REGULATION OF LIPOPROTEIN(A) METABOLISM BY PCSK9 IN ANIMAL MODELS

T. T. T. Tran[1], E. Villard[1], B. Poirier[1], E. Guillot[1], JC. Le Bail[1], C. Boixel[2], D. Boulay[3], J. Blankenstein[4], A. Parkar[1], T. Muslin[1] and P. Janiak[1]

[1] Cardiovascular and fibrosis unit, Sanofi Research and Development, Chilly-Mazarin, France; [2] DSAR, Sanofi, Alfortville, France; [3] Translational Science Unit, Sanofi Research and Development, Chilly-Mazarin, France
[4] DSAR-DD-ICMS, Sanofi, Vitry-Sur-Seine, France.

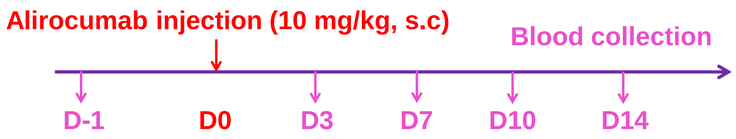
INTRODUCTION

- ***Low density lipoprotein-cholesterol (LDL-C), a major risk factor for coronary heart disease, is a primary interventional target for lipid-lowering PCSK9 antibodies such as alirocumab. In contrast to statins, alirocumab also significantly reduces plasma lipoprotein (a) [Lp(a)] in patients, another independent risk factor of cardiovascular diseases including aortic valve stenosis***
- ***Recent in vitro studies suggest that PCSK9 may regulate Lp(a) production and/or assembly in human primary hepatocytes***
- ***We aimed to consolidate these findings in liver humanized mice as well as in monkeys***

Our objective was to benchmark these two Lp(a)-expressing models for PCSK9 neutralization by alirocumab

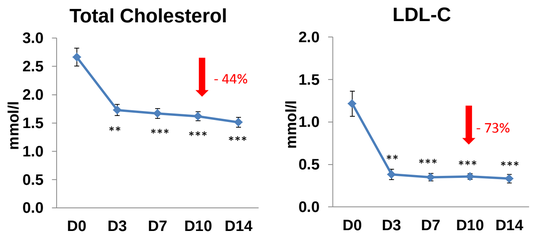
RESULTS

Cynomolgus monkeys

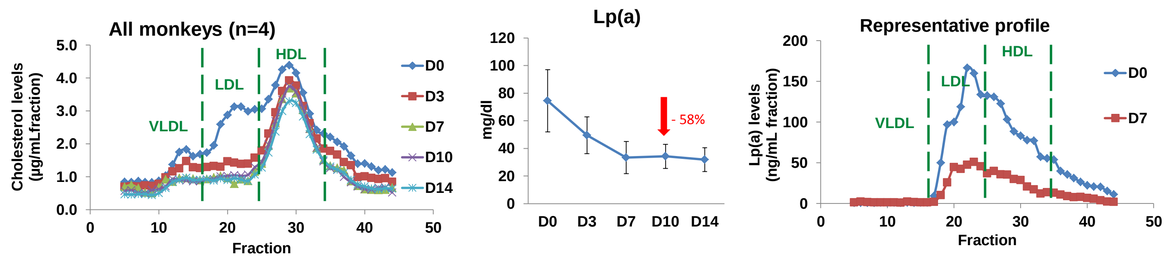


4 cynomolgus monkeys (2 males, 2 females)

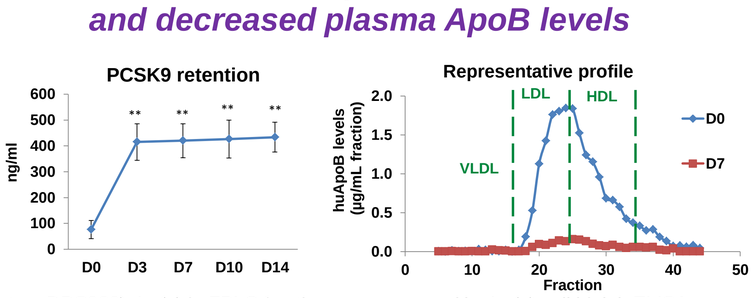
Alirocumab decreased plasma total cholesterol and LDL-C



Alirocumab decreased plasma Lp(a) levels



Alirocumab increased plasma PCSK9 retention and decreased plasma ApoB levels



Cholesterol, LDL-C, HDL-C was measured using ABX kits, Plasma Lp(a) was measured using Randox LP3403 on Pentra 400 analyser, Plasma PCSK9 was measured using hPCSK9 ELISA kit (R&D system DPC900), Lp(a) in FPLC fraction was measured by Lp(a) cell biolab ELISA.
T-test, 4 animals; ** p<0.01, *** p< 0.001 D3, D7, D10, D14 vs D0.

Alirocumab decreased rapidly and efficiently plasma cholesterol and Lp(a) levels in monkeys

Liver humanized FRG mice model

Description & benchmarking of the model

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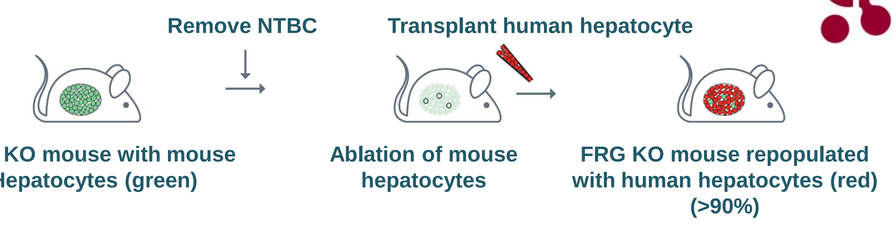
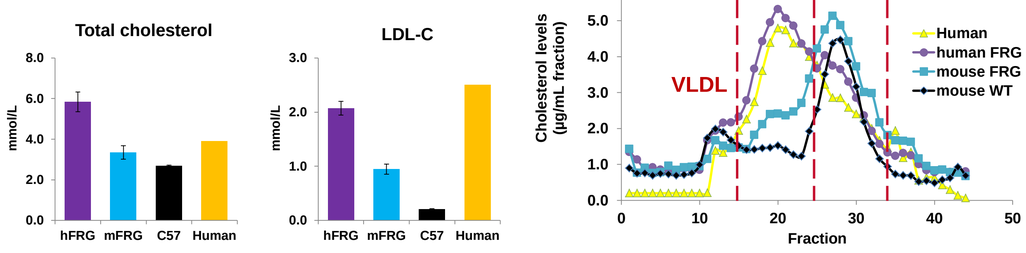


Fumarylacetoacetate hydrolase > Mouse hepatocyte death driver
Rag2 > T and B cell deficiency
Gamma-chain of IL2 receptor > NK cell deficiency
(**NTBC = Nitisinone** [2-(2-nitro--4-fluoromethylbenzoyl) cyclohexane-1,3-dione], inhibitor of enzyme in Tyrosine Catabolic Pathway)

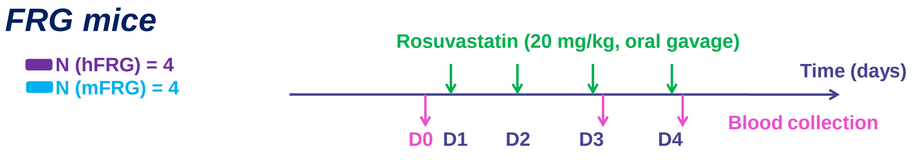
NTBC cycling

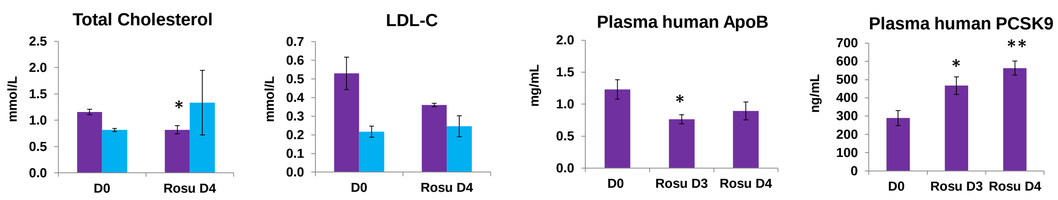
Human FRG mice displayed human-like lipid profile and expressed human liver proteins



Protein	Lp(a)	Human ApoB	Human PCSK9	Mouse PCSK9
Plasma levels	1 µg/mL	1000 µg/mL	75 ng/mL	750 ng/mL

Rosuvastatin expected properties translated only in liver humanized FRG mice

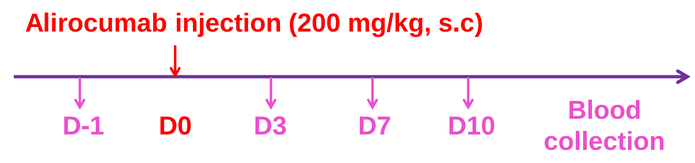




T-test, 3 animals/group; * p<0.05, D3, D4 vs D0

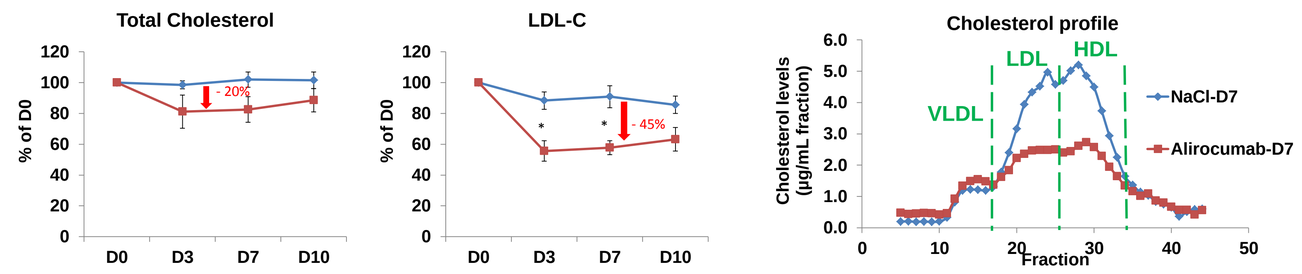
FRG mice is a relevant model to investigate human liver cholesterol metabolism

Alirocumab effect on:

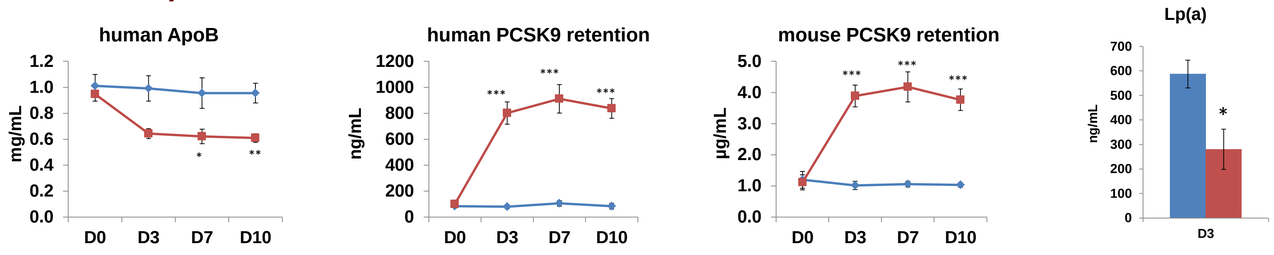


- ***Lipid profile in FRG mice***

NaCl N=5
Alirocumab N=5



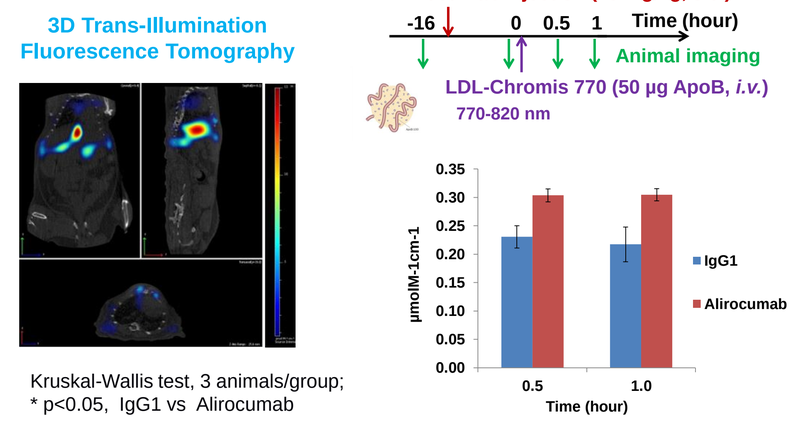
- ***Plasma proteins***



Plasma levels of Lp(a), human ApoB, human PCSK9 was measured by ELISA kits.
T-test, 5 animals/group; * p<0.05, ** p<0.01, *** p<0.001 NaCl vs Alirocumab

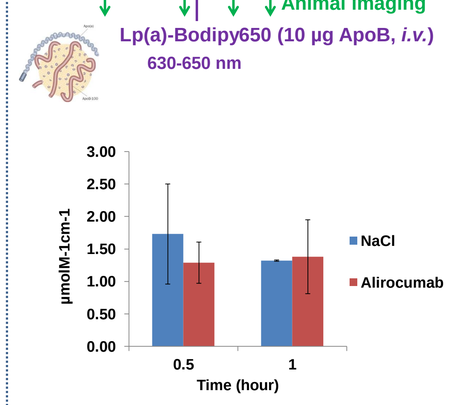
- ***Liver uptake of labelled lipoproteins***

Liver uptake assay validation in WT mice (LDL-chromis)



Kruskal-Wallis test, 3 animals/group;
* p<0.05, IgG1 vs Alirocumab

Lp(a)-uptake in FRG mice (Lp(a)-bodipy)



Alirocumab decreased plasma LDL-C and Lp(a) levels, and seemed inactive on Lp(a) hepatic uptake in FGR mice

CONCLUSION

Liver humanized FRG mice displayed a "human-like" lipid profile and a response to statin similar to human liver

- ***Alirocumab decreased plasma LDL-C and Lp(a) levels both in liver humanized FRG mice and in monkeys***
- ***Alirocumab did not seem to modify liver specific Lp(a) uptake in human FRG mice***

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