PMG-AD: Personalized medicine approach for novel microglia-associated genetic variants in Alzheimer's disease (AD)

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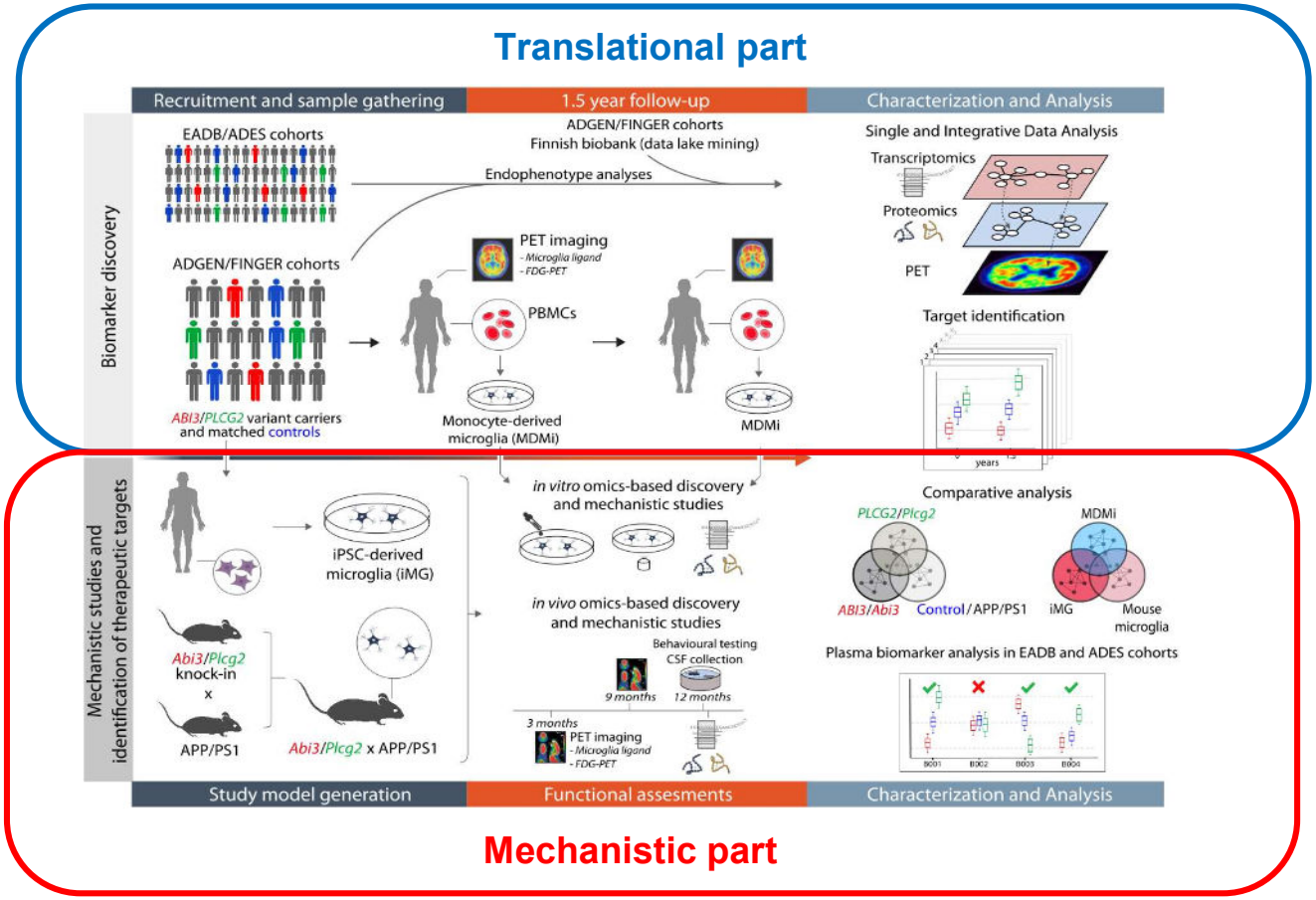
<u>Aim:</u> To establish a personalized medicine-based approach for identification of early biomarkers and therapeutic targets in AD by focusing on AD-associated genetic variants in *ABI3* (S209F) and *PLCG2* (P522R) genes



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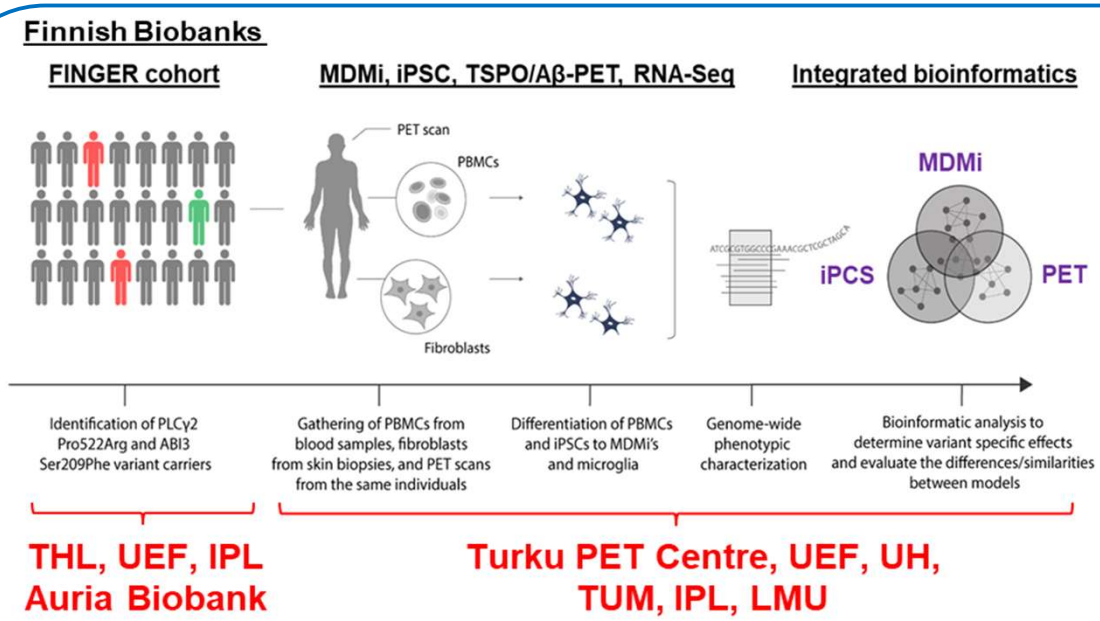
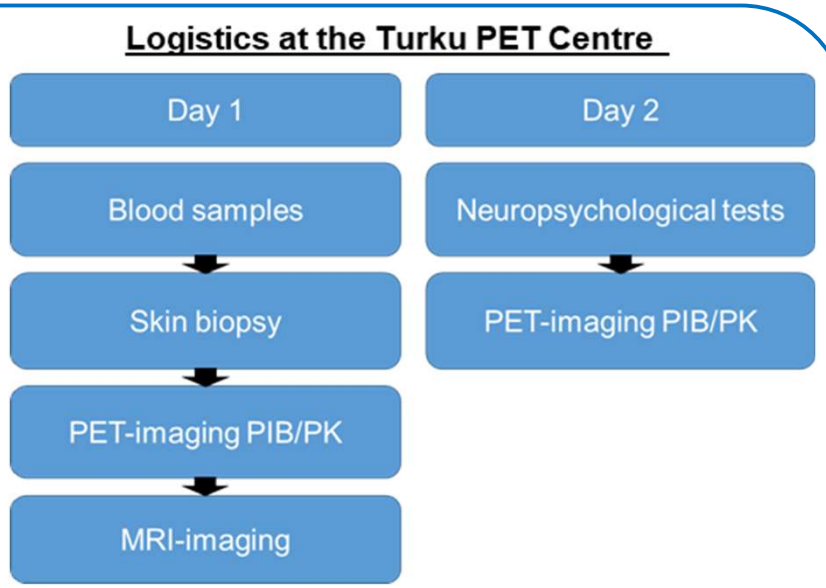
Concepts and applications in the translational part

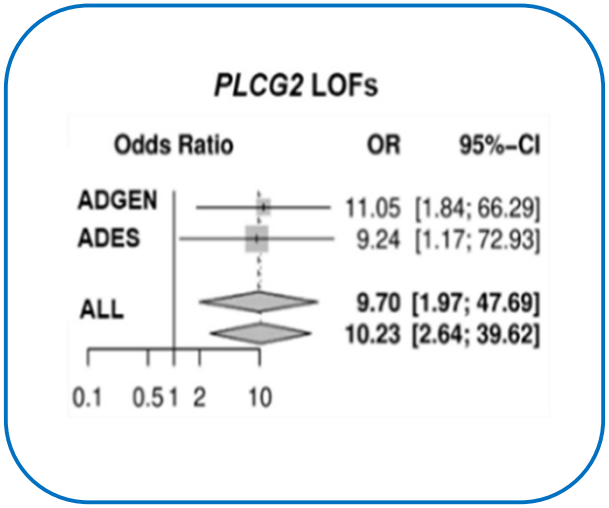
- To build **a multimodal translational research platform** for assessing microglia-specific genetic risk variants in AD
- To evaluate similarities/dissimilarities **between the different human microglial models** to be later applied in the personalized medicine approaches (monocyte-derived microglia-like cells (MDMi) vs. iPSC-derived microglia (iMG))
- To assess the capacity of PET-imaging to **identify individuals in the asymptomatic phase** by focusing on microglia-specific disease-mechanisms

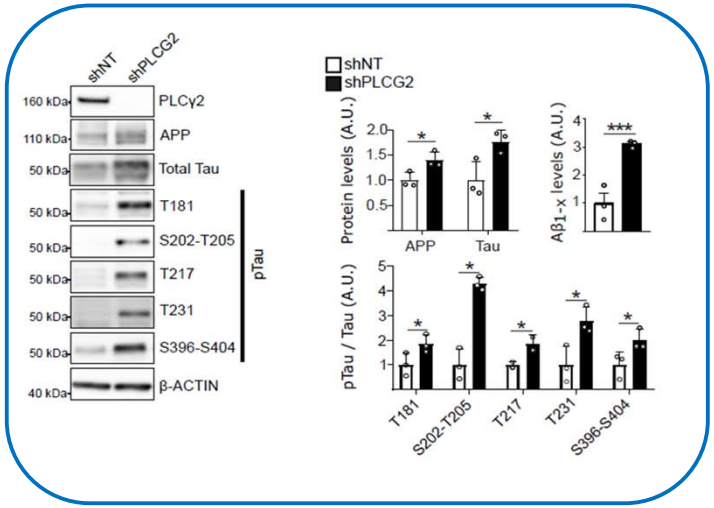




The key outcomes in the translational part

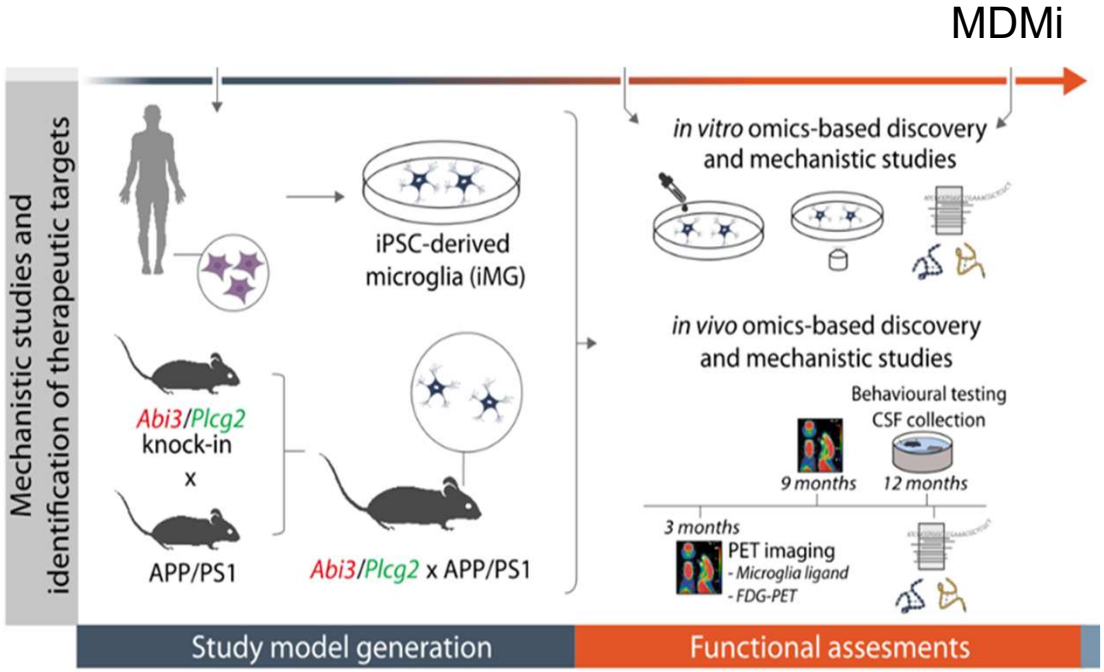
- **Rare loss-of-function (LOF) variants in *PLCG2* significantly increased the risk of AD** in different cohorts (ADGEN and ADES) via nonsense-mediated mRNA decay → 50% reduced levels of PLCγ2 (Coulon et al., in revision)
- **High content screening identified PLCγ2 as a potential strong modulator of synapse density** → Downregulation of PLCγ2 in human neuronal cultures **increases both TAU phosphorylation and the levels of Aβ** (Coulon et al., in revision)
- **Identification of ghrelin as a potential upregulated plasma-based biomarker target in the individuals carrying the protective *PLCG2*-P522R variant in the FINGER cohort** → Plasma analysis of Plcγ2-P522R KI female mice showed **significant increase in the ghrelin levels** (Jeskanen et al., manuscript in preparation)
- MRI and PET ([11C]-(R)-PK11195 and PiB) imaging, and neuropsychological analyses **in asymptomatic individuals with or without *ABI3*-S209F variant did not show significant changes between groups** (*ABI3*-S209F/*APOE*4- *vs.* *ABI3*-S209/*APOE*4-)

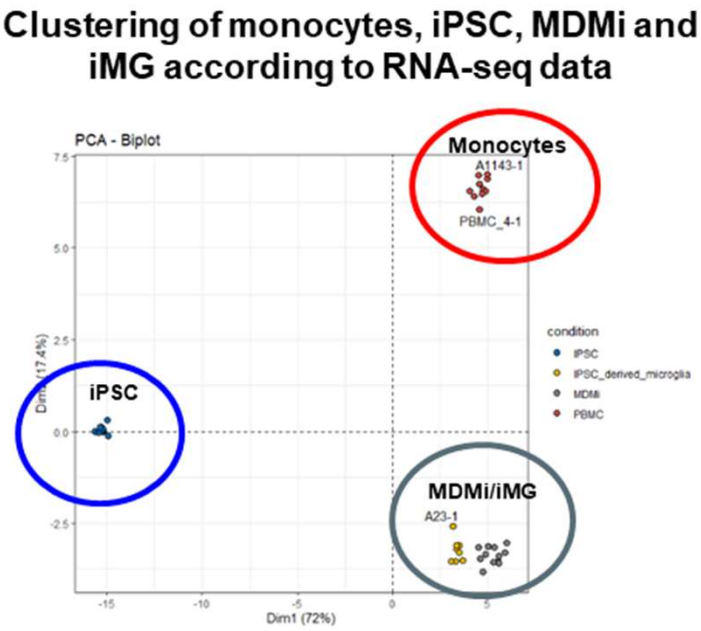




Concepts and applications in the mechanistic part

- To characterize the **microglia-specific mechanisms** of *ABI3* and *PLCG2* variants **in the cellular processes relevant for AD** in *Abi3*-S212F and *Plcg2*-P522R knock-in mouse models crossed with transgenic AD mice (APP/PS1 mice) as well as in human MDMi and iMG cells from genetically defined *ABI3* and *PLCG2* individuals

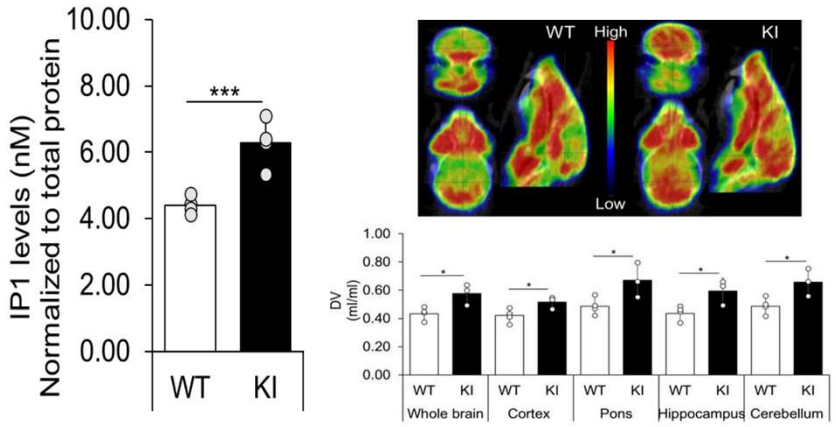




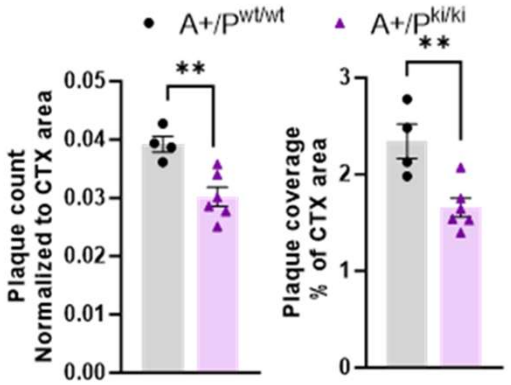
The key outcomes in the mechanistic part

- Macrophages derived from homozygous Plcγ2-P522R KI mice and wild type littermates revealed that the **P522R variant potentiates the primary function of Plcγ2 as a Pip2-metabolizing enzyme** (Takalo et al. 2020)
- Brain mRNA signature together with microglia-related PET imaging **suggested enhanced microglial functions in Plcγ2-P522R KI and Plcγ2-P522R KI x APP/PS1 mice** (Takalo et al. 2020, Takalo et al. bioRxiv 2024.09.12.612207)
- PLCγ2-P522R variant decreased **β-amyloid burden and dystrophic neurites, increased microglia clustering around β-amyloid plaques, and alleviated inflammation** in the brain of APP/PS1 mice (Takalo et al. bioRxiv 2024.09.12.612207)

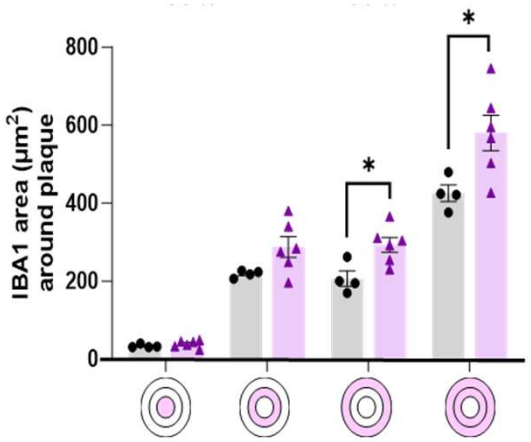
Activity of PLCγ2 and ^{18}F-FEPPA uptake in PET



β-amyloid pathology



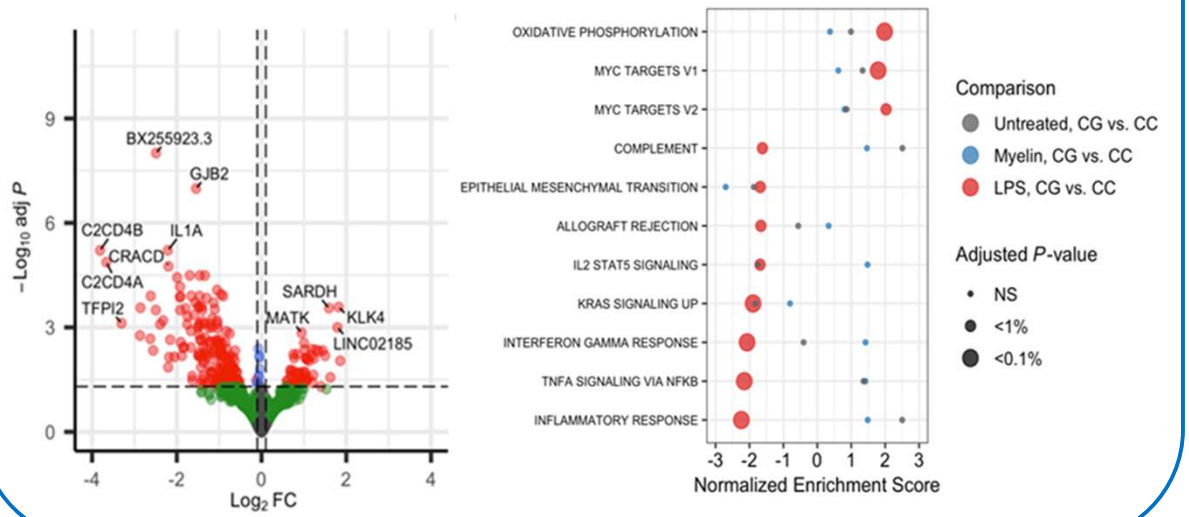
Microglia clustering around β-amyloid plaques



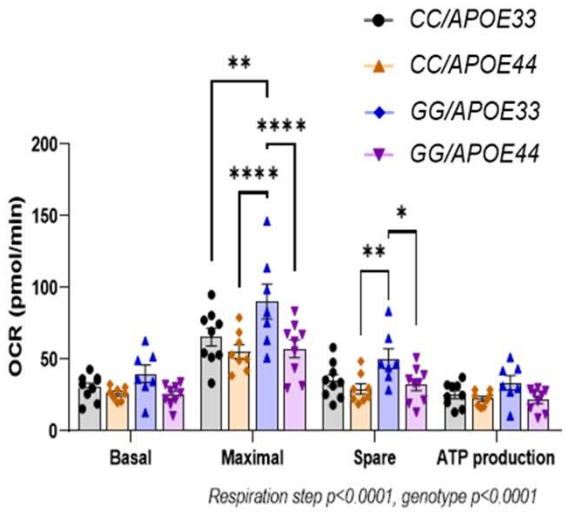
The key outcomes in the mechanistic part

- Myelin-induced changes between *ABI3*-S209F and control iMGs → **Genotype differences in 181 proteins**
- RNA-sequencing findings from the MDMi cells revealed **a significant upregulation of oxidative phosphorylation-related pathway targets**, which coincided with **reduced inflammatory response** due to *PLCG2*-P522R variant
- Assessment of mitochondrial functions **in human iMG revealed a significant improvement in the maximal respiration capacity** owing to *PLCG2*-P522R variant in *APOE*33, but not in *APOE*44 background

DEG/pathways analyses upon induction of inflammation in MDMi cells
***PLCG2*-P522R (CG) vs. *PLCG2*-P522 (CC)**



Mitochondrial respiratory capacity in iMG cells
***PLCG2*-P522R (GG) vs. *PLCG2*-P522 (CC)**



Dissemination, PPI activities and challenges

- PMG-AD organized **regular meetings**, in which the PIs and early-stage researchers (ESRs) presented the progression reports, potential problems and results → **Several PhD theses (3), scientific articles (7), as well as press releases and commentaries related to outcomes in PMG-AD (AlzForum, web-pages, different social media platforms)**

- PMG-AD organized remotely on 8th of June 2021 a one-day **mid-term meeting**, focusing on both **1) scientific progress of the subprojects given by ESRs** and **2) patient and public involvement (PPI) activities** → PMG-AD invited **clinicians** working in the field of neurodegenerative diseases, **medical geneticists** as well as **researchers working on the field of neuro-ethics and law** from the participating countries

- PMG-AD organized **a side event exclusively focusing on PPI activities** related to different **EU-funded JPND projects** during the 9th **Kuopio Alzheimer's disease Symposium** (August 23rd, 2022) → Event included participants from **patient organizations, clinicians, students, patients, lay audience and companies**

- Due to COVID19 lock-down and the subsequent restrictions, recruitments and further analyses of *ABI3*-S209F and *PLCG2*-P522R variants carriers and respective controls **were significantly slower than initially planned** → Translational part was delayed

Tackling the societal challenges and the added value for AD patients

- PMG-AD has determined the **translational applicability** of novel AD-associated microglia-specific risk gene variants by assessing the effects of *PLCG2* P522R and *ABI3* S209F on microglial functions and activity → **Novel variant-specific mechanisms and biomarker targets were discovered**

- PMG-AD has set up a **translational research platform** for assessing microglia-specific genetic risk variants in AD → **Scientific renewal by promoting movement away from the neuron-centric view in aiming to understand molecular mechanisms in AD**

- PMG-AD has significantly promoted **international collaboration between researchers, clinicians and patient organizations as well as established best practices in the personalized medicine by combining basic research, clinics and biobanks for the benefit of AD patients**

- **Expertise and translational research concepts developed during PMG-AD** have been further applied in the **Brain Research Unit, Kuopio, Finland** to study asymptomatic individuals with specific genetic profiles retrieved from Finnish biobanks → **Platform is available for academic partners and companies**