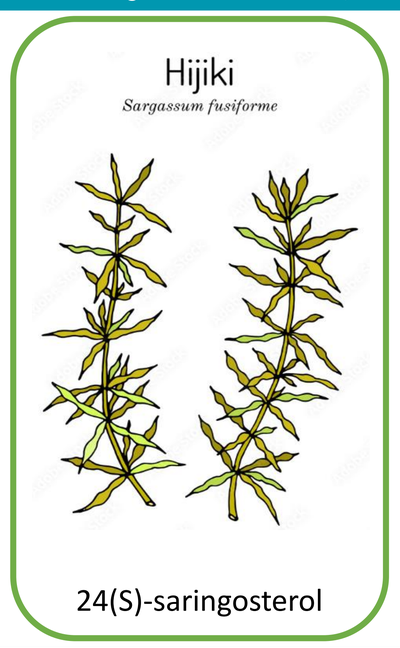
Identification of New Side Chain Oxidized Sterols as Novel Liver X Receptor Agonists with Therapeutic Potential in the Treatment of Cardiovascular Diseases

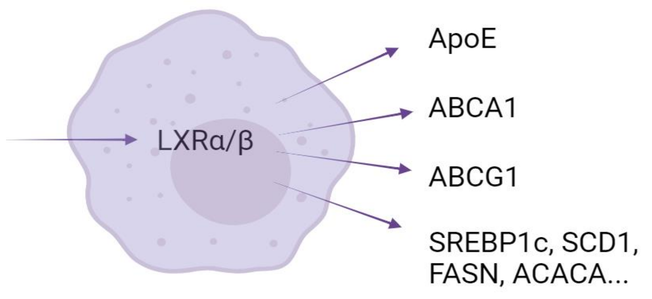
Marcella Palumbo 1, Na Zhan 2 3, Boyang Wang 2, Nikita Martens 3 4, Yankai Liu 2, Shangge Zhao 2, Gardi Voortman 3, Jeroen van Rooij 3, Frank Leijten 3, Tim Vanmierlo 4 5, Folkert Kuipers 6 7, Johan W Jonker 6, Vincent W Bloks 6, Dieter Lütjohann 8, Francesca Zimetti 1, Maria Pia Adorni 9, Hongbing Liu 2, Monique T Mulder 3

marcella.palumbo@unipr.it

1Department of Food and Drug, University of Parma, 43124 Parma, Ital; 2Key Laboratory of Marine Drugs, Ministry of Education, School of Medicine and Pharmacy, Ocean University of China, Qingdao 266003, China; 3Department of Internal Medicine, Erasmus Medical Center, 3015 CN Rotterdam, The Netherlands; 4Department of Neuroscience, Biomedical Research Institute, Hasselt University, 3500 Hasselt, Belgium; 5School for Mental Health and Neuroscience, Maastricht University, 6229 ER Maastricht, The Netherlands; 6Department of Pediatrics, University Medical Center Groningen, University of Groningen, 9713 GZ Groningen, The Netherlands; 7European Research Institute for the Biology of Ageing (ERIBA), University Medical Center Groningen, University of Groningen, 9713 GZ Groningen, The Netherlands; 8Institute of Clinical Chemistry and Clinical Pharmacology, University Hospital Bonn, 53105 Bonn, Germany; 9Unit of Neurosciences, Department of Medicine and Surgery, University of Parma, 43125 Parma, Italy.

Poster presented: Convegno monotematico SIF, GdL Cardivascolare e Metabolico, Ottobre 16-17,2023; Parma, Italia





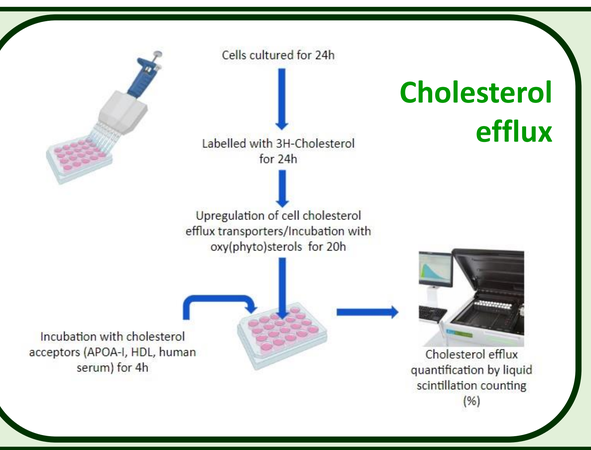
Background

The **nuclear liver X receptors** (LXR) α and β, may be potential **therapeutic targets** in cardiovascular diseases because of their key role in the regulation of lipid homeostasis and inflammatory processes.

Among the mechanisms involved in the maintenance of macrophage cholesterol homeostasis, **cholesterol efflux** plays a crucial role.

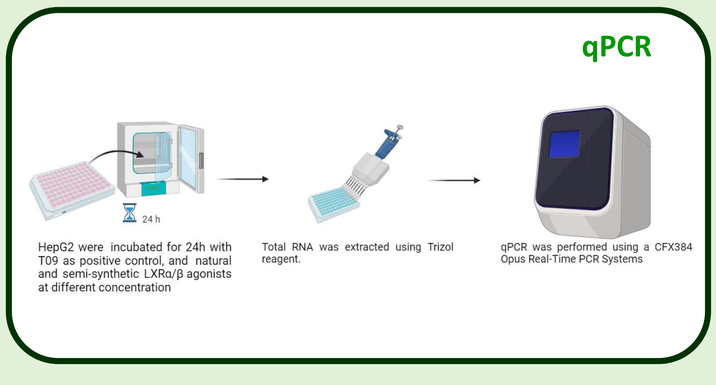
Specific **oxy(phyto)sterols** differentially modulate the transcriptional activity of **LXRs**, providing opportunities to develop **new therapies**. However, this development is precluded by unwanted side effects, such as hypertriglyceridemia and hepatic steatosis due to hepatic LXRα activation.

The aim of this study was to investigate the effect of newly isolated oxyphytosterols from ***Sargassum Fusiforme*** and new synthesized side chain oxidized sterols analogs on cholesterol efflux and on expression of genes involved in this process.



Methods

Cellular cholesterol efflux was evaluated with a radioisotopic cell-based assay on human hepatocellular carcinoma cell line (HepG2) after treatment with n=5 new LXRα/β agonists at different concentrations. Gene expression was assessed in HepG2 by qPCR. T0901317 1µM was used as positive control.



Results

Figure 1

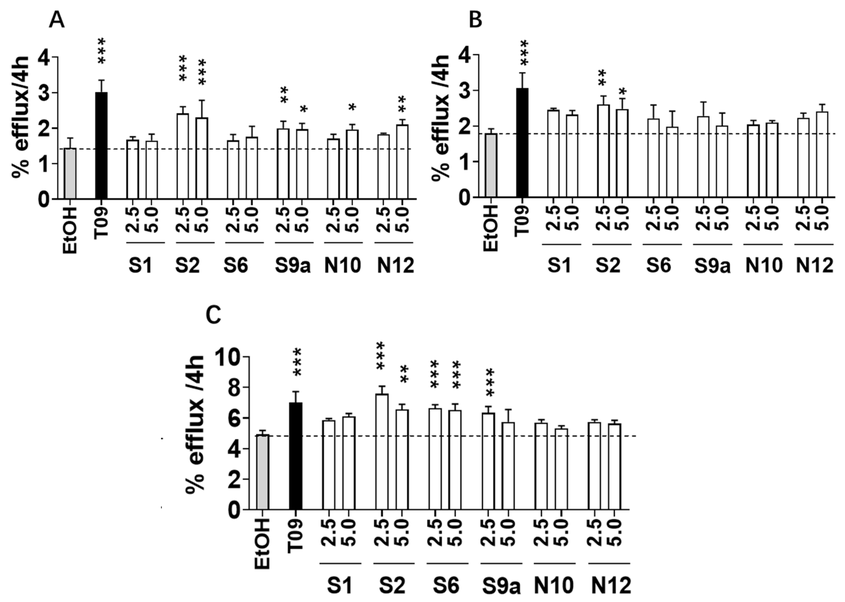
Cell line	HepG2	
LXR	α	β
Compounds	5.0µM	
S1	2.56	2.33
S2	2.40	2.55
S3	1.05	1.39
S4	0.82	0.80
S5	0.67	0.71
S6	3.22	2.73
S7	0.83	1.45
S8a	2.64	3.70
S8b	1.54	2.01
S9a	2.34	2.75
S9b	1.37	1.14
N10	2.88	4.15
N11	1.87	2.30
N12	1.38	4.23
N13	0.37	0.59

Fold change
3.5 < X
3.0 < X ≤ 3.5
2.5 < X ≤ 3.0
2.0 < X ≤ 2.5
1.5 < X ≤ 2.0
1.0 < X ≤ 1.5
≤1.0

Among the 5 natural and semi-synthetic 24-oxidized sterols with high potency for LXRα/β activation (**Figure 1**), we identified two synthetic side chain compounds (**S2** and **S6**) that also regulate cholesterol homeostasis.

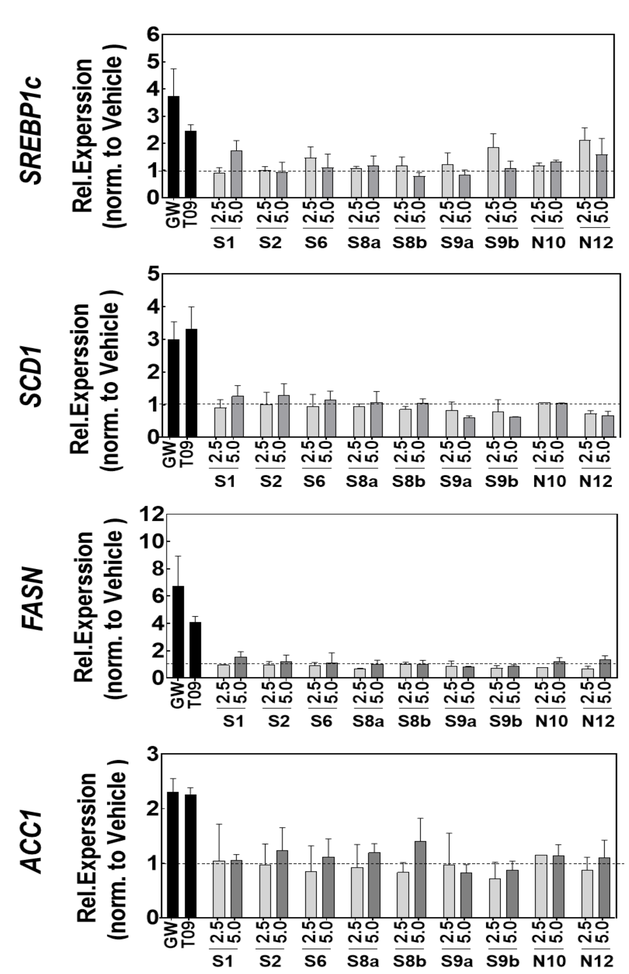
In detail, **S2** increased cholesterol efflux from HepG2 by 54.3%, 15.2%, and 24.5% using isolated APOA-I (**A**) and HDL (**B**), and human serum (**C**) as cholesterol acceptors, respectively, while **S6** increased cholesterol efflux by 9.3% and 6.7% in presence of APOA-I (**A**), and human serum (**C**) (**Figure 2**).

Figure 2



Regarding the impact of these compounds on gene expression, in HepG2 cells **S2** and **S6** did not upregulate the expression of the main genes involved in cholesterol efflux **ABCA1** and **ABCG1** (**data not shown**), in addition, they did not affect the expression of **SREBP1c**, **SCD1**, **FASN,** or **ACC1** (**Figure 3**), responsible for the hepatic side effects which are usual for synthetic pan-LXR agonist.

Figure 3



Conclusions

The positive effect of these compounds on cholesterol efflux put the premises to identify and develop novel LXR-activating 24-oxidized sterols as potential therapeutic options in cardiovascular diseases.

SOCIETÀ ITALIANA DI FARMACOLOGIA

EUROPEAN LIPOPROTEIN CLUB

EAS

SOCIETA' ITALIANA PER LO STUDIO DELL'ARTERIOSCLEROSI

ALIFAR DIPARTIMENTO DI ECCELLENZA MUR 2023-2027 UNIVERSITÀ DI PARMA

Erasmus MC University Medical Center Rotterdam