

SUPPLEMENTARY DATA

Glucagon-like peptide-1 acts in the dorsal raphe and modulates central serotonin to reduce appetite and body weight.

Anderberg et al.

Supplementary Table 1. TaqMan probe and primer sets for target genes examined in the hypothalamus.

Gene symbol	Gene name	Assay ID
Htr1a	5-hydroxytryptamine (serotonin) receptor 1A	Rn00561409_s1
Htr2a	5-hydroxytryptamine (serotonin) receptor 2A	Rn00568473_m1
Htr2c	5-hydroxytryptamine (serotonin) receptor 2C	Rn00562748_m1
Htr3a	5-hydroxytryptamine (serotonin) receptor 3A	Rn00667026_m1

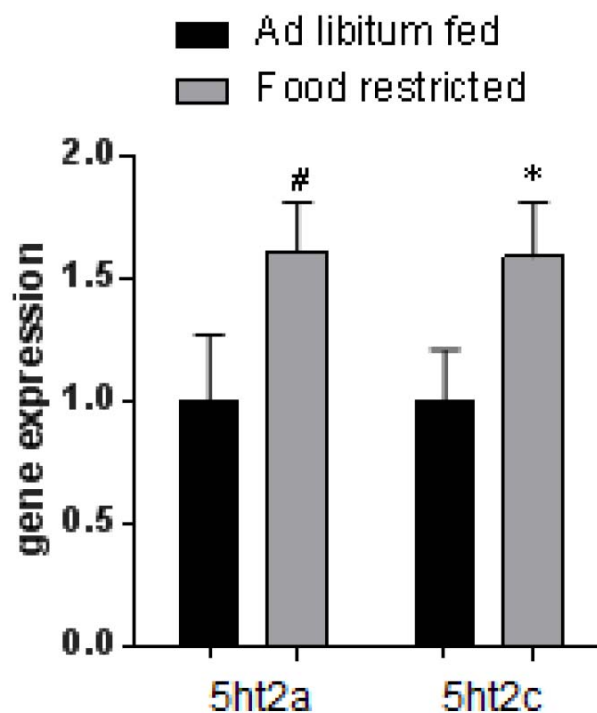
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Supplementary Table 2. Antibodies, manufacturers, and dilutions. Grey background indicates antibodies used in the YFP-mice. In yellow antibodies used to confirm serotonergic identity of neurons after whole-cell electrophysiology in rats.

Antibody name	Manufacturer	Lot #	Dilution
YFP	Abcam, Cambridge, UK	ab13970	1/600
TPH	Sigma-Aldrich, USA	T0678	1/500
Alexa Fluor 488®	Abcam	ab150169	1/400
Alexa Fluor 568®	Abcam	ab175700	1/800
Cy3-donkey anti-mouse	Jackson Immuno Research	715-165-150	1/200
TPH	Sigma-Aldrich, USA	T0678	1/500
Alexa Fluor 488-streptavidin	Invitrogen	S11223	1/200

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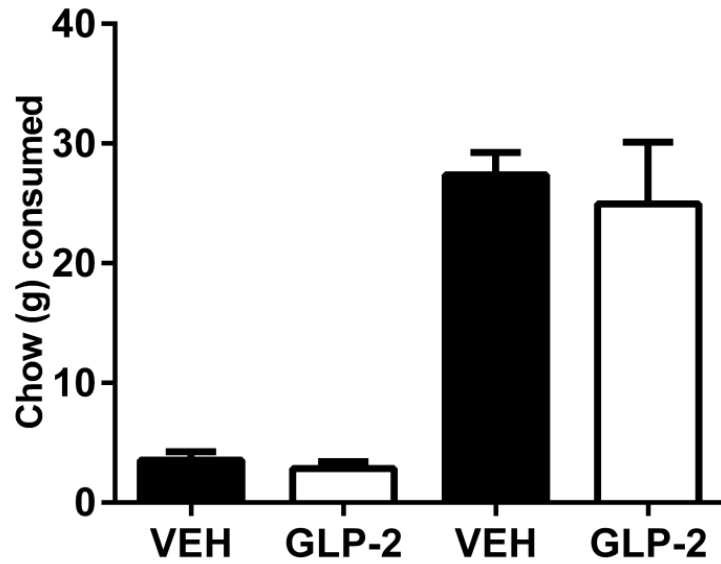
Supplementary Figure S1. The food restricted rats pair-fed to the amount eaten by the EX4-treated rats did not receive central injections. In order to confirm that the difference in serotonin receptor expression persists if the cannulation/injection state of the rats is matched an extra control group of rats not injected or cannulated was tested along with the rats whose intake was restricted to the amount eaten by EX4-injected rats. The new data show that the increased expression of 5ht2a and 5ht2c receptors can still be found when both the control group and the food-restricted group do not receive injections or cannulation surgery. The 50% increase in mRNA expression of the 2A and 2C receptors is similar to that presented in Figure 1B. This control experiment suggests that the change in serotonin receptor expression is not brain injection-dependent. # $p=0.06$, * $p<0.05$



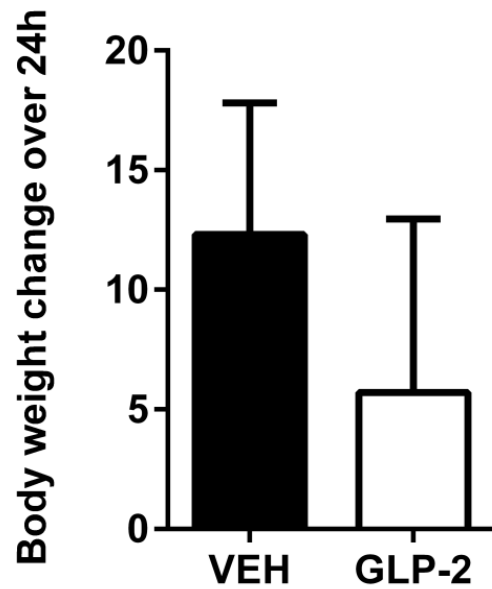
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Supplementary Figure S2. Dorsal raphe GLP-2 injections did not change food intake or body weight. Neither 1h nor 24h food intake was significantly altered by DR-directed GLP-2 microinjection (A). Body weight was also not changed by the treatment (B). GLP-2 was injected at a dose of 2 μ g/0.2 μ L.

A



B



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Supplementary Figure S3 (Video file). Z-line three dimensional reconstruction of a confocal image stack shown an example of a YFP-innervated neuron. Displayed here Z-stack was collected from 6 layers taken at 1uM apart. The movie shows a three dimensional spin around the vertical axis.

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Supplementary Figure S4. Immunohistochemical labeling for YFP in YFP–PPG neurons (green) and 5-HT in serotonin neurons (red) in coronal sections through the DR nucleus of YFP–PPG mice. Low magnification micrographs showing the rostro-caudal extent of the DR examined (A-C) with a rat atlas based overlay indicating subnuclei of the DR. Groups of red 5-HT-immunoreactive cell bodies are shown at higher magnification in D-F. Little to no immunoreactive YFP fibers are found in the ventral sections of the DR (G-I), however the few individual neurons determined to receive YFP-labeled innervation are displayed in J-M.

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