

## **Central Lancashire Online Knowledge (CLoK)**

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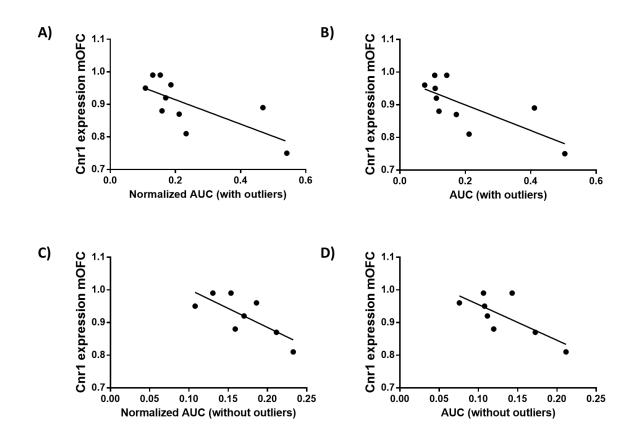
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## **Supplementary Information:**

Alternative analyses of the delay-discounting task with area under the curve (AUC) and normalized AUC

There is a debate in the preclinical literature on impulsivity about the best way to analyze impulsivity using delay-discounting curves (the two debated options are curve-fitting approaches or to compute the area under the delay-discounting curve). In addition to extracting the k parameter (see Methods section) we have also computed the AUC (normalized and non-normalized). When using these AUCs as input variables, the clustering approach was inadequate as it created two groups with a very unequal number of animals, even after removing two outliers (see SI Fig.1). In spite of this, the correlation obtained for *Cnr1* and the k parameter had an equivalent correlation between both the AUC (Magnard et al., 2018) and the normalized AUC (Myerson et al., 2001), with or without two identified outliers, and *Cnr1*, supporting the robustness of our results.



SI. Fig 1. The relationship between *Cnr1* gene expression and impulsive behavior measured (alternative analysis)

Impulsive choice was positively correlated with impulsivity measured by both measures of AUC, with or without outliers (a smaller AUC indicates a faster switch in the preference in favor of the immediate lever i.e. higher impulsivity) A) Impulsive choice (as defined by the normalized AUC, without removing outliers) was positively correlated with Cnr1 (CB<sub>1</sub> cannabinoid receptor) gene expression in the mOFC (Kendall's  $\tau$ =-0.584; p=0.02). B) Impulsive choice (as defined by the AUC, without removing the outliers) was positively

correlated with Cnr1 (CB<sub>1</sub> cannabinoid receptor) gene expression the mOFC (Kendall's  $\tau$ -0.629=; p=0.012). C) Impulsive choice (as defined by the normalized AUC without the two outliers) was positively correlated with Cnr1 (CB<sub>1</sub> cannabinoid receptor) gene expression the mOFC (Pearson's r=-0.747; p=0.033). D) Impulsive choice (as defined by the AUC without the outliers) was positively correlated with Cnr1 (CB<sub>1</sub> cannabinoid receptor) gene expression in the mOFC (Pearson's r=-0.740; p=0.036).

SI Table 1. Primer sets used for RT-qPCR amplification.

Gene	Description	Forward primer	Reverse primer
Grin1	glutamate ionotropic receptor NMDA type subunit 1	AACCTGCAGAACCGCAAG	GCTTGATGAGCAGGTCTATGC
Grin2a	glutamate ionotropic receptor NMDA type subunit 2A	TGTGAAGAAATGCTGCAAGG	GAACGCTCCTCATTGATGGT
Gria1	glutamate ionotropic receptor AMPA type subunit 1	AGAGGCTGGTGGTTGACT	ACCCTGGTATGGTCTCGGGA
Gria2	glutamate ionotropic receptor AMPA type subunit 2	GGCGTGTAATCCTGGACTGT	ACACCAGGGAATCGTCGTAG
Gabrg2	gamma-aminobutyric acid type A receptor gamma 2 subunit	CGGAAACCAAGCAAGGATAA	ACAGTCCTTGCCATCCAAAC
Gabrd	gamma-aminobutyric acid type A receptor delta subunit	GCTGGACCTGGAGAGCTATG	CCGAAGCTGGAAGTGTAAGC
Gabra1	gamma-aminobutyric acid type A receptor alpha 1 subunit	TTGACTGTGAGAGCCGAATG	AAACGTGACCCATCTTCTGC
Gabra2	gamma-aminobutyric acid type A receptor alpha 2 subunit	CCATGCACTTGGAGGACTT	ACTGGCCCAGCAAATCATAC
Cnr1	cannabinoid receptor 1	GTCGATCCTAGATGGCCTTGC	GTCATTCGAGCCCACGTAGAG
Dagla	diacylglycerol lipase, alpha	CTTTGCTGAATTTTTCCGTGACC	TTGTTTGCCTCATCCAGCAC
Mgll	monoacylglycerol lipase	CTACCTGCTCATGGAATC	GACACCCACGTATTTATTTC
Napepld	N-acyl phosphatidylethanolamine phospholipase D	AGATATGGACTCAAGAGTGAAGACTTC	TCCTCAAAGGCTTTGTCATCGG
Faah	fatty acid amide hydrolase	GTTACAGAGTGGAGAGCTGTCC	GTCTCACAGTCGGTCAGATAGG
Gapdh	glyceraldehyde-3-phosphate dehydrogenase	TCCCTGTTCTAGAGACAG	CCACTTTGTCACAAGAGA