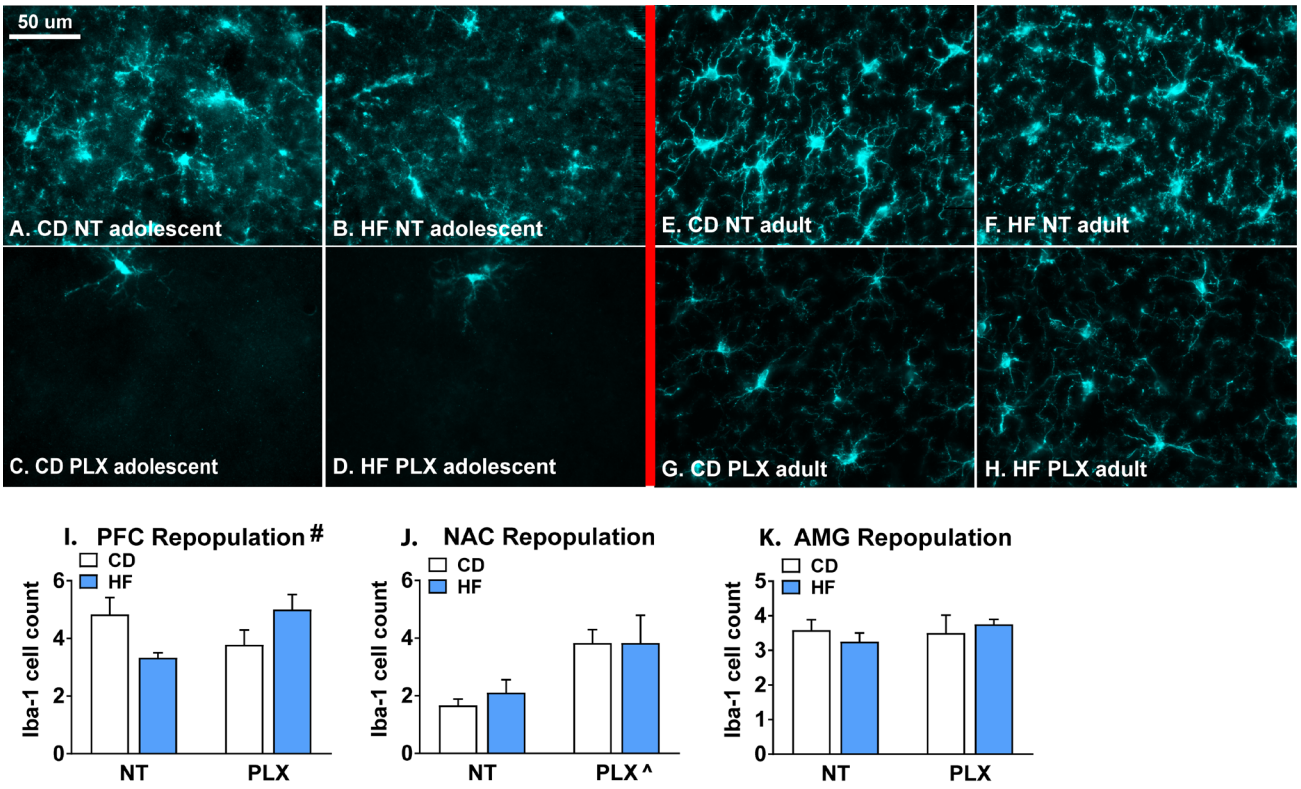

Figure S1: Representative 40x images of Iba-1 immunohistochemistry in the medial PFC of (**A-D**) adolescent males on the last day of PLX treatment, with microglia present in (**A**) CD NT and (**B**) HF NT but nearly depleted from (**C**) CD PLX and (**D**) HF PLX tissue. (**E-H**) Adult males after 3 weeks recovery from PLX, at the age when experimental animals started operant training, depicting (**E**) CD NT (**F**) HF NT microglia and repopulated microglia in (**G**) CD PLX and (**H**) HF PLX tissue. (I) In the PFC after repopulation, HF diet differentially affected male Iba1 cell counts based on PLX treatment. (J) In the NAC after repopulation, PLX treatment increased male Iba1 cell counts. (K) In the AMG after repopulation, male Iba1 cell counts were not affected by HF diet or PLX treatment. Tissue was labeled with Iba-1 antibody from (A-D) Synaptic Systems and (E-K) Wako. (#$p < 0.05$ diet x treatment interaction; ^ $p < 0.05$ main effect of PLX; n = 3/group) (HF = offspring of maternal high fat diet; CD = offspring of maternal control diet; PLX = adolescent PLX3397 treatment; NT = not treated during adolescence)

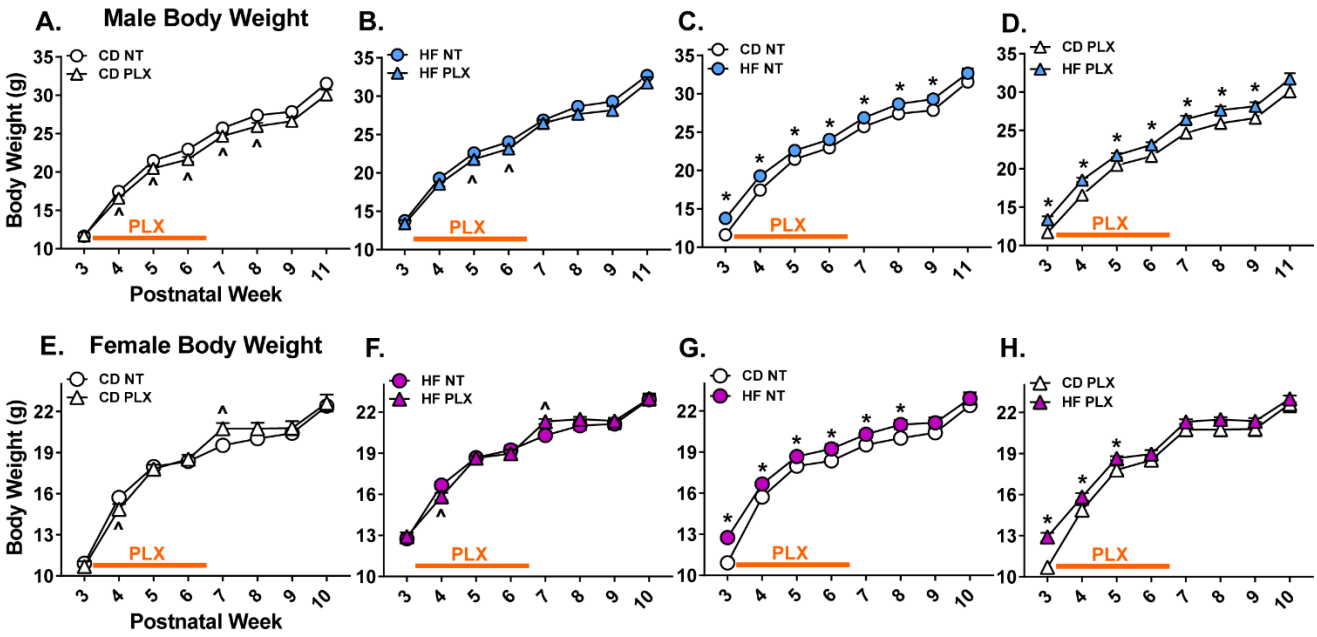

Figure S2: (**A-D**) For clarity, data are presented in two ways; (1) demonstrating the PLX effect (A, B, E, F) and (2) demonstrating the HF diet effect (C, D, G, H). Male offspring body weights from weaning until the start of food restriction at 11 weeks of age showed a significant between-subjects main effect of treatment [$F_{(1,58)}$ = 7.1; p = 0.01] and diet [$F_{(1,58)}$ = 19.9; p < 0.001] and a significant within-subjects effect of week [$F_{(7, 406)}$ = 2892.1; p < 0.001]. PLX treatment decreased male body weight at postnatal weeks (**A**) 4-8 in CD and (**B**) 5-6 weeks in HF offspring (p < 0.05). HF male offspring were heavier than their CD counterparts at 3-9 weeks of age, both in (**C**) NT and (**D**) PLX groups (p < 0.05). By 11 weeks of age when males initiated food restriction for operant testing, there were no differences between groups. (**E-H**) Female offspring body weights revealed a significant between-subjects main effect of diet [$F_{(1,58)}$ = 11.3; p = 0.001] and significant within-subjects effects of week [$F_{(7, 406)}$ = 1507.8; p < 0.001], week*treatment [$F_{(7, 406)}$ = 10.2; p < 0.001], and week*diet [$F_{(7, 406)}$ = 7.4; p < 0.001] interactions. PLX decreased female body weight at postnatal week 4 and increased it on week 7 in both the (**E**) CD and (**F**) HF groups (p < 0.05). HF female offspring were heavier than the respective CD mice at postnatal (**G**) weeks 3-8 in NT groups and (**H**) weeks 3-5 in PLX groups (p < 0.05). By 10 weeks of age when female mice initiated food restriction for operant testing, there were no differences between groups. (*p < 0.05 vs. respective CD group; ^ p < 0.05 vs. respective NT group; n = 15-16/group) (HF = offspring of maternal high fat diet; CD = offspring of maternal control diet; PLX = adolescent PLX3397 treatment; NT = not treated during adolescence)

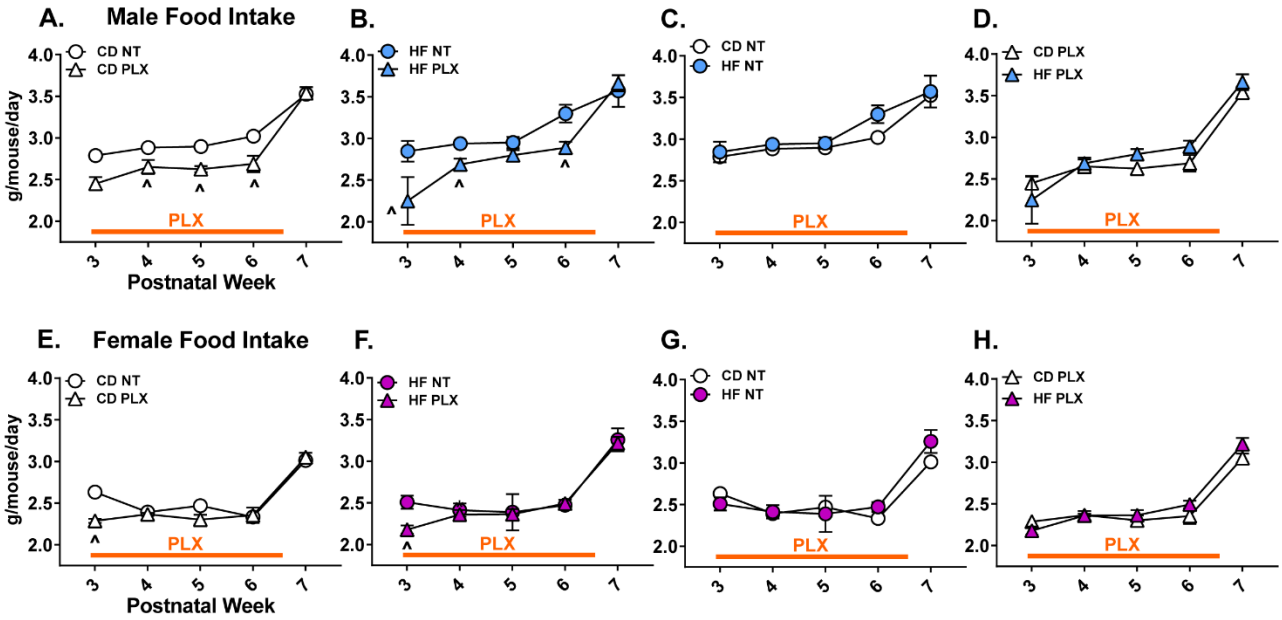

Figure S3: (**A-D**) For clarity, data are presented in two ways; (1) demonstrating the PLX effect (A, B, E, F) and (2) demonstrating the HF diet effect (C, D, G, H). For food intake in the males, there were significant within-subjects effects of week [$F_{(4, 48)}$ = 62.5; $p < 0.001$] and week*treatment interaction [$F_{(4, 48)}$ = 4.2; $p = 0.02$]. There was also a between-subjects effect of treatment [$F_{(1, 12)}$ = 12.6; $p = 0.004$]. PLX treated males ate significantly less on (**A**) weeks 4-6 in the CD group and on (**B**) weeks 3, 4, and 6 in the HF group ($p < 0.05$). There were no significant differences in male food intake on week 7 when all mice were switched to house chow, suggesting that PLX decreased intake due to taste and not a side effect of the drug. (**C, D**) There were no effects of maternal diet on food intake. (**E-H**) For food intake in the females, there was a significant within-subjects effect of week [$F_{(4, 48)}$ = 71.7; $p < 0.001$] and week*treatment interaction [$F_{(4, 48)}$ = 3.4; $p = 0.043$]. PLX females consumed significantly less food on postnatal week 3 only, both in (**E**) CD and (**F**) HF groups ($p < 0.05$). This further suggests that the decrease in food intake is due to the taste of PLX and not a side effect. (**G, H**) There were again no effects of maternal diet on food intake. Overall, perinatal HF diet and adolescent PLX treatment had separate effects on food intake and body weight. Perinatal HF diet increased offspring body weight without increasing food intake, an effect that was most pronounced at weaning and dissipated over time. PLX treatment decreased food intake, more dramatically initially and more pronounced in the males, resulting in a decreased body weight that normalized quickly after switching to house chow. (*$p < 0.05$ vs. respective CD group; ^ $p < 0.05$ vs. respective NT group; n = 3-4 cages of 4 mice/group) (HF = offspring of maternal high fat diet; CD = offspring of maternal control diet; PLX = adolescent PLX3397 treatment; NT = not treated during adolescence)

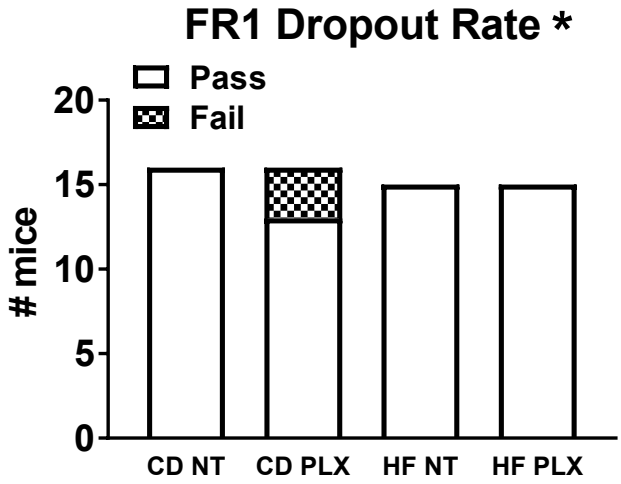

Figure S4: In male fixed ratio 1 (FR1), Chi^2 analysis revealed a significant difference in pass vs. fail outcomes for the number of mice that passed or failed the 70 trial criterion after 14 days. All mice in the CD NT (n = 16), HF NT (n = 15), and HF PLX (n = 15) groups passed criterion but 3 out of 16 mice in the CD PLX group failed to reach criterion. (*$p < 0.05$) (HF = offspring of maternal high fat diet; CD = offspring of maternal control diet; PLX = adolescent PLX3397 treatment; NT = not treated during adolescence)

Description	Abet II schedule name
FR1 habituation	Robbins: 5 choice habituation 2 v2
FR1	Cam: Center Only – Mouse Must Touch training v2
PR	Cam: Progressive Ratio (custom)
5CSRTT habituation	Robbins: 5 Choice Mouse Must Touch
5CSRTT (16, 8, 4 s stimuli)	Robbins: 5-choice Mouse Touch basic v3 (session value 10, 11, 12)
Titration	Robbins: Titration 5-choice mouse touch basic v3

Table S1. List of Abet II pre-programmed schedules used for male operant training and testing.

Gene name	Function	TaqMan assay ID
Actb	HK	Ref: 4352663
C1qa	Complement C1q A chain: complement recognition, predominantly microglial[1]	Mm00432142_m1
Cnr1	Cannabinoid receptor 1: mostly neuronal	Mm1212171_s1
Csf1r	Colony stimulating factor 1: trophic factor needed for microglial survival[2,3]	Mm1266652_m1
Cx3cl1	Chemokine ligand 1: CXC chemokine expressed by neurons and endothelial cells to recruit neutrophils[4]	Mm00436454_m1
Cx3cr1	Fractalkine receptor: expressed in immune cells and microglia to recognize cx3cl1[5,6]	Mm02620111_s1
Dlg4 (PSD95)	Discs Large MAGUK Scaffold Protein 4: postsynaptic density protein 95	Mm00492193_m1
Dnmt1	DNA Methyltransferase 1: maintains DNA methylation[7]	Mm01151063_m1
Dnmt3a	DNA Methyltransferase 3 Alpha: de novo DNA methylation[7]	Mm00432881_m1
Drd1a	Dopamine receptor D1	Mm01353211_m1
Drd2	Dopamine receptor D2	Mm00438545_m1
Fos	Proto-oncogene c-Fos: immediate early gene	Mm00487425_m1
Gad1	Glutamate Decarboxylase 1: catalyzes GABA production	Mm04207432_g1
Gadd45b	Growth Arrest And DNA Damage Inducible Beta: DNA demethylation[8]	Mm00435123_m1
Gapdh	HK	Ref: 4352661
Gphn	Gephyrin: inhibitory neurotransmission	Mm00556895_m1
Grin2b	GluN2B subunit: variants involved in neurodevelopmental disorders[9]	Mm00433820_m1
Hprt1	HK	Mm01324427_m1
Itgam	Integrin Subunit Alpha M (CD11b; CR3): macrophage/microglial adhesion for phagocytosis	Mm01271250_m1
Mecp2	Methyl-CpG binding protein 2: chromatin regulation and development of neuronal networks[10]	Mm01193537_g1
Oprd1	Opioid Receptor Delta 1	Mm01180757_m1
Oprk1	Opioid Receptor Kappa 1	Mm01230885_m1
Oprm1	Opioid Receptor Mu 1	Mm01188089_m1
Penk1	Preproenkephalin	Mm01212875_m1
Ppia	HK	Mm02342429_g1
Ppp1r1b	Protein Phosphatase 1 Regulatory Inhibitor Subunit 1B (DARPP-32): neuronal phosphoprotein regulated by dopamine[11]	Mm00454892_m1
Setd7	SET Domain Containing 7, Histone Lysine Methyltransferase: regulates DNMT1 activity[12]	Mm00499823_m1
Slc17a7 (vglut1)	Solute Carrier Family 17 Member 7 (Vesicular glutamate transporter 1): presynaptic glutamate uptake	Mm00812886_m1
Slc6a3 (DAT)	Solute Carrier Family 6 Member 3 (Dopamine Transporter): dopamine clearance/reuptake from synapses	Mm00438388_m1
Syp	Synaptophysin: neuronal synaptic vesicle glycoprotein	Mm00436850_m1
TH	Tyrosine Hydroxylase: catalyzes dopamine precursor production	Mm00447557_m1
Ywhaz	HK	Mm03950126_s1

Table S2. List of Fluidigm targets, functions, and assay ID's (HK = housekeeping gene)

Gene	AMG	NAC	PFC
C1qa	# Diet x Treatment [$F_{(1, 28)}$ = 4.3; p = 0.047] * Treatment [$F_{(1, 28)}$ = 8.1; p = 0.008] (CD PLX ↑ vs. CD NT)	^ Treatment [$F_{(1, 24)}$ = 5.6; p = 0.027] (PLX ↑ vs. NT)	^ Treatment [$F_{(1, 29)}$ = 15.6; p < 0.001] (PLX ↑ vs. NT)
Csf1r	n.s.	# Diet x Treatment [$F_{(1, 24)}$ = 4.9; p = 0.036] (HF PLX ↑ vs. CD PLX)	n.s.
Cx3cr1	^ Treatment [$F_{(1, 28)}$ = 4.2; p = 0.0498] (PLX ↓ vs. NT)	* Diet [$F_{(1, 24)}$ = 4.3; p = 0.0497] (HF ↑ vs. CD)	n.s.
Dlg4 (Psd95)	* Diet [$F_{(1, 28)}$ = 4.3; p = 0.048] (HF ↑ vs. CD) ^ Treatment [$F_{(1, 28)}$ = 4.7; p = 0.038] (PLX ↓ vs. NT)	n.s.	n.s.
Dnmt1	^ Treatment [$F_{(1, 28)}$ = 8.7; p = 0.006] (PLX ↓ vs. NT)	n.s.	n.s.
Drd1a	# Diet x Treatment [$F_{(1, 28)}$ = 4.9; p = 0.036] (HF PLX ↓ vs. CD PLX)	n.s.	n.s.
Fos	^ Treatment [$F_{(1, 28)}$ = 6.4; p = 0.018] (PLX ↑ vs. NT)	* Diet [$F_{(1, 24)}$ = 5.4; p = 0.028] (HF ↑ vs. CD)	n.s.
Gadd45b	n.s.	^ Treatment [$F_{(1, 24)}$ = 5.9; p = 0.022] (PLX ↑ vs. NT)	n.s.
Grin2b	# Diet x Treatment [$F_{(1, 28)}$ = 7.7; p = 0.0098] (HF PLX ↑ vs. CD PLX) (CD PLX ↓ vs. CD NT)	n.s.	n.s.
Oprk1	* Diet [$F_{(1, 28)}$ = 4.7; p = 0.038] (HF ↓ vs. CD)	n.s.	n.s.
Ppp1r1b (Darpp-32)	# Diet x Treatment [$F_{(1, 28)}$ = 4.5; p = 0.043] (HF PLX ↓ vs. CD PLX)	n.s.	n.s.
Setd7	* Diet [$F_{(1, 28)}$ = 6.3; p = 0.018] (HF ↑ vs. CD)	n.s.	n.s.
Slc6a3 (DAT)	n.s.	^ Treatment [$F_{(1, 24)}$ = 6.3; p = 0.019] (PLX ↓ vs. NT)	n.s.
Slc17a7 (Vglut1)	* Diet [$F_{(1, 28)}$ = 9.1; p = 0.0054] (HF ↑ vs. CD)	n.s.	n.s.
Syp	n.s.	n.s.	* Diet [$F_{(1, 29)}$ = 6.3; p = 0.018] (HF ↑ vs. CD)
Th	# Diet x Treatment [$F_{(1, 28)}$ = 4.3; p = 0.046] * Diet [$F_{(1, 28)}$ = 6.9; p = 0.014] (HF PLX ↓ vs. CD PLX)	^ Treatment [$F_{(1, 24)}$ = 7.1; p = 0.014] (PLX ↓ vs. NT)	n.s.

Table S3. Statistical results for high-throughput qPCR from male brains after the final day of 5CSRTT behavioral testing, after all mice performed the titration schedule for at least two consecutive days. Of the 27 targets, these 16 targets presented in the table revealed significant differences in at least one brain region. The remaining 11 targets with no significant differences are not shown (cnr1, cx3cl1, dnmt3a, drd2, gad1, gephryin, cr3, mecp2, oprd1, oprm1, and penk). (n = 7-8/group) (AMG = amygdala; NAC = nucleus accumbens; PFC = prefrontal cortex; HF = offspring of maternal high fat diet; CD = offspring of maternal control diet; PLX = adolescent PLX3397 treatment; NT = not treated during adolescence; n.s. = no significant results)

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