Supplementary figure S1

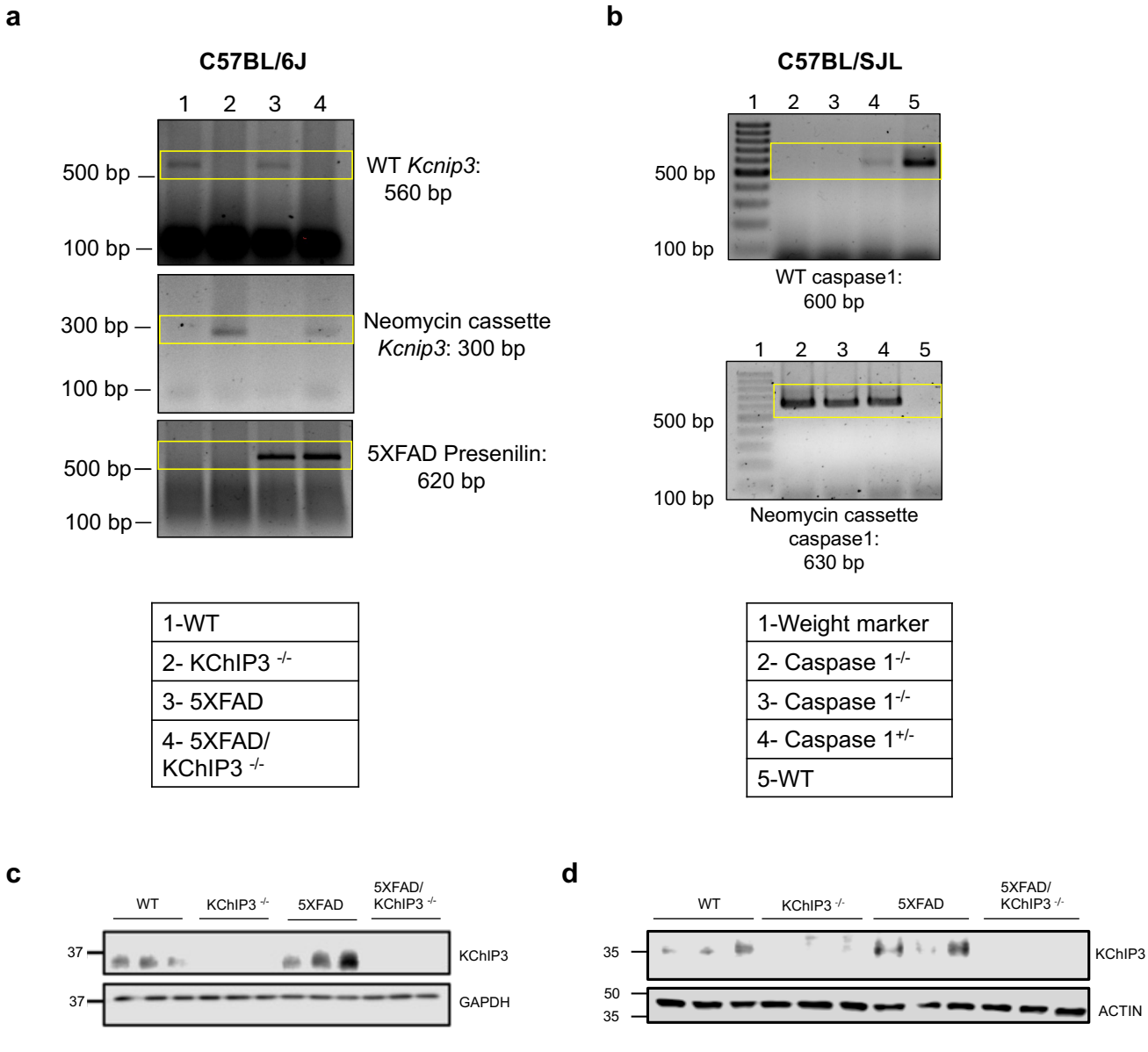


Fig. S1 5XFAD genotype. a Representative agarose gel showing the genotype of WT KChIP3, KChIP3$^{-/-}$ (neomycin cassette), 5XFAD (presenilin) and 5XFAD/KChIP3 $^{-/-}$ mice in the C57BL/6J genetic background. **b** Representative agarose gel showing the genotype of WT Caspase 1, Caspase 1$^{+/-}$ (neomycin cassette), and Caspase 1$^{-/-}$ mice in the C57BL/SJL genetic background. **c-d** Representative western blots showing KChIP3 protein levels in WT, KChIP3$^{-/-}$, 5XFAD and 5XFAD/KChIP3$^{-/-}$ mice in the C57BL/6J genetic background. Hippocampal tissue (30 µg) from male mice of 11 months old, GAPDH and ACTIN were used as a loading control, respectively.

Supplementary figure S2

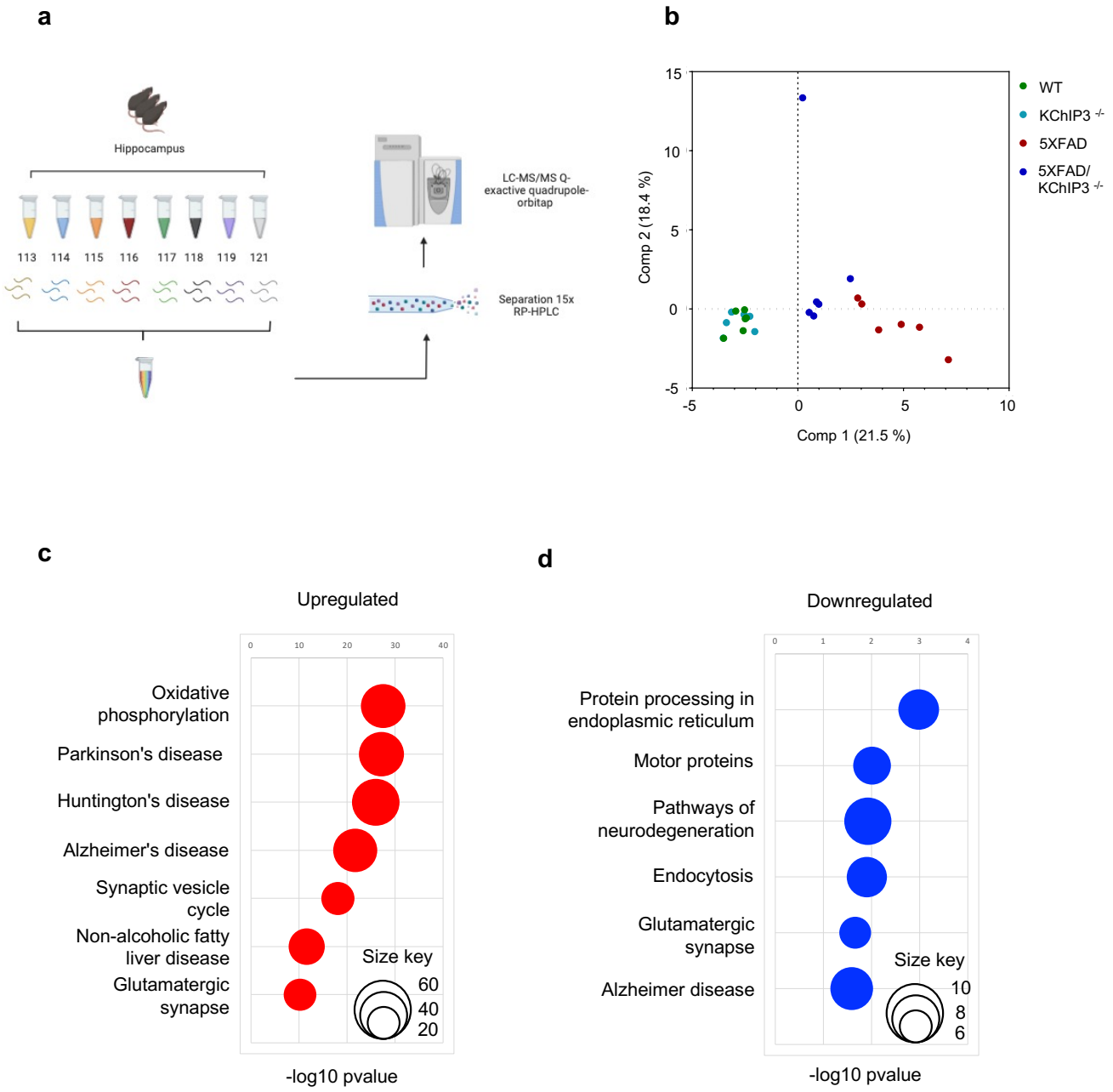


Fig. S2 Hippocampal proteome analysis. **a**, Experimental design of the proteomic study. Hippocampi of three mice per genotype were used. 30 µg of protein extract was taken and digested with trypsin. After digestion, proteins were tagged with an iTRAQ 8 plex. The peptide mixture was submitted to pre-fractionated step by reverse chromatography (RP-HPLC). Fifteen final fractions were obtained. Subsequently, the samples were injected into an LC-MS/MS Q-exactive quadrupole Orbitrap. **b**, Principal component analyses of all replicates was performed with proteins showing an absolute log2 fold change > 0.5 and an FDR < 0.05. **c**, Enrichment pathways analysis of upregulated proteins in 5XFAD *vs.* 5XFAD/KChIP3$^{-/-}$ (PC1 positive). **d**, Analysis of enriched pathways of downregulated proteins in 5XFAD/KChIP3$^{-/-}$ *vs.* 5XFAD (PC1 negative).

Supplementary figure S3

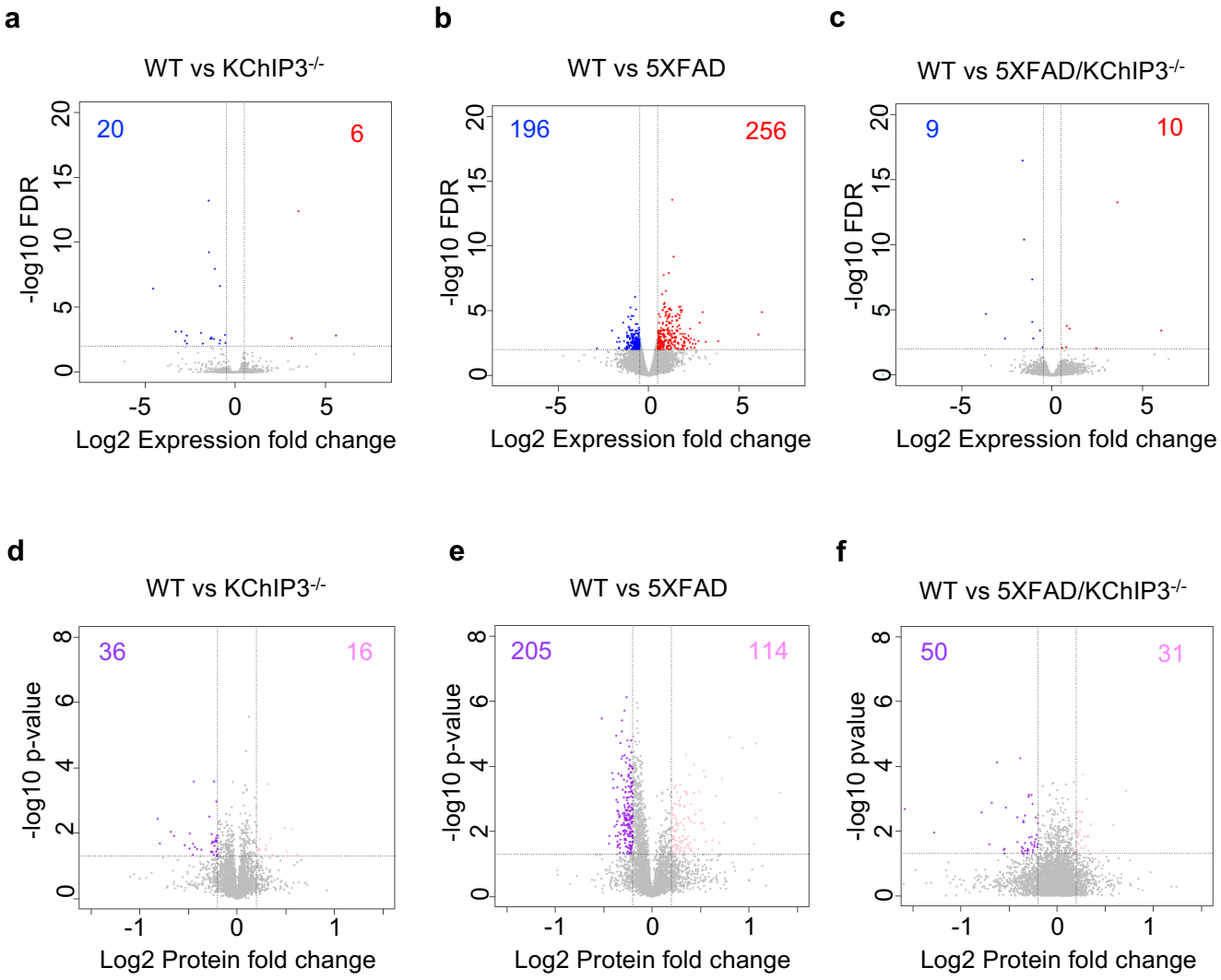
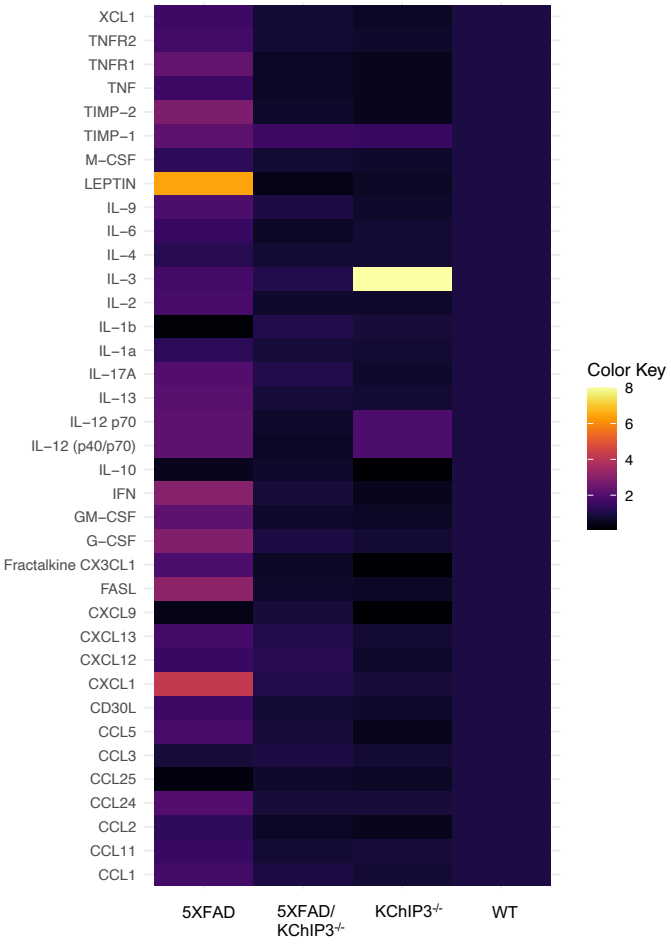


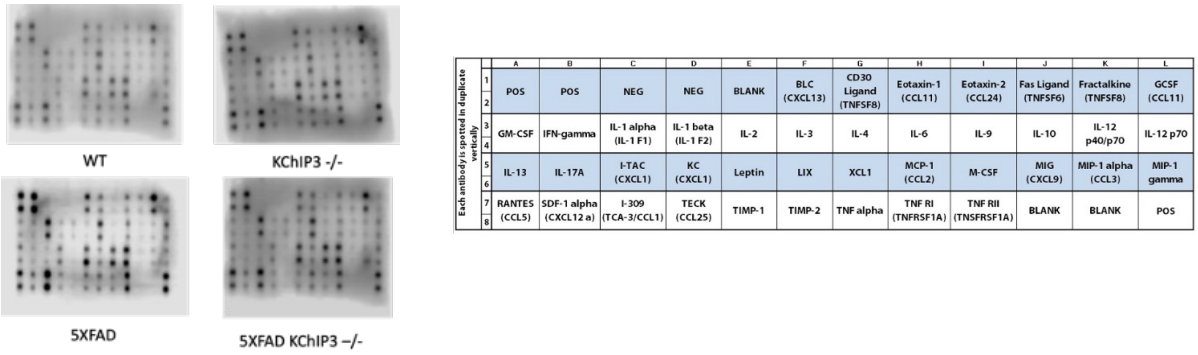
Fig. S3 KChIP3$^{-/-}$ partially rescues gene expression defects of 5XFAD mice. Volcano plots comparing gene expression (**a-c**) and protein levels (**d-f**) in the hippocampus of KChIP3$^{-/-}$ *vs*. WT mice (left), 5XFAD *vs.* WT (center) and 5XFAD/KChIP3$^{-/-}$ *vs.* WT (right). The number of statistically significant down- and up-regulated genes/proteins is shown in blue/purple and red/pink, respectively. Proteome data were analyzed using an unpaired Student's *t*-test.

Suplementary figure S4

a



b



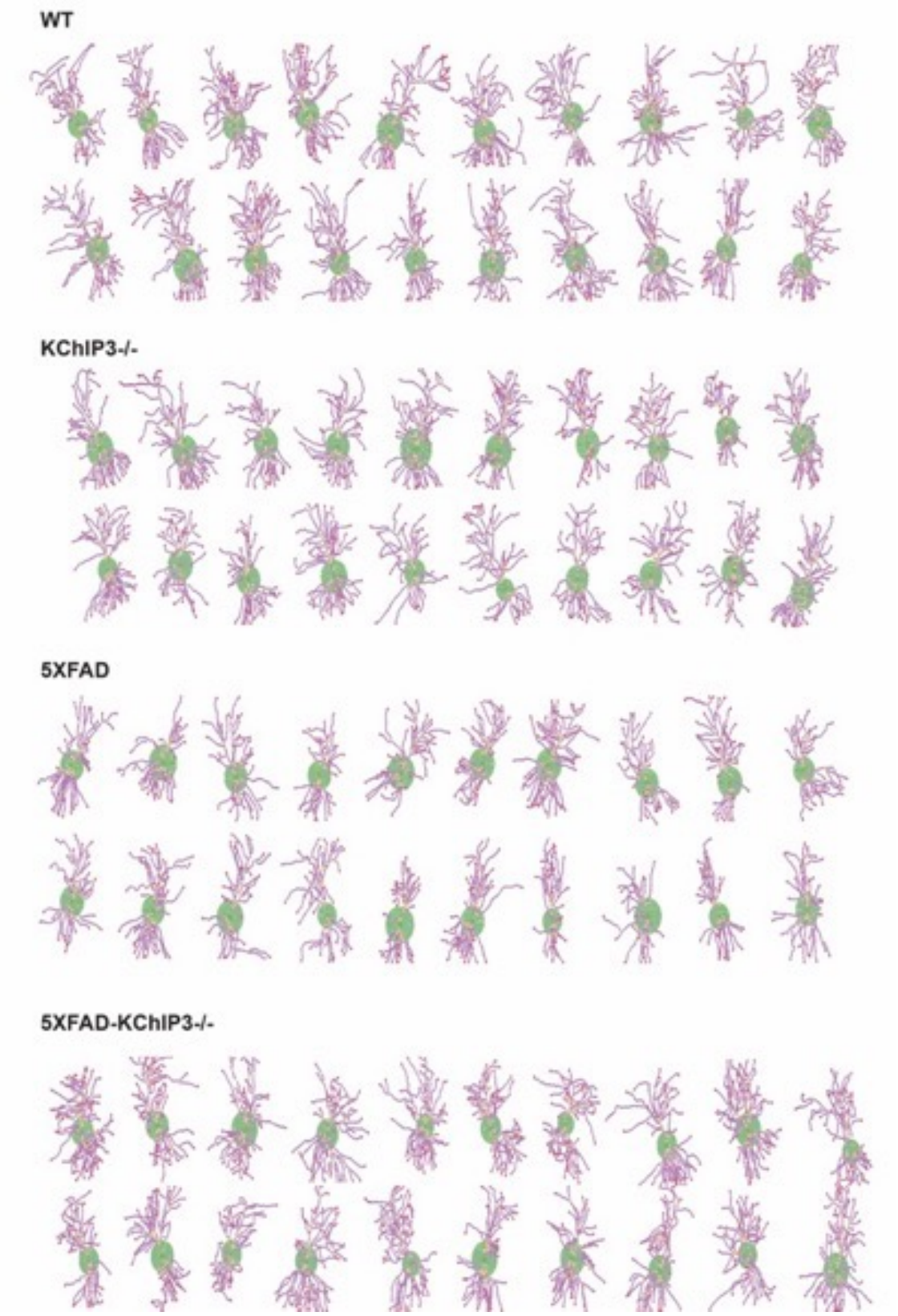
Each antibody is spotted in duplicate vertically

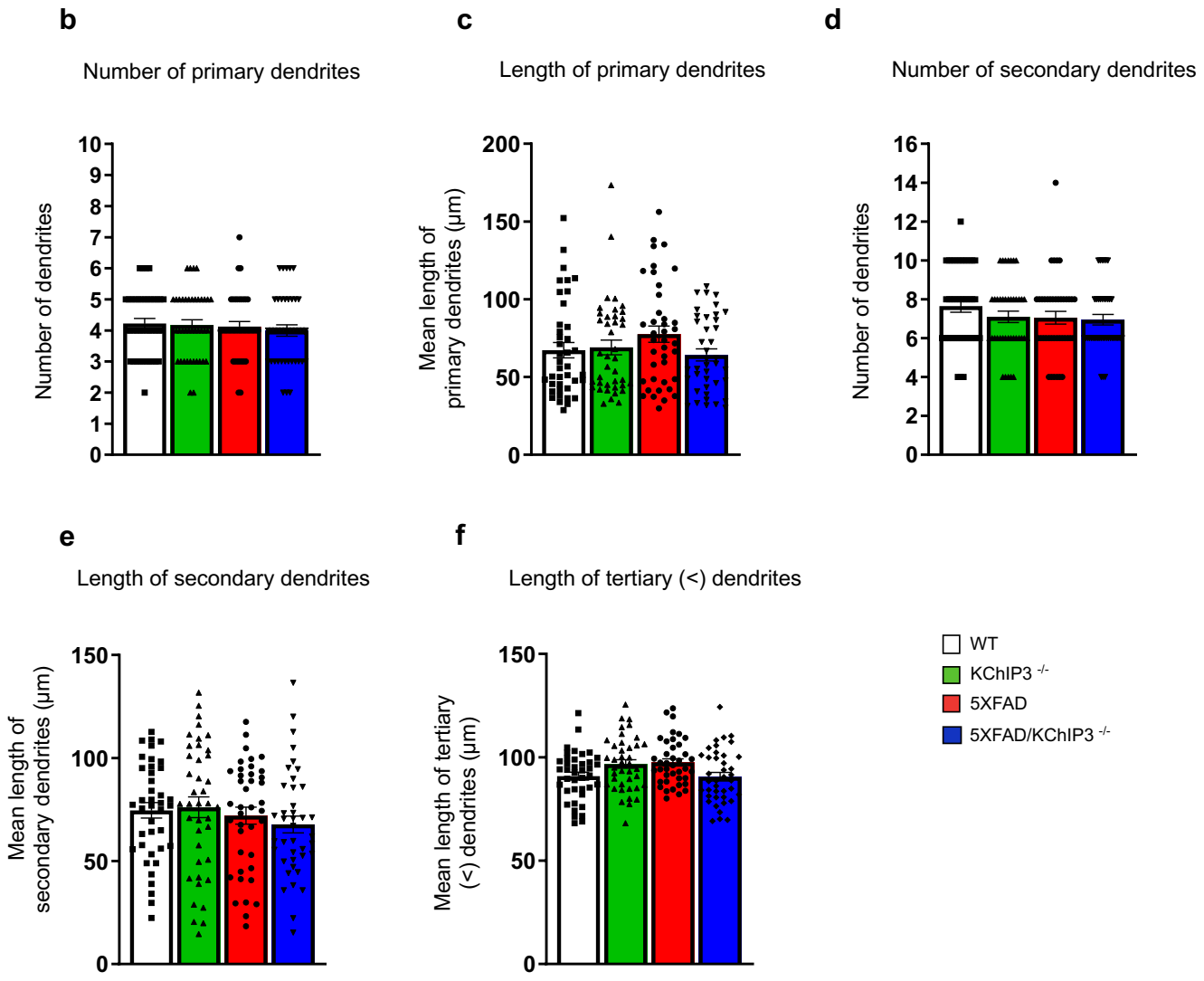
	A	B	C	D	E	F	G	H	I	J	K	L
1	POS	POS	NEG	NEG	BLANK	BLC (CXCL13)	CD30 Ligand (TNFSF8)	Eotaxin-1 (CCL11)	Eotaxin-2 (CCL24)	Fas Ligand (TNFSF6)	Fractalkine (TNFSF8)	GCSF (CCL11)
3	GM-CSF	IFN-gamma	IL-1 alpha (IL-1 F1)	IL-1 beta (IL-1 F2)	IL-2	IL-3	IL-4	IL-6	IL-9	IL-10	IL-12 p40/p70	IL-12 p70
5	IL-13	IL-17A	I-TAC (CXCL1)	KC (CXCL1)	Leptin	LIX	XCL1	MCP-1 (CCL2)	M-CSF	MIG (CXCL9)	MIP-1 alpha (CCL3)	MIP-1 gamma
7	RANTES (CCL5)	SDF-1 alpha (CXCL12 a)	I-309 (TCA-3/CCL1)	TECK (CCL25)	TIMP-1	TIMP-2	TNF alpha	TNF RI (TNFRSF1A)	TNF RII (TNSFRSF1A)	BLANK	BLANK	POS

Fig. S4 Deletion of KChIP3 decreases cytokines and chemokine levels in the hippocampus of the 5XFAD mice. Protein extracts were prepared from hippocampal tissue of WT, KChIP3$^{-/-}$, 5XFAD, and 5XFAD/KChIP3 $^{-/-}$ mice. Cytokines and chemokines levels were determined using an antibody array (C1 RayBio® C-Series). **a**, Heatmap showing the cytokines and chemokines accumulation profile. Densitometry values represent the fold change normalized to 1 (WT). **b**, Membranes showing signals for cytokines and chemokines present in the antibody array (right panel).

Supplementary figure S5

a




b
Number of primary dendrites
Number of dendrites
c
Length of primary dendrites
Mean length of primary dendrites (µm)
d
Number of secondary dendrites
Number of dendrites
e
Length of secondary dendrites
Mean length of secondary dendrites (µm)
f
Length of tertiary (<) dendrites
Mean length of tertiary (<) dendrites (µm)
WT
KChIP3 -/-
5XFAD
5XFAD/KChIP3 -/-

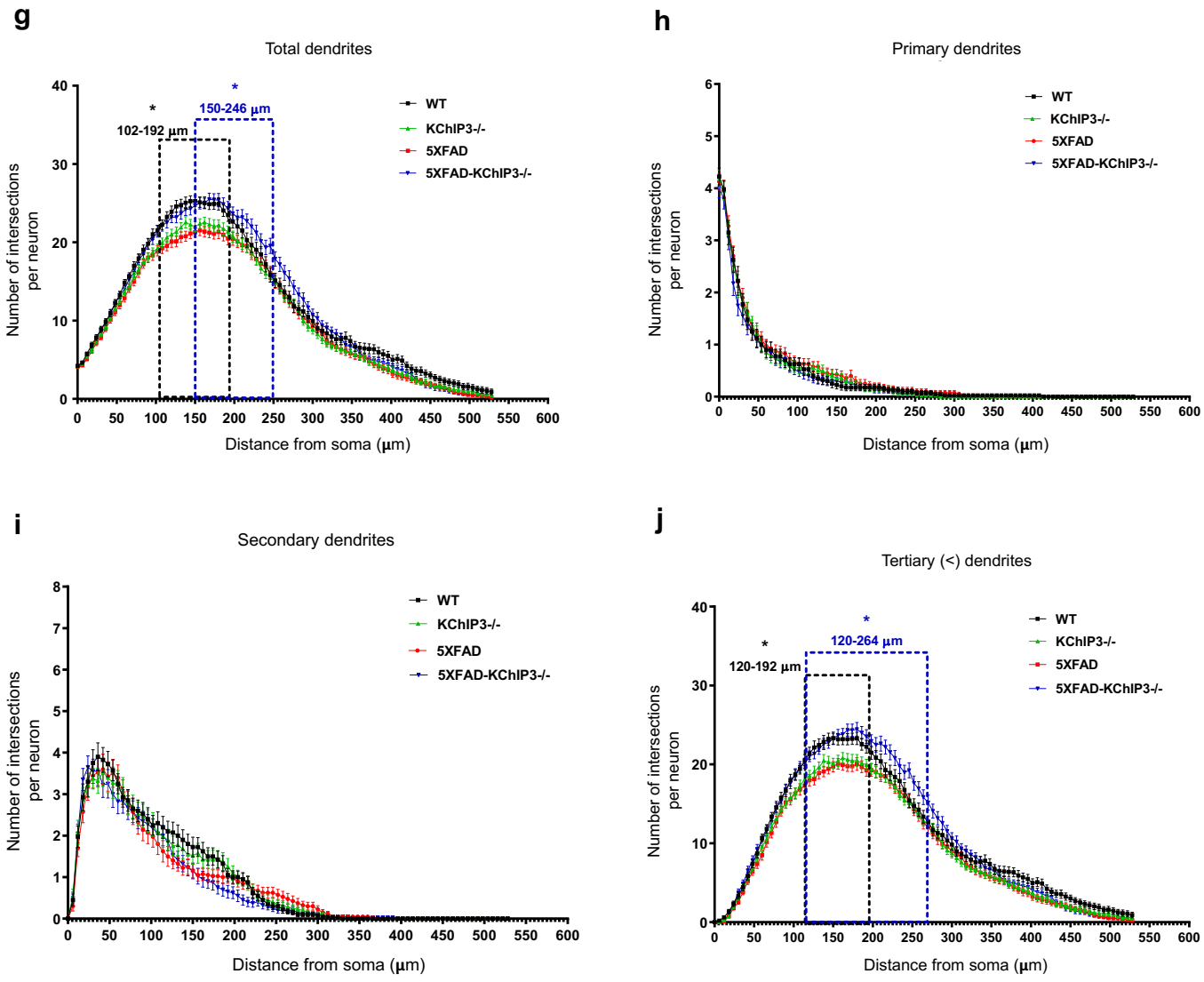


Fig. S5 Evaluation of the dendritic tree branching pattern of hippocampal neurons. **a**, The representative reconstructions of twenty CA1 neurons obtained from Sholl analysis are displayed for WT, KChIP3$^{-/-}$, 5XFAD, and 5XFAD/KChIP3$^{-/-}$ mice. The soma position is shown in green and dendrites in pink. The number of primary (**b**) and secondary (**d**) dendrites is shown for each group. The length of primary (**c**), secondary (**e**) and tertiary < (**f**) dendrites is shown for each group. The number of intersections as a function of the distance from the soma in total (**g**), primary (**h**), secondary (**i**), and tertiary < (**j**) dendrites is shown for each group. **b-j** Results are expressed as mean ± SEM. n= 40 neurons per group, ten neurons per animal. Two-way ANOVA, post hoc Tukey test: *p < 0.05. Black box: *p < 0.05, 5XFAD and KChIP3$^{-/-}$ *vs.* WT group. Blue box: *p < 0.05, 5XFAD and KChIP3$^{-/-}$ *vs.* 5XFAD/KChIP3$^{-/-}$ group.

Supplementary figure S6

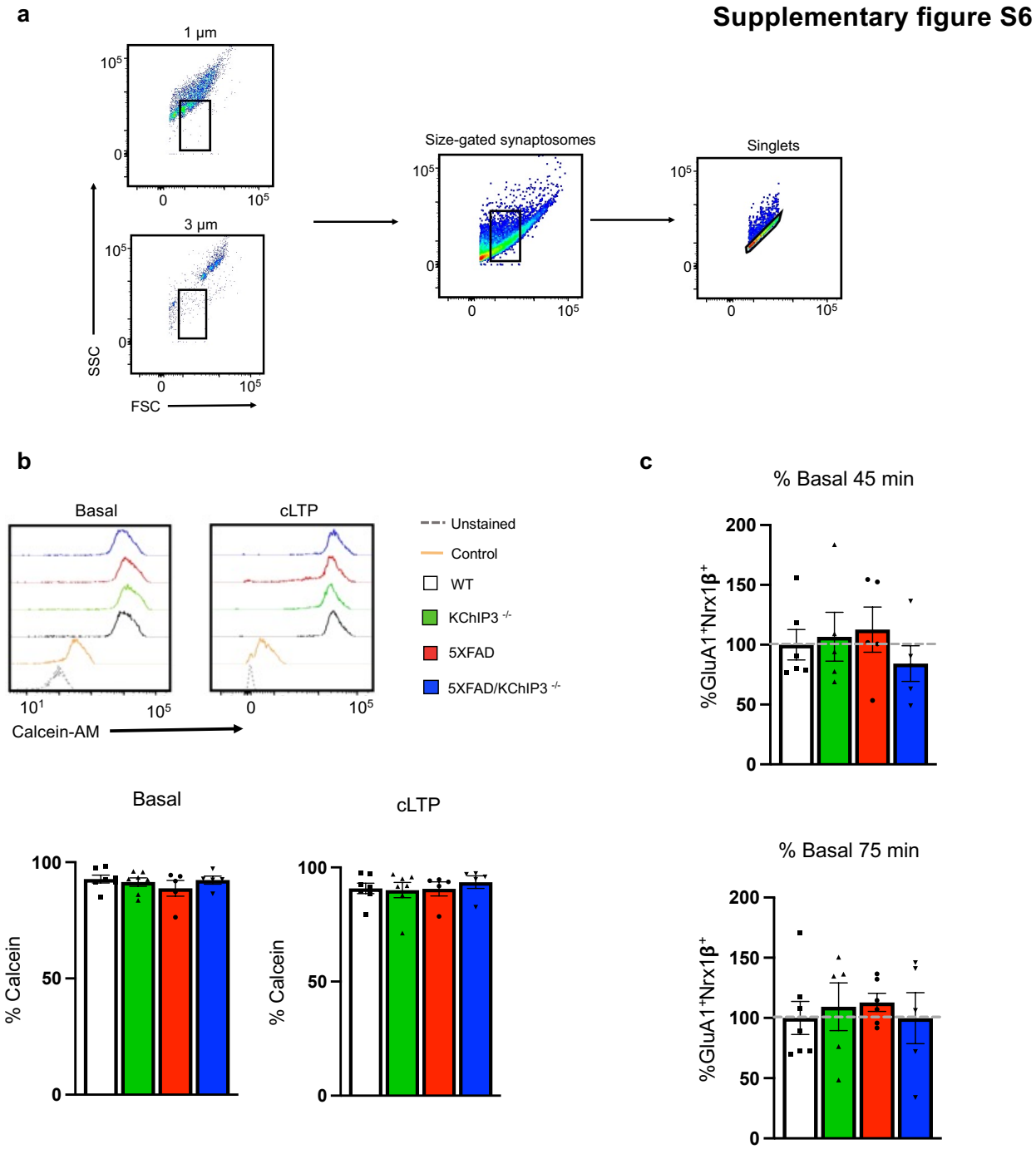


Fig. S6 Size-gated synaptosomes identification and evaluation of synaptosomal membrane integrity. **a**, Size-gated synaptosome were identified on FSC-SSC plots based on calibrated bead of 1µm to 3 µm. Then, singlets profile was select using FSC-A *vs.* FSC-H bivariate plots. **b**, Membrane integrity of hippocampal size-gated synaptosomes were test by calcein-AM stain after FASS-LTP protocol. Graph data represent both stimulation times (45 and 75 minutes) at basal or cLTP conditions of WT, KChIP3$^{-/-}$, 5XFAD, 5XFAD/KChIP3$^{-/-}$ mice. **c**, Percentage of GluA1^{+}/Nrx1β^{+} in basal state at 45 minutes and 75 minutes from KChIP3$^{-/-}$, 5XFAD, 5XFAD/KChIP3$^{-/-}$ mice compared to WT mice. n= 5-7 mice.

Supplementary figure S7

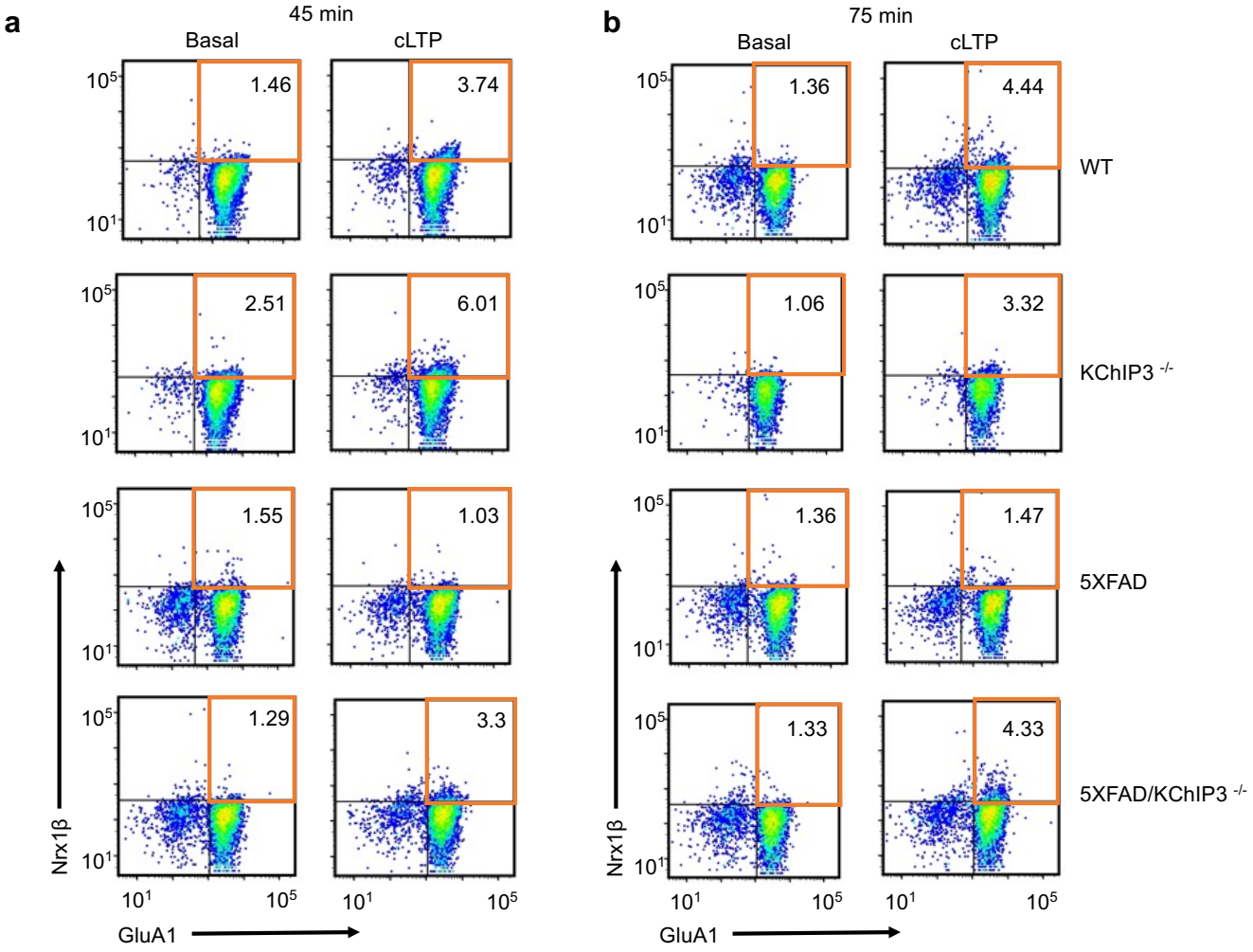


Fig. S7 Deletion of KChIP3 promotes Long-Term Potentiation (LTP) in 5XFAD mice. Representative dot plots of basal controls and cLTP responses at 45 and 75 minutes of WT, KChIP3$^{-/-}$, 5XFAD, 5XFAD/KChIP3$^{-/-}$ mice. The GluA1^{+}/Nrx1β^{+} double-positive of size-gated synaptosomes were visualized in upper right quadrant. **a** Dot Plots at 45 minutes under basal condition and after cLTP induction. **b** Dot Plots at 75 minutes under basal condition and after cLTP induction.

Supplementary figure S8

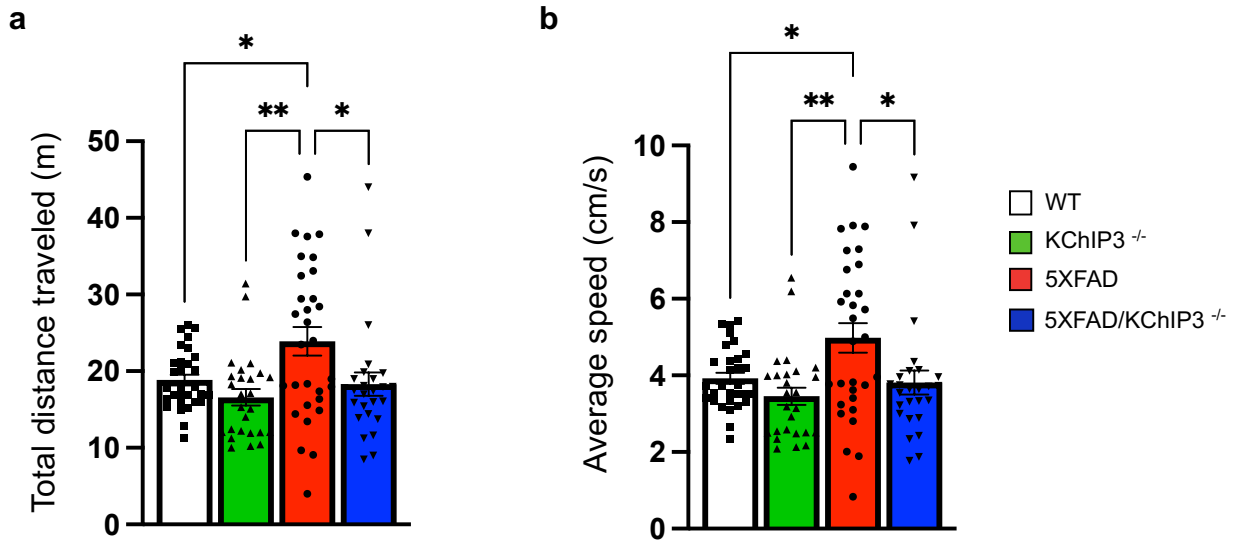


Fig. S8 Deletion of KChIP3 does not cause locomotor activity changes respect to wild-type mice.

Total distance (**a**) and average speed (**b**) in Y-maze were determined in WT, KChIP3$^{-/-}$, 5XFAD and 5XFAD/KChIP3$^{-/-}$ mice. Mean ± SEM. One-way ANOVA followed by Turkey Multiple Comparison test. * $p<0.05$ ** $p<0.01$ mice of 9-12 months old.

Supplementary STable 1. **Animals used in each technique**

Technique		Number of mice				
	Genotype	Age (months)				
		9	10	11	12	Total
Immunoblots C57BL/6J	WT	0	3	3	3	9
	KChIP3$^{-/-}$	0	3	3	3	9
	5XFAD	0	3	3	3	12
	5XFAD/KChIP3$^{-/-}$	0	3	4	4	12
ELISA assays (All cytokines)	WT	0	2	2	2	6
	KChIP3$^{-/-}$	0	2	2	2	6
	5XFAD	0	2	2-3	2-3	7-8
	5XFAD/KChIP3$^{-/-}$	0	2	2-3	2-3	7-8
Immunolabeling	WT	7	0	0	0	7
	KChIP3$^{-/-}$	7	0	0	0	7
	5XFAD	7	0	0	0	7
	5XFAD/KChIP3$^{-/-}$	7	0	0	0	7
	Genotype	Age (months)				
		9	10	11	12	Total
RNAseq Proteome	WT	3	0	0	0	3
	KChIP3$^{-/-}$	3	0	0	0	3
	5XFAD	3	0	0	0	3
	5XFAD/KChIP3$^{-/-}$	3	0	0	0	3
Dendritic tree	WT	0	0	3	1	4
	KChIP3$^{-/-}$	0	0	2	2	4
	5XFAD	0	0	1	3	4
	5XFAD/KChIP3$^{-/-}$	0	0	1	3	4
FASS-LTP (L-LTP)	WT	0	2	2	3	7
	KChIP3$^{-/-}$	0	2	2	2	6
	5XFAD	0	2	2	2	6
	5XFAD/KChIP3$^{-/-}$	0	2	2	2	6
	Genotype	Age (months)				
		9	10	11	12	Total
Behavior test (Y maze-alternation)	WT	3	7	6	14	30
	KChIP3$^{-/-}$	3	7	6	10	26
	5XFAD	3	6	4	13	26
	5XFAD/KChIP3$^{-/-}$	3	7	6	14	30
Behavior test (RON)	WT	12	9	0	0	21
	KChIP3$^{-/-}$	8	14	0	0	21
	5XFAD	7	8	0	0	15
	5XFAD/KChIP3$^{-/-}$	9	6	0	0	15
Immunoblots (C57BL/SJL)	WT	0	2	2	2	6
	Caspase1$^{-/-}$	0	2	2	2	6
	5XFAD	0	2	2	3	7
	5XFAD/Caspase1$^{-/-}$	0	2	2	3	7

Supplementary STable 2. **Common RNA and Protein regulated in the hippocampus of WT, KChIP3$^{-/-}$, 5XFAD and 5XFAD/KChIP3$^{-/-}$ mice**

Downregulated

Protein levels					RNA levels				
Gene	**WT**	**KCHIP3 KO**	**5xFAD**	**KCHIP3 KO/5xFAD**	**Gene**	**WT**	**KCHIP3 KO**	**5xFAD**	**KCHIP3 KO/5xFAD**
Cacnb4	-0.056572	-0.145847758	-0.394667745	-0.015132229	Cacnb4	4.288654438	4.203433774	3.434326986	3.78521903
Cd200	0.146245122	0.017782569	-0.146354318	-0.072096944	Cd200	6.275110882	6.119610608	5.503940261	5.838361611
Cd47	0.015554865	0.00547874	-0.430268407	-0.114846985	Cd47	5.720568046	5.731696051	5.003868872	5.504719474
Gda	0.057059765	0.034967184	-0.261004527	-0.059122006	Gda	6.248025055	6.036965077	5.23083159	5.675483297
Mapk8	0.044030309	0.029388428	-0.206225276	-0.283804655	Mapk8	4.356829973	4.319987443	3.655776735	4.16670992
Negr1	-0.004671971	0.013588111	-0.354668617	-0.178506374	Negr1	4.768477942	4.460300023	3.653900235	4.326397328
Pkia	-0.069955548	-0.037191192	-0.324156245	0.055828452	Pkia	5.875446839	5.737244063	4.8894178	5.571748912
Plcb1	0.008297284	0.02808404	-0.273258289	-0.164627473	Plcb1	4.588603531	4.62618209	3.797930704	4.418129526

Upregulated

Protein levels					RNA levels				
Gene	**WT**	**KCHIP3 KO**	**5xFAD**	**KCHIP3 KO/5xFAD**	**Gene**	**WT**	**KCHIP3 KO**	**5xFAD**	**KCHIP3 KO/5xFAD**
Apoe	-0.461029649	-0.414539059	1.257875562	0.874520262	Apoe	12.14681795	11.99575275	12.92039943	12.50177846
C1qa	-0.113000453	-0.140226305	0.439179242	0.566701114	C1qa	8.187477359	7.763352649	9.881081084	9.177304526
C1qb	-0.104554558	-0.129721355	0.696012974	0.495683193	C1qb	8.243281619	7.958119206	9.8576694	9.27001342
C1qc	-0.090371788	-0.089108288	0.568316042	0.389075458	C1qc	7.349838217	6.938040212	8.966207902	8.308643426
Cd9	-0.19537735	-0.14087081	0.190361261	0.116476138	Cd9	6.229240832	6.257549943	7.457359118	7.119266129
Clu	-0.262458603	-0.173057119	0.808144848	0.512906988	Clu	10.1296961	10.08835733	10.81131129	10.47316728
Cst3	-0.084436973	-0.095543226	0.1225365	0.014667988	Cst3	12.20309616	12.00609613	13.0072676	12.56756377
Ctsd	-0.040079117	-0.051360011	0.205940247	0.07163775	Ctsd	9.23404979	9.004694812	10.81449889	10.16921281
Dbi	-0.028868079	-0.07001698	0.233253201	0.259279132	Dbi	8.800208517	8.717650158	9.301066058	9.225820927
Dhrs1	-0.071300983	-0.100473324	0.32679232	0.086446524	Dhrs1	6.697559452	6.646353694	7.093269736	6.937878192
Fxyd1	-0.055032055	-0.076212128	0.429891229	0.237775127	Fxyd1	5.99635705	6.164708958	6.963891549	6.557194212
Gfap	-0.325485229	-0.199614922	1.60783871	0.981987238	Gfap	8.818521038	8.875994402	10.77211974	10.30979288
Gltp	-0.275189479	-0.148683389	-0.036259015	0.168067455	Gltp	6.133098021	6.310042592	6.688376349	6.487365465
Gsn	-0.073927482	-0.074991544	0.165619532	0.075981935	Gsn	6.531336528	6.732080355	7.016942585	6.983743862
Hexa	-0.221623468	-0.124676752	0.150062704	0.072384405	Hexa	7.262180382	6.930212128	8.106773959	7.658693907
Hexb	-0.230438153	-0.287318865	0.319529374	0.273292859	Hexb	7.385902591	7.196738629	8.487898318	8.118832007
Lgals1	0.027273774	0.01638337	0.37824746	0.374525587	Lgals1	6.500762929	6.562868247	6.969750083	6.811709555
Mdk	-0.441714406	-0.511006872	1.310821335	0.976404309	Mdk	5.503106741	5.551797388	6.007961065	5.76174102
Padi2	-0.006261015	-0.083743143	0.220160913	0.052185965	Padi2	4.512457612	4.954370248	4.919722669	5.249406382
Prdx6	-0.140333494	-0.069347143	0.290874879	0.243605852	Prdx6	7.467136366	7.520398218	8.087389851	7.879099585
Rhog	-0.226116419	-0.123067061	-0.000365257	0.328319391	Rhog	5.924517022	6.085602458	6.639558325	6.420816296
S100a13	-0.114111026	-0.035435677	0.094161669	0.111557166	S100a13	6.760672203	6.771061021	7.271003292	7.07991355
S100a6	-0.101034085	-0.093285561	0.975600878	0.121423086	S100a6	5.911116203	5.427009431	6.787342089	6.57019182
Sdc4	-0.046481133	-0.065690517	0.22142601	-0.198841413	Sdc4	6.854919849	6.854728175	7.336675823	7.026386144
Slc14a1	-0.096636629	-0.039981222	0.622224474	0.275040293	Slc14a1	3.592248127	3.770095918	4.45389327	4.242564468

Supplementary STable 3. **Predicted genes containing DRE motifs in their promotor region**
The proteins products of these genes are involved in proinflammatory/amyloidogenic processes.

Gene	Total DRE motifs	Associated functions
Apoe	4	Regulates processes of lipid metabolism, cholesterol and the biogenesis of ßA peptide in AD[96–98]
Clu	1	Proinflammatory glycoprotein. Increased levels in serum and brain in AD[99,100]
Mdk	3	Proinflammatory cytokine, increases expression of chemokines. Increased levels in serum, brain and promotes ßA accumulation in AD[101]
S100a13	2	Calcium binding protein associated with proinflammatory processes and AD[102]
Fxyd1	7	Membrane protein that controls Na^+, K^+ -ATPase (NKA). Increased levels compromise dendritic arborization in the frontal cortex in Rett syndrome[103]

Transcripts and proteins products of these genes are reduced in hippocampus of 5XFAD/KChIP3$^{-/-}$ *vs.* 5XFAD.
ßA: Beta Amyloid; AD: Alzheimer's disease

Supplementary STable 4. **Predicted genes containing DRE motifs in their promotor region**
The proteins products of these genes are involved in synaptic/LPT processes.

Gene	Total DRE motifs	Associated functions
Cd47	1	Protects neurons from synaptic pruning in development brain and ageing[73,78,79]
Cacnb4	1	Subunit of VGCCs which are present at specific presynaptic sites. Involved in dendritogenesis processes in psychiatric disorders[104–106]
Gda	1	Interacts with PSD95. Promotes dendritic growth[107]

Transcripts and proteins products of these genes are rescued in hippocampus of 5XFAD/KChIP3$^{-/-}$vs 5XFAD.
Cacnb4: voltage-gated calcium channel subunit β4; VGCCs: Voltaje-gated Calcium Channels

Supplementary STable 5. **Genes enriched in the 5XFAD hippocampus associated with the DAM-1 microglial phenotype are reduced in 5XFAD/KChIP3$^{-/-}$ mice**

Gene	Associated functions
Apoe	Risk factor associated with sporadic AD[96–98]
B2m	B2M is a subunit of MHC. Increased levels in serum and CSF associated with AD[108]
Ctsb	Cathepsin B. Increased levels in plasma and CSF associated with cognitive dysfunction in AD and TBI[109]
Ctsd	Cathepsin D, a plasma biomarker for AD[110]
Fth1	Involved in astrocytes reactivity in AD[111]
Lyz2	Biomarker in AD[112]
Tyrobp	Involved in proinflammatory microglia activation in AD. Associated with synaptic integrity markers [113]
H2-D1	MHC-I component. Increased expression in microglia in AD and aging[114]

AD: Alzheimer's disease; TBI: traumatic brain injury; Beta2-microglobulin: B2M; MHC-I: Major Histocompatibility Complex I; CSF: cerebrospinal fluid