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HAL Authorization

1 **Early memory deficits and extensive brain network disorganization in the *App^{NL-F}/MAPT* double**
2 **knock-in mouse model of familial Alzheimer's disease**

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33 **Abstract:**

34 A critical challenge in current research on AD is to clarify the relationship between early neuropathology
35 and network dysfunction associated to the emergence of subtle memory alterations which announce
36 disease onset. In the present work, the new generation *App^{NL-F}/MAPT* double knock in (dKI) model was
37 used to evaluate early stages of AD. The initial step of tau pathology was restricted to the perirhinal-
38 entorhinal region, sparing the hippocampus. This discrete neuropathological sign was associated with
39 deficits in the object-place associative memory, one of the earliest recognition memories affected in
40 individuals at risk for developing AD. Analyses of task-dependent c-Fos activation was carried out in 22
41 brain regions across the medial prefrontal cortex, claustrum, retrosplenial cortex, and medial temporal
42 lobe. Initial hyperactivity was detected in the entorhinal cortex and the claustrum of dKI mice. The
43 retention phase was associated to reduced network efficiency especially across cingulate cortical regions,
44 which may be caused by a disruption of information flow through the retrosplenial cortex. Moreover, the
45 relationship between network global efficiency and memory performance in the WT could predict
46 memory loss in the dKI, further linking reduced network efficiency to memory dysfunction. Our results
47 suggest that early perirhinal-entorhinal pathology is associated with local hyperactivity which spreads
48 towards connected regions such as the claustrum, the medial prefrontal cortex and ultimately the key
49 retrosplenial hub which is needed to relay information flow from frontal to temporal lobes. The similarity
50 between our findings and those reported in the earliest stages of AD suggests that the *App^{NL-F}/MAPT* dKI
51 model has a high potential for generating key information on the initial stage of the disease.

52 **Abbreviations**

AD – Alzheimer’s disease

CLA – claustrum

DH – dorsal hippocampus

dKI - *App^{NL-F}/MAPT* double knock-in mouse

FC – functional connectivity

mPFC – medial prefrontal cortex

MTC – medial temporal cortex

MTL – medial temporal lobe

OP – object-place association

PS – Pattern Separation

RSC – retrosplenial cortex

53 **Introduction**

54 Current diagnosis and treatment for Alzheimer's disease (AD) occurs too late, when physiological
55 symptoms such as overt neurodegeneration have already reached an irreversible state (Selkoe, 2012).
56 Hallmark pathologies of AD include insoluble amyloid plaques, initially formed in the medial prefrontal
57 and posterior medial cortices indicative of the default mode network (DMN), and neurofibrillary tangles,
58 initially formed in the medial temporal lobe (MTL). Combining functional neuroimaging with amyloid or
59 tau PET reveals that spatial patterns of deposition induce functional network perturbations (Myers et al.,
60 2014; Jones et al., 2017), and accompany cognitive decline (Sepulcre et al., 2017; Pereira et al., 2019).
61 However, soluble amyloid peptide (A β) and hyperphosphorylated tau precursors are generally more toxic
62 and accumulate in the brain decades prior to the onset of typical symptoms (Chen et al., 2017; Hill et al.,
63 2020). This highlights the importance of focusing on the earliest stages to understand the origin of this
64 devastating disease and to develop more proactive detection methods and therapies.

65 Amnesic mild cognitive impairment (aMCI) indicates a higher risk of progressing to AD and has been
66 extensively studied over the last decade as a potential prodromal stage of AD (Petersen et al., 2009).
67 Resting state functional connectivity (FC) has been evaluated in aMCI patients through the temporal
68 correlation of signal fluctuations in fMRI, EEG, or MEG. In order to achieve a more rigorous
69 understanding of FC disruption, graph theory has been used to evaluate the topology of functional
70 networks. Healthy cognition likely requires efficient small world topology, combining local segregation
71 between anatomically and functionally similar regions, and good integration between distant regions with
72 the help of strongly connected hubs (Filippi et al., 2013, Bassett and Bullmore., 2006). In regards to
73 aMCI, many studies have reported functional network perturbations within and between the DMN and the
74 MTL, with reduced network integration along with reduced strength of cortical hubs (Drzezga et al.,
75 2011; Wang et al., 2013a; Bai et al., 2013, Lin et al., 2019). However, these results are not entirely
76 conclusive, as others have reported opposing or null results (Gardini et al., 2015; Grajski and Bressler,
77 2019; Liang et al., 2020, Liu et al., 2012). This inconsistency may be indicative of the heterogeneity of
78 aMCI staging, where the sense of perturbation (hyperconnectivity vs hypoconnectivity) is shown to
79 change depending on the severity of the pathological state (Pusil et al., 2019; Jones et al., 2016). This
80 highlights the importance of identifying earlier biomarkers to more precisely detect preclinical stages of
81 AD, even preceding aMCI. Recent progress has led to the development of sensitive recognition memory
82 paradigms, such as pattern separation and associative memory tasks, which can detect slight cognitive
83 defects in subjective cognitive impairment and cognitively healthy elderly (Naveh-Benjamin, 2000, Sinha
84 et al., 2018; Reagh et al., 2018). These tasks depend on specific interactions across DMN and MTL
85 structures sensitive to early AD (Reagh and Yassa., 2014; Miller et al., 2014; Hales and Brewer, 2011;

86 Caviezel et al., 2020; Ritchey et al., 2020), and have been implemented to detect the earliest
87 “asymptomatic” stages of preclinical AD (Rentz et al., 2011; Maass et al., 2019). By evaluating functional
88 networks directly responsible for the earliest subtle memory deficits, we may achieve a clearer
89 understanding of the neural networks first affected in the initial stages of AD.

90 Initial stages of AD are inevitably linked to discrete physiological and neuroanatomical perturbations,
91 which should be more easily investigated in mouse models of AD (Searce-Levie et al, 2020). Using
92 fMRI on young AD transgenic mice, increased soluble A β (Shah et al, 2016; Shah et al., 2018), and
93 regionally specific increases in neuroinflammation and phospho-tau (Degiorgis et al., 2020) have been
94 linked to disruptions in resting state functional networks. As fMRI/EEG/MEG methods are currently
95 difficult to implement in freely moving mice, ex-vivo imaging based on quantification of activity
96 regulated expression of immediate early genes (IEG; c-fos, Egr1 and arc) may be used to evaluate
97 memory related neural activation (Kinnavane et al, 2015). Using this technique, early-stage
98 pathophysiology has been linked to individual changes in memory driven regional activity (Hamm et al.,
99 2017), but not to functional networks. However, outside of AD, across-subjects correlations of activity
100 regulated IEG levels have been used to assess functional networks directly related to memory (Tanimizu
101 et al., 2017; Wheeler et al., 2013; Vetere et al., 2017). This approach can prove useful for evaluating
102 memory driven FC in mouse models of AD.

103 Recently, knock-in mouse models have been created to express AD-related genes under endogenous
104 mouse promoters (Saito et al., 2014). This removes gene overexpression and resulting potential artificial
105 phenotypes, allowing for a more careful assessment of the initial stages of AD (Sasaguri et al., 2017). In
106 the current study, we detected object-place (OP) associative memory deficits as the earliest sign of
107 cognitive decline in the humanized *App*^{NL-F}/*MAPT* double knock-in (dKI) mouse model. This model
108 specifically expresses all six isoforms of tau and overexpresses A β ₄₂ which leads to marked amyloid
109 deposition and typical pathological hyperphosphorylation of tau at an advanced age of 24 months (Saito
110 et al., 2019). Given our specific interest in the early stages of AD, this model was chosen rather than the
111 *App*^{NL-G-F}/*xMAPT* dKIs because of the slower development of its neuropathology. This option seemed
112 fortunate as the first memory deficits detected in the OP task coincided with the emergence of the earliest
113 stages of abnormal tau phosphorylation within the entorhinal-perirhinal region and not in the
114 hippocampus, indicative of an early pathological state. Task-dependent activity was then measured in
115 regions implicated with this form of associative memory, including primarily the DMN/MTL, and FC
116 was assessed using graph theory techniques. Our results show that initial memory decline in AD could be
117 linked to specific topological changes in memory dependent functional networks.

118 **Results**

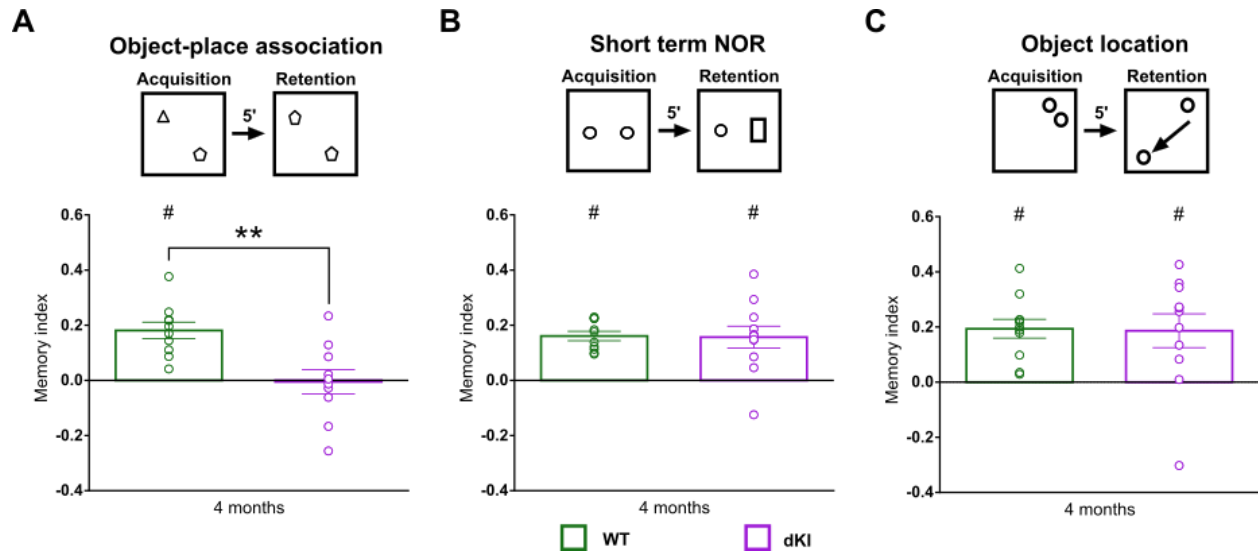
119 **Behavioural phenotyping reveals early alterations in object-place associative memory**

120 Pattern separation (PS) and OP associative memory performances are commonly affected in prodromal
121 stages of AD and cognitively normal elderly (Yeung et al., 2018, Hampstead et al., 2018, Reagh et al.,
122 2018, Reagh and Yassa, 2014), and in mouse models at pre-plaque deposition stages (Zhu et al., 2017;
123 Hamm et al., 2017). In order to detect earliest sensitive recognition memory deficits, a preliminary study
124 was made to evaluate OP and PS performance, as well as long term object recognition performance at 2,
125 4, and 6 months of age (Figure 1—figure supplement 1). For OP and PS tasks, a short inter-trial-interval
126 (ITI) of 5 minutes was chosen to ensure that deficits did not arise from a confounding perturbation of
127 long-term memory consolidation. Given the high number of ages being tested and the difficult nature of
128 the tasks, no results were significant in this preliminary experiment with multiple comparisons
129 corrections. All groups succeeded in the long-term object recognition task and the PS task remained
130 inconclusive because dKI's performances were highly variable (Figure 1—figure supplement 1A and 1B,
131 respectively). On the other hand, a robust potential deficit could be seen at 4 months in the OP task, and
132 persisted at 6 months of age (Figure 1—figure supplement 1C).

133 With a second independent cohort of mice, we confirmed that dKI mice were unable to detect a new OP
134 association at 4 months of age (Figure 1A) (two sample t-test, WT vs. dKI: $t(20) = 2.85$, $P = 0.0098$; one
135 sample t-test, against chance: dKI $t(10)=0.3095$, $p = 0.763$). On the other hand, 4-month-old mice did not
136 show any deficits in short term novel object recognition (Figure 1B) (two sample t-test, WT vs. dKI: $t(19)$
137 $= .0934$, $P = 0.9265$; one sample t-test, against chance: all $ts>3.94$, $ps<0.003$) and object location tasks
138 (Figure 1C) (two sample t-test, WT vs. dKI: $t(20) = 1.052$, $P = 0.9173$; one sample t-test, against chance:
139 all $ts>3.03$, $ps<0.013$). These results confirmed that the OP deficit was due to specific perturbations in
140 associative memory rather than any separate impairments in the object recognition or spatial recognition.

141 We then evaluated early AD pathophysiological changes in 4-month-old mice using western blotting
142 (Figure 1—figure supplement 2A). Antibodies targeting the amyloid precursor protein, APP-cleaved
143 carboxy terminal fragments, phosphorylated tau (Thr181) and total tau proteins were used. Wild-type
144 mice expressed mainly APP α -CTF fragments whereas dKI expressed mainly APP β -CTF fragments.
145 Moreover, an increase in β -CTF/ α -CTF ratio was seen in both the hippocampus (Figure 1—figure
146 supplement 2B) and the medial temporal cortex (MTC; including entorhinal, perirhinal, postrhinal
147 cortices) (Figure 1—figure supplement 2C), indicating upregulated amyloidogenic APP processing. In
148 addition, an increase of the tau phosphorylation degree on Thr181 was only seen in the MTC of dKI mice
149 compared to wild-type mice while both models expressed similar levels of total tau proteins. This is

150 consistent with general consensus of the MTC as the initial area of tau staging (Braak and Braak, 1995).
151 These results highlight that OP deficits occur in conjunction with considerably light pathophysiological
152 changes, akin to an early preclinical AD stage.



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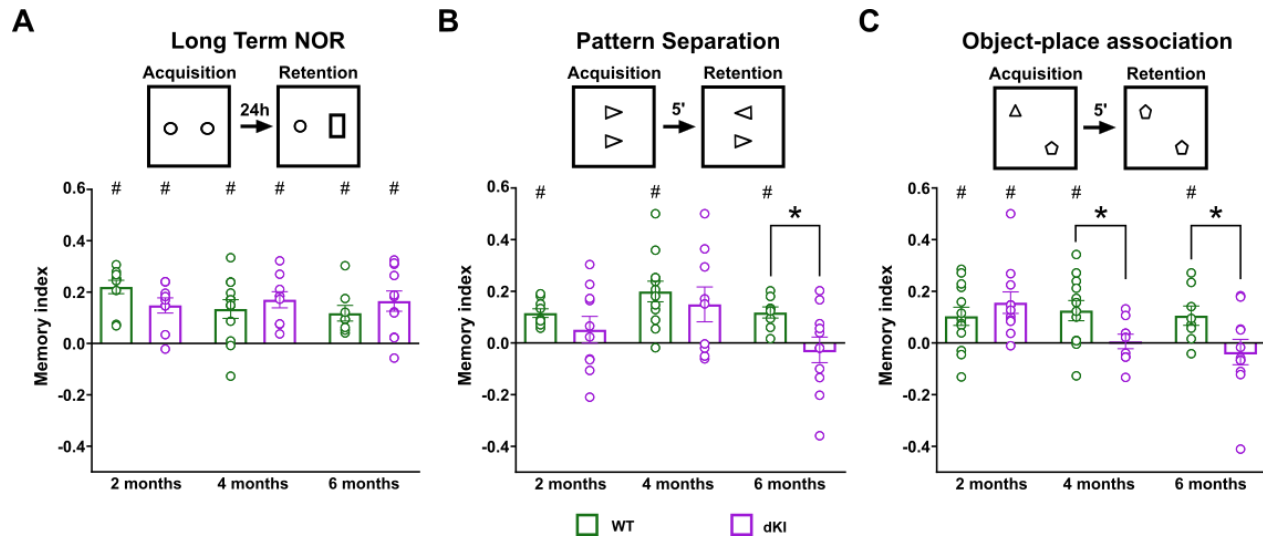
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Figure 1. Behavioral characterization of young dKI mice (WT n = 11, dKI n = 11). (A) At 4 months of age, dKI mice reproduced the potential deficit seen in the object-place association task during preliminary phenotyping (two sample t-test, WT vs. dKI: $t(20) = 2.85$, $P = 0.0098$; one sample t-test, against chance: dKI $t(10) = 0.3095$, $p = 0.763$). At the same age, dKI mice were normal in (B) object recognition (two sample t-test, WT vs. dKI: $t(19) = .0934$, $P = 0.9265$; one sample t-test, against chance: all $t_s > 3.94$, $p_s < 0.003$) (one WT mouse was removed due to being dropped before testing) and (C) spatial recognition (two sample t-test, WT vs. dKI: $t(20) = 1.052$, $P = 0.9173$; one sample t-test, against chance: all $t_s > 3.03$, $p_s < 0.013$), indicating that the loss in associative memory was not due to any singular loss in the short term object recognition or object location memory domains. Bar graphs represent the mean density (\pm SEM) * difference between genotypes ** $p < .01$ (two sample t-test); #, difference from chance, # $p < .05$ (one sample t-test).



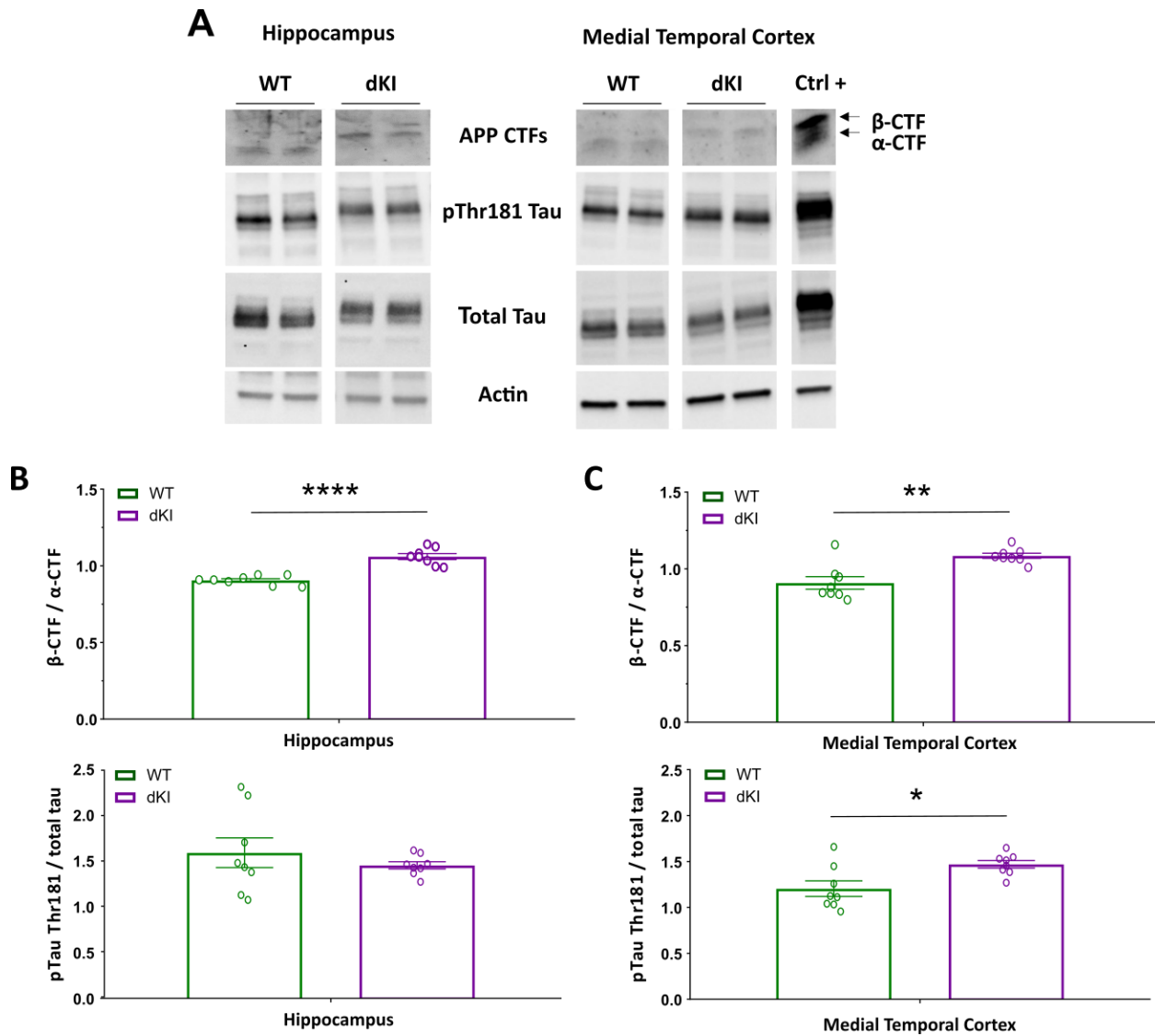
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168 **Figure 1 - figure supplement 1.** Preliminary phenotyping of young dKI mice with sensitive recognition memory paradigms.
169 Mice were tested at 2 months (WT n = 10, dKI n = 10), 4 months (WT n = 12, dKI n = 9) and at 6 months (WT n = 8, dKI n =
170 11). An additional cohort of (WT n = 3, dKI n = 1) was tested at 2 months in object-place association as the initial group did not
171 reach significance above 0. (A) In the long term object recognition task, dKI mice were completely unaffected. (B) In the pattern
172 separation task the dKI mice were highly variable making it so that they were not significant against chance at any age (one
173 sample t-test, against chance: all dKI $t_s < .996$, $p_s > 0.055$). However, between group significance was only obtained at 6 months of
174 age (WT vs. dKI: $t(17) = 2.364$, $P = 0.030$). This variability may represent a variety of factors including perhaps affected DG
175 neurogenesis (Scopa et al., 2020). (C) The earliest significant deficit in dKI mice appeared at 4 months in the object-place
176 association task, a deficit which was conserved at 6 months (two sample t-test, WT_4mo vs. dKI_4mo: $t(20) = 2.325$, $P = 0.0313$;
177 one sample t-test, against chance: dKI $t(8) = 0.2086$, $P = 0.8399$; two sample t-test, WT_6mo vs. dKI_6mo: $t(17) = 2.132$, $P =$
178 0.0479 ; one sample t-test, against chance: dKI $t(10) = 0.7196$, $P = 0.4883$). Although the variability seen during pattern
179 separation may be interesting, we chose to continue our study with the more robust object-place deficit. Bar graphs represent the
180 mean density (\pm SEM). difference between genotypes * $p < .05$ (two sample t-test); difference from chance, # $p < .05$ (one sample t-
181 test). No multiple comparisons corrections were performed for these preliminary comparisons.

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186 **Figure 1 - figure supplement 2.** Early stage of Alzheimer-like disease in 4 month old male dKI mice. (A) Representative blots
187 of APP-cleaved fragments, phosphorylated tau proteins on the threonine 181 site and of total tau proteins in the hippocampus and
188 medial temporal cortex from WT and dKI mice. Analysis of the ratio of the β -CTF on α -CTF fragments and the degree of tau
189 phosphorylation on Thr181 in the (B) hippocampus and in the (C) medial temporal cortex. Results showed that dKI mice
190 expressed mainly the β -CTF whereas the WT mice expressed mainly the α -CTF and the β -CTF/ α -CTF ratio increased in both
191 brain regions. Only the MTC displayed an increase of phosphorylated tau proteins. Bar graphs represent the mean density (\pm
192 SEM). * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ and **** $p < 0.0001$ (two sample t-test).

193

194 **Assessing OP dependent changes in brain activity**

195 While encoding processes are known to be perturbed in AD patients (Granholm and Butters, 1988), recent
196 studies in animal models also find dysfunctional retrieval of engrams as a mechanism for memory loss in
197 early stages of AD-like pathology (Roy et al., 2016; Etter et al., 2019). We thus aimed to evaluate
198 encoding versus recall dependent changes in brain activity in relation to the OP deficit, through
199 independent evaluation of the two test-phases. Thus c-Fos activation was quantified in brains of mice that
200 had underwent either the acquisition phase (*encoding*) (Figure 2A) or the retention phase (*recall*) (Figure
201 2B) of the OP task. In order to account for the c-Fos protein expression curve, the ITI was extended to 3
202 hours, the shortest delay that could permit definite isolation between test-phases. As with a 5-min ITI,
203 dKI mice that underwent the retention phase after a 3-h ITI were likewise unable to detect the novel OP
204 association (two sample t-test, WT vs. dKI: $t(26) = 3.14$, $P = 0.004$; one sample t-test, against chance: WT
205 $t(13) = 3.93$, $P = 0.002$; dKI $t(13) = 0.139$, $P = 0.891$). (Figure 2C). We then chose to evaluate 22 regions
206 of interest (ROIs) encompassing subregions of the medial prefrontal cortex (mPFC), claustrum (CLA),
207 retrosplenial cortex (RSC), dorsal hippocampus (DH) and medial temporal cortex (MTC) (Figure 2D-J).
208 Neuronal hyperactivity is common feature in early AD and in young pre-plaque mice (Zott et al., 2019).
209 We therefore first assessed test-phase and genotype dependent changes in regional activity.

210 **Acquisition induces higher regional activity than retention**

211 All regions except the DH (phase effect: DH $F(1, 192) = 1.85$, $P = 0.175$; subregion x phase effect: $F(3,$
212 $192) = 5.32$, $p = 0.002$; post-hoc Tukey: Acq vs Ret: CA1 $P = .052$, CA3 $P = .383$, DG_ip $P = .597$,
213 DG_sp $P = .513$) showed higher levels of activity in the acquisition phase, although in the LEC this
214 increase in activity was noticeably less drastic (phase effect: LEC $F(1, 96) = 5.22$, $P = 0.025$; all other Fs
215 > 25.6 , $P_s < 0.001$). This can be expected in the initial phase as the mice are subjected to stronger changes
216 with two new objects in previously unoccupied locations in respect to the retention phase, where the only
217 change is a new OP association. During acquisition, the mice are actively encoding several new features,
218 new objects and their unique location in the open-field. Moreover, this helps confirm the absence of
219 residual c-Fos in the retention groups. If residual c-Fos lingered from encoding until perfusion after the
220 retention phase, one would expect similar or increased c-Fos density during the retention phase, not a
221 decrease.

222 **Hyperactivity in the claustrum and entorhinal cortex of dKI mice**

223 The CLA and LEC showed increased activation in dKI mice regardless of the test phase (genotype effect:
224 CLA $F(1, 96) = 8.29$, $P = 0.005$; LEC $F(1, 96) = 10.36$, $P = 0.001$). The MEC revealed increased
225 activation in dKI mice specifically during the acquisition phase (MEC: genotype effect: $F(1, 96) = 25.64$,

226 $P < 0.001$; genotype x phase effect: $F(1, 96) = 7.46$, $P = 0.007$; post-hoc Tukey: WT vs dKI during
 227 acquisition $P < .001$, WT vs dKI during retention $P = 0.252$).

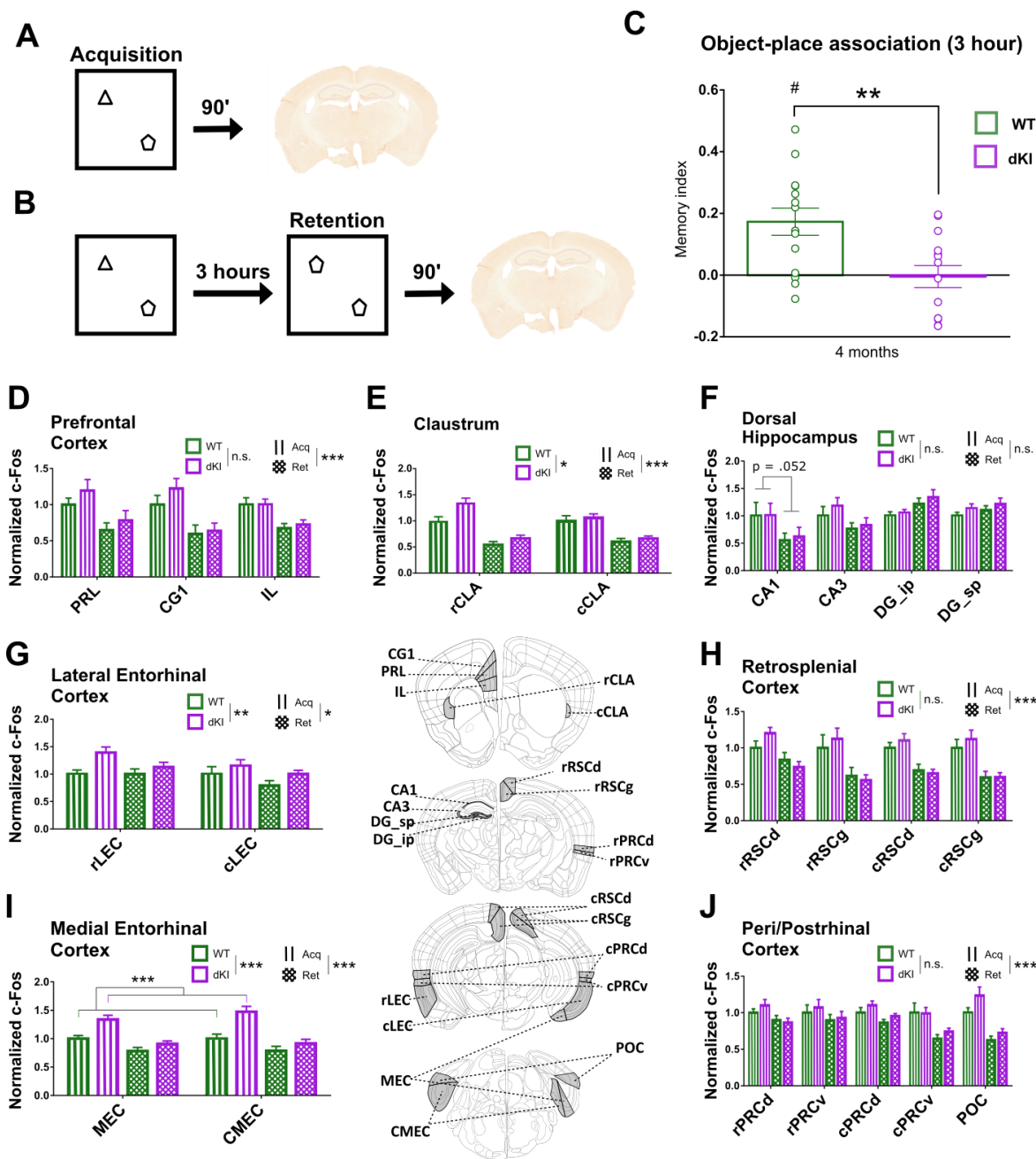
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Region	Acronym
Prelimbic cortex	PRL
Cingulate cortex	CG1
Infralimbic cortex	IL
Rostral claustrum	rCLA
Caudal claustrum	cCLA
Corpus ammoniss 1	CA1
Corpus ammoniss 3	CA3
Dentate gyrus suprapyramidal blade	DG_sp
Dentate gyrus infrapyramidal blade	DG_ip
Rostral retrosplenial cortex dysgranular (RSD)	rRSCd
Rostral retrosplenial cortex granular (RSG)	rRSCg
Caudal retrosplenial cortex dysgranular (RSD)	cRSCd
Caudal retrosplenial cortex granular (RSG)	cRSCg
Rostral perirhinal cortex dorsal (Ect)	rPRCd
Rostral perirhinal cortex ventral (PRh)	rPRCv
Caudal perirhinal cortex dorsal (Ect)	cPRCd
Caudal perirhinal cortex ventral (PRh)	cPRCv
Rostral lateral entorhinal cortex (DLEnt,DIEnt,VIent)	rLEC
Caudal lateral entorhinal cortex (DLEnt,DIEnt,VIent)	cLEC
Medial entorhinal cortex (MEnt)	MEC
Caudal medial entorhinal cortex (CEnt)	CMEC
Postrhinal cortex (Ect,PRh)	POC

229

230 **Table 1. List of evaluated regions.** Regions were chosen a priori based on their relevance to early AD pathology and to
 231 associative memory processing. In parentheses are region identifications according to the third edition Franklin and Paxinos
 232 Mouse Brain Atlas.

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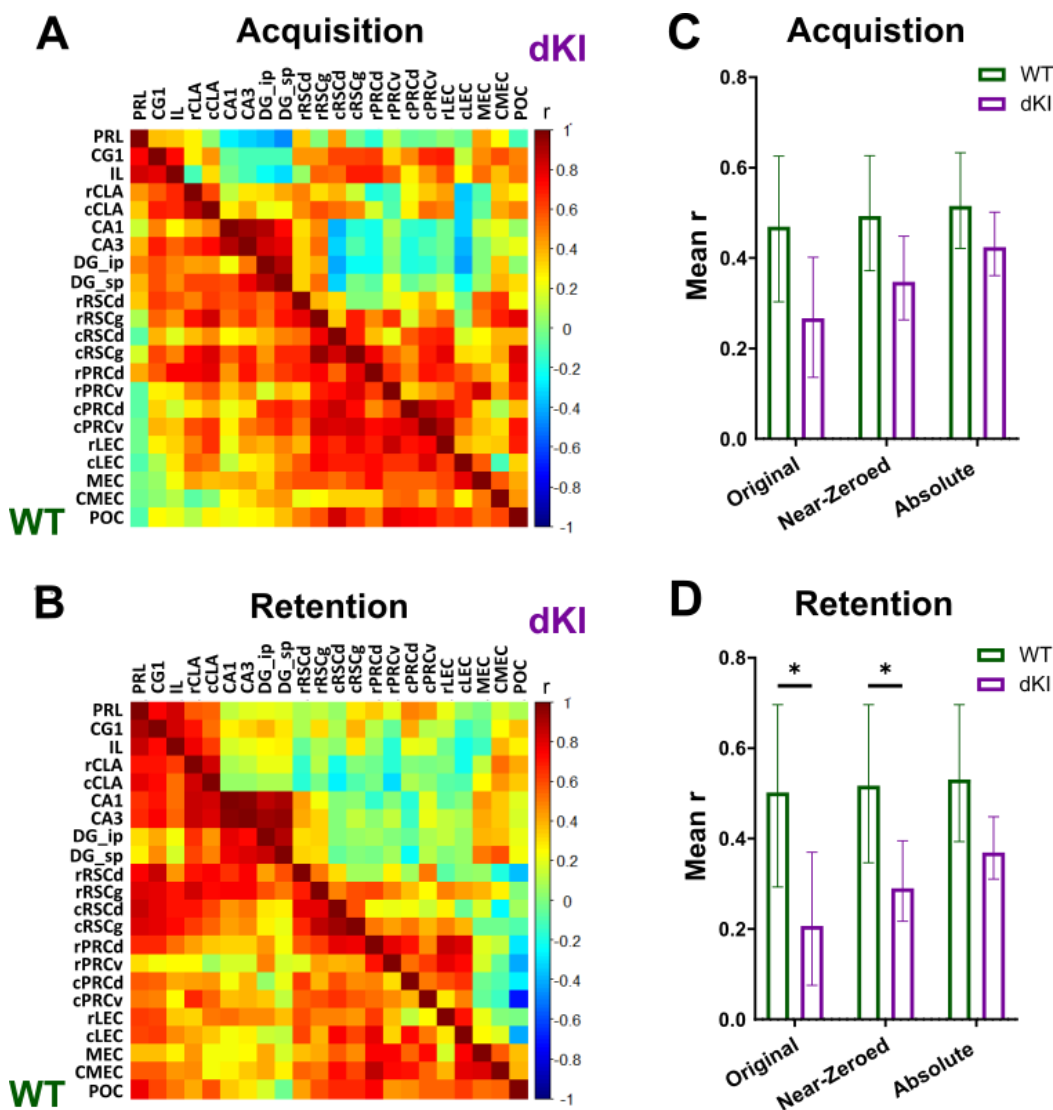
235 **Figure 2.** Regional activity during both phases of OP. Separate cohorts of WT and dKI mice were tested in either the (A)
 236 acquisition phase only or (B) the entire OP3h task, i.e., acquisition and retention phases. Ninety minutes following one test
 237 phase, brains were perfused and expression of the activity-regulated-gene, c-Fos, was evaluated immunohistochemically. (C) The
 238 dKI mice that underwent the whole OP3h task reproduced a deficit in object- place associative memory. difference between
 239 genotypes, $**p < .01$ (two sample t-test); difference from chance, $\#p < .05$ (one sample t-test). (D-J) Graphs that illustrate c-Fos
 240 counts normalized to the WT-Acquisition group. For each region group a 3-factor ANOVA was performed for test-phase
 241 (Acquisition-Lines, Retention-Grid), genotype (WT-green, dKI-purple) and subregion effects. Significance for test-phase
 242 and genotype effects are depicted above each region graph; $*p < .05$, $**p < .01$, $***p < .001$. (D-E,G-J) There was an increase in activity
 243 during the acquisition phase across all regions, except the (F) dorsal hippocampus. The (E) claustrum and (G) lateral entorhinal
 244 cortex presented increases in activity in dKI mice regardless of the test-phase. The (I) medial entorhinal cortex however, was
 245 hyperactive specifically during the acquisition phase $***p < .001$ (Tukey post hoc). Bar graphs represent the mean density (\pm
 246 SEM). Subregion prefixes – r, rostral; c, caudal; suffixes – RSCd, dysgranular; RSCg, granular; PRCd, dorsal; PRCv, ventral; ip,
 247 infrapyramidal; sp, suprapyramidal. Error bars indicate the SEM. Schematics indicating the general locations of brain regions
 248 were adapted from Allen Mouse Brain Atlas derived vector images (Lein et al., 2007).

249 **Computing functional couplings**

250 Brain regions with correlated activity can be said to exhibit functional connectivity (FC), as co-
251 modulations of activity can be a marker of inter-regional information sharing. In humans or head-
252 restrained/anesthetized rodents recorded with EEG, MEG (for electrical signals) or fMRI (for metabolic
253 rate), FC is computed from the covariance across time. In methods using regional expression of c-Fos, FC
254 can be modeled by the covariance of regional activity across subjects to study memory driven networks in
255 mice (Wheeler et al., 2013; Tanimizu et al., 2017). The validity of this approach has been confirmed using
256 chemo-genetic techniques (Vetere et al., 2017).

257 From the c-Fos signal, we assessed FC by computing the inter-regional Spearman correlation coefficients
258 (r) for each Genotype-Phase group. Correlation matrices were used to visualize all possible correlations
259 within each group (Figure 3A-B). We first assessed global FC strength by taking the mean r value of each
260 matrix. Most FC couplings were positive, however we found a few weakly anti-correlated pairs of
261 regions. In evaluating FC, there were 3 ways to consider these negative correlations; as disruptive, as
262 very-weak or as contributing equally to positive correlations. All three cases were evaluated by taking the
263 mean r with retained, near-zeroed, and absolute valued negative correlations respectively. In the near-
264 zeroed case, negative correlations were reduced to a value of .006, the smallest positive correlation
265 observed.

266 During acquisition, we found no significant change in global FC strength between WT and dKI groups,
267 for any of the three ways to treat negative correlations in FC (Figure 3C). However, during retention,
268 there was a decrease in global FC strength in dKI mice with respect to WT. This decrease could be proved
269 significant when considering negative correlations as disruptive or as very weak, but not when their
270 absolute value was taken (Figure 3D). This indicates that two phenomena coexist: first, a reduction of
271 positive inter-regional correlations, corresponding to decreased “cooperation” between some regions;
272 second, an increase in absolute strength of inter-regional negative correlations, corresponding to increased
273 “conflict” between some regions.



274

275 **Figure 3.** Functional connectivity is depicted with matrices showing across subjects inter-regional spearman correlations for c-
 276 Fos expression during (A) retention and (B) acquisition. Colors reflect correlation strength (scale, right). We assessed global FC
 277 strength by taking the mean r value of each matrix with retained, near-zeroed, and absolute valued negative correlations. In the
 278 near-zeroed case, negative correlations were reduced to a value of .006, the smallest positive correlation observed. (C) We found
 279 no change in global FC strength for any of the three cases during acquisition. (D) During retention however, there was a
 280 significant decrease in global FC strength with the original and near-zeroed correlations, but not when they are absolute valued.
 281 Bar graphs represent the mean bootstrap value, and the error bars represent the bootstrapped 95% confidence interval. * -the 95%
 282 CI for the difference ≥ 0

283

284 **Generating functional networks**

285 Each functional connectivity matrix can be considered as the adjacency matrix of a weighted undirected
286 network (Rubinov and Sporns, 2010), and, as such, its organization can be assessed through the analysis
287 of functional networks, using techniques arising from graph theory. From each matrix a functional
288 network was generated as a fully connected weighted graph. In these graphs, the regions are represented
289 as nodes and the functional connections between regions are represented by edges. The strength of a
290 connection between two regions, as represented by the edge weight, is determined by their inter-regional
291 correlation strength. As negative correlations are weak and pose difficulties of interpretation when dealing
292 with many graph theory techniques, we decided for all following analyses to replace them with near-
293 zeroed values when constructing our functional networks, in line with other studies of c-Fos derived
294 functional networks in-vivo (Vetere et al., 2017). It has been shown that community organization of
295 structural networks can change between healthy individuals and different stages of MCI (Pereira et al.,
296 2016). We decided to evaluate community organization of each functional network to see if any
297 consistent or phase specific changes could be detected between genotypes. This could also aid us in
298 obtaining a qualitative understanding of organization of information flow in each network. Network
299 communities were detected through a modularity maximization procedure (see Materials and Methods).
300 This unsupervised algorithm separates the nodes into distinct groups (the communities), such that nodes
301 within a community are more strongly interacting between them than with nodes in other communities.
302 The separation in communities will partly stochastically vary for different runs of the algorithm and
303 different bootstrap instances of the FC matrix. To extract a robust consensus set of communities we then
304 computed allegiance matrices (Bassett et al., 2015), which depict the percentage of times that any given
305 pair of regions is attributed to the same community among bootstraps (Figure 4).

306 **WT networks differentially engage the DH and mPFC**

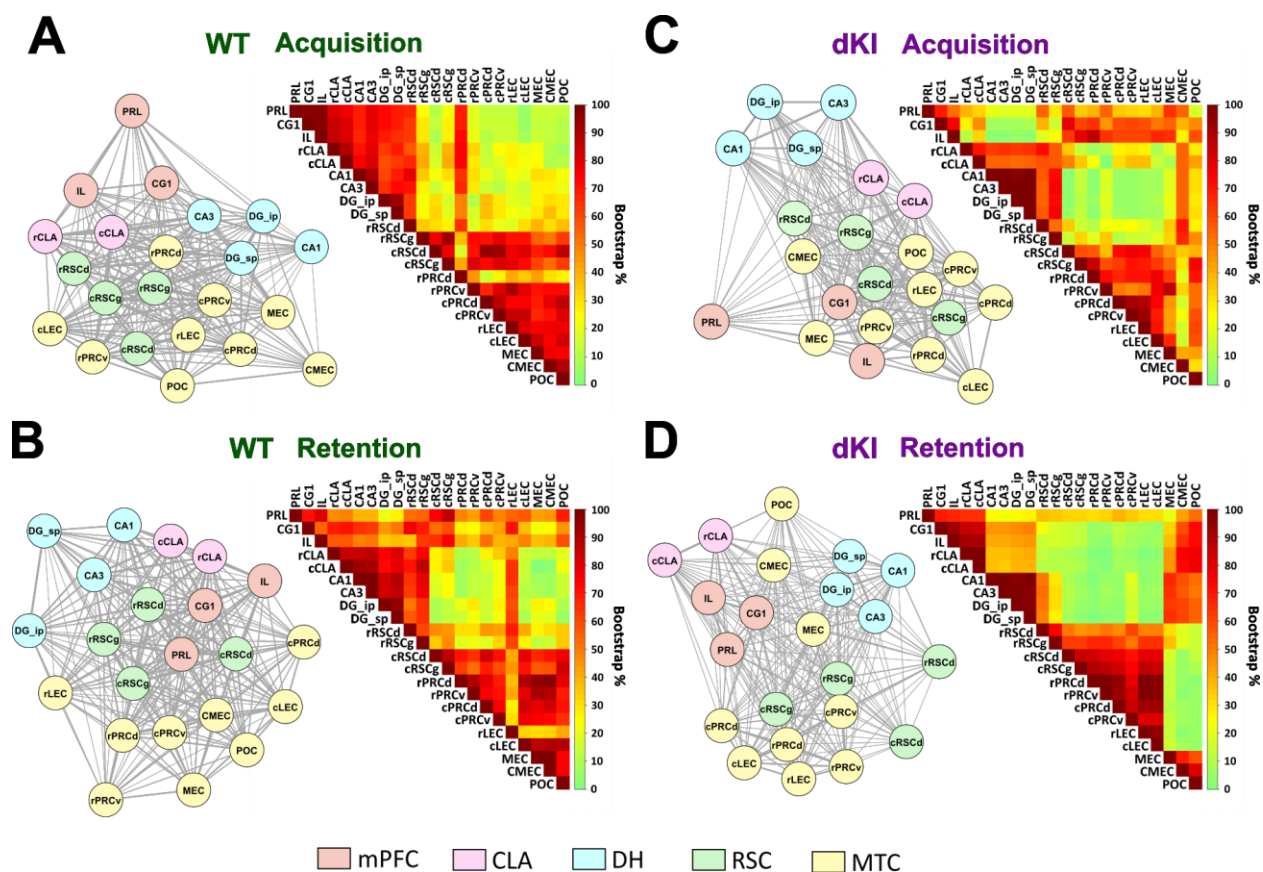
307 A stable community is found in both task phases encompassing most of the MTC and the cRSC (Figure
308 4A,B; red area at the bottom right of each WT allegiance matrix). The main difference between the WT-
309 Acquisition and WT-Retention groups was the differential involvement of the DH or mPFC with this
310 MTC community. During acquisition, the DH shared a community primarily with the mPFC (Figure 4A;
311 red area at top left the allegiance matrix). During retention, the mPFC shared allegiance across the
312 network (Figure 4B; red area along the mPFC rows across the top of the allegiance matrix). This more
313 global incorporation of the mPFC appeared to be driven largely through the RSC (Figure 4B; darker red
314 square area located at the middle of the top three mPFC rows).

315 **dKI networks reveal consistent departures from WT community structure**

316 During acquisition, the DH community largely disengaged with the mPFC as compared to the WT-
317 acquisition group (Figure 4C; green square at the top left of the allegiance matrix). Moreover, the DH
318 appeared to be more strongly aligned to the CMEC, MEC, and POC as compared to the rest of the MTC
319 (Figure 4C; the more red/orange area at the right of the DH rows).

320 During retention, both the mPFC and the DH disengaged with the MTC. This led to distinct communities
321 containing mPFC/CLA, DH and the MTC (Figure 4D; three separate red squares of the allegiance
322 matrix). Similarly to the acquisition phase, the MEC/CMEC/POC regions were heavily recruited by the
323 mPFC and DH communities (Figure 4D; red area at top right), at the cost of disengagement with the MTC
324 community (Figure 4D; green area at bottom right of the allegiance matrix). The consistent recruitment of
325 the MEC/CMEC/POC in both dKI groups elucidates a possible role for these regions as compensatory
326 hubs in this mouse model.

327



328

329 **Figure 4.** Functional network structure and community organization. Functional networks were computed as fully connected
 330 weighted graphs, with regions as nodes (circles) and inter-regional correlation strengths as edge weights (line thickness). For
 331 visual examination of networks, nodes were placed so that they lie closer to nodes with which they are strongly connected, and
 332 were color coded according to a priori region subfields. Communities were detected through modularity maximization, which
 333 finds communities of regions that have stronger connections with each other relative to the rest of the network. Communities
 334 were detected across bootstrapped networks, and allegiance matrices were utilized to depict community stability as the
 335 percentage of bootstraps that contain any given pair of regions in the same community. (A,B) In both WT groups there is a stable
 336 MTC/cRSC community (red area at the bottom right of each WT allegiance matrix). (A) In the WT-Acq there is a stable
 337 DH/mPFC community (red area at the top left of the allegiance matrix). (B) In the WT-Ret the mPFC shares community
 338 allegiance with all of the network (red area along the mPFC rows across the top of the allegiance matrix) as mediated through the
 339 RSC (darker red square in the middle of the top three mPFC rows). (C,D) Across both dKI groups, the MEC/CMEC/POC regions
 340 appear to display a consistently modified community allegiance in respect to the WT, associating more with the DH/mPFC and
 341 sometimes less with the rest of the MTC (red area at top right, some green areas at bottom right of each dKI allegiance matrix).

342

343 **Information flow in functional networks**

344 Efficient integration of information flow across functional networks is shown to aid in cognitive function
345 (Wang et al., 2013a; Martinez et al., 2018). Interpreting network links as “pipes”, how well can a network
346 allow information to flow will depend not only on how wide individual pipes are, but also on how pipes
347 are disposed and aligned to form pipelines between the nodes that must communicate, without too many
348 steps and bottlenecks. The general capacity for a network to sustain efficient flows is quantified in graph
349 theory by metrics such as *global efficiency* (see Materials and Methods; Latora and Marchiori, 2001). It is
350 important to note that even if a network has reduced global FC strength, its global efficiency may rest un-
351 affected if there are well placed “hub” regions to facilitate indirect communication. To evaluate
352 information flow at the regional level, *nodal strength* and *nodal efficiency* metrics are used. Nodal
353 strength measures the degree to which each specific region can exchange information directly with all
354 other regions of the network. A region with low strength may still be able to communicate with its
355 network indirectly, again likely through hub regions. This indirect communication can be measured using
356 nodal efficiency. Organization of information flow in a network can be examined through strength and
357 efficiency distributions. Regions with higher strength and efficiency can be considered to contribute more
358 to their network. High direct connectivity of a region, such as high strength, is also shown to describe
359 potential hub regions (Vetere et al., 2017) which, as we have described, are needed to facilitate indirect
360 information flow and maintain global efficiency. We compared information flow between WT and dKI
361 networks within each test-phase. Organization of information flow was first evaluated through a
362 qualitative examination of node strength and nodal efficiency distributions. To best visualize
363 distributions, the strength/efficiency values were squared and normalized to the highest value within each
364 network, and were displayed as necklace diagrams (acquisition - Figure 5A,B) (retention – Figure 5F,G).
365 We then directly compared global efficiency and nodal strength/efficiency between genotypes in
366 subsequent analyses (acquisition - Figure 5C,D,E) (retention - Figure 5H,I,J).

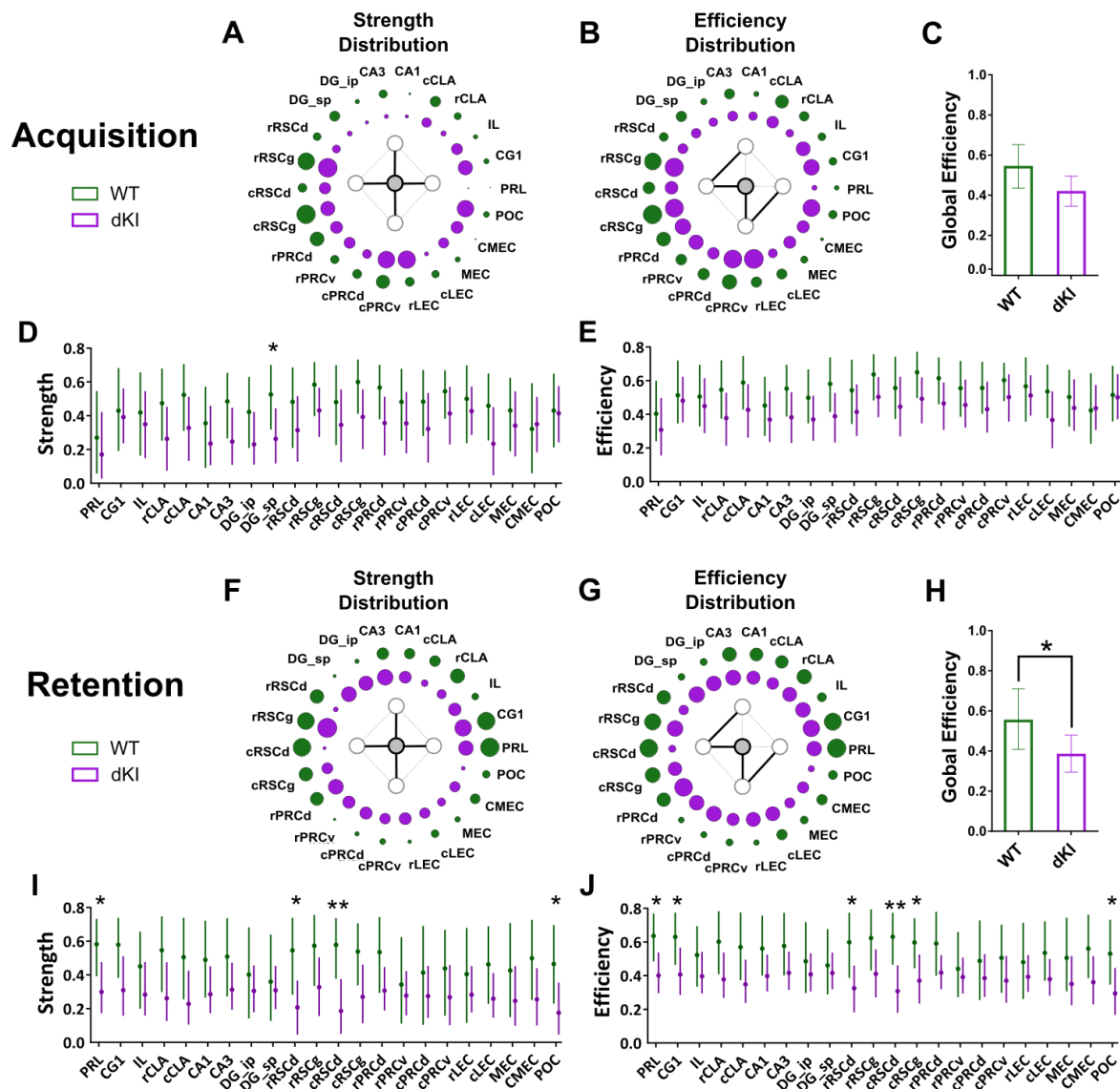
367 **Acquisition– the DG_sp maintains efficiency despite a drop in strength**

368 During acquisition, strength and efficiency distributions were largely conserved between genotypes with
369 strong emphasis on RSC and MTC regions. Although, in the dKI there appeared to be additional
370 involvement of mPFC, MEC and POC (Figure 5A,B). Nevertheless, global efficiency was not severely
371 affected in the dKI network (Figure 5C). The unique significant drop in nodal strength was seen in the
372 DG_sp (Figure 5D), but as there was no drop in nodal efficiency (Figure 5E), it could still effectively
373 communicate with the rest of the network indirectly. This maintenance of indirect communication, despite
374 drops in direct communication, reflects the potential importance of the rRSC or MEC/CMEC/POC
375 compensatory hubs.

376 **Retention – severe losses in network efficiency, especially across the cingulate cortex**

377 During retention, heavy emphasis was placed on mPFC and RSC regions in the WT network. In the dKI
378 network this emphasis was largely lost and the distributions take on a more homogenous structure, though
379 the rRSCg and mPFC subregions still appeared to be among the most involved (Figure 5F,G). Global
380 efficiency was significantly reduced in the dKI network (Figure 5H), with sharp drops in strength seen in
381 the PRL and the POC, but most severely in both subregions of the RSCd (Figure 5I). Loss in nodal
382 efficiency was even more prevalent, with additional reductions in the CG1 and the cRSCg (Figure 5J).
383 These results suggest that retention dependent functional integration across the cingulate cortex is
384 severely disrupted in the dKI, and may be linked to reduced hub strength of the RSCd.

385



386

387 **Figure 5.** Information flow in functional networks. Network organization of information flow was assessed through examination
 388 of node strength and nodal efficiency distributions in both test-phases (acquisition, retention). These distributions were visualized
 389 using necklace diagrams, where circle size reflects the within network normalized (A,G) strength² and (B,H) efficiency². Global
 390 efficiency was then compared directly between genotypes to assess (C,F) network integration. (D,I) Nodal strength and (E,J)
 391 nodal efficiency were compared between genotypes to assess region dependent changes in direct and indirect information flow,
 392 respectively. (A,B) During acquisition, strength and efficiency distributions were roughly conserved between genotypes, with
 393 strong emphasis on the RSC and MTC regions, although in the dKI, mPFC, MEC and POC subregions were much more
 394 involved. (C) Global efficiency was not severely affected in dKI mice. (D) A drop in region strength was seen in the DGsp, (E)
 395 but its efficiency was unaffected. (G,H) During retention, heavy emphasis was placed on mPFC and RSC regions in the WT
 396 network. In the dKI, this emphasis is largely lost. (F) Global efficiency of the dKI network was significantly reduced as
 397 compared to the WT network. (I) Reductions in region strength were seen in the POC, PRL, and most severely in both subregions
 398 of the RSCd. (J) Reductions in efficiency was retained among these regions, with additional losses in the CG1 and the cRSCg.
 399 Bar graphs and dot graphs represent the mean bootstrap value, and the error bars represent the bootstrapped 95% confidence
 400 interval. * -the 95% CI for the difference ≥ 0 ; ** -the 99% CI for the difference ≥ 0

401

402 **Relating network efficiency and memory**

403 Finally, we addressed the question of whether the reduction of OP memory performance in dKI mice
404 could be accounted for by the reduction in network efficiency observed during retention. Our aim was
405 thus to determine if a relationship could be found between global efficiency and memory index in the WT
406 and be used to predict dKI memory loss. This was assessed through correlation testing across sub-
407 sampled WT populations, where each sub-sample corresponded to the removal of one mouse. From each
408 sub-sample the average memory index and resulting global efficiency were calculated, and correlation
409 significance was tested with Pearson and Spearman correlation coefficients.

410 **Global efficiency positively correlates with an exploration adjusted memory index, and predicts** 411 **memory deficiency in the dKI**

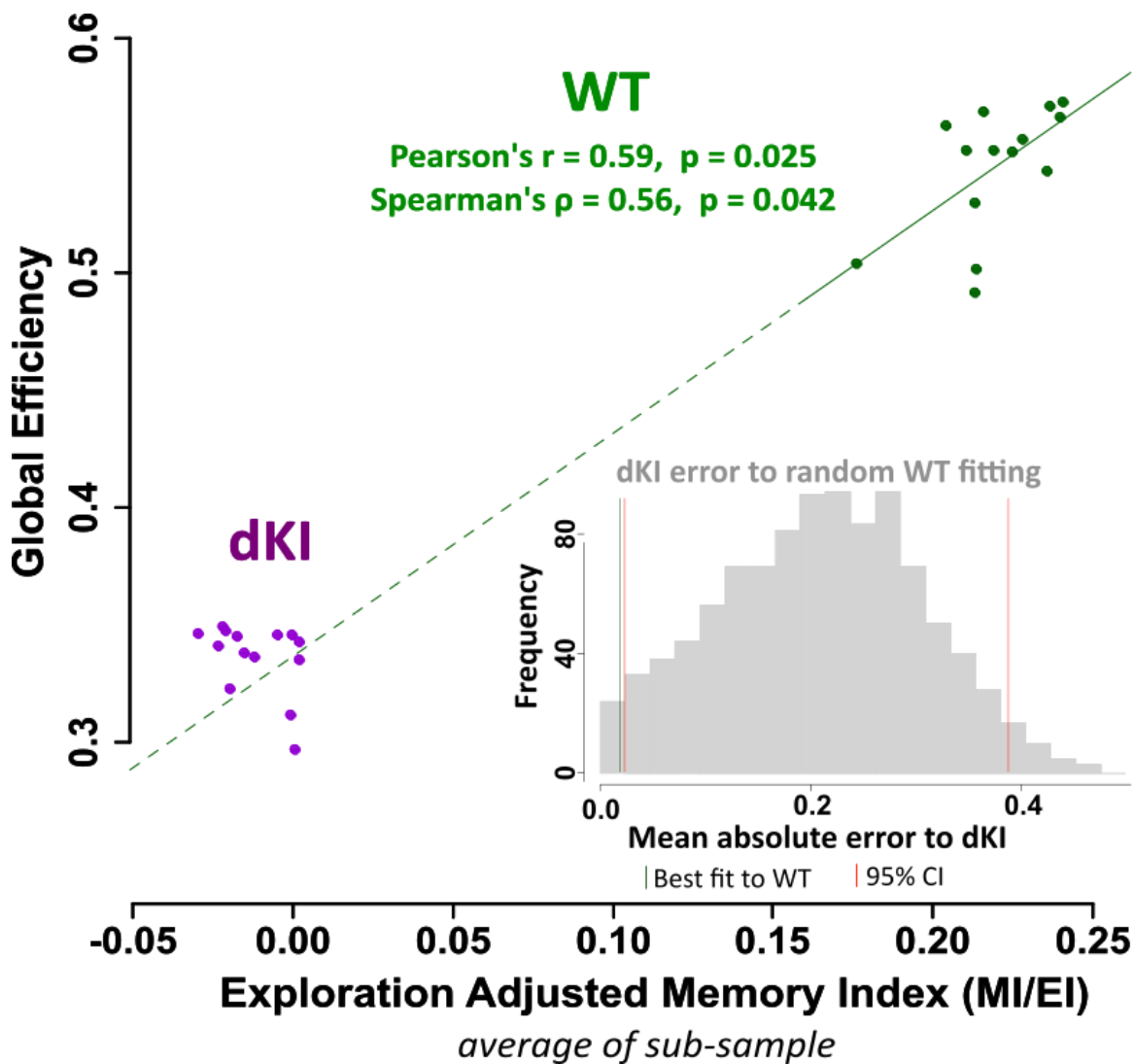
412 No significant correlation was found between global efficiency and memory index across WT sub-
413 samples (Figure 6 - figure supplement 1A). However, we noticed that of the mice with high memory
414 index, those with greater exploration times (total time exploring both objects) appeared to have a weaker
415 contribution to global efficiency. This hinted a potential dampening effect of general object exploration
416 on memory driven FC. Note that increases in exploration time themselves may be interpreted as a further
417 sign of memory function alteration, beyond reductions in the previously defined memory index MI. We
418 thus defined an exploration index EI, quantifying the exploration time of the mouse normalized to the
419 group and used it to compute as well an exploration adjusted memory index as the memory index divided
420 by the exploration index (MI/EI). Decrease of such an exploration adjusted memory index may reflect
421 both MI decrease and EI increase, thus summarizing both probed facets of memory-related behavior
422 alteration. As in the case of MI alone, we did not find a significant correlation between global efficiency
423 and EI index (Figure 6 - figure supplement 1B). A significant positive correlation was found however
424 between global efficiency and the adjusted memory index MI/EI across the WT sub-sampled networks
425 (Figure 6).

426 Moreover, we could show that extrapolating the best fit line for the WT ensemble to drops in global
427 efficiency as large as the ones observed in the dKI ensemble also predicted the observed dKI decrease of
428 the MI/EI behavioral performance index. This is illustrated in Figure 6 where the dashed continuation of
429 the green line (the extrapolation of the best fit line to the WT ensemble) intersects the cloud of purple
430 points (the ensemble of dKI sub-samples). The prediction of dKI sub-samples MI/EI index based on
431 extrapolating the law fitted on WT sub-samples yielded errors way smaller than what expected at chance
432 level, as we could verify via comparison with a null distribution of prediction errors constructed from
433 1000 randomly fit models to the WT ensemble. This is shown in the inset histogram of Figure 6, where

434 the green vertical line (prediction error of best fit to WT) lies to the left of the leftmost red vertical line
435 (lower end of the null distribution 95% CI).

436 This suggests that subjects with a high memory index but low total exploration time are thus more likely
437 to have higher global efficiency. Contrary to memory index, there is no detectable significant difference
438 in total object exploration time between the WT and dKI groups (Figure 6 - figure supplement 1C).
439 Memory index is thus a more sensitive metric of subtle and complex behavioral changes, which are
440 tracked by a decrease of global efficiency from WT to dKI. These results support reduced network
441 integration as a potential factor for impairment of OP memory recall in the dKI.

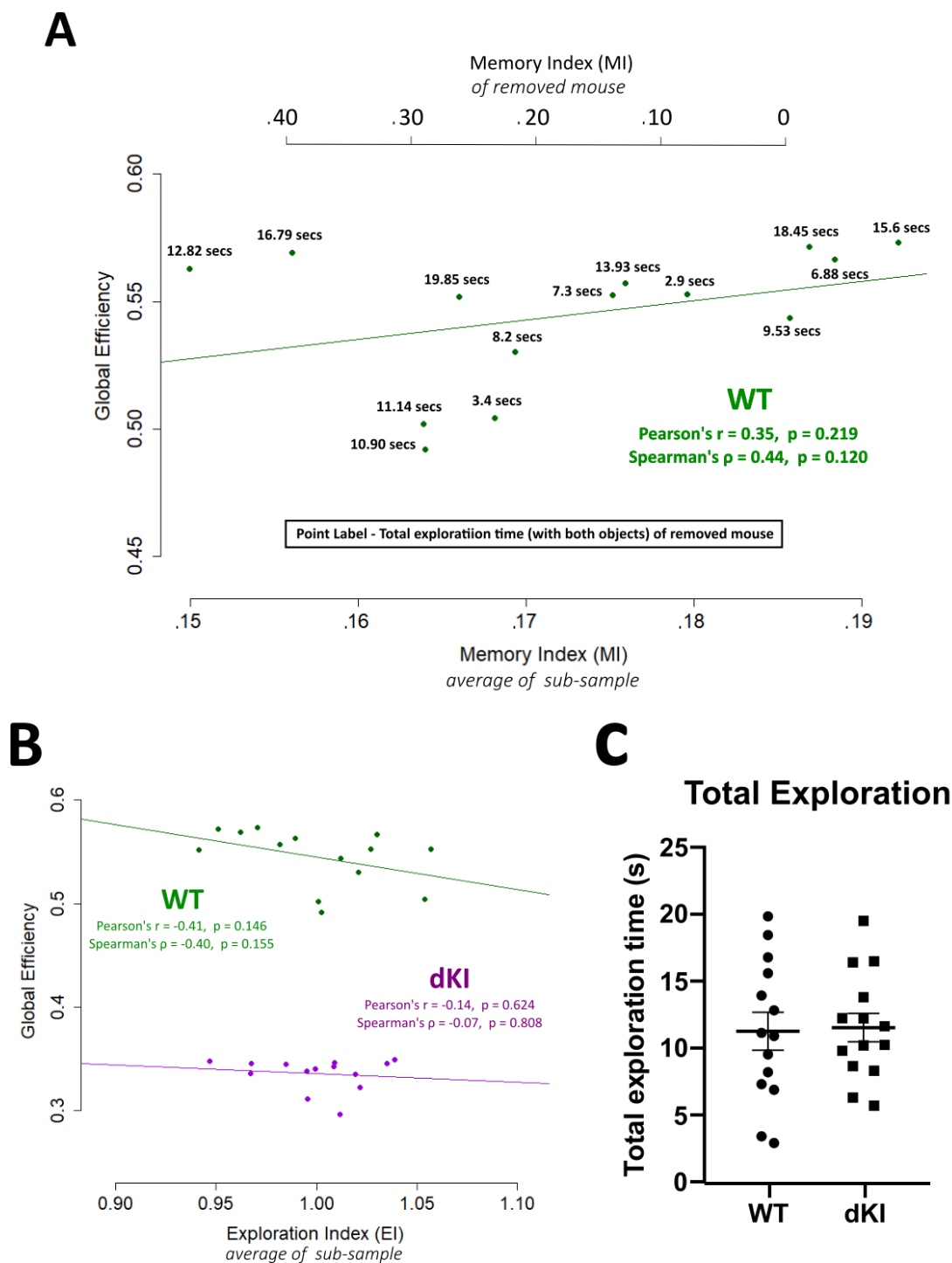
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443

444 **Figure 6.** Association between network efficiency and exploration adjusted memory index "MI/EI". Sub-samples were
445 generated from each retention group by resampling $n-1$ mice without replacement. From each sub-sample, the average MI/EI and
446 the network global efficiency were computed, where each sub-sample is depicted as point on the plot. The relationship between
447 global efficiency and MI/EI was evaluated across only the WT sub-samples (solid green line), and a significant positive linear
448 relationship was found. Interestingly, when this linear law across WT sub-samples was extrapolated down (dashed green line) to
449 the reduced global efficiency of the dKI sub-samples, it could predict their reduction in MI/EI (the intersection of the dashed
450 green line to the cloud of purple points). The prediction of dKI sub-samples MI/EI index based on extrapolating the law fitted on
451 WT sub-samples yielded errors way smaller than what expected at chance level, as we could verify via comparison with a null
452 distribution of prediction errors constructed from 1000 randomly fit models. See inset histogram: the green vertical line (dKI
453 error to best fit of WT) lies to the left of the leftmost red vertical line (lower end of the null distribution 95% CI).

454



455

456 **Figure 6-figure supplement 1** Associations of memory index “MI” and exploration index “EI” to network efficiency. (A) There
 457 was no relationship found between global efficiency and memory index. However, we noticed that of the mice with high memory
 458 index, those with greater exploration times (total time exploring both objects) appeared to have a weaker contribution to global
 459 efficiency. To take into account an exploration disruption effect on memory related FC, an exploration adjusted memory index
 460 was computed as the memory index divided by the exploration index (MI/EI), where the exploration index (EI) is the exploration
 461 time of the mouse normalized to the group. (B) We verified that there was no direct relationship found between global efficiency
 462 and EI. (C) There was also no difference in total exploration between WT and dKI mice.

463 **Discussion**

464 The current necessity to learn more about initial steps of AD pathology prompted us to investigate brain
465 network alterations associated to the encoding and retrieval phases of the first recognition memory
466 paradigm affected in the late-onset *App^{NL F}/MAPT* dKI mouse model of the disease (Saito et al, 2019).
467 Here we show that the neuropathology expressed by 4-month-old dKI mice at the time of their earliest
468 recognition deficit is reminiscent of an early preclinical stage. Quantification of c-Fos activation
469 highlighted an abnormally increased activity in the entorhinal cortex and the claustrum, two entities
470 known to be vulnerable to AD pathology. Finally, extensive analyses of network connectivity point to a
471 disorganization of internal communication between and within the MTL and interconnected regions
472 where the RSC seems to play a pivotal “hub” role which could be disrupted in dKI mice.

473 **Object-place recognition deficit as an early marker of emerging AD neuropathology**

474 In the battery of five recognition memory tasks, the object-place associative memory task was the first to
475 detect a deficit in dKI mice at 4 months of age. This recognition deficit occurred 2 months before spatial
476 pattern separation deficits and with intact long term object recognition memory up to 6 months of age.
477 The object-place task seems to be highly sensitive to early amyloid pathology because it was also
478 impaired at pre-plaque stages in more aggressive models of AD (Bonardi et al, 2016; Hamm et al., 2017).
479 In TgCRND8 mice it was specifically associated with increased levels of β -CTF in the hippocampus
480 (Bonardi et al, 2016; Hamm et al., 2017). $A\beta$ levels of *App^{NL F}/MAPT* dKI were not evaluated in this
481 study, but the seminal work characterizing the parental single KI *App^{NL F}* mice already showed increased
482 cortical levels of the neurotoxic species $A\beta_{42}$ at the age of 2 months (Saito et al, 2014). This early
483 accumulation of $A\beta$ in 2-month *App^{NL F}* mice was associated with significant cell loss and hyperexcitation
484 in the LEC, whereas the same alterations appeared several months later in the CA1 (Petrache et al., 2019).
485 Here, we show for the first time evidence for early onset of AD pathology in 4-month-old dKI mice with
486 specific increases in levels of β -CTF and phosphorylated tau labeled with AT270 antibodies in the MTC,
487 but not in the dorsal hippocampus. These results suggest that early abnormal levels of tau phosphorylation
488 are still restricted to the MTC at the age of 4 months. This is of particular importance because the PRC-
489 EC pathway has long been seen as a primary site of neurodegenerative event and dysfunction that
490 characterizes the earliest preclinical Braak stage I of AD tau deposition (Kahn et al, 2013; Braak and
491 Braak, 1995; Hyman et al, 1984). Interestingly, significant worsening of neuropsychological markers of
492 PRC and EC functionality were detected more than 10 years before AD diagnosis (Hirni et al, 2016).
493 More recently, Yeung et al., 2018, showed that object-place recognition performance was predicted by
494 anterolateral EC volume in the elderly. This result is in agreement with studies attributing a key role of

495 the equivalent rodent LEC in the network supporting association of an object with a place or a scene
496 (Deshmukh et al, 2012; Wilson et al, 2013a,b; Chao et al 2016). It must be noted that deficits in the
497 pattern separation task in 6-month-old dKIs suggest subsequent development of functional alterations
498 within the DG-CA3 region which is in agreement with studies in aMCI patients and transgenic mouse
499 models of AD (Yassa et al, 2010; Zhu et al, 2017). All these findings strongly suggest that 4-month-old
500 dKI mice recapitulate early impairment in recognition memory and specific vulnerability of the PRH-EC
501 region to both amyloid and tau pathologies as found in preclinical stages of AD (Kahn et al, 2014).

502 **Distributed increase in c-Fos activation possibly reflecting spreading of pathological hyperactivity**

503 We detected high level of c-Fos expression in the CLA and the LEC of dKI mice regardless of the test-
504 phase suggesting that both structures would be hyperactivated during memory encoding and recall. On the
505 other hand, the hyperactivation of the MEC restricted to the acquisition phase could be have been
506 triggered by active encoding of the new environment. Well documented in mouse models of AD, early
507 hyperactivity within the LEC appears as a consequence of local increase in amyloid pathology and it is
508 considered as a major factor driving propagation of both A β and tau pathology to its main outputs
509 especially the hippocampus (Xu et al., 2015; Nuriel et al., 2017; Rodriquez et al., 2020). The CLA has
510 been less extensively studied, but a few studies show evidence of A β accumulation and
511 neurodegeneration in this region (Qin et al, 2013; Ogomori et al., 1989; Gustafson et al., 1998).
512 Interestingly, an early study in Alzheimer's patients found neuronal loss only within a CLA subregion
513 strongly connected with the EC (Morys et al., 1996). Thus, EC may also propagate hyperactivity,
514 exporting amyloid and tau pathology to the CLA as well as other limbic cortices in preclinical stages of
515 AD (Bonthius et al., 2005). Note that an alternative hypothesis has been proposed with a central role of
516 the CLA in the spreading of AD pathology in the brain (Avila and Perry, 2021). Being largely
517 interconnected with most cortical areas (Wang et al, 2017), hyperactivity of CLA may initiate or
518 aggravate spontaneous the cortical network hypersynchrony and task-induced hyperactivation found in
519 elderly at risk and in early MCI (Mueller and Weiner, 2017; Corriveau-Lecavalier et al., 2020).

520 Early-stage hyperactivity has also been proposed to reflect compensatory mechanisms which would first
521 help maintaining and then worsen cognitive function through the spreading of neuropathological
522 processes and a deleterious impact on cognitive-related network organization and functioning (Corriveau-
523 Lecavalier et al., 2020). There is some indication for a reorganization of network information flow in the
524 dKI mice. We saw through the community organization of dKI functional networks during both phases,
525 that the MEC and POC are more integrated with mPFC and DH communities (Figure 4C,D). This change
526 in community allegiance may be a consequence of an initial hyperactive state within the EC, much like

527 artificial stimulation of brain structures can enforce specific functional networks (Warren et al., 2019). In
528 certain cases, this shift towards stronger outbound allegiance may help facilitate alternative
529 communication between the MTC and interconnected regions, as seen with the maintenance of indirect
530 information flow of the DG_sp during acquisition (Figure 5D,E). The apparition of compensatory hubs
531 may be one way through which hyperactivity could initially help memory processing during the
532 preclinical AD.

533 **mPFC and RSC are disrupted during object-place associative memory retention**

534 The mPFC and RSC were the most heavily utilized regions in WT mice during retention phase, and direct
535 and indirect information flow through these regions was significantly reduced in dKI mice (Figure 5F).
536 The specificity of this dysfunction to the retention phase is not surprising as the mPFC and RSC are
537 shown to be more involved in the retrieval and/or editing of memory traces rather than encoding *per se*
538 (Mitchell et al., 2018). Lesioning the RSC disrupts object-place associative memory in rodents (Parron
539 and Save., 2004), and communication between the mPFC-MTC is also shown to be essential (Chao et al.,
540 2016; Hernandez et al., 2017). The RSC is proposed to play a pivotal “hub” role in facilitating this
541 communication as it exhibits strong structural connections with both the mPFC and the MTC (Sugar et
542 al., 2011, Vann et al., 2009). In humans, the posterior cingulate cortex (PCC) and RSC, close equivalents
543 to the rodent RSC (Lu et al., 2012, Stafford et al., 2014, Vogt and Paxinos, 2014), are shown to support
544 structural connectivity between the mPFC and MTL, essential to associative memory networks and the
545 DMN (Greicius et al., 2009, Miller et al., 2014). The hubness of the RSC during retrieval is clearly
546 supported by the community organization of our functional networks, where it is well positioned to
547 facilitate communication between the mPFC and MTL communities (Figure 4B). The severe loss in
548 strength seen in the dysgranular RSC may therefore indicate an object-place recall-dependent roadblock
549 for mPFC-MTL communication.

550 One question lies as to why the dysgranular RSC was more severely disrupted than the granular part. The
551 dysgranular cortex is comparatively more connected to visual and sensory processing areas, making it
552 ideal for processing and identifying local and distal cues for allocentric orientation. Lesioning the
553 dysgranular RSC alone is enough to have rats shift from allocentric to egocentric strategies in a spatial
554 memory task (Vann and Aggleton., 2005). The dysgranular RSC contains landmark dependent head
555 direction cells, which may help orient the mice in the open field with respect to the objects (Jacob et al.,
556 2016), and decreased strength of the dysgranular RSC may contribute to their dysfunction.

557 Our findings also corroborate those of human studies showing that the mPFC and the PCC/RSC are often
558 disrupted in evaluations of functional and structural connectivity in aMCI (Catheline et al., 2010; Wang et

559 al., 2013c). The especially severe disruption of the RSC reflects clinical observations of the human PCC
560 as one of the most, if not the most, consistently disrupted regions in evaluations of resting state FC of
561 aMCI (Badhwar et al., 2017; Eyler et al., 2019). The mPFC and the PCC/RSC are also the first regions to
562 display amyloid deposition (Palmqvist et al., 2017). The PCC/RSC shows reduced connectivity in pre-
563 plaque APOE ϵ 4 carriers at risk for developing AD (Wang et al., 2013b; Jones et al., 2016), and very
564 early increases in PCC amyloid deposition correlate with face-name associative memory deficits in
565 subjective cognitive decline (Sanabria et al., 2017). Initial pathologies of amyloid in the mPFC/PCC/RSC
566 and tau in the MTC may thus have a combined negative effect on associative memory through reducing
567 memory-recall dependent FC.

568 **Decreased global efficiency during retention predicts memory deficits in dKI mice**

569 In WT mice, we detected a positive relationship between global efficiency of the retention network and
570 memory performance when taking into account a disruptive effect of total object exploration. The
571 exploration disruption on memory driven FC can perhaps be explained by increased focus and a more
572 conscious exploration of objects for mice with a low exploration index, in contrast to a more haphazard
573 exploration of objects for mice with high exploration index. This may also indicate a disruptive effect of
574 simultaneous sensory or motor network activation. Regardless, dKI mice presented no difference in object
575 exploration as compared to the WT mice, making it an unlikely behavioral link to reduced global
576 efficiency in these mice.

577 Global efficiency of c-Fos derived fear memory networks has already been shown as a reliable measure
578 for predicting memory performance in mice (Vetere et al., 2017). Such findings are consistent with
579 clinical observations linking increased global efficiency to better cognitive function (Li et al., 2009;
580 Stanley et al., 2015). The relationship across the WT group can be extrapolated down to predict memory
581 performance in the dKI. This provides further evidence that their drop in memory performance is directly
582 linked to deficient information transfer of the memory retention network. There is increasing evidence
583 that memory loss in AD pathology is related to dysfunctional recall, as it can be rescued by aiding
584 retrieval processes (i.e., activating the “silent” engram; Roy et al., 2016, Perusini et al., 2017). Perhaps
585 early associative memory deficits of dKI mice could be rescued by activating “silent” inefficient
586 functional networks. In humans, noninvasive brain stimulation has been extensively studied as a potential
587 therapeutic tool for AD on regions such as the dorsolateral mPFC with mixed results (Weiler et al., 2020).
588 However, electromagnetic stimulation of the parietal cortex was shown to support associative memory by
589 increasing memory-retrieval dependent FC strength of an associated parietal-RSC-hippocampal network
590 (Wang et al., 2014; Warren et al., 2019). Taken together with our study, these results suggest that the

591 PCC/RSC, or more laterally accessible and functionally similar regions such as the parietal cortex, may
592 be more effective targets for improving associative memory in AD.

593 **Contrast with resting state fMRI in other mouse models of AD**

594 Contrary to our results, studies that evaluate resting state networks in pre-plaque or early-NFT mouse
595 models of AD predominantly find cortical and hippocampal-cortical hyperconnectivity, while
596 hypoconnectivity appears at later stages (Asaad and Lee, 2018). This suggests that perturbations in resting
597 state FC may predict memory FC perturbations, but do not directly mirror them. Moreover, the mouse
598 models used to evaluate pre-aggregate resting state FC presented either amyloid pathology (Shah et al.,
599 2016; Shah et al., 2018) or tau pathology (Degiorgis et al., 2020) but not both concurrently. It has been
600 shown with combined resting state fMRI and PET imaging in humans that increased A β alone is
601 associated with hyperconnectivity of the DMN while combined A β and Tau pathologies reveal
602 hypoconnectivity (Schultz et al., 2017). Whether these contrasting results reflect differences between
603 “resting state vs memory driven FC” or differences in “amyloid/tau pathological staging” will be more
604 thoroughly understood once resting state fMRI is directly measured in young dKI mice.

605 In conclusion, our results suggest that the present dKI model was caught at the very beginning of its
606 neuropathology as it was restricted to the MTC region, leaving the dorsal hippocampus quite preserved.
607 The local MTC pathology of these mice was associated with EC and CLA hyperactivity which would
608 most likely spread towards densely interconnected regions such as the mPFC, the DH and the RSC.
609 Retrieval dependent communication between cingulate areas and the MTL was disrupted, and can be
610 potentially linked to reduced dysgranular RSC hub strength. The similarity between our findings in the
611 dKI model and those reported in the earliest stages of the disease suggests that the *App*^{NL-F} version of the
612 dKI model has a high potential for generating new discoveries on the earliest stage of AD.

613

614

615 **Methods**

616 **Animals**

617 The *App*^{NL-F}/*MAPT* double knock-in (dKI) mice were produced through crossing homozygotes of two
618 single knock-in (KI) mouse lines for the humanized *App*^{NL-F} and *MAPT* genes, and then through the
619 crossing of the resulting doubly heterozygote mice to obtain dKI and non-knock-in WT line founders.
620 After three generation of homozygous breeding, the dKI mouse line was backcrossed with C57BL/6J
621 mice (Janvier Laboratories, Le Genest Saint Isle, France) in order to limit genetic drift between dKI mice
622 and their WT controls. The *App*^{NL-F} gene contains a humanized A β fragment with Beyreuther/Iberian and
623 Swedish FAD mutations, and the human *MAPT* gene expresses all 6 isoforms of tau found in humans.
624 Both single KI mouse lines were produced by T Saido and T Saito (RIKEN Brain Science Institute,
625 JAPAN) and sent to us by the RIKEN BioResource Center. Mice were group-housed with food and water
626 ad libitum, nesting material, and additional food pellets on bedding to promote natural behavioral
627 patterns. The animal room was under controlled temperature (23 °C \pm 1°C) and a 12/12-hour light/dark
628 cycle (lights on at 8.00 AM). Procedures were in compliance with rules of the European Community
629 Council Directive 2010-63 and French Department of Agriculture Directive 2013-118 and approved by
630 the local review board (CREMEAS: APAFIS#9848). Animal facilities were approved for animal
631 experimentation (H 67-482-13).

632 **Behavioral Testing**

633 Behavioral testing took place during the light phase. Mice were single-housed for 1 week before testing.
634 Spontaneous object exploration tests were carried out in an open field (100cmx100cmx50cm, Ugo Basile,
635 Italy) with dark grey acrylic walls and a grey metal floor. The open field was evenly illuminated by three
636 indirect halogen lights (open field center, 15 lux), and a radio gave background noise from 1.5m away
637 (open field center, 45 \pm 5 dB). Nine different sets of objects were used: two for the habituation phases,
638 two for the long-term novel object recognition task, two for the OP task, two for the short-term novel
639 object recognition task, and one for the object location task. These objects differed in size (10 to 20 cm),
640 material (metal, glass, or plastic), shape, and color. Each object was available in duplicate or triplicate.
641 Ethanol (30%) was used to clean the objects and the open field between each trial. Object exploration
642 time was recorded and defined as the nose pointing toward the object within 2 cm. Gnawing and climbing
643 of objects were not counted as exploration time.

644 **Habituation**

645 Before testing, all mice received two days of habituation. On the first day, they were given a habituation
646 trial of 10 min with two identical objects placed in the open field. On the second day they were given two
647 10 min trials with a different set of two identical objects, with the trials separated by an inter trial interval
648 (ITI) of 5' that the mice spent in their home cage. For the OP task with a 3-hour ITI, the second day
649 habituation procedure was applied with an ITI of 3 hours.

650 **Preliminary testing cohorts**

651 Mice were tested at 2 months (WT n = 10, dKI n = 10), 4 months (WT n = 12, dKI n = 9) and at 6 months
652 (WT n = 8, dKI n = 11). An additional cohort of (WT n = 3, dKI n = 1) was tested at 2 months in object-
653 place association as the initial WT group did not reach significance above 0. For pattern separation one
654 two month old WT mouse was removed due to the wrong objects accidentally being placed during the
655 retention phase.

656 **Short term, pattern separation and long-term novel object recognition**

657 Mice were tested at 4 months (WT n = 11, dKI n = 11) in the short term OR task. One WT mouse was
658 removed from the analysis due to being dropped before the task. Mice explored two identical objects
659 during a 10-min acquisition trial, and were returned to their home cage for an ITI of 5 min (short term,
660 pattern separation) or 24 hours (long term). Thereafter, mice were given a 10-min retention trial, where
661 one of the familiar objects was replaced by an unfamiliar new one. For the pattern separation task, objects
662 were made of legos, the novel object had the same composition of colored lego blocks but a different
663 pattern. Exploration of the objects was recorded during the 6 minutes (4 minutes for pattern separation)
664 following the initial exploration of object for each mouse. The memory index was calculated as:

$$\text{Memory index} = \frac{\text{Time at replaced object} - \text{Time at unchanged object}}{\text{Time at both objects}}$$

665 **Short term object-place association**

666 To validate the potential deficit seen during preliminary phenotyping, an additional cohort (WT n = 11,
667 dKI n = 11) was tested in the object-place association task. Mice explored two different objects during a
668 10-min acquisition trial, and were returned to their home cage for an ITI of 5 minutes. During the 10-min
669 retention trial, one of the objects was replaced by a copy of the other. The mice had to detect the
670 mismatch between one object and its actual location in the open field. Exploration of the objects was
671 recorded during the 4 minutes following initiation of object exploration episode for each mouse. The
672 memory index was calculated as with the long-term object recognition task.

673 **Short term novel object location**

674 Mice were tested at 4 months (WT n = 11, dKI n =11). Mice explored two identical objects during a 10-
675 min acquisition trial, and were returned to their home cage for an ITI of 5 minutes. During a 10-min
676 retention trial, one of the objects was moved 55 cm from its original position for the object location task.
677 Exploration of the objects was evaluated during the 6 minutes following the initiation of object
678 exploration for each mouse. During both phases, all objects were placed equidistant from the walls. The
679 memory index was calculated as:

$$\text{Memory index} = \frac{\text{Time at moved object} - \text{Time at unmoved object}}{\text{Time at both objects}}$$

680 **Western Blotting**

681 Four-month-old male mice were sacrificed by cervical dislocation (n=8 per group), and their brains were
682 carefully dissected on ice. Hippocampi and medial temporal cortex were quickly removed, frozen in
683 liquid nitrogen and then stored at -80°C until their use. Tissues were homogenized in 10 volumes of ice-
684 cold radioimmunoprecipitation assay buffer containing protease inhibitor cocktail (Sigma-Aldrich, St.
685 Louis, Missouri), phosphatase inhibitor cocktail (PhosStop, Roche Life Science, Penzberg, Germany),
686 and 1 mM phenylmethylsulfonyl fluoride (Sigma-Aldrich). After centrifugation at 20,000g for 20 min at
687 4°C , supernatants were aliquoted for immunoblot analysis. Brains extracts of Tg2576 and Thy-tau22 mice
688 were homogeneized and separated in parallel to our samples as a positive control for APP proteins and
689 cleaved fragments and for tau proteins, respectively. Protein concentration was measured using the Bio-
690 Rad Protein Assay (Bio-Rad, Hercules, California). Thirty or 20 microgramms were respectively loaded
691 on 4-20% precast gel (Mini-Protean TGX precast gels, Bio-Rad) for APP and tau proteins. After
692 electrophoresis and transfer to nitrocellulose membranes using the Trans-Blot Turbo System (Bio-Rad),
693 membranes were incubated with 5% skimmed milk for 1 hour at room temperature and then with primary
694 antibodies diluted in 2% bovine serum albumin (Sigma-Aldrich) in tris-buffered saline 0.05% Tween 20
695 (Sigma-Aldrich) overnight at 4°C . After washes, membranes were incubated with anti-mouse or anti-
696 rabbit immunoglobulins conjugated to horseradish peroxidase (Jackson ImmunoResearch, West Grove,
697 Pennsylvania) for development with enhanced ECL chemiluminescence detection kit (Thermo Fisher
698 Scientific, Waltham, Massachusetts). After detection, all membranes were re-probed with anti-actin
699 antibody for normalization of total protein. The primary antibodies used were the rabbit polyclonal anti-
700 APP, C terminus (Sigma-Aldrich), the rabbit polyclonal anti-tau (B19, generously gifted by JP Brion,
701 ULB, Belgium), the mouse monoclonal anti-phospho-Tau Thr181 (AT270, ThermoScientific), the mouse
702 monoclonal anti-actin (Sigma-Aldrich) and the rabbit polyclonal anti-actin (Sigma-Aldrich). The
703 secondary antibodies used were the peroxidase-conjugated AffiniPure goat anti-mouse and goat anti-
704 rabbit (Jackson ImmunoResearch). The quantification of the band intensity acquired with the ChemiDoc

705 Imaging system (Bio-Rad) was performed by densitometry analysis using the ImageJ program. For each
706 mouse, the phosphorylation degree was calculated as the ratio of total phosphorylated Thr181 tau proteins
707 on total tau proteins. The ratio of APP-cleaved fragments was calculated as the total of APP β -CTF on α -
708 CTF fragments.

709 **Object-place association with a 3-hour ITI for ex-vivo imaging**

710 The same protocol as with the short-term object-place task was followed, except with an ITI of 3-hours.
711 The “Acquisition” set of WT (N=12) and dKI (N=12) mice were only tested in the acquisition phase. The
712 “Retention” set of WT (N=14) and dKI (N=14) mice were tested for the entire task, acquisition and
713 retention phases included. At the end of testing mice were left in their home cage in a quiet dim room (9
714 lux, 35 ± 5 dB) next to the testing room for 90 minutes, after which they were taken for brain perfusion
715 and removal.

716 **Perfusion and tissue preparation.**

717 Mice were killed with an overdose of sodium pentobarbital (105 mg/kg intraperitoneally) and
718 transcardially perfused in 0.1% heparin phosphate-buffered saline (PBS) and 4% paraformaldehyde [PFA;
719 in phosphate buffer (PB) pH7.4; 4°C]. Brains were removed, postfixed in a 4% PFA solution for 24h, and
720 cryoprotected in a saccharose solution (20% in PB, 0.1 M; pH 7.4; 4°C) for 48 hours before being frozen
721 with isopentane (-40°C) and subsequently stored at -80°C . Forty μm coronal cryostat sections were cut
722 from the anterior to the posterior of the brain. For the medial prefrontal cortex (mPFC), claustrum (CLA),
723 dorsal hippocampus (DH), medial entorhinal cortex (MEC), and postrhinal cortex (POC) every 4th section
724 created a set (spacing of 160 μm), and for the retrosplenial cortex (RSC), lateral entorhinal cortex (LEC),
725 and perirhinal cortex (PRC) every 6th section (spacing of 240 μm).

726 **Immunohistochemistry**

727 Brain sections were given three 10 min washes in 0.1 M phosphate-buffered saline (PBS), followed by a
728 30 min incubation in 1% H_2O_2 . They were washed for 5 min with ultra-pure water and again three times
729 for 10 min in 0.1 M PBS. This was followed by a 45 min blocking incubation in a 5% natal goat serum
730 (NGS) diluted in a “diluent” solution consisting of 0.1 M PBS, and 0.5% triton. The sections were
731 incubated at room temperature for 2 days in a 1/15000 dilution of rabbit-anti-cFos (Synaptic Systems,
732 Göttingen, Germany) primary antibody in diluent containing 2% NGS. After 2 days the sections were first
733 given two 10 min washes in 0.1 M PBS, and then incubated for 2 hours at room temperature in a 1/500
734 dilution of biotinylated mouse-anti-rabbit (Vector Laboratories, Burlingame, California) secondary
735 antibody in diluent containing 2% NGS. The sections were given two 10 min washes in PBS, followed by

736 a 45 min incubation in the avidin/biotin (Vector Laboratories) solution. This was followed by three 10
737 min washes in 0.1 M PBS and a 10 min wash in phosphate buffer. The sections were finally revealed with
738 a 10 min incubation in 3,3-diaminobenzidine (Vector Laboratories). Images of whole brain sections were
739 taken at 20x magnification using a Hamamatsu NanoZoomer S60 digital slide scanner (Hamamatsu
740 Photonics K.K., Hamamatsu City, Japan) for offline quantification of c-Fos expression.

741 **c-Fos Imaging**

742 Neuronal activation is associated with increases in intracellular calcium levels, which in turn leads to the
743 rapid up-regulation of immediate early genes such as c-fos. The quantification of c-Fos protein levels can
744 thus be used to derive a measure of neuronal activity (Tischmeyer and Grimm, 1999). c-Fos expression
745 was analyzed in 22 regions of interest (ROIs) (see Table S1), including sub-regions of the PFC, CLA,
746 DH, RSC, PRC, POC and EC. Image processing was done using ImageJ (National Institute of Health,
747 Bethesda, MD). ROIs were anatomically defined according to the atlas of Franklin and Paxinos (2008).
748 For c-Fos quantification, the images were transformed into 8-bit grayscale. A grayscale threshold was set
749 at a consistent level for each region by an experimenter blind to group condition. Only c-Fos positive
750 nuclei with a grayscale intensity below the threshold and an area between 25–300 μm^2 were counted. At
751 least three brain sections were processed per ROI. Mean c-Fos density was calculated for each ROI as the
752 quantity of c-Fos marked nuclei per mm^2 , normalized to the WT-Acquisition group. ROIs were grouped
753 into region subfields whenever anatomically and functionally justified. These groupings reduce the
754 number of comparisons and, thereby, restrict Type 1 errors. Three-way ANOVAs compared test-phases
755 (acquisition or retention), genotypes (WT or dKI) and subregions for each subfield. When an interaction
756 was significant, the simple effects were examined.

757 **Functional Connectivity**

758 From the c-Fos signals, functional connectivity (FC) was assessed by computing the between subject
759 inter-regional spearman correlations for each Genotype-Phase group. Spearman correlations, rather than
760 Pearson correlations, were used to account for potential outlier effects and the relatively small sample
761 sizes. Correlation matrices were used to visualize all possible pairwise inter-regional correlations within
762 each group. To assess global FC strength the mean r was calculated with retained, near-zeroed, and
763 absolute valued negative correlations respectively. In the near-zeroed case, negative correlations were
764 reduced to a value of .006, the smallest positive correlation observed.

765 **Generating functional networks as fully connected weighted graphs**

766 From each correlation matrix, a functional network was generated as a fully connected weighted graph.
767 The edges weights of the graph reflect inter-regional spearman correlation strengths and the nodes reflect
768 regions. Negative correlations can be interesting, but complicate considerably graph analyses as various
769 algorithm variants exist to handle them (e.g. in community detection) and there is no obvious criterion to
770 choose one variant over others. For the sake of clarity, we thus treated negative correlations in most
771 analyses (unless explicitly mentioned) as near-zero positive value of correlation (minimum edge weight
772 of 0.006, see above). In efficiency analyses, this corresponds to interpreting negative correlations as open
773 but difficult paths for information transfer. All graph construction and graph analysis were done through
774 the igraph (Csardi G and Nepusz T, 2006) package on R (R Core Team, 2017).

775 **Bootstrapping confidence intervals**

776 For all network metrics, confidence intervals were computed through bootstrapping. This involves
777 resampling subjects with replacement 1000 times, each time regenerating a functional network, then
778 recalculating the estimate of interest. The 95% quartile of the bootstrap distribution was taken as the 95%
779 confidence interval. Confidence intervals for the difference were used to test for differences between
780 genotype groups (Wright et al., 2011). Groups were considered different to a $P < .05$ if the 95 %
781 confidence interval for the difference ≥ 0 , and to a $P < .01$ if the 99 % confidence interval for the difference
782 ≥ 0 .

783 **Community Analysis**

784 Networks with high modularity, Q , have strong connections between the nodes within communities and
785 relatively weaker connections between nodes of different communities.

$$Q = \frac{1}{2m} \sum_{i \neq j \in N} \left[w_{ij} - \frac{s_i s_j}{2m} \right] \delta(c_i, c_j)$$

786 where N denotes the set of all nodes in the network, m denotes the total number of edges in the network,
787 $w(i, j)$ denotes the edge weight between a node i and another node j , s denotes the sum of a node's edge
788 weights, c denotes the community to which a node belongs, and $\delta(c_i, c_j)$ indicates if the compared nodes
789 are in the same community ($\delta(c_i, c_j)$ is 1 if $c_i = c_j$, and 0 otherwise). Communities were detected in
790 each bootstrap through finding the maximum modularity across all possible community partitions. This
791 modularity maximization computation was done through the *cluster_optimal* function of the igraph
792 package. This transforms modularity maximization into an integer programming problem, and calls the
793 GNU Linear Programming Kit (GLPK) to solve that. See Brandes et al, 2008 for more details. This
794 computationally expensive detection method of evaluating all possible community partitions for

795 modularity maximization (in contrast to less expensive methods that infer modularity, as through greedy
796 optimization, i.e. the Louvain method), could be permitted due to the relatively small number of nodes in
797 our network. Allegiance matrices were used to assess community stability across bootstraps, by depicting
798 the percentage of bootstraps ($n = 1000$) that contain any given pair of regions within the same
799 community.

800 **Information flow**

801 Nodal strength, \mathbf{s} , is traditionally calculated as the sum of a node's edge weights. In a fully connected
802 network this is directly proportional to the average of a node's edge weights. The average, $\mathbf{s}_{average}$, was
803 used in our case for better comparison with nodal efficiency.

$$s_{average}(i) = \frac{1}{n-1} \sum_{j \in N} w_{ij}$$

804 where N denotes the set of all nodes in the network, n denotes the total number of nodes in the network,
805 and w_{ij} denotes the edge weight between a node i and another node j .

806 For efficiency metrics, edge lengths were first computed as inverted edge weights. Nodal efficiency,
807 E_{nodal} , was calculated as the average inverse shortest path length between a region and all other regions
808 of the network.

$$E_{nodal}(i) = \frac{1}{n-1} \sum_{j \in N} \frac{1}{d_{ij}}$$

809 where d_{ij} denotes the length of the shortest path (lowest sum of edge lengths) between a node i and
810 another node j . Global efficiency, E_{global} , was calculated as the average inverse shortest path length of
811 the network.

$$E_{global} = \frac{1}{n(n-1)} \sum_{i \neq j \in N} \frac{1}{d_{ij}}$$

812

813 **Assessing the impact of global efficiency on memory deficits**

814 Subsamples were generated from each retention group by resampling $n-1$ mice without replacement. This
815 procedure generates a set of subsamples of size $n-1$, where the number of subsamples is the number of
816 mice, n . From each subsample, the average memory index and the resulting network global efficiency

817 were computed. Correlation significance between memory index and global efficiency was computed
818 through both Pearson and Spearman correlation coefficients. To compute the exploration adjusted
819 memory index, first the exploration index was defined for each mouse as total time exploring both objects
820 normalized to the group average:

$$\text{Exploration Index (EI)} = \frac{\text{Time exploring both objects of mouse}}{\text{Average time exploring both objects across the group}}$$

821 The exploration adjusted memory index was then defined as the memory index divided by the exploration
822 index.

$$\text{Exploration adjusted memory index (MI/EI)} = \frac{\text{Memory Index (MI)}}{\text{Exploration Index (EI)}}$$

823 To assess the significance of the intersection of the WT linear fit with the dKI subsample population,
824 1000 randomly fit linear models to the WT were generated by permuting the y labels of the WT
825 subsample population and recalculating the best fit line each time. The mean absolute error to the dKI
826 subsamples was calculated for each random model and their distribution is presented as a histogram. The
827 95% confidence interval was considered as the 95% quartile of the distribution, and compared to the dKI
828 mean absolute error to the original WT fit.

829

830 References

- 831 Asaad M, Lee JH. 2018. A guide to using functional magnetic resonance imaging to study Alzheimer's disease in
832 animal models. *Dis Model Mech* **11**. doi:[10.1242/dmm.031724](https://doi.org/10.1242/dmm.031724)
- 833 Avila J, Perry G. 2021. A Multilevel View of the Development of Alzheimer's Disease. *Neuroscience* **457**:283–293.
834 doi:[10.1016/j.neuroscience.2020.11.015](https://doi.org/10.1016/j.neuroscience.2020.11.015)
- 835 Bai L, Zhang M, Chen S, Ai L, Xu M, Wang D, Wang Fei, Liu L, Wang Fang, Lao L. 2013. Characterizing
836 acupuncture de qi in mild cognitive impairment: relations with small-world efficiency of functional brain
837 networks. *Evid Based Complement Alternat Med* **2013**:304804. doi:[10.1155/2013/304804](https://doi.org/10.1155/2013/304804)
- 838 Bassett DS, Bullmore E. 2006. Small-world brain networks. *Neuroscientist* **12**:512–523.
839 doi:[10.1177/1073858406293182](https://doi.org/10.1177/1073858406293182)
- 840 Bassett DS, Yang M, Wymbs NF, Grafton ST. 2015. Learning-induced autonomy of sensorimotor systems. *Nat*
841 *Neurosci* **18**:744–751. doi:[10.1038/nn.3993](https://doi.org/10.1038/nn.3993)
- 842 Bergmann E, Zur G, Bershadsky G, Kahn I. 2016. The Organization of Mouse and Human Cortico-Hippocampal
843 Networks Estimated by Intrinsic Functional Connectivity. *Cereb Cortex* **26**:4497–4512.
844 doi:[10.1093/cercor/bhw327](https://doi.org/10.1093/cercor/bhw327)
- 845 Bonardi C, Pardon M-C, Armstrong P. 2016. Deficits in object-in-place but not relative recency performance in the
846 APPsw/PS1dE9 mouse model of Alzheimer's disease: Implications for object recognition. *Behavioural Brain*
847 *Research* **313**:71–81. doi:[10.1016/j.bbr.2016.07.008](https://doi.org/10.1016/j.bbr.2016.07.008)
- 848 Braak H, Braak E. 1995. Staging of Alzheimer's disease-related neurofibrillary changes. *Neurobiol Aging* **16**:271–
849 278; discussion 278-284. doi:[10.1016/0197-4580\(95\)00021-6](https://doi.org/10.1016/0197-4580(95)00021-6)
- 850 Brandes U, Delling D, Gaertler M, Gorke R, Hofer M, Nikoloski Z, Wagner D. 2008. On Modularity Clustering.
851 *IEEE Trans Knowl Data Eng* **20**:172–188. doi:[10.1109/TKDE.2007.190689](https://doi.org/10.1109/TKDE.2007.190689)
- 852 Catheline G, Periot O, Amirault M, Braun M, Dartigues J-F, Auriacombe S, Allard M. 2010. Distinctive alterations
853 of the cingulum bundle during aging and Alzheimer's disease. *Neurobiology of Aging* **31**:1582–1592.
854 doi:[10.1016/j.neurobiolaging.2008.08.012](https://doi.org/10.1016/j.neurobiolaging.2008.08.012)
- 855 Caviezel MP, Reichert CF, Sadeghi Bahmani D, Linnemann C, Liechti C, Bieri O, Borgwardt S, Leyhe T, Melcher
856 T. 2020. The Neural Mechanisms of Associative Memory Revisited: fMRI Evidence from Implicit
857 Contingency Learning. *Front Psychiatry* **10**. doi:[10.3389/fpsy.2019.01002](https://doi.org/10.3389/fpsy.2019.01002)
- 858 Chao OY, Huston JP, Li J-S, Wang A-L, Silva MA de S. 2016. The medial prefrontal cortex—lateral entorhinal
859 cortex circuit is essential for episodic-like memory and associative object-recognition. *Hippocampus* **26**:633–
860 645. doi:[10.1002/hipo.22547](https://doi.org/10.1002/hipo.22547)
- 861 Chen G, Xu T, Yan Y, Zhou Y, Jiang Y, Melcher K, Xu HE. 2017. Amyloid beta: structure, biology and structure-
862 based therapeutic development. *Acta Pharmacologica Sinica* **38**:1205–1235. doi:[10.1038/aps.2017.28](https://doi.org/10.1038/aps.2017.28)
- 863 Corriveau-Lecavalier N, Duchesne S, Gauthier S, Hudon C, Kergoat M, Mellah S, Belleville S, for the Consortium
864 for the Early Identification of Alzheimer's Disease-Quebec (CIMA-Q). 2020. A quadratic function of
865 activation in individuals at risk of Alzheimer's disease. *Alzheimer's & Dementia: Diagnosis, Assessment*
866 *& Disease Monitoring* **12**. doi:[10.1002/dad2.12139](https://doi.org/10.1002/dad2.12139)
- 867 Csardi G, Nepusz T. 2006. The igraph software package for complex network research. *InterJournal Complex*
868 *Systems*:1695. doi:<https://igraph.org>
- 869

- 870 Degiorgis L, Karatas M, Sourty M, Faivre E, Lamy J, Noblet V, Bienert T, Reisert M, von Elverfeldt D, Buée L,
871 Blum D, Boutillier A-L, Armspach J-P, Blanc F, Harsan L-A. 2020. Brain network remodelling reflects tau-
872 related pathology prior to memory deficits in Thy-Tau22 mice. *Brain*. doi:[10.1093/brain/awaa312](https://doi.org/10.1093/brain/awaa312)
- 873 Deshmukh SS, Johnson JL, Knierim JJ. 2012. Perirhinal cortex represents nonspatial, but not spatial, information in
874 rats foraging in the presence of objects: Comparison with lateral entorhinal cortex. *Hippocampus* **22**:2045–
875 2058. doi:[10.1002/hipo.22046](https://doi.org/10.1002/hipo.22046)
- 876 DiCiccio, T. J. and B. Efron. (1996). Bootstrap Confidence Intervals. *Statistical Science*. 11(3): 189–212.
877 <https://doi.org/10.1214/ss/1032280214>
- 878 Drzegza A, Becker JA, Van Dijk KRA, Sreenivasan A, Talukdar T, Sullivan C, Schultz AP, Sepulcre J, Putcha D,
879 Greve D, Johnson KA, Sperling RA. 2011. Neuronal dysfunction and disconnection of cortical hubs in non-
880 demented subjects with elevated amyloid burden. *Brain* **134**:1635–1646. doi:[10.1093/brain/awr066](https://doi.org/10.1093/brain/awr066)
- 881 Etter G, van der Veldt S, Manseau F, Zarrinkoub I, Trillaud-Doppia E, Williams S. 2019. Optogenetic gamma
882 stimulation rescues memory impairments in an Alzheimer’s disease mouse model. *Nature Communications*
883 **10**:5322. doi:[10.1038/s41467-019-13260-9](https://doi.org/10.1038/s41467-019-13260-9)
- 884 Eyler LT, Elman JA, Hatton SN, Gough S, Mischel AK, Hagler DJ, Franz CE, Docherty A, Fennema-Notestine C,
885 Gillespie N, Gustavson D, Lyons MJ, Neale MC, Panizzon MS, Dale AM, Kremen WS. 2019. Resting State
886 Abnormalities of the Default Mode Network in Mild Cognitive Impairment: A Systematic Review and Meta-
887 Analysis. *J Alzheimers Dis* **70**:107–120. doi:[10.3233/JAD-180847](https://doi.org/10.3233/JAD-180847)
- 888 Franklin K, Paxinos G. 2008. The mouse brain in stereotaxic coordinates. San Diego: Academic Press.
889
- 890 Filippi M, van den Heuvel MP, Fornito A, He Y, Hulshoff Pol HE, Agosta F, Comi G, Rocca MA. 2013.
891 Assessment of system dysfunction in the brain through MRI-based connectomics. *The Lancet Neurology*
892 **12**:1189–1199. doi:[10.1016/S1474-4422\(13\)70144-3](https://doi.org/10.1016/S1474-4422(13)70144-3)
- 893 Gardini S, Venneri A, Sambataro F, Cuetos F, Fasano F, Marchi M, Crisi G, Caffarra P. 2015. Increased functional
894 connectivity in the default mode network in mild cognitive impairment: a maladaptive compensatory
895 mechanism associated with poor semantic memory performance. *J Alzheimers Dis* **45**:457–470.
896 doi:[10.3233/JAD-142547](https://doi.org/10.3233/JAD-142547)
- 897 Grajski KA, Bressler SL. 2019. Differential medial temporal lobe and default-mode network functional connectivity
898 and morphometric changes in Alzheimer’s disease. *NeuroImage: Clinical* **23**:101860.
899 doi:[10.1016/j.nicl.2019.101860](https://doi.org/10.1016/j.nicl.2019.101860)
- 900 Granholm E, Butters N. 1988. Associative encoding and retrieval in Alzheimer’s and Huntington’s disease. *Brain*
901 *Cogn* **7**:335–347. doi:[10.1016/0278-2626\(88\)90007-3](https://doi.org/10.1016/0278-2626(88)90007-3)
- 902 Greicius MD, Supekar K, Menon V, Dougherty RF. 2009. Resting-State Functional Connectivity Reflects Structural
903 Connectivity in the Default Mode Network. *Cereb Cortex* **19**:72–78. doi:[10.1093/cercor/bhn059](https://doi.org/10.1093/cercor/bhn059)
- 904 Guo Q, Li H, Cole AL, Hur J-Y, Li Y, Zheng H. 2013. Modeling Alzheimer’s Disease in Mouse without Mutant
905 Protein Overexpression: Cooperative and Independent Effects of A β and Tau. *PLOS ONE* **8**:e80706.
906 doi:[10.1371/journal.pone.0080706](https://doi.org/10.1371/journal.pone.0080706)
- 907 Gustafson L, Brun A, Englund E, Hagnell O, Nilsson K, Stensmyr M, Öhlin A-K, Abrahamson M. 1998. A 50-year
908 perspective of a family with chromosome-14-linked Alzheimer’s disease. *Hum Genet* **102**:253–257.
909 doi:[10.1007/s004390050688](https://doi.org/10.1007/s004390050688)
- 910 Hales JB, Brewer JB. 2011. The timing of associative memory formation: frontal lobe and anterior medial temporal
911 lobe activity at associative binding predicts memory. *J Neurophysiol* **105**:1454–1463.
912 doi:[10.1152/jn.00902.2010](https://doi.org/10.1152/jn.00902.2010)

- 913 Hamm V, Héraud C, Bott J-B, Herbeaux K, Strittmatter C, Mathis C, Goutagny R. 2017. Differential contribution of
914 APP metabolites to early cognitive deficits in a TgCRND8 mouse model of Alzheimer's disease. *Sci Adv*
915 **3**:e1601068. doi:[10.1126/sciadv.1601068](https://doi.org/10.1126/sciadv.1601068)
- 916 Hampstead BM, Stringer AY, Stilla RF, Amaraneni A, Sathian K. 2011. Where did I put that? Patients with
917 amnesic mild cognitive impairment demonstrate widespread reductions in activity during the encoding of
918 ecologically relevant object-location associations. *Neuropsychologia* **49**:2349–2361.
919 doi:[10.1016/j.neuropsychologia.2011.04.008](https://doi.org/10.1016/j.neuropsychologia.2011.04.008)
- 920 Hampstead BM, Towler S, Stringer AY, Sathian K. 2018. Continuous measurement of object location memory is
921 sensitive to effects of age and mild cognitive impairment and related to medial temporal lobe volume.
922 *Alzheimers Dement (Amst)* **10**:76–85. doi:[10.1016/j.dadm.2017.10.007](https://doi.org/10.1016/j.dadm.2017.10.007)
- 923 Hernandez AR, Reasor JE, Truckenbrod LM, Lubke KN, Johnson SA, Bizon JL, Maurer AP, Burke SN. 2017.
924 Medial prefrontal-perirhinal cortical communication is necessary for flexible response selection. *Neurobiology*
925 *of Learning and Memory* **137**:36–47. doi:[10.1016/j.nlm.2016.10.012](https://doi.org/10.1016/j.nlm.2016.10.012)
- 926 Hirni DI, Kivisaari SL, Krumm S, Monsch AU, Berres M, Oeksuez F, Reinhardt J, Ulmer S, Kressig RW, Stippich
927 C, Taylor KI. 2016. Neuropsychological Markers of Medial Perirhinal and Entorhinal Cortex Functioning are
928 Impaired Twelve Years Preceding Diagnosis of Alzheimer's Dementia. *JAD* **52**:573–580. doi:[10.3233/JAD-150158](https://doi.org/10.3233/JAD-150158)
- 930 Hyman B, Van Hoesen G, Damasio A, Barnes C. 1984. Alzheimer's disease: cell-specific pathology isolates the
931 hippocampal formation. *Science* **225**:1168–1170. doi:[10.1126/science.6474172](https://doi.org/10.1126/science.6474172)
- 932 Jacob P-Y, Casali G, Spieser L, Page H, Overington D, Jeffery K. 2017. An independent, landmark-dominated head-
933 direction signal in dysgranular retrosplenial cortex. *Nature Neuroscience* **20**:173–175. doi:[10.1038/nn.4465](https://doi.org/10.1038/nn.4465)
- 934 Jones DT, Graff-Radford J, Lowe VJ, Wiste HJ, Gunter JL, Senjem ML, Botha H, Kantarci K, Boeve BF, Knopman
935 DS, Petersen RC, Jack CR. 2017. Tau, amyloid, and cascading network failure across the Alzheimer's disease
936 spectrum. *Cortex*, Special Section dedicated to the temporal and parietal lobes **97**:143–159.
937 doi:[10.1016/j.cortex.2017.09.018](https://doi.org/10.1016/j.cortex.2017.09.018)
- 938 Jones DT, Knopman DS, Gunter JL, Graff-Radford J, Vemuri P, Boeve BF, Petersen RC, Weiner MW, Jack CR Jr,
939 on behalf of the Alzheimer's Disease Neuroimaging Initiative. 2016. Cascading network failure across the
940 Alzheimer's disease spectrum. *Brain* **139**:547–562. doi:[10.1093/brain/awv338](https://doi.org/10.1093/brain/awv338)
- 941 Khan UA, Liu L, Provenzano FA, Berman DE, Profaci CP, Sloan R, Mayeux R, Duff KE, Small SA. 2014.
942 Molecular drivers and cortical spread of lateral entorhinal cortex dysfunction in preclinical Alzheimer's
943 disease. *Nat Neurosci* **17**:304–311. doi:[10.1038/nn.3606](https://doi.org/10.1038/nn.3606)
- 944 Kinnavane L, Albasser MM, Aggleton JP. 2015. Advances in the behavioural testing and network imaging of rodent
945 recognition memory. *Behavioural Brain Research*, SI: Object Recognition Memory in Rats and Mice **285**:67–
946 78. doi:[10.1016/j.bbr.2014.07.049](https://doi.org/10.1016/j.bbr.2014.07.049)
- 947 Latora V, Marchiori M. 2001. Efficient Behavior of Small-World Networks. *Phys Rev Lett* **87**:198701.
948 doi:[10.1103/PhysRevLett.87.198701](https://doi.org/10.1103/PhysRevLett.87.198701)
- 949 Lein ES, Hawrylycz MJ, Ao N, Ayres M, Bensinger A, Bernard A, Boe AF, Boguski MS, Brockway KS, Byrnes EJ,
950 Chen Lin, Chen Li, Chen T-M, Chi Chin M, Chong J, Crook BE, Czaplinska A, Dang CN, Datta S, Dee NR,
951 Desaki AL, Desta T, Diep E, Dolbeare TA, Donelan MJ, Dong H-W, Dougherty JG, Duncan BJ, Ebbert AJ,
952 Eichele G, Estin LK, Faber C, Facer BA, Fields R, Fischer SR, Fliiss TP, Frensley C, Gates SN, Glattfelder KJ,
953 Halverson KR, Hart MR, Hohmann JG, Howell MP, Jeung DP, Johnson RA, Karr PT, Kawal R, Kidney JM,
954 Knapik RH, Kuan CL, Lake JH, Laramée AR, Larsen KD, Lau C, Lemon TA, Liang AJ, Liu Y, Luong LT,
955 Michaels J, Morgan JJ, Morgan RJ, Mortrud MT, Mosqueda NF, Ng LL, Ng R, Orta GJ, Overly CC, Pak TH,
956 Parry SE, Pathak SD, Pearson OC, Puchalski RB, Riley ZL, Rockett HR, Rowland SA, Royall JJ, Ruiz MJ,

- 957 Sarno NR, Schaffnit K, Shapovalova NV, Sivisay T, Slaughterbeck CR, Smith SC, Smith KA, Smith BI, Sodr
958 AJ, Stewart NN, Stumpf K-R, Sunkin SM, Sutram M, Tam A, Teemer CD, Thaller C, Thompson CL, Varnam
959 LR, Visel A, Whitlock RM, Wohnoutka PE, Wolkey CK, Wong VY, Wood M, Yaylaoglu MB, Young RC,
960 Youngstrom BL, Feng Yuan X, Zhang B, Zwingman TA, Jones AR. 2007. Genome-wide atlas of gene
961 expression in the adult mouse brain. *Nature* **445**:168–176. doi:[10.1038/nature05453](https://doi.org/10.1038/nature05453)
- 962 Liang J, Li Y, Liu H, Zhang S, Wang M, Chu Y, Ye J, Xi Q, Zhao X. 2020. Increased intrinsic default-mode
963 network activity as a compensatory mechanism in aMCI: a resting-state functional connectivity MRI study.
964 *Aging (Albany NY)* **12**:5907–5919. doi:[10.18632/aging.102986](https://doi.org/10.18632/aging.102986)
- 965 Lin S-Y, Lin C-P, Hsieh T-J, Lin C-F, Chen S-H, Chao Y-P, Chen Y-S, Hsu C-C, Kuo L-W. 2019. Multiparametric
966 graph theoretical analysis reveals altered structural and functional network topology in Alzheimer’s disease.
967 *NeuroImage: Clinical* **22**:101680. doi:[10.1016/j.nicl.2019.101680](https://doi.org/10.1016/j.nicl.2019.101680)
- 968 Liu Z, Zhang Y, Yan H, Bai L, Dai R, Wei W, Zhong C, Xue T, Wang H, Feng Y, You Y, Zhang X, Tian J. 2012.
969 Altered topological patterns of brain networks in mild cognitive impairment and Alzheimer’s disease: A
970 resting-state fMRI study. *Psychiatry Research: Neuroimaging* **202**:118–125.
971 doi:[10.1016/j.psychres.2012.03.002](https://doi.org/10.1016/j.psychres.2012.03.002)
- 972 Lu H, Zou Q, Gu H, Raichle ME, Stein EA, Yang Y. 2012. Rat brains also have a default mode network. *PNAS*
973 **109**:3979–3984. doi:[10.1073/pnas.1200506109](https://doi.org/10.1073/pnas.1200506109)
- 974 Maass A, Berron D, Harrison TM, Adams JN, La Joie R, Baker S, Mellinger T, Bell RK, Swinnerton K, Inglis B,
975 Rabinovici GD, Düzel E, Jagust WJ. 2019. Alzheimer’s pathology targets distinct memory networks in the
976 ageing brain. *Brain* **142**:2492–2509. doi:[10.1093/brain/awz154](https://doi.org/10.1093/brain/awz154)
- 977 McAvoy K, Besnard A, Sahay A. 2015. Adult hippocampal neurogenesis and pattern separation in DG: a role for
978 feedback inhibition in modulating sparseness to govern population-based coding. *Front Syst Neurosci* **9**.
979 doi:[10.3389/fnsys.2015.00120](https://doi.org/10.3389/fnsys.2015.00120)
- 980 Miller AMP, Vedder LC, Law LM, Smith DM. 2014. Cues, context, and long-term memory: the role of the
981 retrosplenial cortex in spatial cognition. *Front Hum Neurosci* **8**. doi:[10.3389/fnhum.2014.00586](https://doi.org/10.3389/fnhum.2014.00586)
- 982 Mitchell AS, Czajkowski R, Zhang N, Jeffery K, Nelson AJD. 2018. Retrosplenial cortex and its role in spatial
983 cognition. *Brain Neurosci Adv* **2**. doi:[10.1177/2398212818757098](https://doi.org/10.1177/2398212818757098)
- 984 Morys J, Bobinski M, Wegiel J, Wisniewski HM, Narkiewicz O. 1996. Alzheimer’s disease severely affects areas of
985 the claustrum connected with the entorhinal cortex. *J Hirnforsch* **37**:173–180.
- 986 Myers N, Pasquini L, Göttler J, Grimmer T, Koch K, Ortner M, Neitzel J, Mühlau M, Förster S, Kurz A, Förstl H,
987 Zimmer C, Wohlschläger AM, Riedl V, Drzezga A, Sorg C. 2014. Within-patient correspondence of amyloid-
988 β and intrinsic network connectivity in Alzheimer’s disease. *Brain* **137**:2052–2064. doi:[10.1093/brain/awu103](https://doi.org/10.1093/brain/awu103)
- 989 Naveh-Benjamin M. 2000. Adult age differences in memory performance: Tests of an associative deficit hypothesis.
990 *Journal of Experimental Psychology: Learning, Memory, and Cognition* **26**:1170–1187. doi:[10.1037/0278-
991 7393.26.5.1170](https://doi.org/10.1037/0278-7393.26.5.1170)
- 992 Nuriel T, Angulo SL, Khan U, Ashok A, Chen Q, Figueroa HY, Emrani S, Liu L, Herman M, Barrett G, Savage V,
993 Buitrago L, Cepeda-Prado E, Fung C, Goldberg E, Gross SS, Hussaini SA, Moreno H, Small SA, Duff KE.
994 2017. Neuronal hyperactivity due to loss of inhibitory tone in APOE4 mice lacking Alzheimer’s disease-like
995 pathology. *Nat Commun* **8**:1464. doi:[10.1038/s41467-017-01444-0](https://doi.org/10.1038/s41467-017-01444-0)
- 996 Ogomori K, Kitamoto T, Tateishi J, Sato Y, Suetsugu M, Abe M. 1989. Beta-protein amyloid is widely distributed
997 in the central nervous system of patients with Alzheimer’s disease. *Am J Pathol* **134**:243–251.

- 998 Palmqvist S, Schöll M, Strandberg O, Mattsson N, Stomrud E, Zetterberg H, Blennow K, Landau S, Jagust W,
999 Hansson O. 2017. Earliest accumulation of β -amyloid occurs within the default-mode network and
1000 concurrently affects brain connectivity. *Nature Communications* **8**:1214. doi:[10.1038/s41467-017-01150-x](https://doi.org/10.1038/s41467-017-01150-x)
- 1001 Parron C, Save E. 2004. Comparison of the effects of entorhinal and retrosplenial cortical lesions on habituation,
1002 reaction to spatial and non-spatial changes during object exploration in the rat. *Neurobiology of Learning and*
1003 *Memory* **82**:1–11. doi:[10.1016/j.nlm.2004.03.004](https://doi.org/10.1016/j.nlm.2004.03.004)
- 1004 Pereira JB, Mijalkov M, Kakaei E, Mecocci P, Vellas B, Tsolaki M, Kłoszewska I, Soininen H, Spenger C,
1005 Lovestone S, Simmons A, Wahlund L-O, Volpe G, Westman E. 2016. Disrupted Network Topology in
1006 Patients with Stable and Progressive Mild Cognitive Impairment and Alzheimer’s Disease. *Cereb Cortex*
1007 **26**:3476–3493. doi:[10.1093/cercor/bhw128](https://doi.org/10.1093/cercor/bhw128)
- 1008 Pereira JB, Ossenkoppele R, Palmqvist S, Strandberg TO, Smith R, Westman E, Hansson O. 2019. Amyloid and tau
1009 accumulate across distinct spatial networks and are differentially associated with brain connectivity. *eLife*
1010 **8**:e50830. doi:[10.7554/eLife.50830](https://doi.org/10.7554/eLife.50830)
- 1011 Perusini JN, Cajigas SA, Cohensedgh O, Lim SC, Pavlova IP, Donaldson ZR, Denny CA. 2017. Optogenetic
1012 stimulation of dentate gyrus engrams restores memory in Alzheimer’s disease mice. *Hippocampus* **27**:1110–
1013 1122. doi:[10.1002/hipo.22756](https://doi.org/10.1002/hipo.22756)
- 1014 Petersen RC. 2011. Mild Cognitive Impairment. *New England Journal of Medicine* **364**:2227–2234.
1015 doi:[10.1056/NEJMc0910237](https://doi.org/10.1056/NEJMc0910237)
- 1016 Petrache AL, Rajulawalla A, Shi A, Wetzel A, Saito T, Saido TC, Harvey K, Ali AB. 2019. Aberrant Excitatory–
1017 Inhibitory Synaptic Mechanisms in Entorhinal Cortex Microcircuits During the Pathogenesis of Alzheimer’s
1018 Disease. *Cerebral Cortex* **29**:1834–1850. doi:[10.1093/cercor/bhz016](https://doi.org/10.1093/cercor/bhz016)
- 1019 Pusil S, López ME, Cuesta P, Bruña R, Pereda E, Maestú F. 2019. Hypersynchronization in mild cognitive
1020 impairment: the ‘X’ model. *Brain* **142**:3936–3950. doi:[10.1093/brain/awz320](https://doi.org/10.1093/brain/awz320)
- 1021 Qin Y-Y, Li M-W, Zhang S, Zhang Y, Zhao L-Y, Lei H, Oishi K, Zhu W-Z. 2013. In vivo quantitative whole-brain
1022 diffusion tensor imaging analysis of APP/PS1 transgenic mice using voxel-based and atlas-based methods.
1023 *Neuroradiology* **55**:1027–1038. doi:[10.1007/s00234-013-1195-0](https://doi.org/10.1007/s00234-013-1195-0)
- 1024 Reagh ZM, Noche JA, Tustison NJ, Delisle D, Murray EA, Yassa MA. 2018. Functional Imbalance of Anterolateral
1025 Entorhinal Cortex and Hippocampal Dentate/CA3 Underlies Age-Related Object Pattern Separation Deficits.
1026 *Neuron* **97**:1187–1198.e4. doi:[10.1016/j.neuron.2018.01.039](https://doi.org/10.1016/j.neuron.2018.01.039)
- 1027 Reagh ZM, Yassa MA. 2014. Object and spatial mnemonic interference differentially engage lateral and medial
1028 entorhinal cortex in humans. *Proceedings of the National Academy of Sciences* **111**:E4264–E4273.
1029 doi:[10.1073/pnas.1411250111](https://doi.org/10.1073/pnas.1411250111)
- 1030 Rentz DM, Amariglio RE, Becker JA, Frey M, Olson LE, Frishe K, Carmasin J, Maye JE, Johnson KA, Sperling
1031 RA. 2011. Face-name associative memory performance is related to amyloid burden in normal elderly.
1032 *Neuropsychologia* **49**:2776–2783. doi:[10.1016/j.neuropsychologia.2011.06.006](https://doi.org/10.1016/j.neuropsychologia.2011.06.006)
- 1033 Ritchey M, Cooper RA. 2020. Deconstructing the Posterior Medial Episodic Network. *Trends in Cognitive Sciences*
1034 **24**:451–465. doi:[10.1016/j.tics.2020.03.006](https://doi.org/10.1016/j.tics.2020.03.006)
- 1035 Rodriguez GA, Barrett GM, Duff KE, Hussaini SA. 2020. Chemogenetic attenuation of neuronal activity in the
1036 entorhinal cortex reduces A β and tau pathology in the hippocampus. *PLOS Biology* **18**:e3000851.
1037 doi:[10.1371/journal.pbio.3000851](https://doi.org/10.1371/journal.pbio.3000851)
- 1038 Roy DS, Arons A, Mitchell TI, Pignatelli M, Ryan TJ, Tonegawa S. 2016. Memory retrieval by activating engram
1039 cells in mouse models of early Alzheimer’s disease. *Nature* **531**:508–512. doi:[10.1038/nature17172](https://doi.org/10.1038/nature17172)

- 1040 Rubinov M, Sporns O. 2010. Complex network measures of brain connectivity: Uses and interpretations.
1041 *NeuroImage*, Computational Models of the Brain **52**:1059–1069. doi:[10.1016/j.neuroimage.2009.10.003](https://doi.org/10.1016/j.neuroimage.2009.10.003)
- 1042 Saito T, Matsuba Y, Mihira N, Takano J, Nilsson P, Itohara S, Iwata N, Saido TC. 2014. Single App knock-in
1043 mouse models of Alzheimer’s disease. *Nat Neurosci* **17**:661–663. doi:[10.1038/nn.3697](https://doi.org/10.1038/nn.3697)
- 1044 Saito T, Mihira N, Matsuba Y, Sasaguri H, Hashimoto S, Narasimhan S, Zhang B, Murayama S, Higuchi M, Lee
1045 VMY, Trojanowski JQ, Saido TC. 2019. Humanization of the entire murine *Mapt* gene provides a murine
1046 model of pathological human tau propagation. *J Biol Chem* jbc.RA119.009487.
1047 doi:[10.1074/jbc.RA119.009487](https://doi.org/10.1074/jbc.RA119.009487)
- 1048 Sanabria A, Alegret M, Rodriguez-Gomez O, Valero S, Sotolongo-Grau O, Monté-Rubio G, Abdelnour C, Espinosa
1049 A, Ortega G, Perez-Cordon A, Gailhajianet A, Hernandez I, Rosende-Roca M, Vargas L, Mauleon A, Sanchez
1050 D, Martin E, Rentz DM, Lomeña F, Ruiz A, Tarraga L, Boada M. 2018. The Spanish version of Face-Name
1051 Associative Memory Exam (S-FNAME) performance is related to amyloid burden in Subjective Cognitive
1052 Decline. *Scientific Reports* **8**:3828. doi:[10.1038/s41598-018-21644-y](https://doi.org/10.1038/s41598-018-21644-y)
- 1053 Sasaguri H, Nilsson P, Hashimoto S, Nagata K, Saito T, Strooper BD, Hardy J, Vassar R, Winblad B, Saido TC.
1054 2017. APP mouse models for Alzheimer’s disease preclinical studies. *The EMBO Journal* **36**:2473–2487.
1055 doi:[10.15252/embj.201797397](https://doi.org/10.15252/embj.201797397)
- 1056 Searce-Levie K, Sanchez PE, Lewcock JW. 2020. Leveraging preclinical models for the development of Alzheimer
1057 disease therapeutics. *Nature Reviews Drug Discovery* **19**:447–462. doi:[10.1038/s41573-020-0065-9](https://doi.org/10.1038/s41573-020-0065-9)
- 1058 Schultz AP, Chhatwal JP, Hedden T, Mormino EC, Hanseeuw BJ, Sepulcre J, Huijbers W, LaPoint M, Buckley RF,
1059 Johnson KA, Sperling RA. 2017. Phases of hyper and hypo connectivity in the Default Mode and Salience
1060 networks track with amyloid and Tau in clinically normal individuals. *J Neurosci*.
1061 doi:[10.1523/JNEUROSCI.3263-16.2017](https://doi.org/10.1523/JNEUROSCI.3263-16.2017)
- 1062 Selkoe DJ. 2012. Preventing Alzheimer’s Disease. *Science* **337**:1488–1492. doi:[10.1126/science.1228541](https://doi.org/10.1126/science.1228541)
- 1063 Sepulcre J, Sabuncu MR, Li Q, El Fakhri G, Sperling R, Johnson KA. 2017. Tau and amyloid β proteins
1064 distinctively associate to functional network changes in the aging brain. *Alzheimer’s & Dementia* **13**:1261–
1065 1269. doi:[10.1016/j.jalz.2017.02.011](https://doi.org/10.1016/j.jalz.2017.02.011)
- 1066 Shah D, Latif-Hernandez A, Strooper B, Saito T, Saido T, Verhoye M, D’Hooge R, Linden A. 2018. Spatial reversal
1067 learning defect coincides with hypersynchronous telencephalic BOLD functional connectivity in APP NL-
1068 F/NL-F knock-in mice. *Scientific Reports* **8**:6264. doi:[10.1038/s41598-018-24657-9](https://doi.org/10.1038/s41598-018-24657-9)
- 1069 Shah D, Praet J, Latif Hernandez A, Höfling C, Anckaerts C, Bard F, Morawski M, Detrez JR, Prinsen E, Villa A,
1070 De Vos WH, Maggi A, D’Hooge R, Balschun D, Rossner S, Verhoye M, Van der Linden A. 2016. Early
1071 pathologic amyloid induces hypersynchrony of BOLD resting-state networks in transgenic mice and provides
1072 an early therapeutic window before amyloid plaque deposition. *Alzheimer’s & Dementia* **12**:964–976.
1073 doi:[10.1016/j.jalz.2016.03.010](https://doi.org/10.1016/j.jalz.2016.03.010)
- 1074 Sinha N, Berg CN, Tustison NJ, Shaw A, Hill D, Yassa MA, Gluck MA. 2018. APOE ϵ 4 Status in Healthy Older
1075 African Americans is Associated with Deficits in Pattern Separation and Hippocampal Hyperactivation.
1076 *Neurobiol Aging* **69**:221–229. doi:[10.1016/j.neurobiolaging.2018.05.023](https://doi.org/10.1016/j.neurobiolaging.2018.05.023)
- 1077 Stafford JM, Jarrett BR, Miranda-Dominguez O, Mills BD, Cain N, Mihalas S, Lahvis GP, Lattal KM, Mitchell SH,
1078 David SV, Fryer JD, Nigg JT, Fair DA. 2014. Large-scale topology and the default mode network in the
1079 mouse connectome. *PNAS* **111**:18745–18750. doi:[10.1073/pnas.1404346111](https://doi.org/10.1073/pnas.1404346111)
- 1080 Sugar J, Witter MP, van Strien N, Cappaert N. 2011. The Retrosplenial Cortex: Intrinsic Connectivity and
1081 Connections with the (Para)Hippocampal Region in the Rat. An Interactive Connectome. *Front Neuroinform*
1082 **5**. doi:[10.3389/fninf.2011.00007](https://doi.org/10.3389/fninf.2011.00007)

- 1083 Tanimizu T, Kenney JW, Okano E, Kadoma K, Frankland PW, Kida S. 2017. Functional Connectivity of Multiple
1084 Brain Regions Required for the Consolidation of Social Recognition Memory. *J Neurosci* **37**:4103–4116.
1085 doi:[10.1523/JNEUROSCI.3451-16.2017](https://doi.org/10.1523/JNEUROSCI.3451-16.2017)
- 1086 Tischmeyer W, Grimm R. 1999. Activation of immediate early genes and memory formation. *Cell Mol Life Sci*
1087 **55**:564–574. doi:[10.1007/s000180050315](https://doi.org/10.1007/s000180050315)
- 1088 Vann SD, Aggleton JP. 2005. Selective dysgranular retrosplenial cortex lesions in rats disrupt allocentric
1089 performance of the radial-arm maze task. *Behav Neurosci* **119**:1682–1686. doi:[10.1037/0735-7044.119.6.1682](https://doi.org/10.1037/0735-7044.119.6.1682)
- 1090 Vann SD, Aggleton JP, Maguire EA. 2009. What does the retrosplenial cortex do? *Nature Reviews Neuroscience*
1091 **10**:792–802. doi:[10.1038/nrn2733](https://doi.org/10.1038/nrn2733)
- 1092 Vetere G, Kenney JW, Tran LM, Xia F, Steadman PE, Parkinson J, Josselyn SA, Frankland PW. 2017.
1093 Chemogenetic Interrogation of a Brain-wide Fear Memory Network in Mice. *Neuron* **94**:363-374.e4.
1094 doi:[10.1016/j.neuron.2017.03.037](https://doi.org/10.1016/j.neuron.2017.03.037)
- 1095 Vogt BA, Paxinos G. 2014. Cytoarchitecture of mouse and rat cingulate cortex with human homologies. *Brain*
1096 *Struct Funct* **219**:185–192. doi:[10.1007/s00429-012-0493-3](https://doi.org/10.1007/s00429-012-0493-3)
- 1097 Wang J, Zuo X, Dai Z, Xia M, Zhao Z, Zhao X, Jia J, Han Y, He Y. 2013a. Disrupted Functional Brain Connectome
1098 in Individuals at Risk for Alzheimer’s Disease. *Biological Psychiatry*, Disturbances in the Connectome and
1099 Risk for Alzheimer’s Disease **73**:472–481. doi:[10.1016/j.biopsych.2012.03.026](https://doi.org/10.1016/j.biopsych.2012.03.026)
- 1100 Wang JX, Rogers LM, Gross EZ, Ryals AJ, Dokucu ME, Brandstatt KL, Hermiller MS, Voss JL. 2014. Targeted
1101 enhancement of cortical-hippocampal brain networks and associative memory. *Science* **345**:1054–1057.
1102 doi:[10.1126/science.1252900](https://doi.org/10.1126/science.1252900)
- 1103 Wang L, Brier MR, Snyder AZ, Thomas JB, Fagan AM, Xiong C, Benzinger TL, Holtzman DM, Morris JC, Ances
1104 BM. 2013b. Cerebrospinal Fluid A β 42, Phosphorylated Tau₁₈₁, and Resting-State Functional Connectivity.
1105 *JAMA Neurol*. doi:[10.1001/jamaneurol.2013.3253](https://doi.org/10.1001/jamaneurol.2013.3253)
- 1106 Wang Q, Ng L, Harris JA, Feng D, Li Y, Royall JJ, Oh SW, Bernard A, Sunkin SM, Koch C, Zeng H. 2017.
1107 Organization of the connections between claustrum and cortex in the mouse. *J Comp Neurol* **525**:1317–1346.
1108 doi:[10.1002/cne.24047](https://doi.org/10.1002/cne.24047)
- 1109 Wang Y, Risacher SL, West JD, McDonald BC, Magee TR, Farlow MR, Gao S, O’Neill DP, Saykin AJ. 2013c.
1110 Altered default mode network connectivity in older adults with cognitive complaints and amnesic mild
1111 cognitive impairment. *J Alzheimers Dis* **35**:751–760. doi:[10.3233/JAD-130080](https://doi.org/10.3233/JAD-130080)
- 1112 Warren KN, Hermiller MS, Nilakantan AS, Voss JL. 2019. Stimulating the hippocampal posterior-medial network
1113 enhances task-dependent connectivity and memory. *eLife* **8**:e49458. doi:[10.7554/eLife.49458](https://doi.org/10.7554/eLife.49458)
- 1114 Weiler M, Stieger KC, Long JM, Rapp PR. 2020. Transcranial Magnetic Stimulation in Alzheimer’s Disease: Are
1115 We Ready? *eNeuro* **7**. doi:[10.1523/ENEURO.0235-19.2019](https://doi.org/10.1523/ENEURO.0235-19.2019)
- 1116 Wheeler AL, Teixeira CM, Wang AH, Xiong X, Kovacevic N, Lerch JP, McIntosh AR, Parkinson J, Frankland PW.
1117 2013. Identification of a Functional Connectome for Long-Term Fear Memory in Mice. *PLOS Computational*
1118 *Biology* **9**:e1002853. doi:[10.1371/journal.pcbi.1002853](https://doi.org/10.1371/journal.pcbi.1002853)
- 1119 Wilson DIG, Langston RF, Schlesinger MI, Wagner M, Watanabe S, Ainge JA. 2013a. Lateral entorhinal cortex is
1120 critical for novel object-context recognition. *Hippocampus* **23**:352–366. doi:[10.1002/hipo.22095](https://doi.org/10.1002/hipo.22095)
- 1121 Wilson DIG, Watanabe S, Milner H, Ainge JA. 2013b. Lateral entorhinal cortex is necessary for associative but not
1122 nonassociative recognition memory. *Hippocampus* **23**:1280–1290. doi:[10.1002/hipo.22165](https://doi.org/10.1002/hipo.22165)

- 1123 Wright DB, London K, Field AP. 2011. Using Bootstrap Estimation and the Plug-in Principle for Clinical
1124 Psychology Data. *Journal of Experimental Psychopathology* **2**:252–270. doi:[10.5127/jep.013611](https://doi.org/10.5127/jep.013611)
- 1125 Xu W, Fitzgerald S, Nixon RA, Levy E, Wilson DA. 2015. Early hyperactivity in lateral entorhinal cortex is
1126 associated with elevated levels of A β PP metabolites in the Tg2576 mouse model of Alzheimer’s disease. *Exp*
1127 *Neurol* **264**:82–91. doi:[10.1016/j.expneurol.2014.12.008](https://doi.org/10.1016/j.expneurol.2014.12.008)
- 1128 Yassa MA, Stark SM, Bakker A, Albert MS, Gallagher M, Stark CEL. 2010. High-resolution structural and
1129 functional MRI of hippocampal CA3 and dentate gyrus in patients with amnesic Mild Cognitive Impairment.
1130 *NeuroImage* **51**:1242–1252. doi:[10.1016/j.neuroimage.2010.03.040](https://doi.org/10.1016/j.neuroimage.2010.03.040)
- 1131 Yeung L-K, Olsen RK, Hong B, Mihajlovic V, D’Angelo MC, Kacollja A, Ryan JD, Barense MD. 2018. Object-in-
1132 Place Memory Predicted by Anterolateral Entorhinal Cortex and Parahippocampal Cortex Volume in Older
1133 Adults. *bioRxiv* 409607. doi:[10.1101/409607](https://doi.org/10.1101/409607)
- 1134 Zott B, Simon MM, Hong W, Unger F, Chen-Engerer H-J, Frosch MP, Sakmann B, Walsh DM, Konnerth A. 2019.
1135 A vicious cycle of β amyloid–dependent neuronal hyperactivation. *Science* **365**:559–565.
1136 doi:[10.1126/science.aay0198](https://doi.org/10.1126/science.aay0198)
- 1137
- 1138
- 1139
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