many of which lead to reduced lifespan and childhood deaths. Experiments with partial PolG knock-out rat models have shown symptoms of premature aging.

The goal of our study was to screen a sample of healthy population for mutations in the exonuclease domain of the *POLG* gene to determine the prevalence of *POLG* polymorphisms in the general population. We sequenced the exonuclease domain of the *POLG* gene of 165 healthy individuals from Latvia in three age groups (20–45 y.o., 65–75. y.o. and over 85 y.o.) with no know history of neural or muscular pathologies. We also tested for heteroplasmy in the HVS I region of the mitochondrial DNA to compare the impact on mtDNA of different *POLG* polymorphisms, and determined the participants mitochondrial haplogroups.

Only one sample contained a polymorphism in a heterozygous state, corresponding to the missense mutation G268A in PolG, associated with progressive external ophthalmoplegia.

The lack of polymorphisms in PolG could be related to its important role in human cell function. Further research is required to screen the other domains of the *POLG* gene.

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LB-016
Inverse-agonistic mechanisms of thioridazine on Gi protein-coupled D_{2L} dopamine receptors

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G-protein-coupled receptors (GPCR) are the largest superfamily of the signaling molecules. The D_{2L} receptor ($D_{2L}R$) is a subclass of D2-type dopaminergic GPCR and α-subunits of their Gi proteins (Gαi) inhibit the adenylcyclase (AC) enzyme nearby. Overexpression of Gi protein-coupled $D_{2L}R$ in the mesocorticolimbic area of the brain causes schizophrenia although the downregulation or dysfunction of them in the Corpus Striatum area of the brain causes the Parkinson Syndrome through the inhibition of AC enzymes. It causes the inhibition of striatopallidal neurons, whose key role is to stop locomotor behaviors.

The activation mechanisms of Gi protein-coupled receptors by inverse-agonists are still poorly understood despite extensive work in the field. Thioridazine is an antipsychotic (or neuroleptic) drug, which is specifically used in schizophrenia and bipolar disorders. It is also an inverse agonist of D_{2L} dopamine receptor.

In this study, our aim is to elucidate the interaction mechanisms of thioridazine to design more $D_{2L}R$ specific drugs, which have less side effects. For this purpose we investigated inverse-agonistic mechanisms of thioridazine by homology modeling and explicit solvent simulations of human $D_{2L}R$ in complex with a Gi-coupled protein (including the αi, β and γ subunits). The complex receptor was implanted in a membrane system including phosphatidylcholine, phospatidylethanolamine and cholesterol developed by CHARMM-GUI membrane builder.

Our preliminary results indicate that thioridazine favors inactive-state conformation of $D_{2L}R$ by weakening specific interactions between Gα(i) and $D_{2L}R$. It possibly prevents inhibition of its effector and stops the activation of $D_{2L}R$, which have roles in psychiatric and neurodegenerative diseases.

LB-017
Apoptosis induction by 3′,4′-dibenzylflavonol in leukemia cells

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Introduction: In the present study we synthesized a series of flavonols and methylether derivatives, evaluated their effects on viability of three human leukemia cell lines and examined whether the most potent induces apoptosis.

Methods: Flavonoids were obtained by a combination of a Claisen-Schmidt condensation of 2-hydroxyacetophenones and benzaldehydes followed by a cyclization. Cytotoxicity against HL60, U937 and Molt-3 cells was evaluated by the MTT assay. Apoptosis was determined by fluorescent microscopy, DNA fragmentation and flow cytometric analysis. The cleavage of procaspases, cytochrome *c* release and the activation of the mitogen activated protein kinases were studied by western blot. Reactive oxygen species were determined by flow cytometry.

Results: A series of 74 flavonoids were obtained by organic synthesis. Cytotoxicity assays on human leukemia cells revealed that 3',4'-dibenzylflavonol was the most potent of the flavonoids assayed (IC_{50} < 1 μM) and it was 50-fold more toxic than the naturally occurring flavonol quercetin. This compound induced G_1 phase cell cycle arrest and it was a potent apoptotic inducer. Cell death was (i) mediated by the activation and the cleavage of initiator and executioner caspases; (ii) prevented by the pan-caspase inhibitor z-VAD-fmk; (iii) associated with the release of cytochrome c and with the phosphorylation of members of the mitogen activated protein kinases including p38, JNK/SAPK and ERK, and (iv) through a mechanism independent on reactive oxygen species generation.

Conclusion: 3',4'-dibenzylflavonol is a potent cell death inducer and might be useful in the development of new strategies in the fight against cancer.

LB-018
Dehydrodipeptidase 1 expression triggers invasive activity to regulate the EMT/MET switch in colorectal cancer

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Dehydrodipeptidase 1 (DPEP1/EC3.4.13.19) is a zinc-dependent metalloproteinase that the candidate novel marker of colorectal cancer based on an analysis of a gene expression microarray. However, functional roles and mechanism of DPEP1 in metastasis has not been elucidated. In this study, we showed that transcriptional and translational expression level of DPEP1 increase in stage-dependent colon tissues and cell lines, compared with non-tumor tissues. Increased invasiveness and adhesion but not cell proliferation were observed in SW480 (SW480-DPEP1) and HCT-116 (HCT-116-DPEP1) cell lines stably transfected with DPEP1 cDNA, in opposite with DPEP1 siRNA treatment. We also investigated that DPEP1-overexpressing cell lines exhibited increased metastatic activity in a xenograft nude mouse model. Interestingly, expression level of DPEP1 was decreased by TGF-b1 treatment in DPEP1 overexpressed cells but increased Leukotriene D4 (LTD4) secretion and E-cadherin such as epithelial-mesenchymal transition (EMT) regulator. Increased LTD4 in