Supplemental Table 1. Primers used in ChIP-qPCR assays

	Primer Sequence (Forward and reverse pair)
AKT3	CTC TCT GTC TCA GGC GTA GGG TGA AGG CTC CTA GAA GAA GCC GAG AAA GCA CGT GAG AC
FOXO1	CGC GCC GCA CCT CTC TGG GGT TCA GGA TG GGA TCT GCA GCG GGC TCC CCG CCC GCG GCG
FOXO3	GCT TTA GCA TAA GAA AAC TGT TGC CCG CTT C GCA GGG CGG CCG CGG CAG CAG CAC AAA G
GLUT4	CCT TGG GTC CCC TCC AAG AAC CAG TGT AGA G GTG AGA CCC GCT TGA GGG GGA AAG ATG CGG
IGF1	CCA AAA AGC TTT CAC TTC CTG CCT CCC TG CTC GCC TTT GCC ACA GGG AAT GGA GTT CTG ATC
IRS1	CAC CAA GAG AAT GGG TTT ACT CTC ATC AAA TTT G CAT TTT CAG AAT TTA TAA ACA CCC ACA TTC
IGF1R	CGT GCT CGG CTT TGA CCT TCA GCG AGC CGG CCT TTT ATT TGG GAC GAA ATT CCC TTT TCT C
InsR	GGT ATC TGG CGG CAT TTC CAG ACT TTA AAA TTG GGC GGC GGG AGC TAG GGC TCG ACT CCA GCC
LAR	GCA CAC ACA CAC ACT GGC GTA CAC ACA C GCC AGC TCC AGC CGC GCG CGC CCG CCC GCC
GH	CTG GAA GAA CAA CAA GCT CCC TTA TCA TAT AAG GAC TTG GTC CCT TGC GTG CCC TTT TTA TAC
Jak2	CGC AGA AGT GGT GGT TAA CTG AAG TCA GGT AC GAG CGG GGG CCC GCC CAG CCT GTC AGC GCG
Leptin	CTG TTC TGT ATC TGT TTC CTC CCA TTA GG GGG GCT CCA TGC CTG CCT GCC CCT CTT ATA AC
Leptin receptor	CTG TCC TAT TGG AGG GGT AGG GGG TAA GGG CCT CAC CGC TGC AAA TCC TTT TAA AGG AGT GC
PI3K	CCT CAC AGA ACA CTC CTT TTC CTC TAG ATC CTC CCT GAG CGG CGC AGC TCC AGC GCC AGG
PTP1B	GCA TGT CTG TGA TTC CAA GAC GTG GGA GAT G GTC AAG CCG GCA GCC GGA AGT ACG CAC CC
PTEN	CAT CTT TCG GGC AAA TGA ACC CAG CTG CC GAG AGC TGA GCG CAG CCG CGT CCC GAC GCC
EYA2	CGGGACTCTCTCTCATTCCA CACCACAGCGATTCAAACAC
STAT3	CTG TGA GCT GAC CAA TCA GCC TTG CTG TTG CCA ATC CCC ACC CAG CCG CCA CTG GCC AAG

Supplemental Table 2

	PTP1B$_{1-301}$:CPT157633[s]
Data Collection	
Space group	*P* 2_1
Cell dimensions	
a,b,c (Å)	52.17, 71.75, 88.01
α, β, γ (°)	90, 94.3, 90
Resolution (Å)	43.35 – 1.90 (1.97 – 1.90)*
R_{merge}	0.068 (0.307)
I/σI	28.68 (3.38)
Completeness (%)	90.7 (49.4)
Redundancy	7.0 (4.4)
Refinement	
Resolution (Å)	43.35 – 1.90
No. reflections	46442
R_{work}[d]/R_{free}[e]	0.170/0.208
No. atoms	
Protein	4751
Water	539
Ligands	85
R.m.s. deviations	
Bond length (Å)	0.007
Bond angle (°)	1.030
Ramachandran plot	
Outliers	0.34
Allowed	1.72
Favored	97.93
PDB Code	4Y14

[s]1 crystal

*highest resolution shell is shown in parentheses

Methods

ChIP and quantitative PCR. For ChIP three whole mouse brains from WT male mice were used. Briefly, flash-frozen brains were ground and to the powder 1% formaldehyde was added. Fixation was continued for 15 mins at room temperature and terminated with 0.125 M glycine solution. The cells were pelleted and homogenized in a dounce homogenizer. After centrifugation, cells were resuspended in 5 ml of lysis buffer (10 mM Tris pH 8.0, 0.2% NP-40, 10 mM NaCl, Complete protease inhibitors (Roche)). The lysate was passed through a 25-guage needle to remove lumps and incubated for an additional 15 min in 5 ml of lysis buffer. Nuclei were harvested by centrifugation (4,000 x g) for 5 min and the pellet was resuspended in 2 ml of nucleus lysis buffer (50 mM Tris-HCl, 10 mM EDTA, 1% sodium dodecyl sulfate [SDS], protease inhibitors). Nuclei were lysed for 5 min at room temperature and diluted in 1 ml of ChIP dilution buffer (20 mM Tris (pH 8.0), 150 mM NaCl, 2 mM EDTA, 1% Triton, protease inhibitors). Chromatin was sonicated (5 min, duty cycle 50, output 8). Following sonication chromatin was cleared by centrifugation, precleared with protein A-Sepharose (Sata cruz biotechnology), and subjected to overnight immunoprecipitation with anti-MeCP2 antibody. Antibody precipitates were bound to protein A-Sepharose for 1 h and washed once (0.1% SDS, 1% Triton X-100, 2 mM EDTA, 150 mM NaCl, 20 mM Tris-HCl [pH 8]), three times with ChIP wash buffer I (0.1% SDS, 1% Triton X-100, 2 mM EDTA, 500 mM NaCl, 20 mM Tris-HCl [pH 8]), once with ChIP wash buffer III (0.25 M LiCl, 1% NP-40, 1% deoxycholate, 1 mM EDTA, 10 mM Tris-HCl [pH 8]), and three times with Tris-EDTA. DNA-antibody precipitates were eluted twice with 1% SDS, 0.1M sodium bicarbonate, and cross links were reversed at 65 C for 6 hours. DNA was purified with QIAGEN pcr purification kit. From the final eluate 2µl was used for PCRs with appropriate primers (Table S1). For gene expression studies total RNA was extracted from WT and MeCP2-null male mice. RNA extracted was used to synthesize cDNA using the iScript cDNA synthesis kit (Bio-Rad, 170-8890). The cDNA synthesized from WT and MECP2-null brain samples were used for quantitative PCR using an Applied Biosystems 7900HT instrument. RT profiler PCR arrays (Qiagen, Cat. No. 330231-006ZA and 330231-0030ZA) were used to look at changes in gene expression in insulin signaling pathway and glucose metabolism.

Metabolic measurements. Glucose in tail blood was measured using a glucometer (One-Touch Basic; Lifescan, CA). For glucose tolerance tests (GTTs), mice were fasted

for 10 hours and then injected 20% D-glucose (2 mg/g body weight) and the blood glucose was monitored immediately before and at 15, 30, 60 and 120 mins following the injection. For insulin tolerance tests (ITTs), 4-h fasted animals were given insulin (0.75 mU/g) and blood glucose was measured immediately before and at 30, 60 and 120 minutes postinjection. Statistical analysis was performed using ANOVA for both GTT and ITT. Serum insulin, cholesterol, triglycerides (Stanbio labs, TX), BDNF (Abnova), IGF1 and IGFBPs (R&D systems) were determined by enzyme-linked immunosorbent assay.

Enzyme Kinetics

PTP assays were performed in polystyrol 96-well plate using pNPP as substrate. pNPP (0-5 mM) was added to assay buffer (50 mM Hepes, 100 mM NaCl, 0.1 % BSA, 2 mM DTT, 2 mM EDTA pH 6.5) containing 10 nM purified PTP1B in a final volume of 100 µl. The absorbance at 405 nm was monitored continuously for 20 minutes using a spectramax 190 plate reader. For assays using radiolabelled substrate, reduced carboxamidomethylated and maleylated lysozyme (RCML) was phosphorylated on tyrosine to a stoichiometry up to 0.8 mol ^{32}P/mol of protein, by recombinant GST-FER kinase and [γ^{32}P] ATP, and activity was measured against PTP1B in the absence and presence of the inhibitors.

Pup retrieval assay

Male and female CBA/J mice were paired. A single male and a single female were housed together in a standard mouse maternity cage from the mating period until the females were pregnant. Pregnant female mice were subsequently housed with one WT and one *Mecp2*$^{-/+}$ mice in each cage without the father until the pregnant females delivered pups. Both WT and *Mecp2*$^{-/+}$ mice were treated with saline or PTP1B inhibitor two weeks before the pups were delivered. Retrieval testing of all three females was performed during the day cycle in the dark and in sound isolation. The home cage was placed in a test chamber. Mice were initially allowed to habituate to the test chamber for 5 minutes and then the pups were removed from the cage for 2 minutes. Four pups were then selected from the litter and placed individually at sites remote from the nest in the home cage. The trial was monitored and recorded to a computer via a night vision digital camera under infrared illumination. Each subject was allowed to interact freely with the pups for five minutes or until all pups are gathered in the nest. Videos were later scored

blind to experimental condition. We measured gathering latency with a normalized measure that corrects for different numbers of pups in rare cases of small litters and credits for gathering of only some pups:

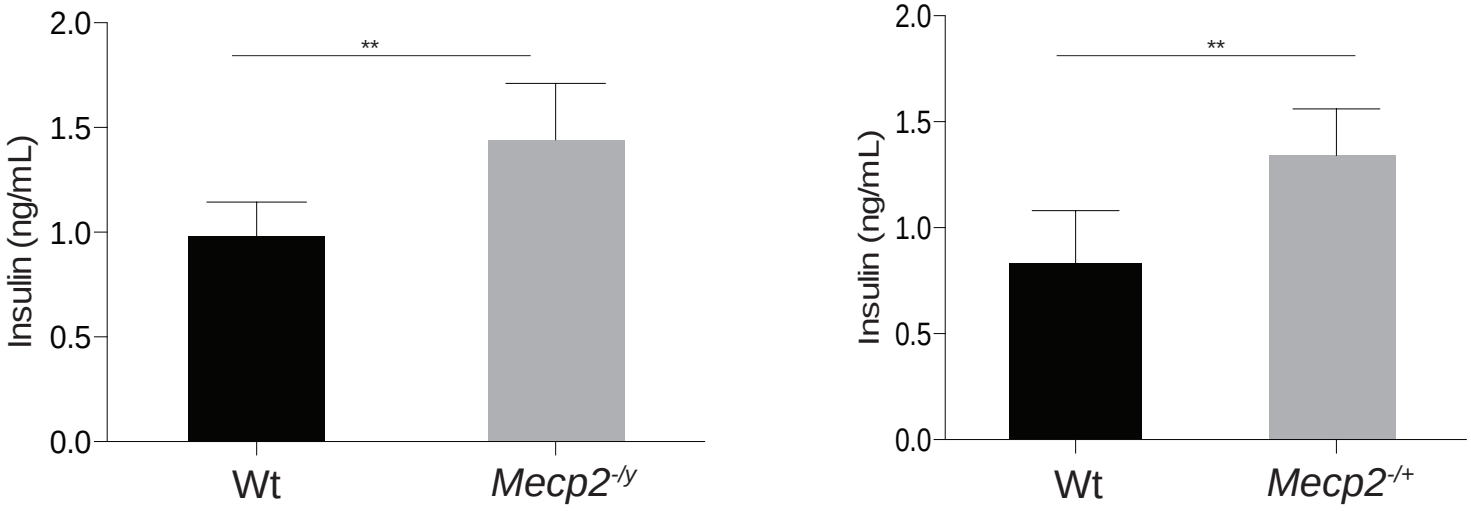
$$\text{latency} = [(t_1 - t_0) + (t_2 - t_0) + \ldots + (t_n - t_0)]/(n*L)$$

(n = # of pups, t_0 = start time, t_n = time of nth gather; L = trial length)

Statistical analysis was performed using Mann-Whitney U test.

Supplemental Figure 1

A.



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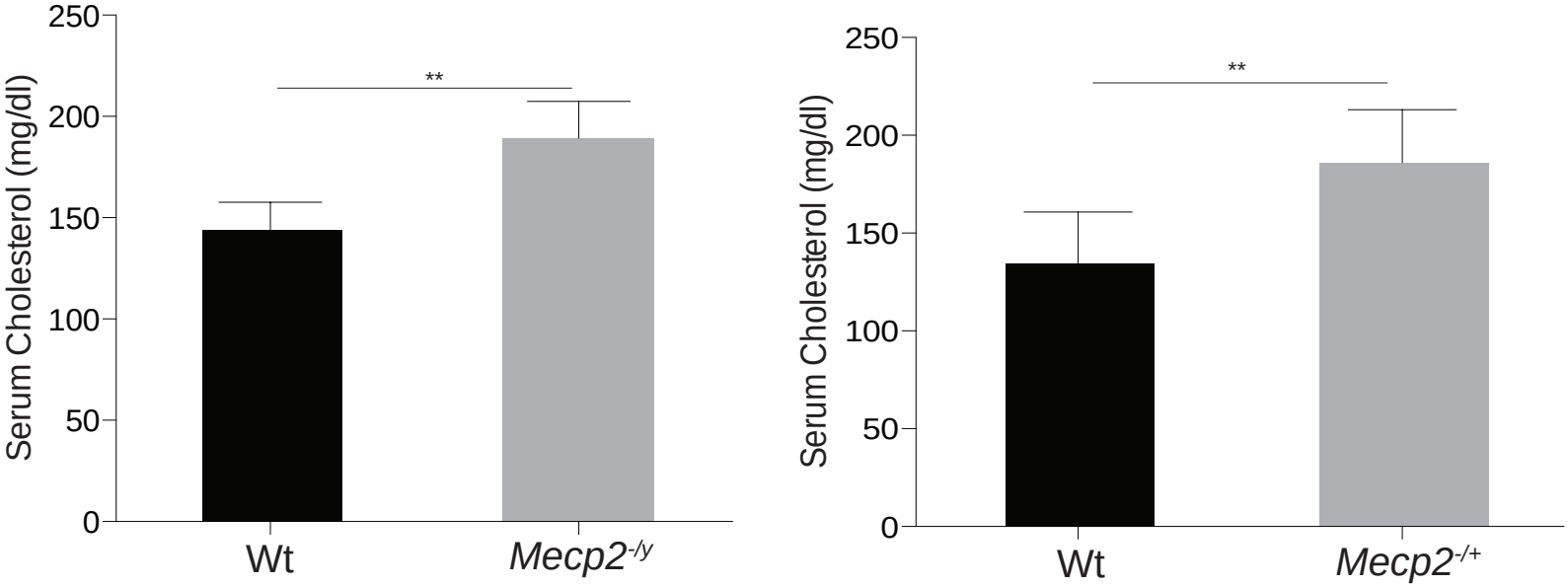
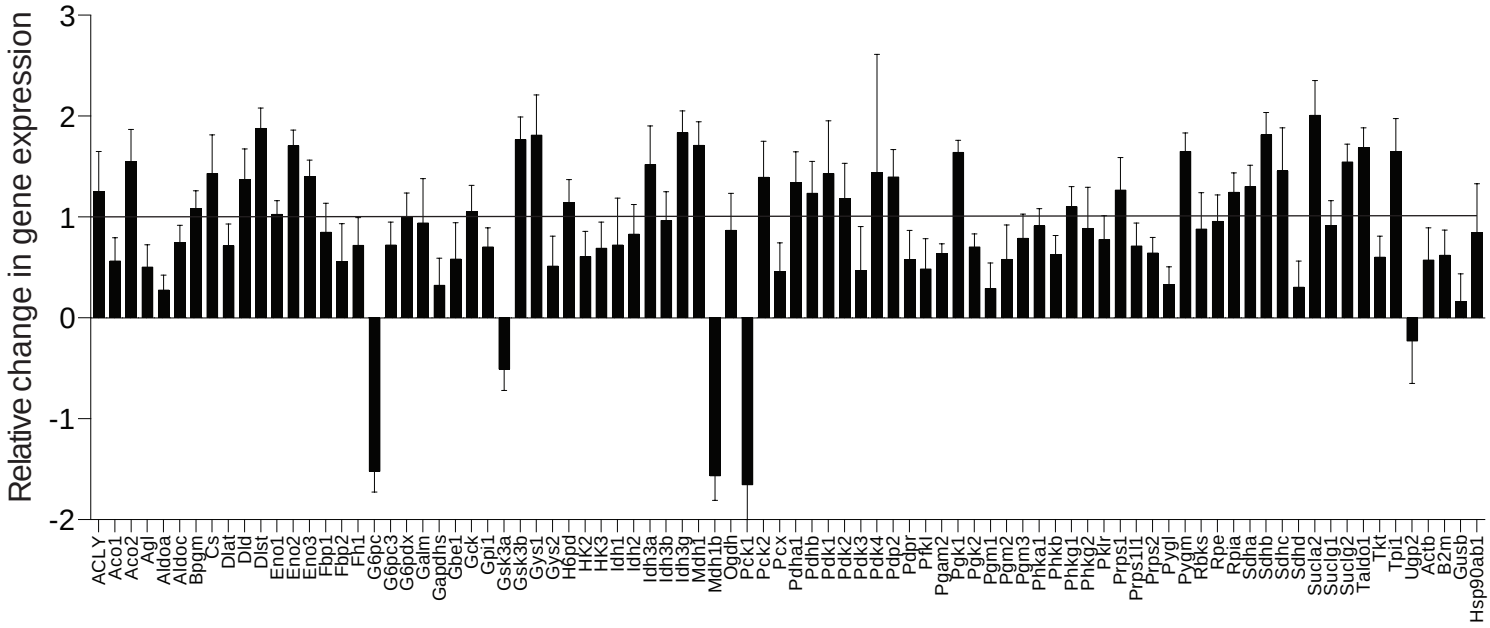


Supplemental Figure 1

A. Serum levels of insulin measured by ELISA in post-natal day 30 *Mecp2*$^{-/y}$ (left panel, gray bar) and 10-week-old *Mecp2*$^{-/+}$ mice (right panel, gray bar) compared with WT animals (black bars) t-test **P<0.001

B. Serum levels of cholesterol measured by ELISA in post-natal day 30 *Mecp2*$^{-/y}$ (left panel, gray bar) and 10-week-old *Mecp2*$^{-/+}$ mice (right panel, gray bar) compared with WT animals (black bars). t-test **P<0.001

A



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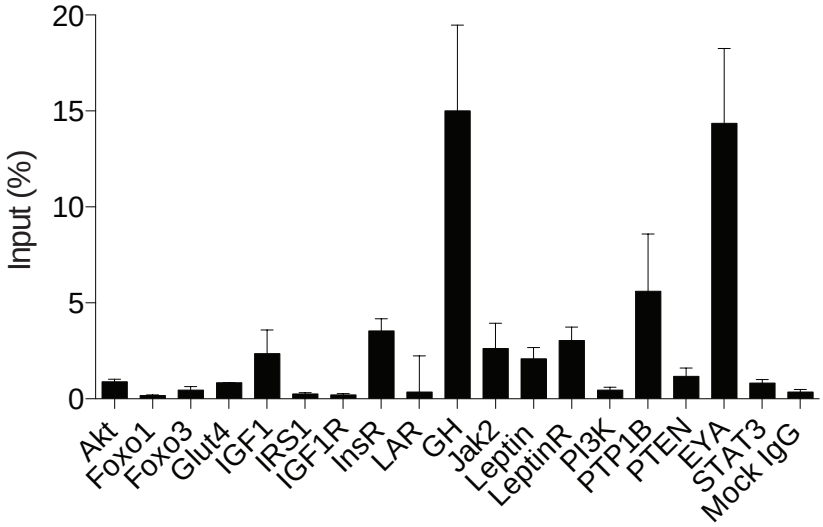

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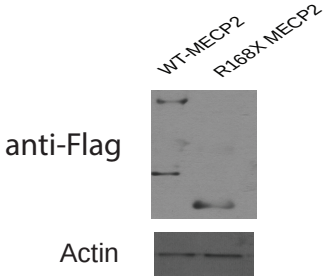
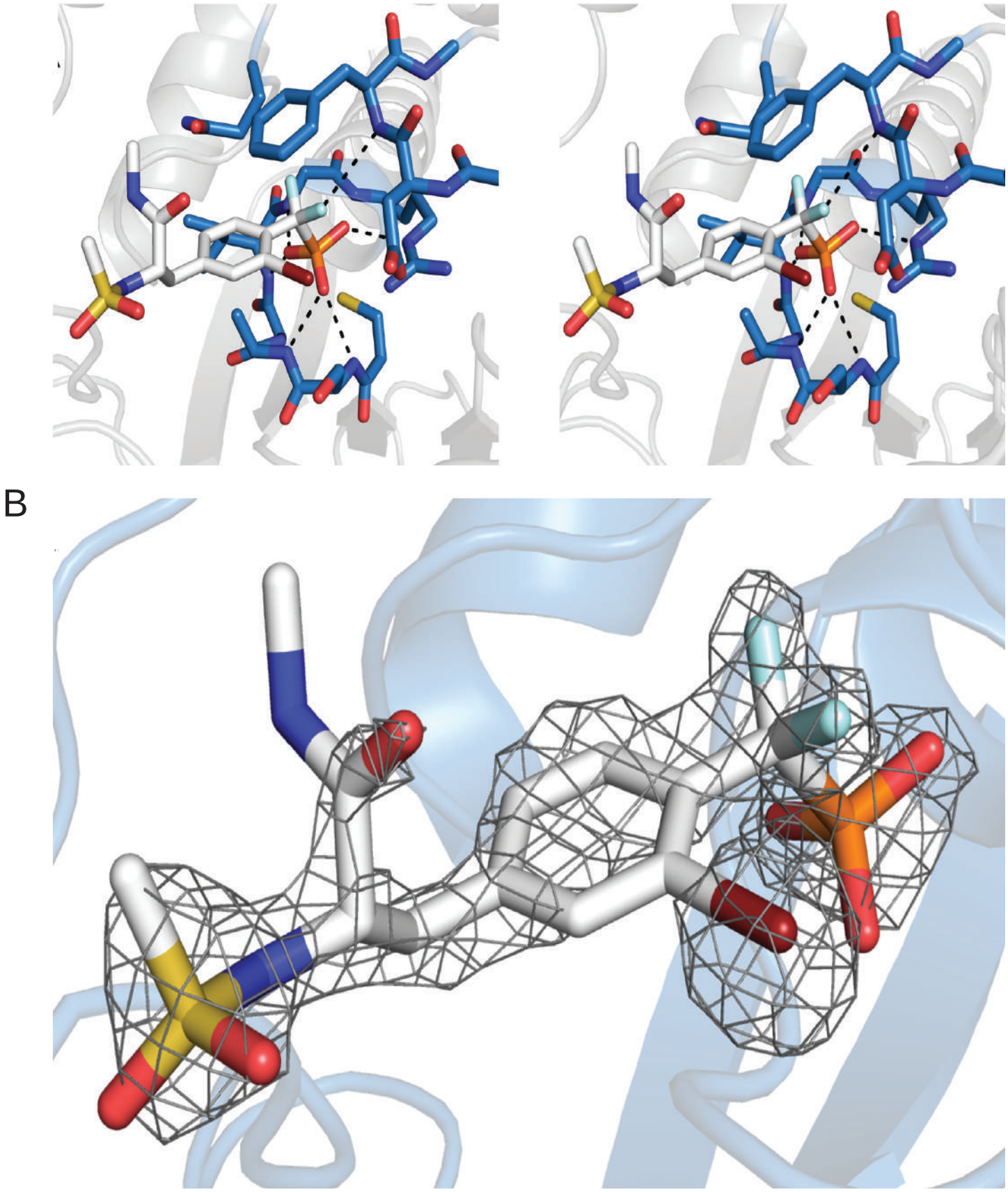


Supplemental Figure 2

A. Total RNA obtained from WT and *Mecp2*$^{-/y}$ mice was reverse transcribed and the cDNA was used in quantitative PCR analysis. The relative change in expression of genes involved in glucose metabolism in *Mecp2*$^{-/y}$ mice forebrain compared to WT was measured and the data was normalized to GAPDH expression. (data represent mean ± s.e.m).

B. Chromatin immunoprecipitation (ChIP) performed with anti-MECP2 on whole-brain samples obtained from post-natal day 30 (data represent mean ± s.e.m).

C. Representative blot to show overexpression of WT and mutant forms of MECP2

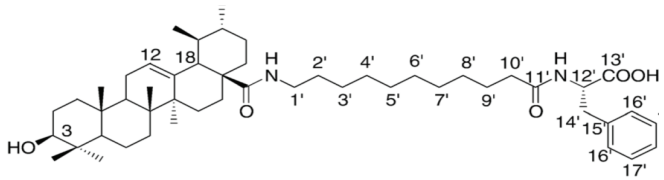


Supplemental Figure 3

A. Stereo image of the PTP1B$_{1\text{-}301}$-CPT157633 crystal structure, illustrating the interaction of CPT157633 (light grey) with PTP1B (blue) active site residues.

B. Electron density (Fo-Fc, Sigma level=3.0) of the CPT157633 inhibitor after molecular replacement and prior to refinement and model building.

A



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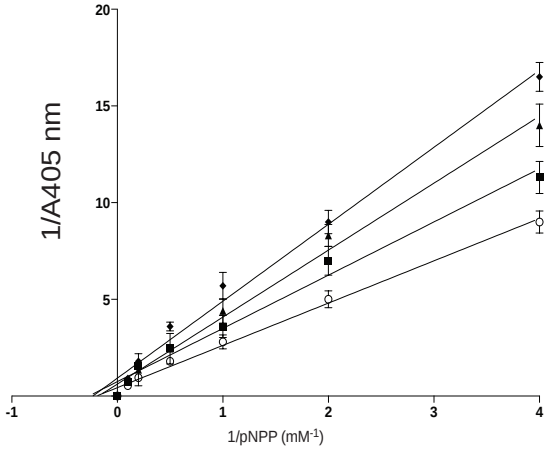

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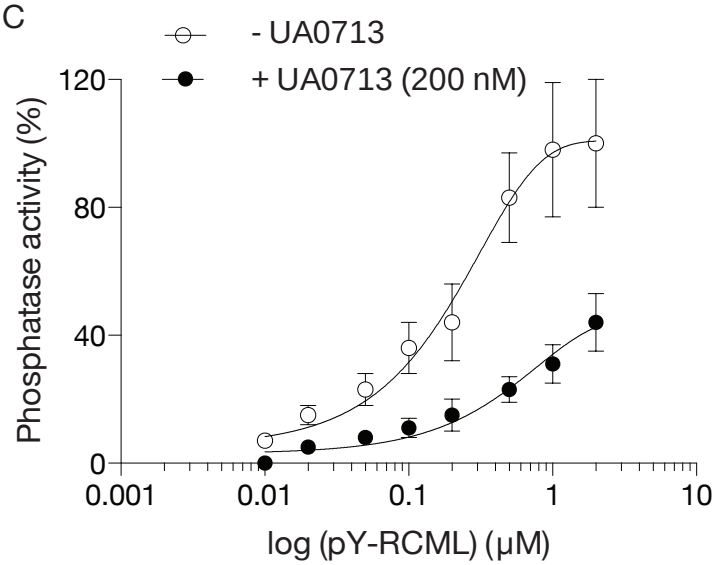

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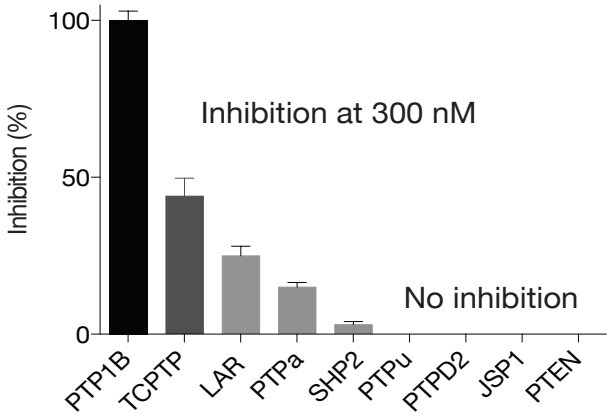


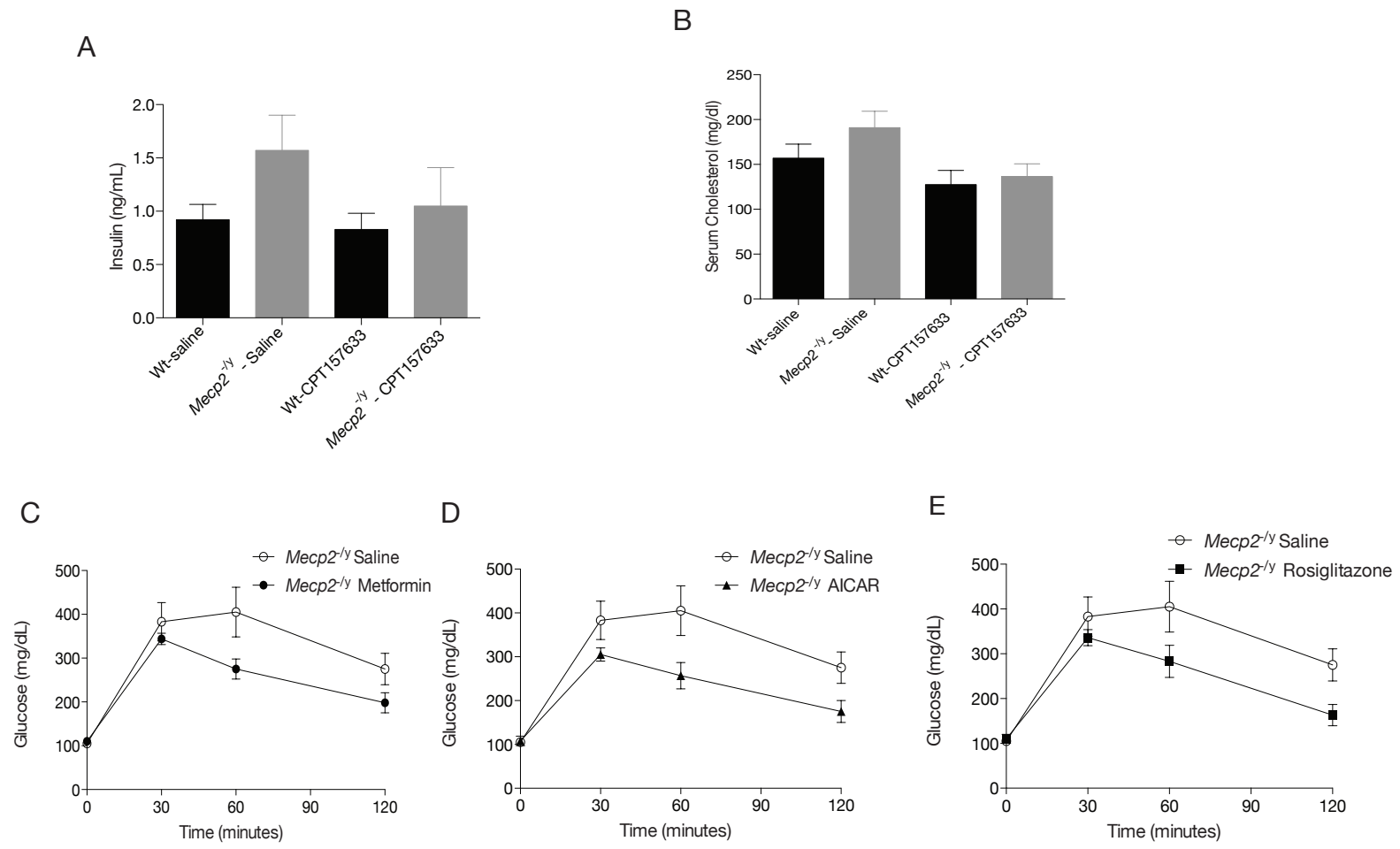
Supplemental Figure 4

A. Chemical structure of UA0713

B. Lineweaver Burk plot for $PTP1B_{1-405}$ showing 1/rate versus 1/substrate at varying concentrations of UA0713 0 (o), 50 nM (■), 100 nM (▲) and 200 nM (◆). The K_i was calculated to be 150 nM. Data are representative of three independent experiments (data represent mean ± s.e.m).

C. PTP1B inhibition by UA0713 was characterized using 32P-RCML as the substrate. [^{32}P]-RCML (0-1μM) was titrated against PTP1B (10 nM) in the absence and presence of UA0713 (200 nM) (data represent mean ± s.e.m).

D. Phosphatase activity of a panel of PTPs (100 nM) was tested in the absence and presence of UA0713 (100 nM) using pNPP (2 mM) as substrate (data represent mean ± s.e.m).



Supplemental Figure 4

A. Serum levels of insulin measured by ELISA using blood samples obtained from saline or CPT157633 treated WT (black) or *Mecp2*$^{-/y}$ (gray) mice.
(two-way ANOVA Wt-saline and Wt-CPT157633, P=0.05; Mecp2-/y-saline and Mecp2-/y-CPT157633, P=0.001)

B. Serum levels of cholesterol measured by ELISA using blood samples obtained from saline or CPT157633 treated WT (black) or *Mecp2*$^{-/y}$ (gray) mice.
(two-way ANOVA Wt-saline and Wt-CPT157633, NS; Mecp2-/y-saline and Mecp2-/y-CPT157633, P=0.01)

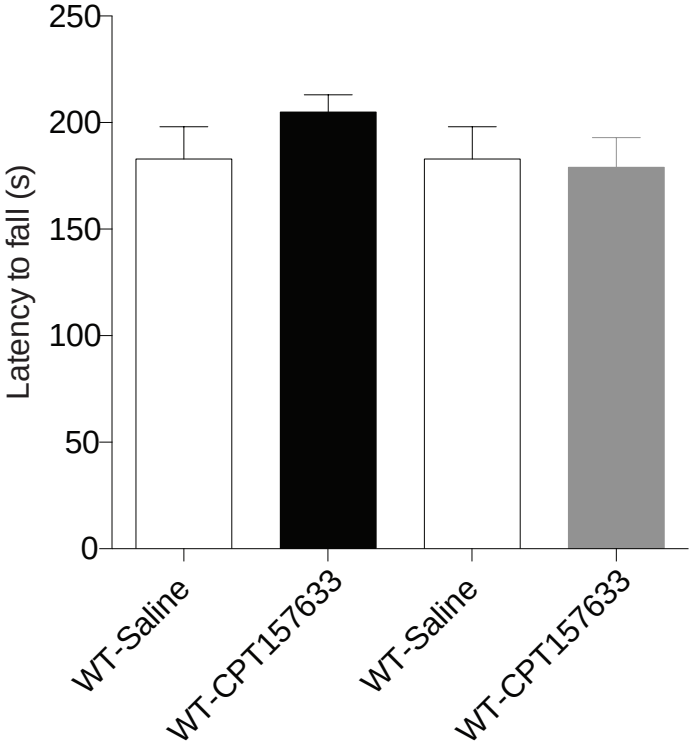
C. GTT results for *Mecp2*$^{-/y}$ mice treated with saline or metformin (50 mg/Kg). (two-way ANOVA, P =0.0075)

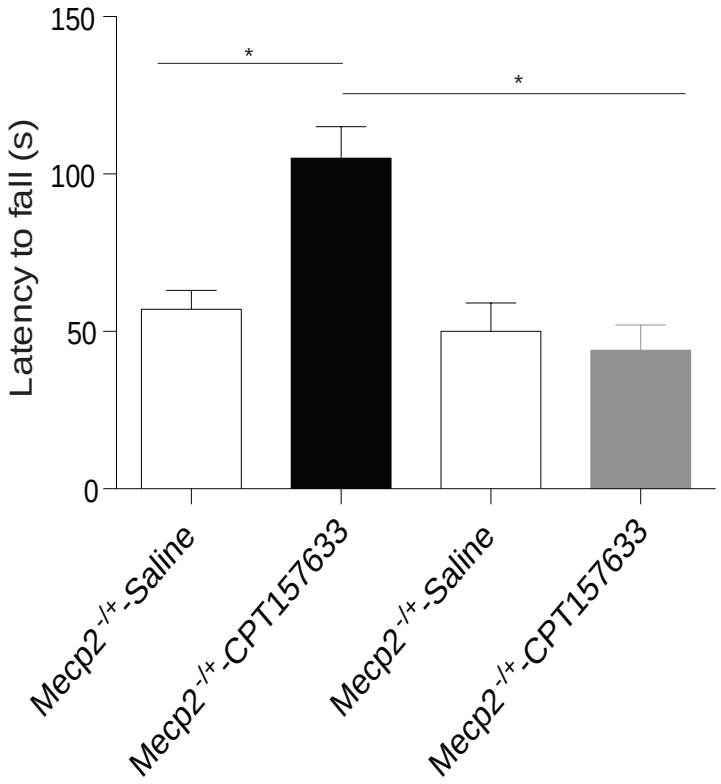
D. GTT results for *Mecp2*$^{-/y}$ mice treated with saline or AICAR (50 mg/Kg). (two-way ANOVA, P=0.005)

E. GTT results for *Mecp2*$^{-/y}$ mice treated with saline or rosiglitazone (25 mg/Kg).(two-way ANOVA, P=0.008)

Supplemental Figure 6

A.





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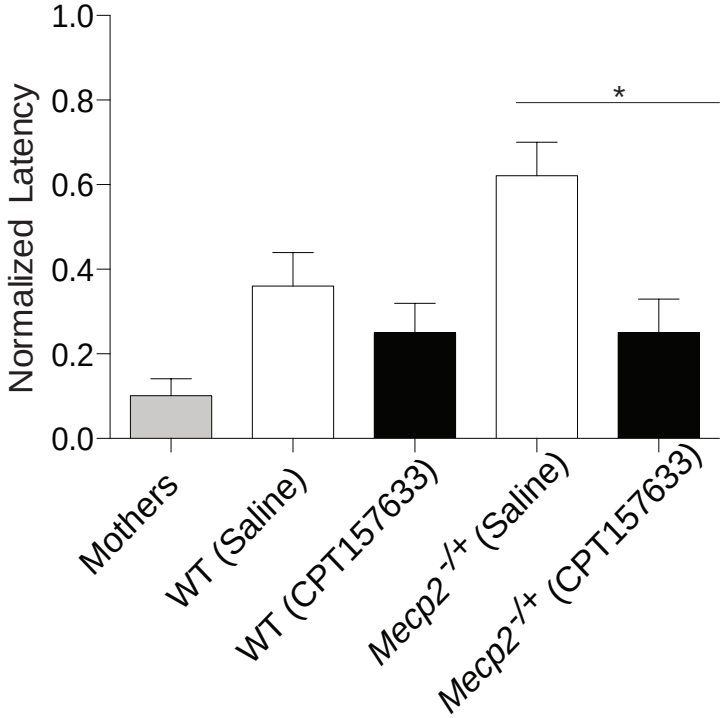

Supplemental Figure 6

A. Latency to fall from the rotarod was studied 7-days after CPT157633 treatment was stopped in WT and *Mecp2*$^{-/+}$ mice. In the WT mice no significant change was observed. However, in *Mecp2*$^{-/+}$ mice the improved motor behavior observed with CPT157633 treatment was lost. (* P=0.01)

B. Mean performance of normalized latency to gather pups averaged over days P0, P3 and P5 (n=6 per genotype per treatment). Saline treated *Mecp2*$^{-/+}$ trend towards longer latency in retrieving pups to the nest, compared to WT (P=0.06). Comparing saline treated WT and *Mecp2*$^{-/+}$ to the mother, WT mice do not have significantly different latencies (P=0.4), unlike *Mecp2*$^{-/+}$ mice (P<0.005) suggesting *Mecp2*$^{-/+}$ display poor pup retrieval behavior. When *Mecp2*$^{-/+}$ are treated with CPT157633, their latency improves significantly (P=0.03)