGFP signal or the mRFP signal and neurons that were unlabeled.

Figure S2 (related to Fig. 3): MCH and Orexin are Markers of Lateral Hypothalamus Neurons Projecting to the Salivary Gland and the Masseter Muscle

To determine the phenotype of LH neurons infected by PRV following injection into SAL and MAS, we used immunohistochemistry against PRV-encoded EGFP and either MCH or orexin. These studies revealed that both MCH^+ (panels **A** and **B**) and $orexin^+$ (panels **C** and **D**) neurons were infected following PRV-injection into the salivary gland (panels **A** and **C** respectively) and masseter muscle (panels **B** and **D** respectively).

Colocalization of PRV and EGFP signals

BAC Driving EGFP Expression	PRV Injection Site	
	MAS	SAL
Nurr1	Ins, Amy	Ins, Amy
Cnr1	-	Ins
Gabrb2	-	Ins
Elav3	-	Rhi
Lhx6	Ins	-
Lrig2	Ins	Ins
Hnrpab	-	-
6430573F11Rik	-	-
BC056494	-	-
Lamb3	-	-
Absb1b	-	-
Htr5b	-	-
Drd1a	-	-
Ppp1cc	-	-
Cdh1	-	-
Sema3c	-	-
AI427653	-	-

Table S1 (related to Fig. 4): PRV Infection of Transgenic EGFP Mouse Lines Reveals Markers for Feeding Neuronal Circuits

List of EGFP transgenic mice strains used in this study. 11 out of 17 transgenic mouse lines studied failed to be useful in defining molecular markers while 6 transgenic mouse lines displayed “yellow” neurons following PRV injections. In terms of sites of PRV injection, in some cases there was colocalization only after PRV injection into the MAS (Lhx6) or the SAL (Cnr1, Gabrb2, Elav3) and in the case of Nurr1 and Lrig2 colocalization was detected after injection in both tissues. In regard to the brain region were colocalization was detected, in some cases only one region was positive (Insular cortex: Lhx6, Lrig2, Gabrb2, Cnr1; Rhinal cortex: Elav3), and in one case two regions displayed “yellow” neurons (Insular cortex and Amygdala: Nurr1).