Synthetic HDL Lipid Scavengers for the Treatment of Niemann-Pick Diseases

Maria V. Fawaz (Advisor: Anna Schwendeman)
Medicinal Chemistry Third Year Seminar
Tuesday, January 23, 2018 at 4:00 pm

Niemann-Pick disease (NPD) is a fatal lipid storage disorder that results in an accumulation of unesterified cholesterol (type C) and sphingomyelin (types A and B) in late endosomes and lysosomes. Inherited defects in *NPC1* (95%) and *NPC2* (5%) are responsible for NPD type C (neurovisceral) while *SMPD1* mutations cause types A (neurovisceral) and B (visceral). To date, NPD management is largely symptomatic with only few therapies in clinical trials. NPA/NPB and NPC are caused by completely different cellular pathways. However, both cholesterol and sphingomyelin defects in NPD patients lead to markedly reduced high-density lipoprotein (HDL) levels (> 2-fold). The body produces HDL as an 8-12 nm nanoparticle composed of a lipid membrane wrapped around by a belt of 243 amino acid apolipoprotein A-I (ApoA-I). Infusion of synthetic HDL (sHDL) in patients with atherosclerosis has been shown to reduce cholesterol in arterial plaques and was found to be safe at doses up to 100 mg/kg. Applying the same strategy to NPD, we propose that sHDL similar to native HDL can serve as a scavenger of accumulated cholesterol and sphingomyelin (Fig. 1) in NPD patients and rescue their phenotypes thus representing a novel therapeutic strategy.

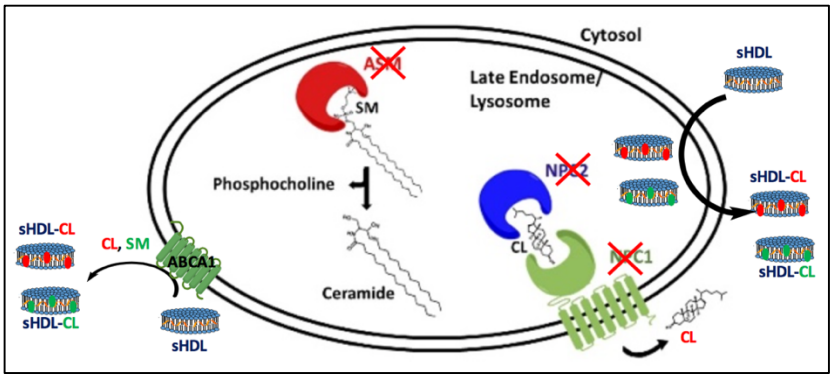

Figure 1. A schematic diagram of sHDL acting as a scavenger of cholesterol (CL) and sphingomyelin (SM) from late endosome/lysosome. ASM (acid sphingomyelinase), ABCA1 (ATP-binding cassette transporter A1).

Composition of sHDL can be customized to modulate its cholesterol and phospholipid binding capacity, pharmacokinetics and safety. We prepared a panel of sHDLs made from short ApoA-I mimetic peptides 5A, 18A, or 22A complexed with phospholipids such as sphingomyelin (SM), palmitoyl-oleoyl phosphatidylcholine (POPC), or dimyristoyl PC (DMPC). Peptides and sHDLs were tested in several primary NPC patient fibroblast cell lines. It was found that all sHDLs were capable of reducing accumulated intracellular cholesterol to varying degrees while peptide alone was not a good scavenger of cholesterol. Trafficking of sHDL was assessed by incubating NPC cells with fluorescently-labeled particles (DiD dye) and lysosomal marker LAMP1. It was revealed that sHDLs were endocytosed into cells and co-localized with LAMP1. Finally, an *in vivo* study was executed to examine effects of our best sHDL formulation 5A-SM in NPC1 I1061T homozygotes and littermate controls treated with vehicle or sHDL (100 mg/kg, i.p., 3x/wk) for 4 weeks, starting at 7 wks of age. The administration of 5A-SM resulted in a significant rescue of body weight ($p<0.01$) in the NPC mice with no toxicity to animals. However, neurocorrection after sHDL treatment in adult mice was not detected indicating alternative delivery routes, treatment durations, or HDL compositions are still needed. Next, sHDLs and peptides were tested against NPA primary patient fibroblast cells. It was found that peptide alone as well as lipid-poor sHDLs were capable of substantially decreasing levels of sphingomyelin accumulations from NPA cells in a time- and dose-dependent manner. Further evaluation of a peptide or sHDL in NPA mouse model is necessary to determine its efficacy and safety *in vivo*. The completion of the proposed experiments will provide a proof of concept showing the effectiveness of using sHDL for the treatment of NPD.