Scientific Reports

Supplementary Information

Developmental transitions in amygdala PKC isoforms and AMPA receptor expression associated with threat memory in infant rats

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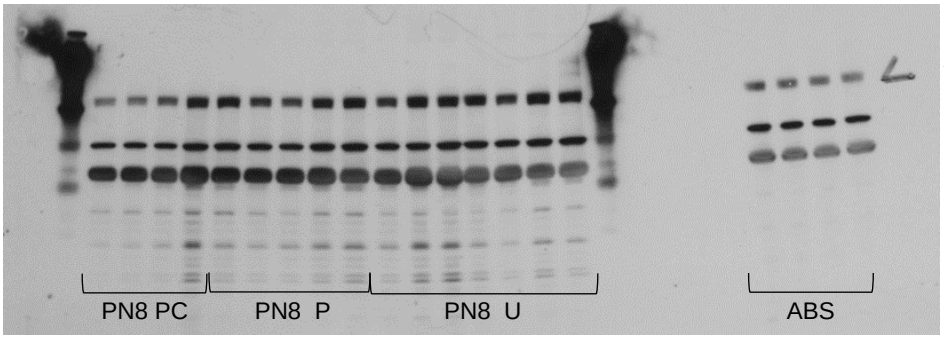
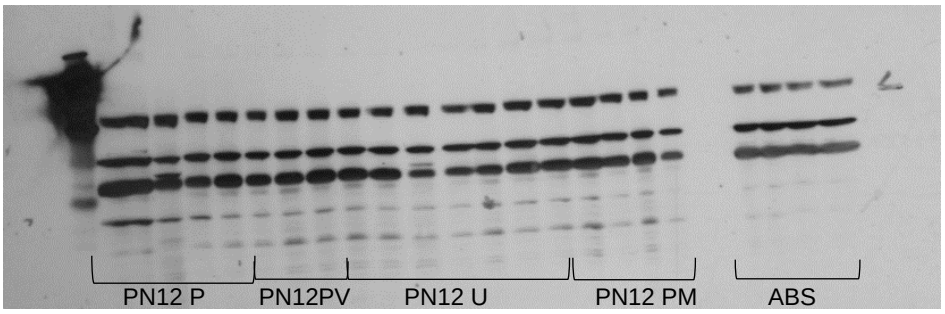
Supplementary Figure 1.

GluA2 Blots (100 kDa)



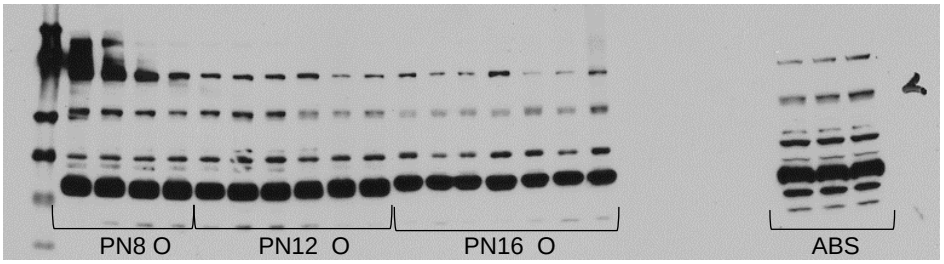
◄ GluA2

α-Tubulin comparison: Fig. 5c



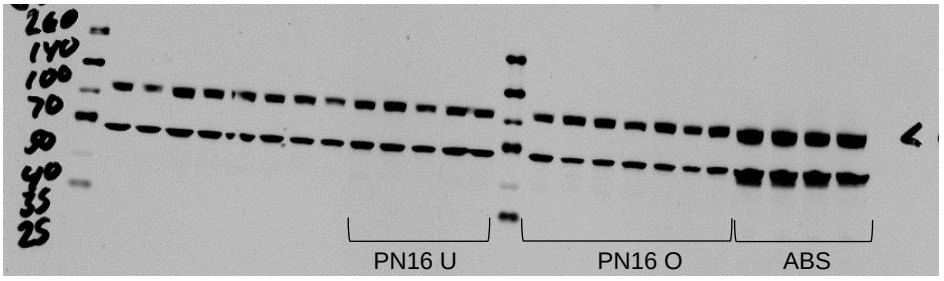
◄ GluA2

α-Tubulin comparison: Fig. 5d



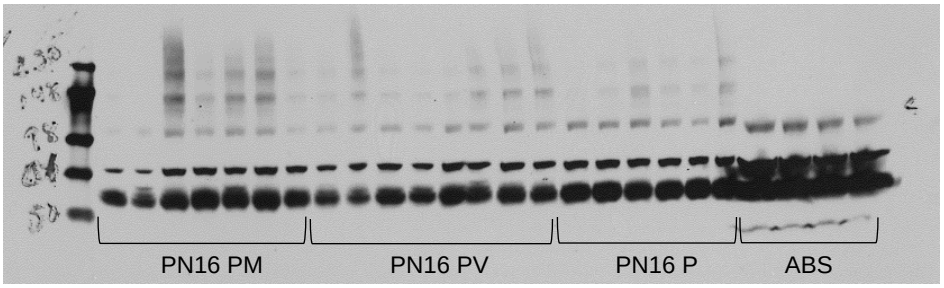
◄ GluA2

α-Tubulin comparison: Fig. 5f



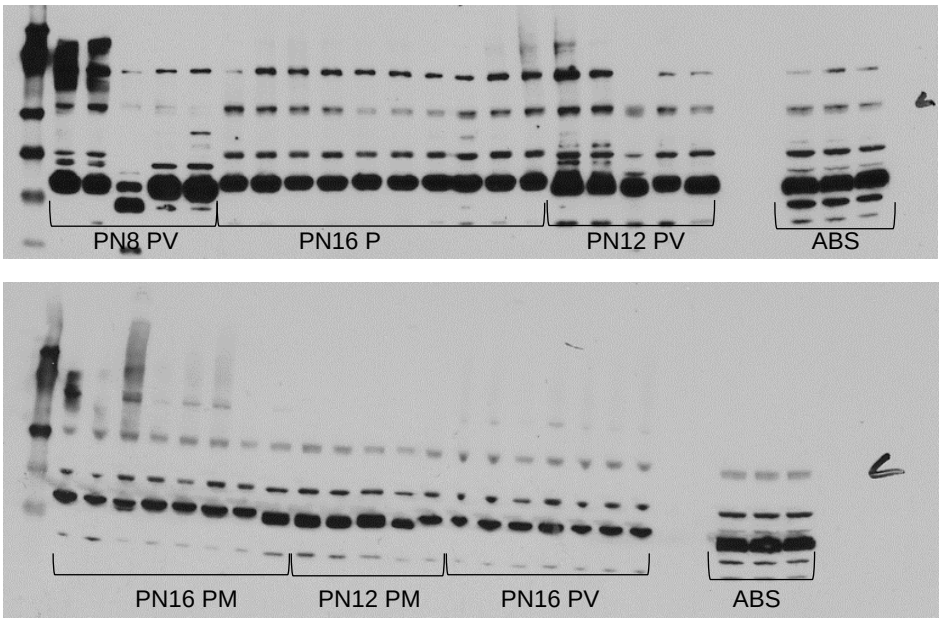
◄ GluA2

α-Tubulin comparison: Fig. 5k



◄ GluA2

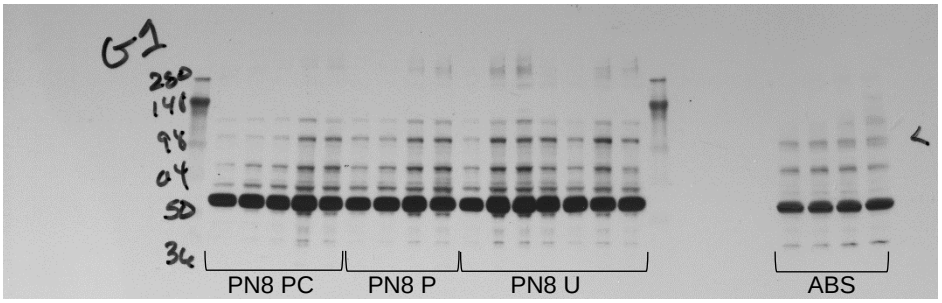
α-Tubulin comparison: Fig. 5l



Supp. Figure 1. Western blot showing GluA2 (100 kDa, arrow) levels in amygdalae of pups conditioned at age postnatal day (PN)8, PN12 and PN16 and sac'd 24 hr later. P/U/O: Paired, Unpaired, Odor only conditioning. PC/PM/PV: Paired+CORT, Paired+Metyrapone, Paired+Vehicle. ABS: All brain sample, positive control. Empty lanes were loaded with 1x Laemmli Buffer. The same tubulin-corrected values were used for all markers probed. Comparisons were made across gels processed in parallel using samples derived from the same experiment. Unmarked blots were run as part of a separate study.

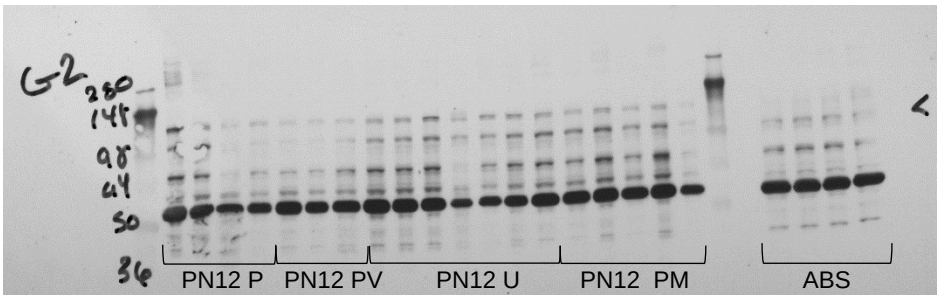
Supplementary Figure 2.

GluA1 Blots (100 kDa)



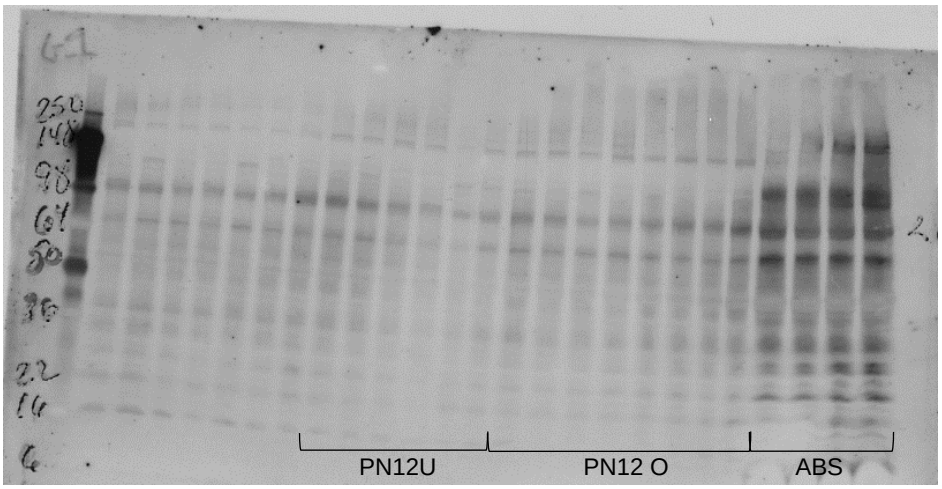
◄ GluA1

α-Tub comparison: Fig. 5c



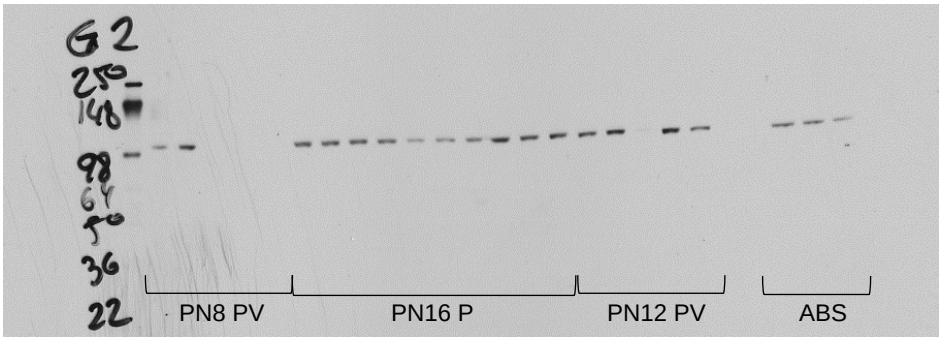
◄ GluA1

α-Tubulin comparison: Fig. 5d



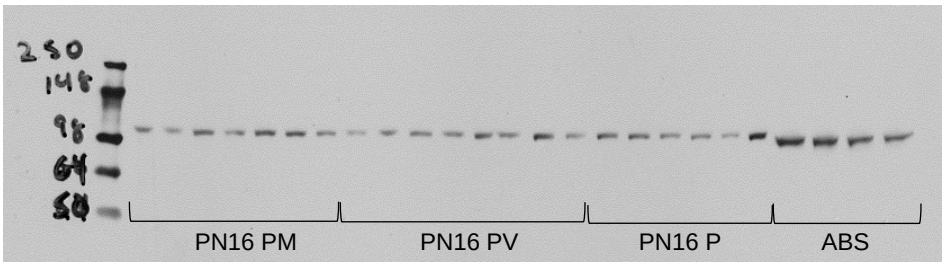
◄ GluA1

α-Tubulin comparison: Fig. 5j



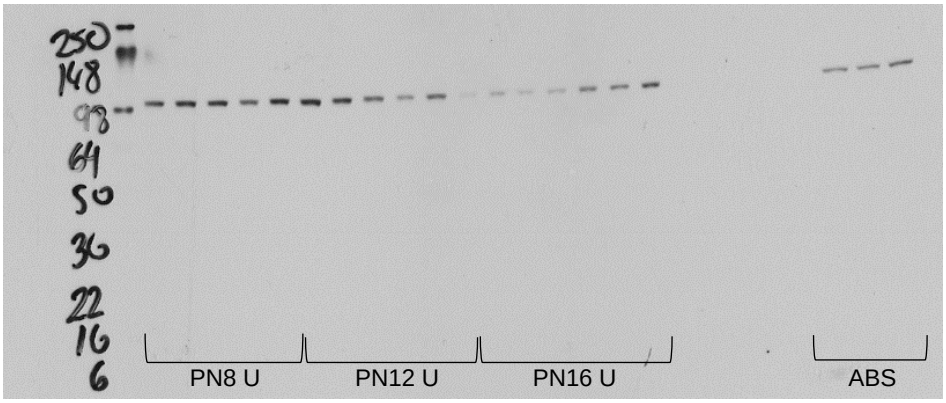
◄ GluA1

α-Tubulin comparison: Fig. 5e



◄ GluA1

α-Tubulin comparison: Fig. 5l



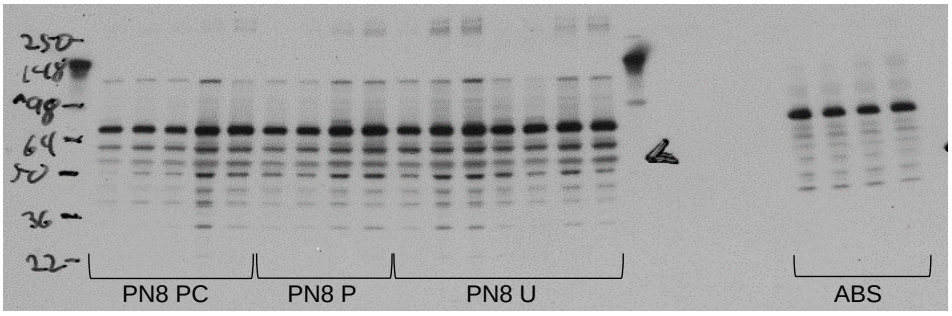
◄ GluA1

α-Tubulin comparison: Fig. 5g

Supp. Figure 2. Western blot showing GluA1 (100 kDa, arrow) levels in amygdalae of pups conditioned at age postnatal day (PN)8, PN12 and PN16 and sac'd 24 hr later. P/U/O: Paired, Unpaired, Odor only conditioning. PC/PM/PV: Paired+CORT, Paired+Metyrapone, Paired+Vehicle. ABS: All brain sample, positive control. Empty lanes were loaded with 1x Laemmli Buffer. The same tubulin-corrected values were used for all markers probed. Comparisons were made across gels processed in parallel using samples derived from the same experiment. Unmarked blots were run as part of a separate study.

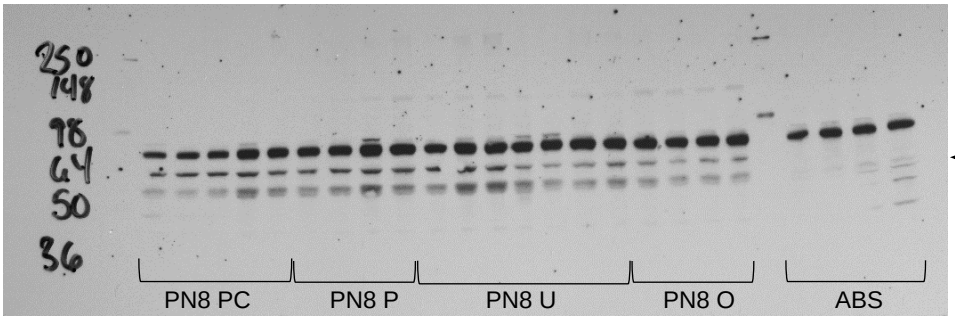
Supplementary Figure 3.

PKMζ Blots (55 kDa)



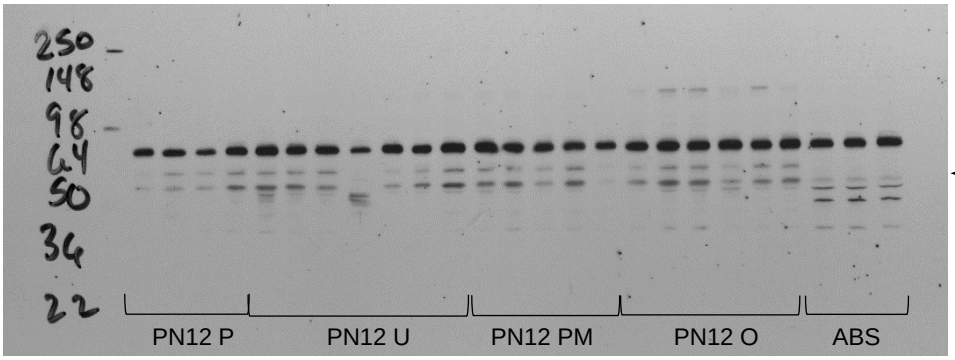
◄ PKMζ

α-Tubulin comparison: Fig. 5c



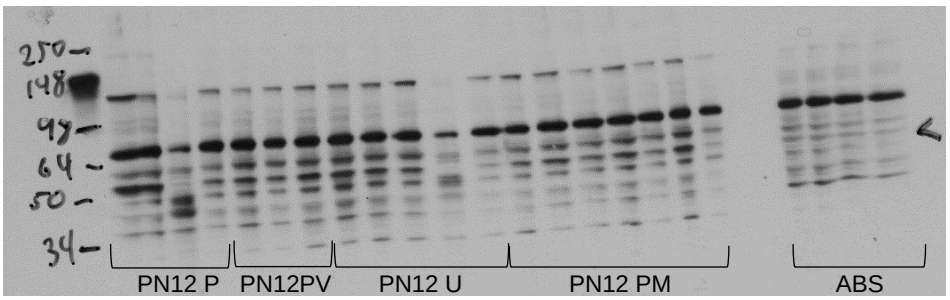
◄ PKMζ

α-Tubulin comparison: Fig. 5b



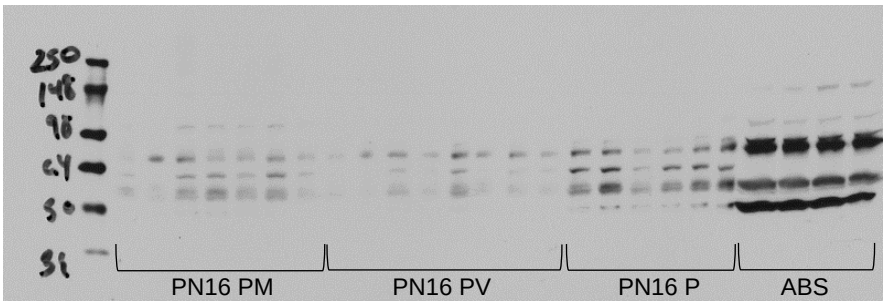
◄ PKMζ

α-Tubulin comparison: Fig. 5a



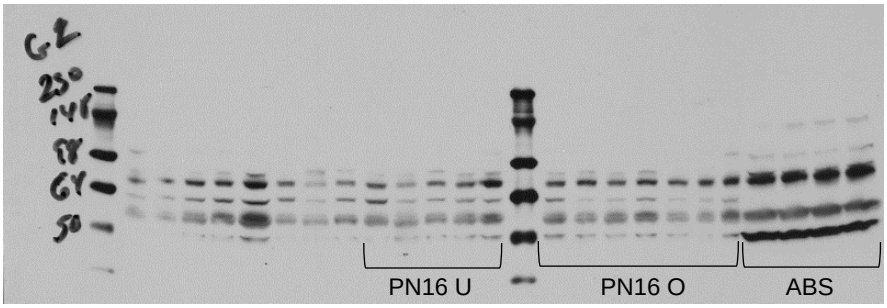
◄ PKMζ

α-Tubulin comparison: Fig. 5d



◄ PKMζ

α-Tubulin comparison: Fig. 5l



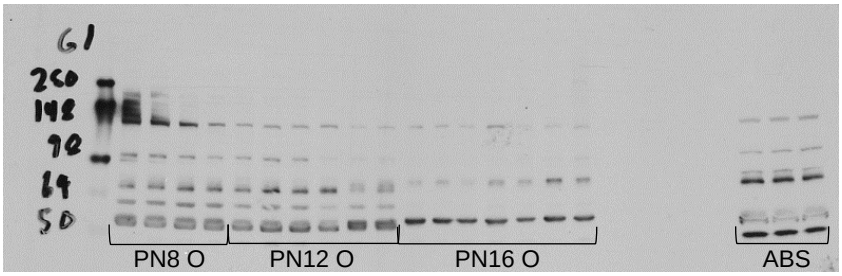
◄ PKMζ

α-Tubulin comparison: Fig. 5k

Supp. Figure 3. Western blot showing PKMζ (55 kDa, arrow) levels in amygdalae of pups conditioned at age postnatal day (PN)8, PN12 and PN16 and sac'd 24 hr later. P/U/O: Paired, Unpaired, Odor only conditioning. PC/PM/PV: Paired+CORT, Paired+Metyrapone, Paired+Vehicle. ABS: All brain sample, positive control. Empty lanes were loaded with 1x Laemmli Buffer. The same tubulin-corrected values were used for all markers probed. Comparisons were made across gels processed in parallel using samples derived from the same experiment. Unmarked blots were run as part of a separate study.

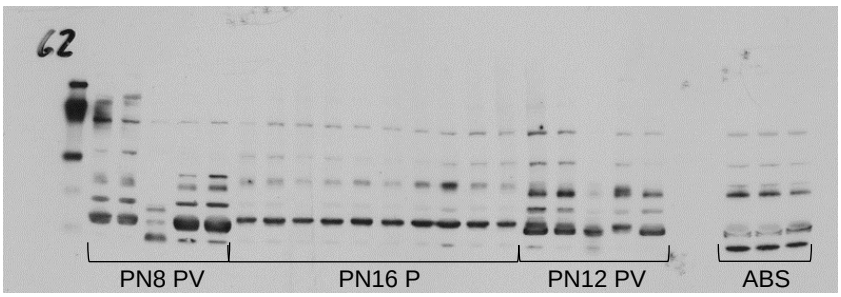
Supplementary Figure 4.

PKC ι/λ Blots (70 kDa)



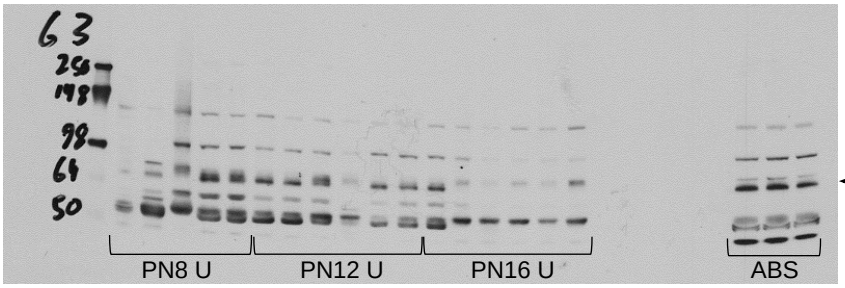
◄ PKC ι/λ

α-Tubulin comparison: Fig. 5f



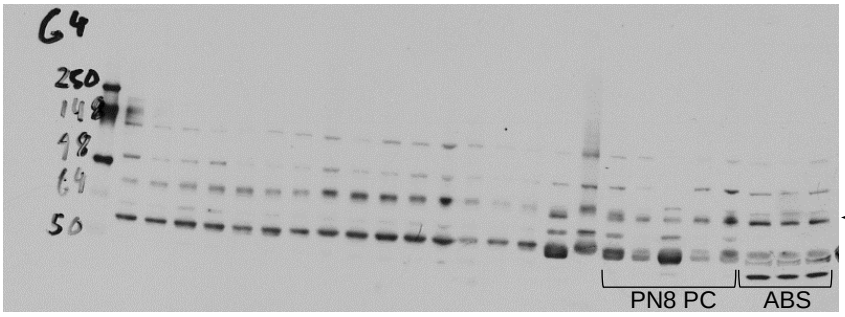
◄ PKC ι/λ

α-Tubulin comparison: Fig. 5e



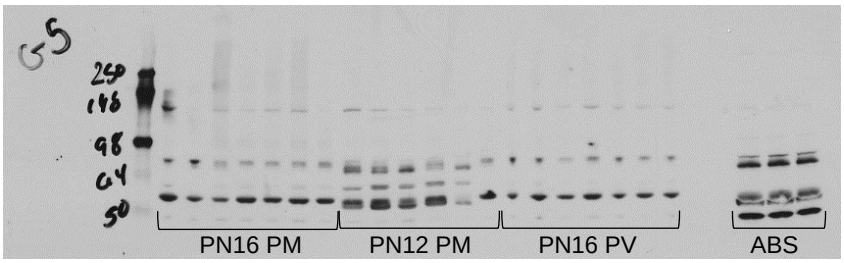
◄ PKC ι/λ

α-Tubulin comparison: Fig. 5g



◄ PKC ι/λ

α-Tubulin comparison: Fig. 5h



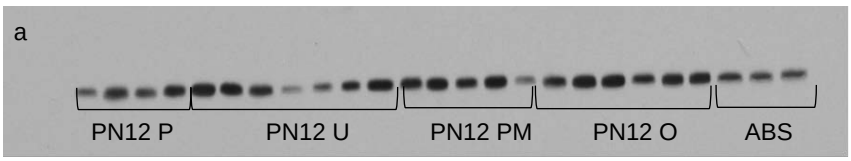
◄ PKC ι/λ

α-Tubulin comparison: Fig. 5i

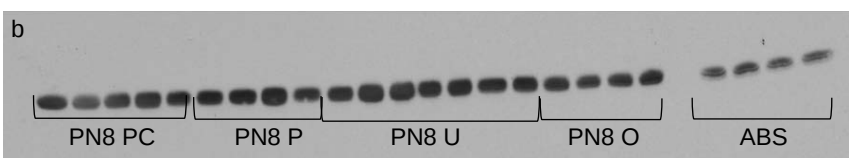
Supp. Figure 4. Western blot showing PKCι/λ (70 kDa, arrow) levels in amygdalae of pups conditioned at age postnatal day (PN)8, PN12 and PN16 and sac'd 24 hr later. P/U/O: Paired, Unpaired, Odor only conditioning. PC/PM/PV: Paired+CORT, Paired+Metyrapone, Paired+Vehicle. ABS: All brain sample, positive control. Empty lanes were loaded with 1x Laemmli Buffer. The same tubulin-corrected values were used for all markers probed. Comparisons were made across gels processed in parallel using samples derived from the same experiment. Unmarked blots were run as part of a separate study.

Supplementary Figure 5.

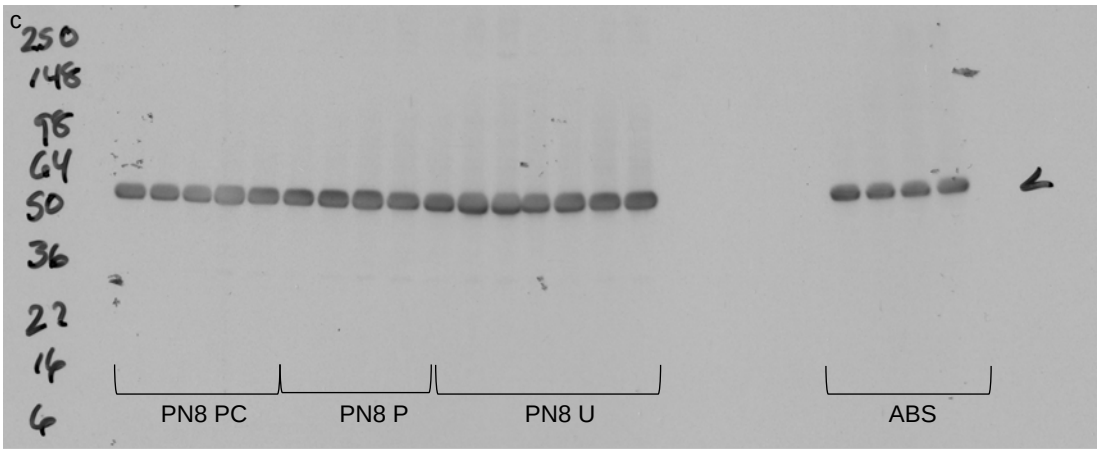
α-Tubulin Blots (50 kDa)



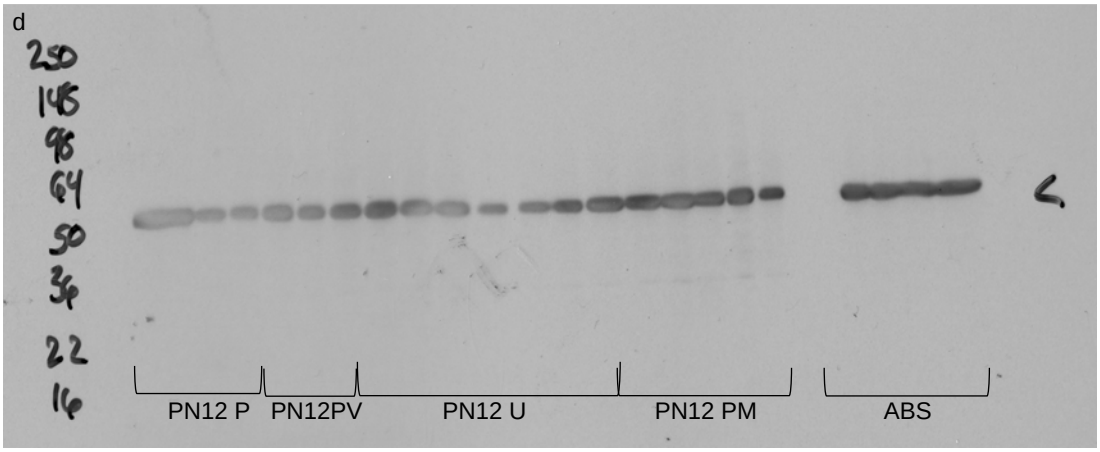
◄ α-Tubulin



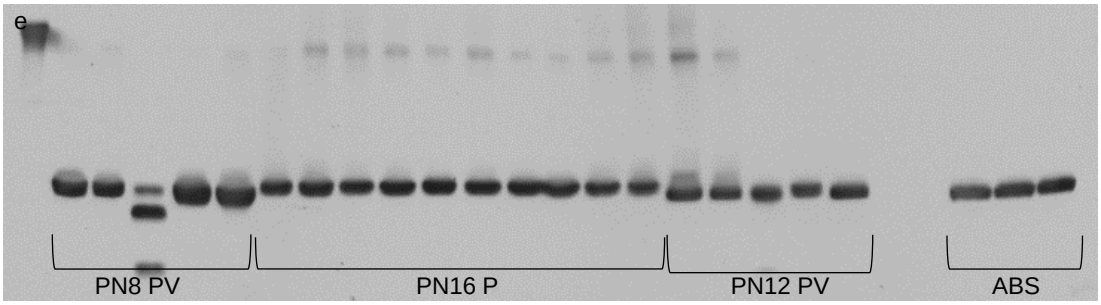
◄ α-Tubulin



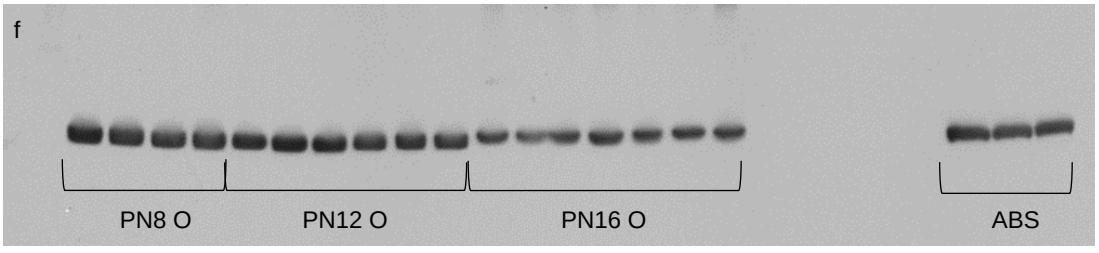
◄ α-Tubulin



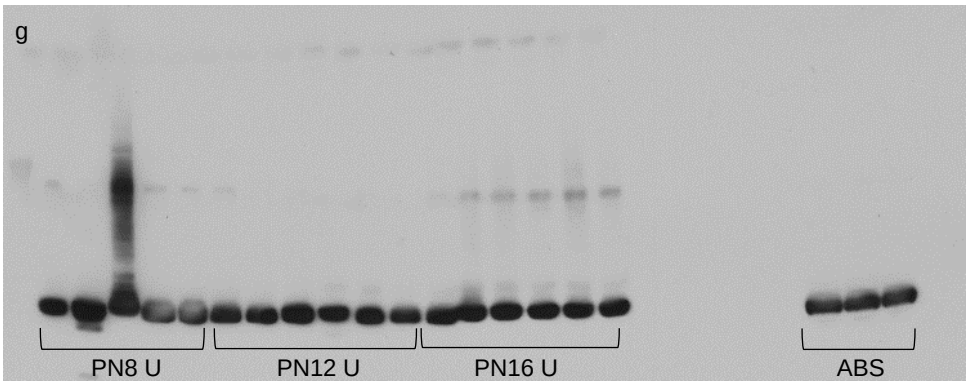
◄ α-Tubulin

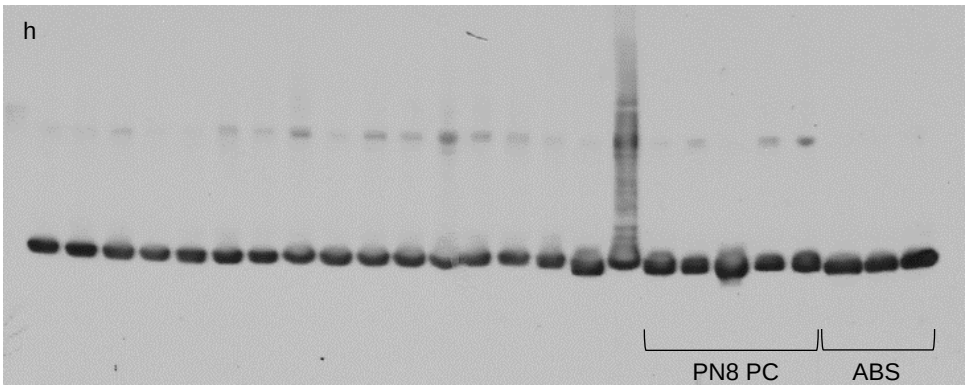


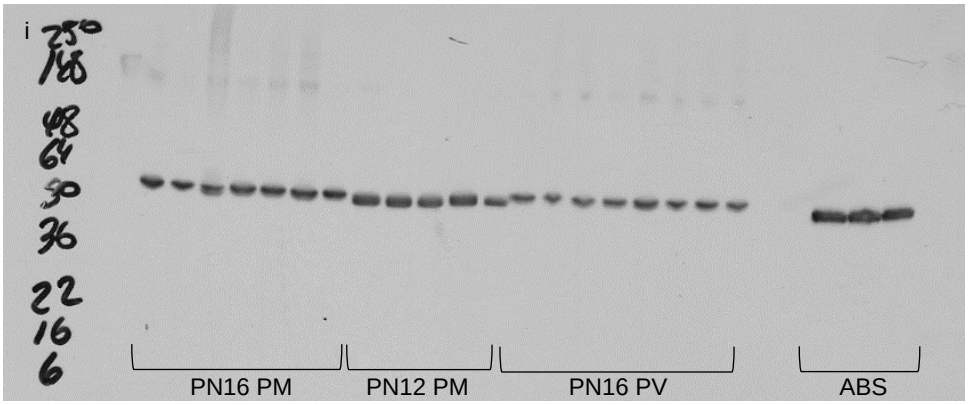
◄ α-Tubulin

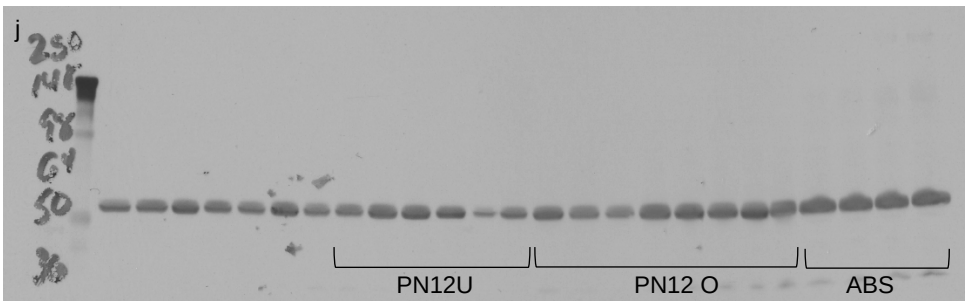


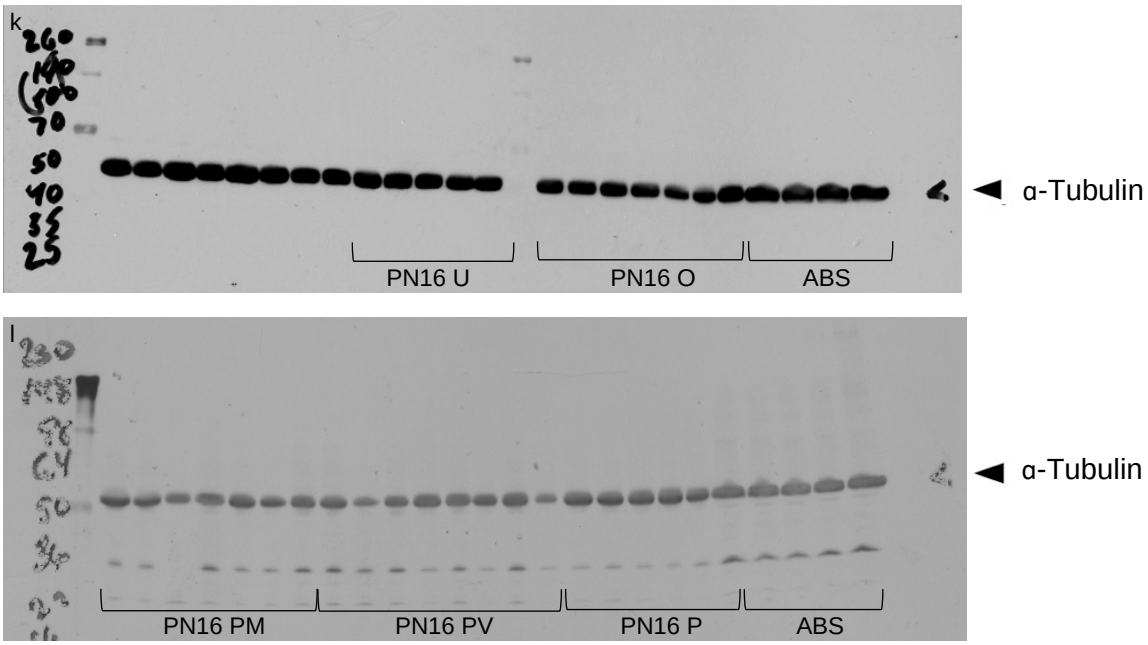
◄ α-Tubulin


g
PN8 U
PN12 U
PN16 U
ABS
◄ α-Tubulin


h
PN8 PC
ABS
◄ α-Tubulin

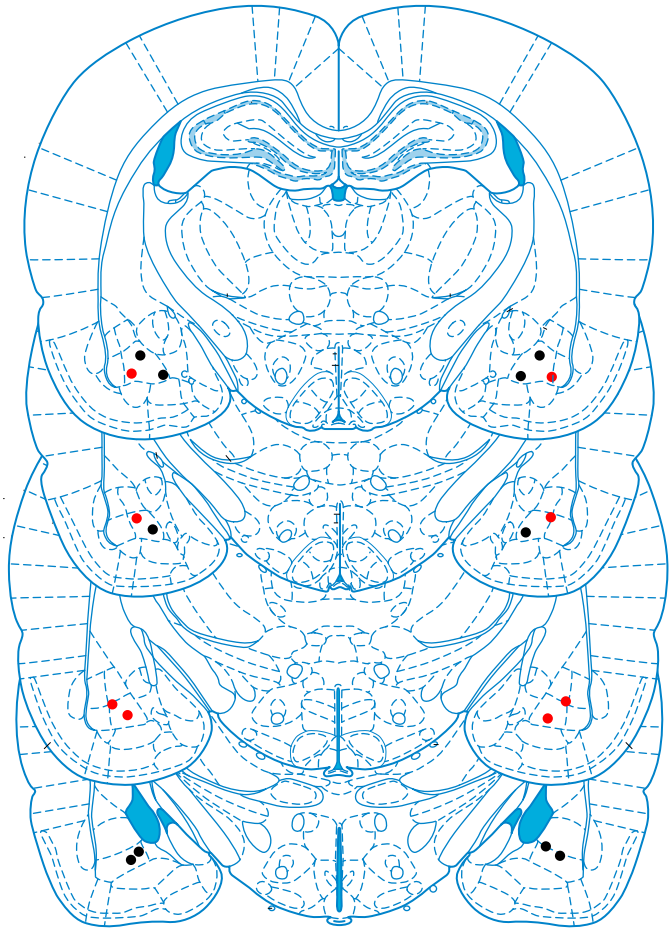

i
250
148
98
64
50
36
22
16
6
PN16 PM
PN12 PM
PN16 PV
ABS
◄ α-Tubulin


j
250
148
98
64
50
36
PN12U
PN12 O
ABS
◄ α-Tubulin

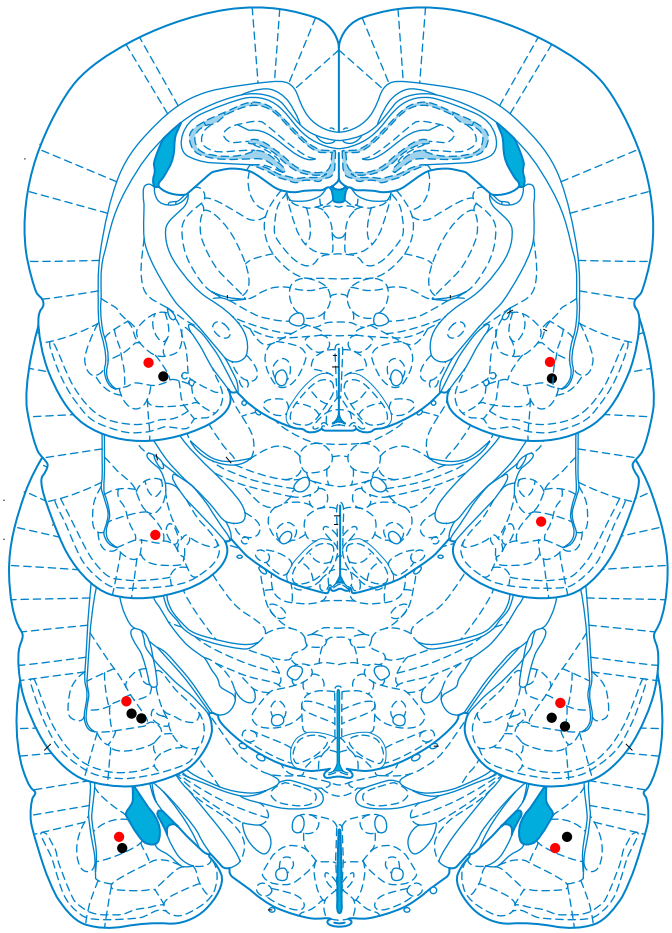


Supp. Figure 5. Western blot showing α-Tubulin (50 kDa, arrow) levels in amygdalae of pups conditioned at age postnatal day (PN)8, PN12 and PN16 and sac'd 24 hr later. P/U/O: Paired, Unpaired, Odor only conditioning. PC/PM/PV: Paired+CORT, Paired+Metyrapone, Paired+Vehicle. ABS: All brain sample, positive control. Empty lanes were loaded with 1x Laemmli Buffer. The same tubulin-corrected values were used for all markers probed. Comparisons were made across gels processed in parallel using samples derived from the same experiment. Unmarked blots were run as part of a separate study.

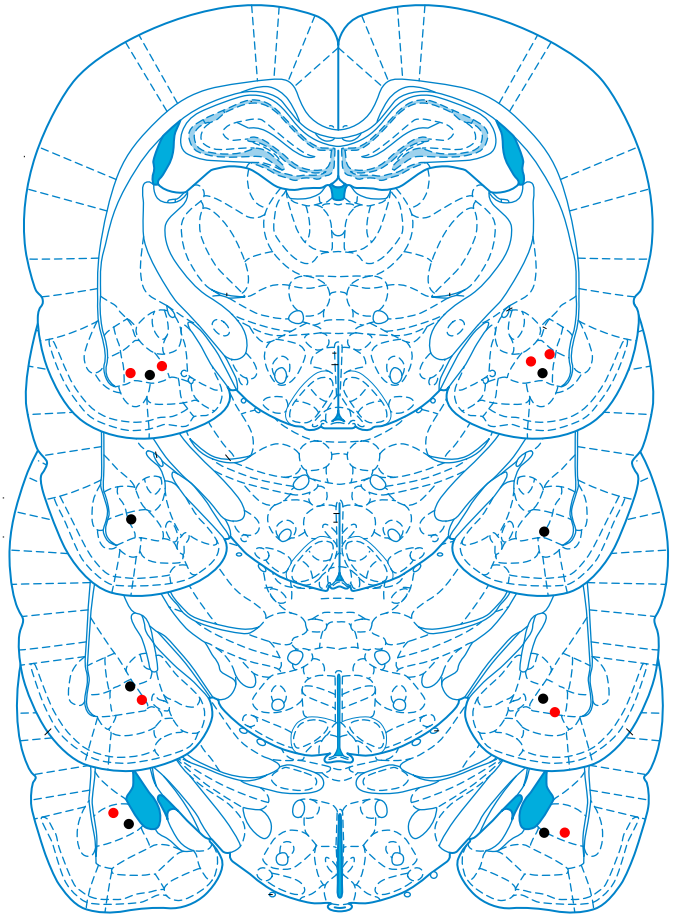
PN8



PN12



PN16



Supplementary Figure 6. Representative histology confirming cannula placement for pups receiving ZIP or Scrambled ZIP in Experiment 3 (Fig 4). Due to the large number of animals used, 4-5 representative pups per age per condition are shown here. Red dots, ZIP; black dots, Scr-ZIP.