Epigenetic mechanisms in cognitive impairment linked to aging and Alzheimer's disease

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Neuropharmacology in Aging and Neurodegeneration
Faculty of Pharmacy and Food Sciences
May 14th, 2019
Alzheimer’s Disease

- The most common cause of dementia
- Neurodegenerative disease
- Progressive and irreversible
- Cognitive impairment and behavioural abnormalities
- Neuropathological alterations: β-amyloid, Tau hyperphosphorylation, neurotransmitter deficits and cell death.

Plaques

Neurofibrillary Tangles

Brain Atrophy in Advanced Alzheimer’s Disease
### Characteristics

<table>
<thead>
<tr>
<th>Characteristics</th>
<th>DONEPEZIL</th>
<th>RIVASTIGMINE</th>
<th>GALANTAMINE</th>
<th>MEMANTINE</th>
</tr>
</thead>
<tbody>
<tr>
<td><strong>Chemical class</strong></td>
<td>piperidine</td>
<td>carbamate</td>
<td>phenantherinealkaloid</td>
<td>Similar to amantadine</td>
</tr>
<tr>
<td><strong>Primary mechanism</strong></td>
<td>AchE inh</td>
<td>AchE inh</td>
<td>AchE inh</td>
<td>NMDA antagonist</td>
</tr>
<tr>
<td><strong>Other mechanism</strong></td>
<td>None</td>
<td>None</td>
<td>Nicotine modulator</td>
<td>HT3 receptor antagonist</td>
</tr>
<tr>
<td><strong>Half life</strong></td>
<td>70 h</td>
<td>90 min</td>
<td>7 h</td>
<td>70 h</td>
</tr>
</tbody>
</table>

**CURRENT PHARMACOLOGICAL THERAPIES DO NOT STOP THE PROGRESSION OF DEMENTIA**
Beta-amyloid remains a major target in all phases of clinical trials, despite high-profile failures in the past few years.
The greatest risk factor for AD is advanced age.
The hallmarks of aging

The hallmarks of aging, López-Otín et al., Cell 2013
What is epigenetics?

- Changes in gene expression or phenotype that don’t involve changes to the DNA sequence
- Its defined as heritable changes in gene activity and expression that occur without alteration in DNA sequence

Conrad Hal Waddington (1905-1975)
Developmental biologist

Epigenetic landscape 1940
The three pillars of epigenetic regulations

http://journals.plos.org/plospathogens/article?id=10.1371/journal.ppat.1003007
Alterations of these epigenetic mechanisms affect the vast majority of nuclear process, including:

- Gene transcription and silencing
- DNA replication and repair
- Cell cycle progression
- Telomere structure and function
- Mitochondrial function
- Inflammation
- Oxidative Stress
- Cell survival

Epigenetic and life
The human brain expresses numerous genes; approximately 80–95%
Neuronal activity *per se* modifies DNA methylation and histone modifications patterns, and further, that learning and memory depend on these epigenetic changes.

Epigenetic mechanisms in aged brain and AD
Epigenetic mechanisms in aged brain and AD

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**Review**

Epigenetic mechanisms in Alzheimer's disease: Implications for pathogenesis and therapy

Jun Wang a, Jin-Tai Yu a, b, c, *, Meng-Shan Tan b, Teng Jiang c, Lan Tan a, b, c, *

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Epigenetics in neurodegeneration: A new layer of complexity

Sueli C.F. Marques a, b, c, *, Catarina R. Oliveira c, d, Claudia M.F. Pereira c, d, Tiago F. Outeiro b, c, *

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Epigenetic regulation in the pathophysiology of Alzheimer's disease

Leonidas Choulamas a, b, c, *, Bart P.F. Rutten a, b, c, *, Gunter Kenis a, * Odette Peerboom a, Pieter Jelle Visser a, b, *, Frans Verhey a, Jim van Os a, b, *, Harry W.M. Steinbusch a, Daniel L.A. van den Hove a, b, c, *

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Epigenetically regulated microRNAs in Alzheimer's disease

Daniel L. Van den Hove a, b, c, *, Konstantinos Kompotis a, Roy Lardenoije a, Gunter Kenis a, Jonathan Mill c, d, Harry W. Steinbusch a, Klaus-Peter Lesch a, b, *, Carlos P. Fitzsimons a, *, Bart De Strooper a, b, *, Bart P.F. Rutten a, b, c, *
Epigenetic mechanisms in aged brain and AD

Related Aberrant Epigenetic Mark and Regulators | Epigenetic Alteration | References
--- | --- | ---
DNA methylation | Global levels of 5-mC and 5-hmC ↓ | [233]
Histone modifications | H3K24, H3K27, H3K36, H3K79, H3R128, H4K20 and H2AR89 ↓ Global acetylation levels of H3 and H4 ↑ | [242]
miRNAs | miR-16, miR-9 and miR-139 ↓ | [246, 247, 249]
DNTMs and HDACs | Dnmt3b ↑ Sirt1, Hdac5 and Hdac6 ↓ | [233, 25]

Griñán-Ferré et al. J Alzheimer Dis. 2018
The aetiology of Alzheimer's disease is multifactorial.

It is produced by a combination of genetic susceptibility factors added to exposure to environmental factors, where risk factors and protective factors interact, in a prolonged temporary sale that includes the aging process and with a different effect on each individual.
New Clinical Trial in Phase I for AD

0 Clinical Trials for Epigenetic therapies

More than 5 Clinical Trials for Epigenetic therapies

Collaboration

Vafidemstat (ORY-2001)
Phase Ia
SATEEN (EM)
ETHERAL (AD)
REIMAGINE (AB)
Dr. Takeda 1968

AKR/J strain

SAM resistant (SAMR) mice
Median survival 16.3 months

SAM prone (SAMP) mice
Median survival 9.7 months
The SAMP8 Mouse Model

### Table 1: Pathobiological phenotypes

<table>
<thead>
<tr>
<th>Strains</th>
<th>Phenotypes</th>
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</thead>
<tbody>
<tr>
<td>SAMP1</td>
<td>Senile amyloidosis, contracted kidney, impaired immune response, hyperinflation of lungs, hearing impairment</td>
</tr>
<tr>
<td>SAMP1 TA</td>
<td>Deficits in learning and memory</td>
</tr>
<tr>
<td>SAMP2</td>
<td>Senile and secondary amyloidosis, contracted kidney, impaired immune response</td>
</tr>
<tr>
<td>SAMP3</td>
<td>Degenerative temporomandibular joint disease</td>
</tr>
<tr>
<td>SAMP6</td>
<td>Senile osteoporosis, secondary amyloidosis</td>
</tr>
<tr>
<td>SAMP8/Ta</td>
<td>Deficits in learning and memory, impaired immune response</td>
</tr>
<tr>
<td>SAMP9</td>
<td>Cataract, thymic lymphoblastic lymphoma, senile amyloidosis</td>
</tr>
<tr>
<td>SAMP10</td>
<td>Brain atrophy, deficits in learning and memory</td>
</tr>
<tr>
<td>SAMP10/Ta</td>
<td>Deficits in learning and memory, emotional disorder (depressive behavior)</td>
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<tr>
<td>SAMP11</td>
<td>Contracted kidney, senile amyloidosis</td>
</tr>
<tr>
<td>SAMR</td>
<td>Nonthymic lymphoma*, histiocytic sarcoma, ovarian cyst</td>
</tr>
<tr>
<td>SAMR1TA</td>
<td>Nonthymic lymphoma*, histiocytic sarcoma, ovarian cyst</td>
</tr>
<tr>
<td>SAMR4</td>
<td>Nonthymic lymphoma*, histiocytic sarcoma</td>
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<tr>
<td>SAMR5</td>
<td>Colitis</td>
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</table>

### Table 2: Histopathological and cellular markers of AD and aging found in SAMP8.

<table>
<thead>
<tr>
<th>Target</th>
<th>Modification</th>
<th>References</th>
</tr>
</thead>
<tbody>
<tr>
<td>Tau protein</td>
<td>Increased levels and phosphorylation</td>
<td>[22, 55, 56]</td>
</tr>
<tr>
<td>Tau kinases</td>
<td>Increased levels or activity in Cdk5/p25 and GSK3δ</td>
<td>[22, 58, 59]</td>
</tr>
<tr>
<td>Receptor for advanced glycation end product (RAGE)</td>
<td>Increased</td>
<td>[75]</td>
</tr>
<tr>
<td>APP protein</td>
<td>Increased</td>
<td>[63–65, 80, 85, 86]</td>
</tr>
<tr>
<td><strong>B-amylloid</strong></td>
<td>Amyloid content increased and aggregates</td>
<td>[26, 61, 62, 69, 70, 74, 87]</td>
</tr>
<tr>
<td>Secretases</td>
<td>ADAM-10 and PS1</td>
<td>[65, 80, 88]</td>
</tr>
<tr>
<td>BBH</td>
<td>Disrupted</td>
<td>[72, 73, 89]</td>
</tr>
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</table>

### Table 3: Molecular and cellular pathways altered in SAMP8.

<table>
<thead>
<tr>
<th>Pathway/protein target</th>
<th>Modification</th>
<th>Reference</th>
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</thead>
<tbody>
<tr>
<td>Sirtuin 1</td>
<td>Decreased</td>
<td>[20, 58, 97]</td>
</tr>
<tr>
<td>Autophagic process</td>
<td>Reduced</td>
<td>[117–119]</td>
</tr>
<tr>
<td>Cathepsin system</td>
<td>Inactivated</td>
<td>[117]</td>
</tr>
<tr>
<td>AMPK</td>
<td>Not determined</td>
<td></td>
</tr>
<tr>
<td>mTOR</td>
<td>Not determined</td>
<td></td>
</tr>
<tr>
<td>mtDNA damage</td>
<td>Increased</td>
<td>[125]</td>
</tr>
<tr>
<td>Mitochondrial electron chain</td>
<td>Diminished efficiency</td>
<td>[20, 21, 29, 126, 127]</td>
</tr>
<tr>
<td>Fusion/fission proteins</td>
<td>Not determined</td>
<td></td>
</tr>
<tr>
<td>Mitochondrial fatty acid oxidation</td>
<td>Altered</td>
<td>[128]</td>
</tr>
</tbody>
</table>
CHARACTERISTICS OF AGE-RELATED BEHAVIORAL CHANGES IN SENESCENCE-ACCELERATED MOUSE SAMP8 AND SAMP10

MASAO MIYAMOTO
Pharmaceutical Research Laboratories 1, Pharmaceutical Research Division, Takeda Chemical Industries, Ltd., 2-17-85, Jusohonnachi, Yodogawa-ku, Osaka 532, Japan

Molecular Genetic Characterization of the Senescence—Accelerated Mouse (SAM) Strains
Haruo Kitado, Keiichi Higuchi, Toshio Takeda
Published: 01 November 1994 Article history

GENETIC CHARACTERIZATION OF SENESCENCE-ACCELERATED MOUSE (SAM)

KEIICHI HIGUCHI
Department of Senesence Biology, Chest Disease Research Institute, Kyoto University, Sakyoku, Kyoto 606 Japan

Research
Mechanisms of aging in senescence-accelerated mice

Address: The Salk Institute for Biological Studies, La Jolla, CA 92037, USA. *Department of Ophthalmology and Visual Sciences, University of Michigan, Ann Arbor, MI 48109, USA. **Department of Human Genetics, University of Michigan, Ann Arbor, MI 48109, USA. ***Kambly Biosciences, San Diego CA 92121, USA. Current address: BrainCells Inc., 10835 Road to the Cure, San Diego, CA 92121, USA.

Mini review
The senescence-accelerated prone mouse (SAMP8): A model of age-related cognitive decline with relevance to alterations of the gene expression and protein abnormalities in Alzheimer’s disease
D. Allan Butterfield*, H. Fai Poon

Senescence-Accelerated Mice P8: A Tool to Study Brain Aging and Alzheimer’s Disease in a Mouse Model
Merce Pallás
Neuropathology 2017, 37, 293–305
doi:10.1111/neu.12373

Occasional Review
SAMP8 mice as a neuropathological model of accelerated brain aging and dementia: Toshio Takeda’s legacy and future directions
Ichiro Akiyuchi, Mercè Pallás, Herbert Budka, Haruhiko Akiyama, Masaki Ueno, Jingxian Han, Hideo Yagi, Tomohumi Nishikawa, Yoichi Chiba, Hiroshi Sugiyama, Ryoya Takahashi, Keiko Unno, Keiichi Higuchi and Masanori Hosokawa
Accelerated senescence and epigenetics

**EPIGENETIC CHANGES**

- TETs
- DNMTs
- HDACs

**INFLAMMATION**

- Oxidative Stress

**NEURODEGENERATION**

- Neuronal Loss
- Gliosis

**COGNITION and BEHAVIOUR alterations**
Effect of Environment enrichment (EE) in SAMP8

Beneficial effects of EE in cognition

Novel Object Recognition

- Discrimination Index (DI) for SAMP8 Ct and SAMP8 EE
- Time platform zone for SAMP8 Ct and SAMP8 EE

Morris Water Maze

- Learning curve showing latency to escape platform over training days
- Distance to platform showing significant difference between SAMP8 Ct and SAMP8 EE
Neuroprotective effects of EE in SAMP8

SAMP8 CT

SAMP8 EE

GFAP + HOECHST

48 kDa

H 37 kDa

NeuN

Relative expression

NeuN in CA1

****

SAMP8 Ct 3 months

SAMP8 EE 3 months

NeuN in CA3

****

SAMP8 Ct 3 months

SAMP8 EE 3 months

NeuN in DG

****

SAMP8 Ct 3 months

SAMP8 EE 3 months

Effects of EE on Epigenetics in SAMP8

Accelerated senescence and epigenetics

Environmental enrichment

Epigenetic changes

- TETs
- DNMTs
- HDACs

Inflammation

Oxidative stress

↑ Neuroprotection, ↑ Neurogenesis

↓ Neuronal loss

↑ Cognition
Environmental Enrichment Improves Cognitive Deficits, AD Hallmarks and Epigenetic Alterations Presented in 5xFAD Mouse Model

Christian Griñán-Ferré1, Vanesa Izquierdo1, Eduard Otero1, Dolors Puigoriol-Illamola1, Rubén Corpas2, Coral Sanfeliú2, Daniel Ortuño-Sahagún2 and Mercè Pallás1*
Epigenetics can explain in part the senescent phenotype that characterizes SAMP8.
Temporal Integrative Analysis of mRNA and microRNAs Expression Profiles and Epigenetic Alterations in Female SAMP8, a Model of Age-Related Cognitive Decline

2-month-old 9-month-old

SAMR1 (n=16) SAMP8 (n=16)

Molecular analysis

WB
qPCR mRNA
qPCR miRNA
ELISA
Array mRNA
Array miRNA

Bioinformatic analysis

Seminari de Recerca 2019
Mecanismos epigenètics en el deterioro cognitivo ligat al envejecim i enfermedad de Alzheimer
Dimarts 14 de maig de 2019
12:30 h
Aula A4, Facultat de Farmàcia i Ciències de l’Alimentació

Christian Griñán-Ferré
Sectió de Farmacologia
Dept. Farmacologia, Toxicologia i Química Terapèutica
Facultat de Farmàcia i Ciències de l’Alimentació
A. SAMR1 SAMP8 SAMR1 SAMP8
   Ac-H3 (17kDa)
   TBP (38kDa)
   2 months 9 months

B. SAMR1 SAMP8 SAMR1 SAMP8
   Ac-H4 (17kDa)
   TBP (38kDa)
   2 months 9 months

C. Ac-H3
   % Vs. SAMR1 2 months
   2 months 9 months

D. Ac-H4
   % Vs. SAMR1 2 months
   2 months 9 months

E. Hdac1
   Relative mRNA expression
   2 months 9 months

F. Hdac2
   Relative mRNA expression
   2 months 9 months

G. Sirt1
   Relative mRNA expression
   2 months 9 months

H. Sirt2
   Relative mRNA expression
   2 months 9 months

I. Sirt6
   Relative mRNA expression
   2 months 9 months

Epigenetic Landscape
Epigenetics in Alzheimer's Disease

- Animal post-mortem studies show:

<table>
<thead>
<tr>
<th>Pathology</th>
<th>Effect</th>
</tr>
</thead>
<tbody>
<tr>
<td>Decreased global levels of 5’-mC in brain</td>
<td>Increased Tau phosphorylation</td>
</tr>
<tr>
<td>Decreased level of H3 acetylation in Temporal lobe</td>
<td>Decreased synaptic plasticity leading to decreased learning and memory</td>
</tr>
</tbody>
</table>
mRNA:miRNA integrative analysis

1,062 mRNAs differentially expressed in SAMP8 vs. SAMR1 at 2 months
1,033 mRNAs differentially expressed in SAMP8 vs. SAMR1 at 9 months
92 mRNAs differentially expressed in both 2 and 9 months
(Figure 1 and Suppl. Material 3)

Integrative analysis using R program algorithm
miRNA targets database

187 putative mRNAs:miRNAs pairs in 2-month-old SAMP8
(Table 1 and 2)

Selection of mRNAs:miRNAs pairs associated with aging, neurodegeneration and AD:
3 putative mRNAs:miRNAs pairs in both 2 and 9 months (Table 1 and 2)

28 miRNAs differentially expressed in SAMP8 vs. SAMR1 at 2 months
17 miRNAs differentially expressed in SAMP8 vs. SAMR1 at 9 months
6 miRNAs differentially expressed in both 2 and 9 months
(Figure 4 and Suppl. Material 4)

61 putative mRNAs:miRNAs pairs in 9-month-old SAMP8
(Table 1 and 2)

GO enrichment analysis

Validation of 8 representative mRNAs:miRNAs pairs by RT-PCR
(Figure 7)

Clustering enrichment analysis
(Figure 6)
We found 2,095 mRNA targets altered in SAMP8

We found 37 miRNAs altered in SAMP8
We found 174 mRNA targets altered in SAMP8

We found 37 miRNAs altered in SAMP8
<table>
<thead>
<tr>
<th>MicroRNAs</th>
<th>Target mRNAs</th>
<th>Top 10 GO Biological process</th>
<th>p-value</th>
<th>Z-score</th>
<th>Combined score</th>
</tr>
</thead>
<tbody>
<tr>
<td>mmu-miR-298-5-p</td>
<td>Ads1, Ctnnd1, Comt1, Cyp7b1, F84, Ltf, Mas1p, P2x1, Pcp4, Npnt, Sepp1, Scl17a1, Apt8a1, Gabra1, Maff, Man1a2, Ndst2, Ser2, Skil, Coro1c, Abca1, Atph5v0d1, Tcp, Pdk4, Pbx3, Hp, Pdlim4, Ykt6, Kox17, Mesdc2, Tmem167, Serpt1, Lipt2, Alg12, Loht12c1, Mrp19, Manba, Trps1, Kremen1.</td>
<td>Regulation of membrane lipid distribution (GO: 0097035) Cellar response to extracellular stimulus (GO: 0031668) Regulation of extrinsic apoptotic signaling pathway via death domain receptors (GO: 1902041) Wnt signaling pathway (GO: 0016055) Brain development (GO: 0007420) Neuron-neuron synaptic transmission (GO: 0007270) Insulin receptor signaling pathway (GO: 0008286) Cellular ion homeostasis (GO: 0006873) Positive regulation of neuron projection development (GO: 0010976) Locomotor behavior (GO: 0007626)</td>
<td>0.002603</td>
<td>-2.53</td>
<td>4.08</td>
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<tr>
<td>mmu-miR-151-3p</td>
<td></td>
<td></td>
<td>0.002990</td>
<td>-2.26</td>
<td>3.66</td>
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<td>mmu-miR-148b-3p</td>
<td></td>
<td></td>
<td>0.005549</td>
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<td>3.33</td>
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<tr>
<td>mmu-let-7b-5p</td>
<td></td>
<td></td>
<td>0.01844</td>
<td>-2.26</td>
<td>3.17</td>
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<tr>
<td>mmu-let-7e-5p</td>
<td></td>
<td></td>
<td>0.01021</td>
<td>-2.25</td>
<td>3.16</td>
</tr>
</tbody>
</table>

**2-month-old SAMP8**

<table>
<thead>
<tr>
<th>MicroRNAs</th>
<th>Target mRNAs</th>
<th>Top 10 GO Biological process</th>
<th>p-value</th>
<th>Z-score</th>
<th>Combined score</th>
</tr>
</thead>
<tbody>
<tr>
<td>mmu-miR-92a-3p</td>
<td>Adem10, Bsn, Crimp, Csfr3, Ncan, Cyp361, Elk4, Gabrg2, Gata5, Gif, Sic6a9, H3f3b, Hhx, Hmg20b, Hyal1, Kif5b, Kpn1, Zbp239, NAD1, Pde7a, Pkt3ca, Pml, Pou3f2, Rab23, Ret, Rng, Sema5a, Snc1, Tarp2, Tbx2r, Tgbfr1, Tfrb, Unc5c, Nrsn1, Zbx1, Neurod6, Nrsnt1, Ndog1, Matn3, Ndufs4, Pkt3ca, Ppara, Cytb3, Ptpre, Sncg, Tfeb, Tgfb, Tgf, Mrps12, Chst4, Add2, Crem, Eif4e2, Mmp9, Lam, Pox1, Sh3yf1, Mrs, Rasa1, Sic63a1, Dkk3, Gaens, Dna1c5, If5, Sic6a3, Chst2, Cldn8, Gribb2, Rbms2, Ptp2, Amotl2, Cyp2d22, Stx5a, C1qtnf1, Ptknd, Insmb, Srf, Nup160, Rhot1, Midn, Tsc1, Ctnm3, Tcct1, Srebp1, Fbxo32, Nacc2, Efcab2.</td>
<td>Embryonic morphogenesis (GO: 0048598) Brain development (GO: 0007420) Synapse organization (GO: 0050808) Neuron migration (GO: 0001764) Neurodevelopment (GO: 0048666) Behavior (GO: 0007610) Regulation of synaptic transmission (GO: 0050804) Response to oxygen levels (GO: 0070482) Cellular senescence (GO: 0090398) Inflammatory response (GO: 006954)</td>
<td>0.007922</td>
<td>-2.40</td>
<td>6.34</td>
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<td>mmu-miR-24-3p</td>
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<td>0.05911</td>
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<td>0.005018</td>
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<td>mmu-miR-30a-5p</td>
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<td>0.00583</td>
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<td>mmu-miR-181a-3p</td>
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<td>0.02114</td>
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<tr>
<td>mmu-miR-181d-5p</td>
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<td>0.01385</td>
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<td>mmu-miR-132-3p</td>
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<td>0.02707</td>
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<td>mmu-miR-146a-5p</td>
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<td>0.01575</td>
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<td>mmu-miR-134-5p</td>
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<td></td>
<td>0.01830</td>
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<td>2.55</td>
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<tr>
<td>mmu-miR-29a-3p</td>
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<td></td>
<td>0.02275</td>
<td>-1.81</td>
<td>2.54</td>
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<tr>
<td>miRNAs up:miRNAs down</td>
<td>Target mRNAs</td>
<td>Top 10 GO Biological process</td>
<td></td>
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<tr>
<td>----------------------</td>
<td>------------------------------------------------------------------------------</td>
<td>------------------------------------------------------------------------------------------------</td>
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</tr>
<tr>
<td>mmu-miR-128-3p</td>
<td>Capza2, Cav1, Chuk, Dll3, Drd4, Pdca3, Gcdh, Gria1, H3f3b, Htr1f, Kcnj8,</td>
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<td>mmu-miR-140-5p</td>
<td>Dnaic3, Ctip1, Ilgα5, Itk, Cytb3, Hsp90b1, Hs2stf, Spry4, Map2k7, Fgf9,</td>
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<tr>
<td>mmu-miR-148b-3p</td>
<td>Atpl11a, Mapk6, Slc3a6, Socs6, Thsd1, B3galtn1, Kcna4, Txnip, Vps50, Arl4d,</td>
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<tr>
<td>mmu-miR-342-3p</td>
<td>Cdp1, Mmachi, Ergic1, Chac2, Ctnnbp21n, Gpr63.</td>
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<tr>
<td>mmu-miR-98-5p</td>
<td>Memory (GO: 0007613)</td>
<td>0.001378 -2.23 4.11</td>
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<tr>
<td>mmu-miR-107-3p</td>
<td>Response to tumor necrosis factor (GO: 0034612)</td>
<td>0.002467 -2.31 3.96</td>
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<tr>
<td></td>
<td>Synaptic transmission (GO: 0007268)</td>
<td>0.003559 -2.32 3.88</td>
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<tr>
<td></td>
<td>Activation of innate immune response (GO: 002218)</td>
<td>0.005741 -2.21 3.70</td>
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<tr>
<td></td>
<td>Learning or memory (GO:0007611)</td>
<td>0.01189 -2.20 3.63</td>
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<tr>
<td>mmu-let-7b-5p</td>
<td>Positive regulation of canonical Wnt signaling pathway (GO: 0090263)</td>
<td>0.01703 -2.16 3.57</td>
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<tr>
<td>mmu-let-7c-5p</td>
<td>Cognition (GO: 0050890)</td>
<td>0.009200 -2.11 3.49</td>
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<tr>
<td>mmu-let-7d-5p</td>
<td>Positive regulation of cell cycle G2/M phase transition (GO:1902751)</td>
<td>0.01849 -2.01 3.32</td>
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<tr>
<td>mmu-let-7e-5p</td>
<td>Positive regulation of G2/M transition of mitotic cell cycle (GO:0010971)</td>
<td>0.008912 -2.01 3.32</td>
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<tr>
<td>mmu-let-7e-5p</td>
<td>Positive regulation of mitotic cell cycle (GO:0045931)</td>
<td>0.04411 -1.89 3.11</td>
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</tr>
</tbody>
</table>

9-month-old SAMP8

<table>
<thead>
<tr>
<th>miRNAs down:miRNAs up</th>
<th>Target mRNAs</th>
<th>Top 10 GO Biological process</th>
</tr>
</thead>
<tbody>
<tr>
<td>mmu-let-7b-5p</td>
<td>Ncoa3, Pbx1, Mxd1, Pald1, Trp53r, Klh13.</td>
<td>Positive regulation of cell cycle G2/M phase transition (GO:1902751) 0.004199 -2.74 9.30</td>
</tr>
<tr>
<td>mmu-let-7c-5p</td>
<td>Positive regulation of G2/M transition of mitotic cell cycle (GO:0010971)</td>
<td>0.004199 -2.74 9.29</td>
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<tr>
<td>mmu-let-7d-5p</td>
<td>Positive regulation of mitotic cell cycle (GO:0045931)</td>
<td>0.02884 -2.15 6.24</td>
</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Intracellular steroid hormone receptor signaling pathway (GO:0030518)</td>
<td>0.02059 -2.07 6.09</td>
</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Negative regulation of sequence-specific DNA binding Transcription factor activity (GO:0043433) 0.03514 -2.10 5.95</td>
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</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Histone acetylation (GO:0016573)</td>
<td>0.02829 -2.05 5.93</td>
</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Developmental growth (GO:0048589)</td>
<td>0.04520 -2.16 5.83</td>
</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Negative regulation of neuron differentiation (GO:0045665)</td>
<td>0.04085 -2.15 5.83</td>
</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Positive regulation of cell cycle process (GO:0090068)</td>
<td>0.06054 -2.29 5.74</td>
</tr>
<tr>
<td>mmu-let-7e-5p</td>
<td>Negative regulation of neurogenesis (GO:0050768)</td>
<td>0.05032 -2.14 5.56</td>
</tr>
</tbody>
</table>
Validation of mRNA:miRNA pairs

The selected mRNA:miRNA pairs are involved in brain aging and neurodegeneration.

- **Abnormal pathways**
  - Purinergic signaling *P2rx1*
  - Inflammatory process *Socs6*
  - Neuron differentiation *Pou3f2, Hmg20b*
  - Neurogenesis *Pbx1*
  - Protein trafficking *Nup160*
Validation of mRNA:miRNA pairs

- **BDNF**
  - % Vs. SAMR1
  - SAMR1 SAMP8
  - 9-month-old

- **miR-191**
  - SAMR1 SAMP8
  - 9-month-old

- **BDNF (14kDa)**
  - GAPDH (37kDa)
  - 9-month-old

- **miR-191**
  - SAMR1 SAMP8
  - 9-month-old

- **AD pathogenesis** → **miR-191**
  - BDNF
  - BDNF
  - Synaptic dysfunction
  - Neurodegeneration
  - Memory impairment

**Figure:**
- Western blot images of BDNF and GAPDH.
- Bar graph showing relative miRNA expression.
Validation of mRNA:miRNA pairs

**Graph:**
- **y-axis:** BDNF protein levels (WB)
- **x-axis:** miR-191 expression
- **Equation:** $r = -0.8095, p=0.0218$

**Diagram:**
- **AD pathogenesis** → miR-191
- **BDNF**
- Synaptic dysfunction
- Neurodegeneration
- Memory impairment
- BDNF mRNA
  - $3'UTR$
  - miR-191
Epigenetic mechanisms underlying cognitive impairment and Alzheimer disease hallmarks in 5XFAD mice

Christian Griñán-Ferré¹, Sara Sarroca³, Aleksandra Ivanova¹, Dolors Puigoriol-Ilamola¹, Fernando Aguado², Antoni Camins³, Coral Sanfeliu³, and Mercè Pallàs¹

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Key words: Alzheimer disease, neurodegeneration, behavior, cognition, deacetylase, methyltransferase

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Tg6799 (5xFAD) mice
New Challenges: G9a inhibition for AD

Unpublished results
Proof-of-Concept (PoC)

UNC0642 an *in vivo* BBB penetrant G9a inhibitor IC$_{50} < 2.5$ nM

Inhibition of EHMT1/2 rescues synaptic and cognitive functions for Alzheimer’s disease

Yan Zheng,1,2,8 Aiyi Liu,1,3,8 Zi-Jun Wang,1,4,8 Qing Cao,1 Wei Wang,1 Lin Lin,1 Kaijie Ma,1,4 Freddy Zhang,1 Jing Wei,1,4 Emmanuel Matas,1 Jia Cheng,1 Guo-Jun Chen,1 Xiaomin Wang1 and Zhen Yan1,4

Unpublished results
1. SAMP8 and 5XFAD are a suitable model to study ageing processes, including AD.

2. EE, as a tool to unveil epigenetic influence in senescence process, supported the hypothesis of epigenetic control in ageing in SAMP8.

3. Our data indicate the reciprocal interaction between non-genetic factors and epigenetic mechanisms that can explain the senescence process in the SAMP8.

4. miRNA have a pivotal role in gene regulation in SAMP8.

5. The different pathological process that suffer SAMP8 and 5XFAD, will allow to use them an *in vivo* model for drug discovery in neurosciences, by using a broad number of different targets related with inflammation, oxidative stress, AD hallmarks and epigenetic mechanisms.
Our lifestyle choices affect our risk of developing dementia
Dr Carmen Escolano
Sònia Abás
Sergio Rodríguez

Dr Mercè Pallàs
Dr Christian Griñán-Ferré
Dolors Puigoriol-Illamola, Julia Companys and Fotini Vasilopoulou

Dr Santi Vázquez
Dr Rosana Leiva
Dr Carles Galdeano

Dr. Diego Muñoz-Torrero

Chemistry and Pharmacology of drugs against neurodegenerative diseases (CHEMPHARNEURO)
THANK YOU!

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