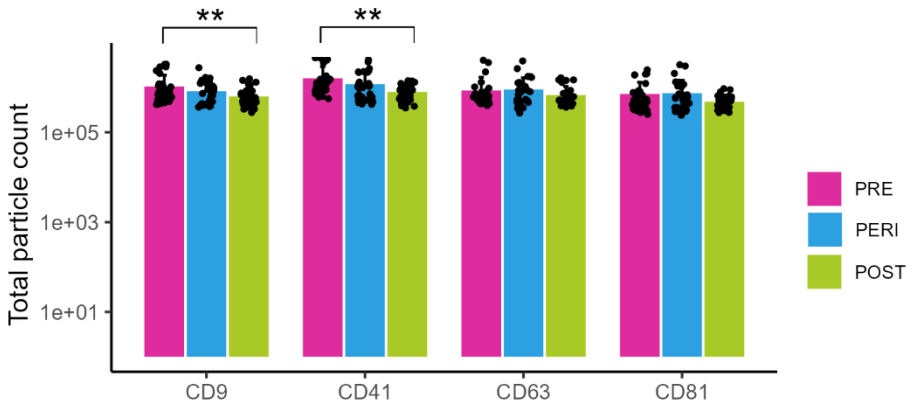
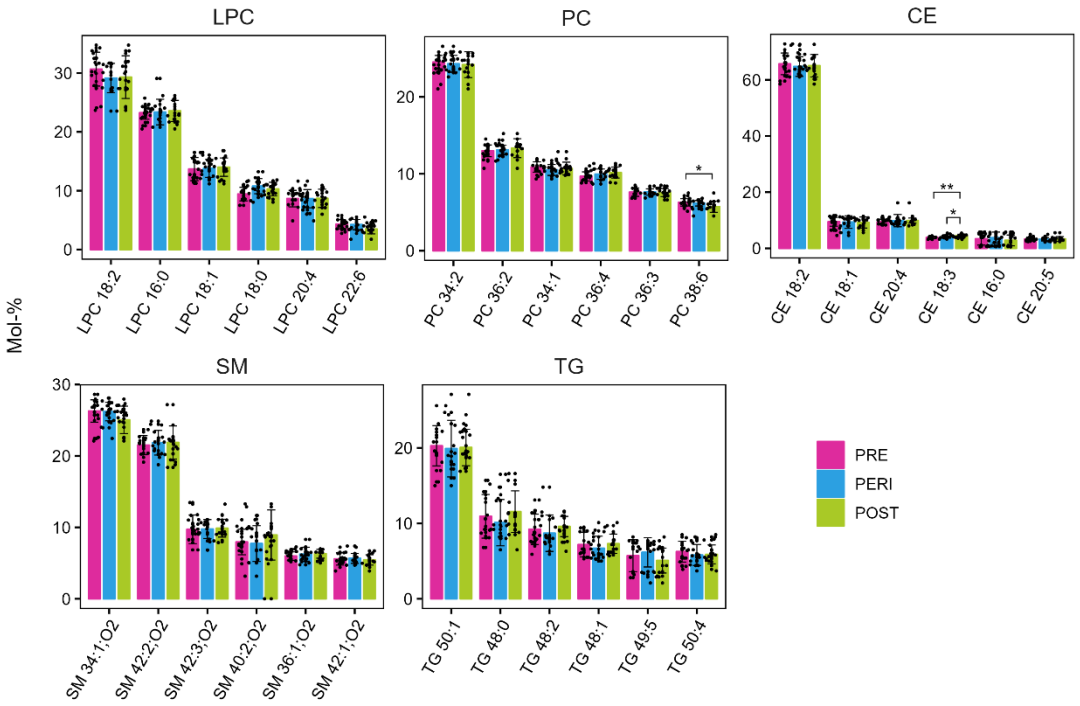
SUPPLEMENTAL FIGURES

Supplemental Figure S1



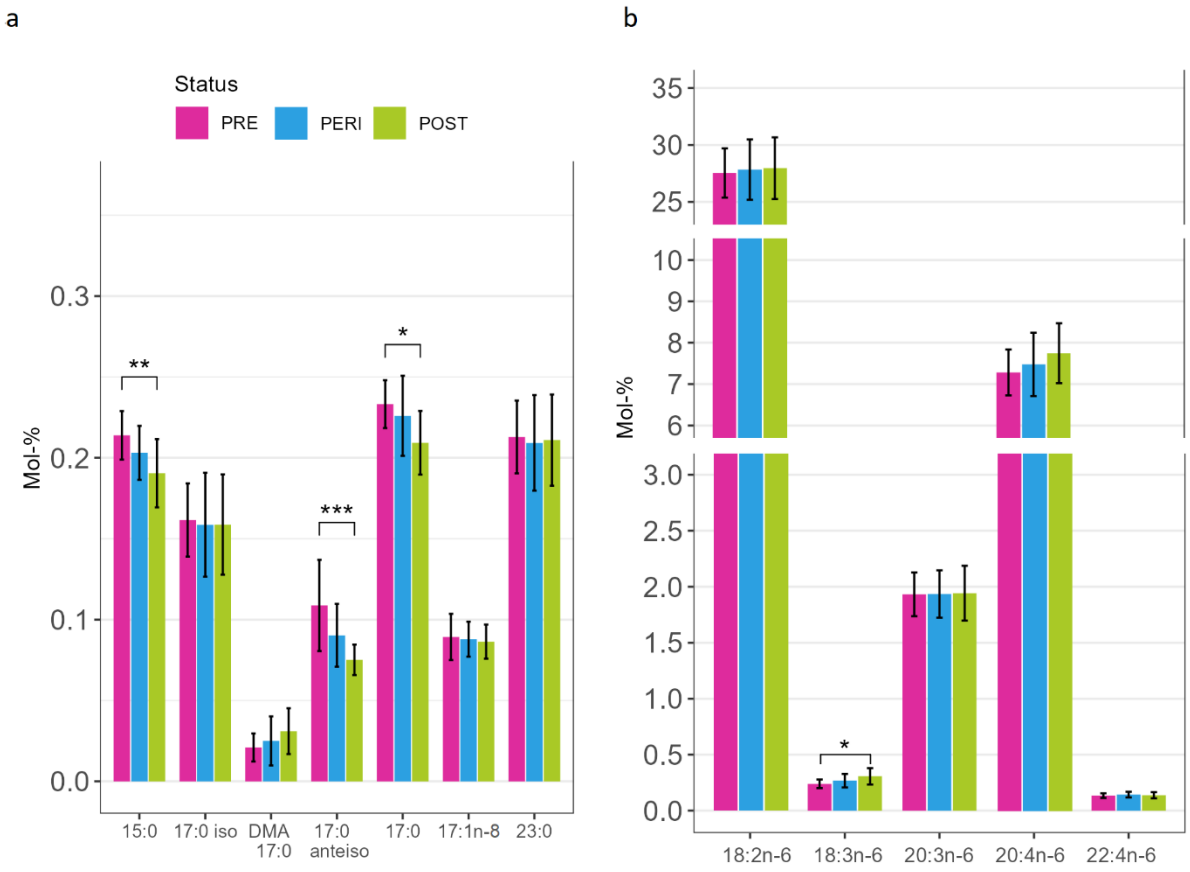
Supplemental Figure S1. Characterization of the size exclusion chromatography -purified extracellular vesicles (EV). Distribution of CD9, CD41, CD63 and CD81 positive EVs across menopausal groups. Statistical significance **<0.01. Abbreviations: PRE, premenopause; PERI, perimenopause; POST postmenopause.

Supplemental Figure S2



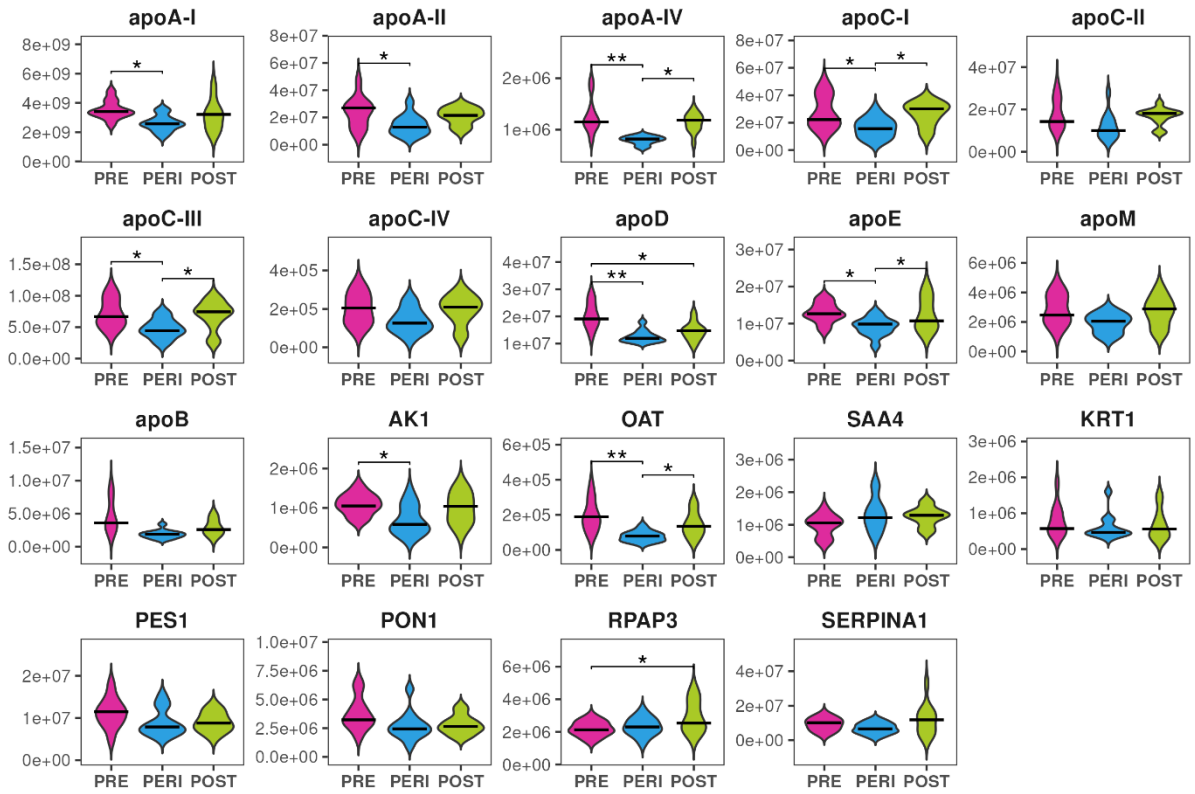
Supplemental Figure S2. The most abundant lipid species observed in the five main lipid classes of the HDL particles. Lipid species abundances are expressed as mol-% within each lipid class. Lipids were extracted from ultracentrifugally isolated HDL and analyzed with ESI-MS. Statistical significance * < 0.05, **<0.01. Abbreviations: HDL high-density lipoprotein, PRE premenopause, PERI perimenopause, POST postmenopause, LPC lysophosphatidylcholine, PC phosphatidylcholine, CE cholesteryl ester, SM sphingomyelin, TG triacylglycerol.

Supplemental Figure S3



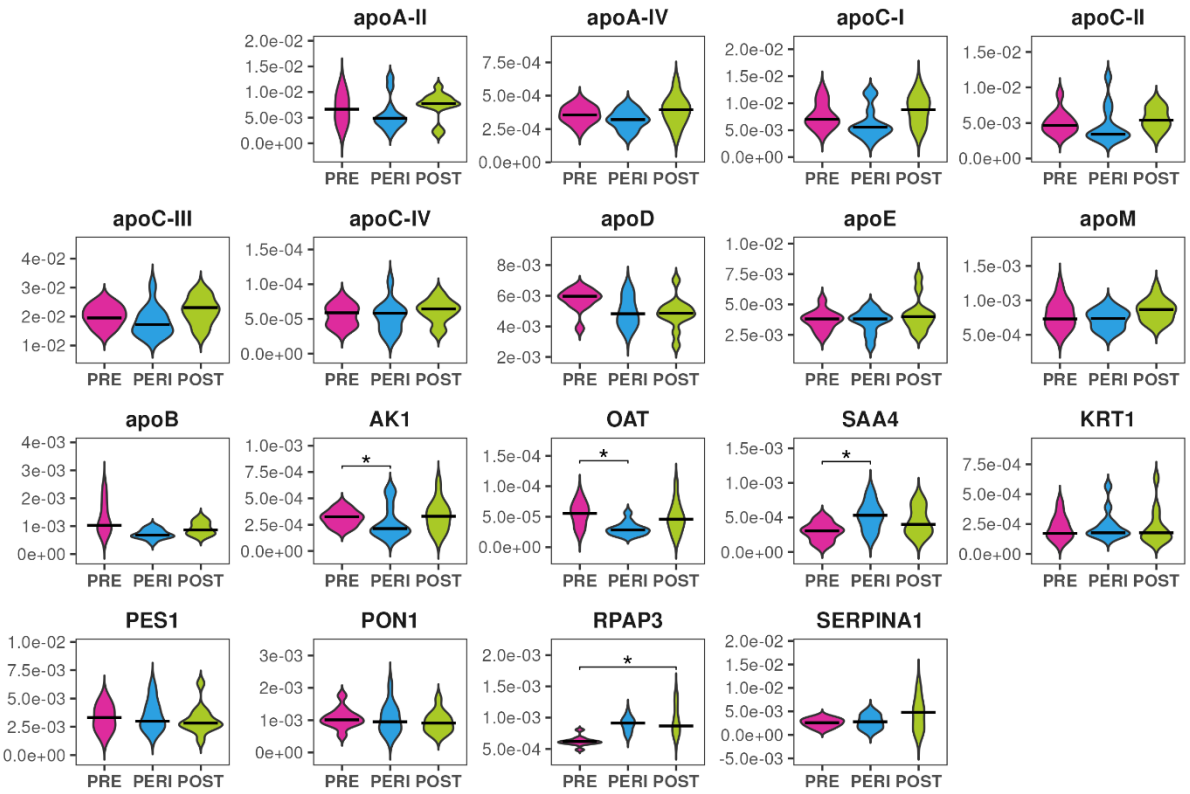
Supplemental Figure S3. Fatty acid composition of HDL lipids in three different menopausal statuses. **(a)** Odd-chain fatty acids of HDL. **(b)** n-6 pathway fatty acids of HDL. Fatty acids were analyzed with GC-FID and GC-MS. Statistical significance * < 0.05, **<0. 01, ***<0.001. Abbreviations: HDL, high-density lipoprotein; PRE, premenopause; PERI, perimenopause; POST, postmenopause; DMA, dimethyl acetal (represents an alkenyl chain from plasmalogen phospholipids, and is formed when methylating fatty acids for gas chromatography); GC-FID, gas chromatography with flame ionization detector; GC-MS gas, chromatography-mass spectrometry.

Supplemental Figure S4



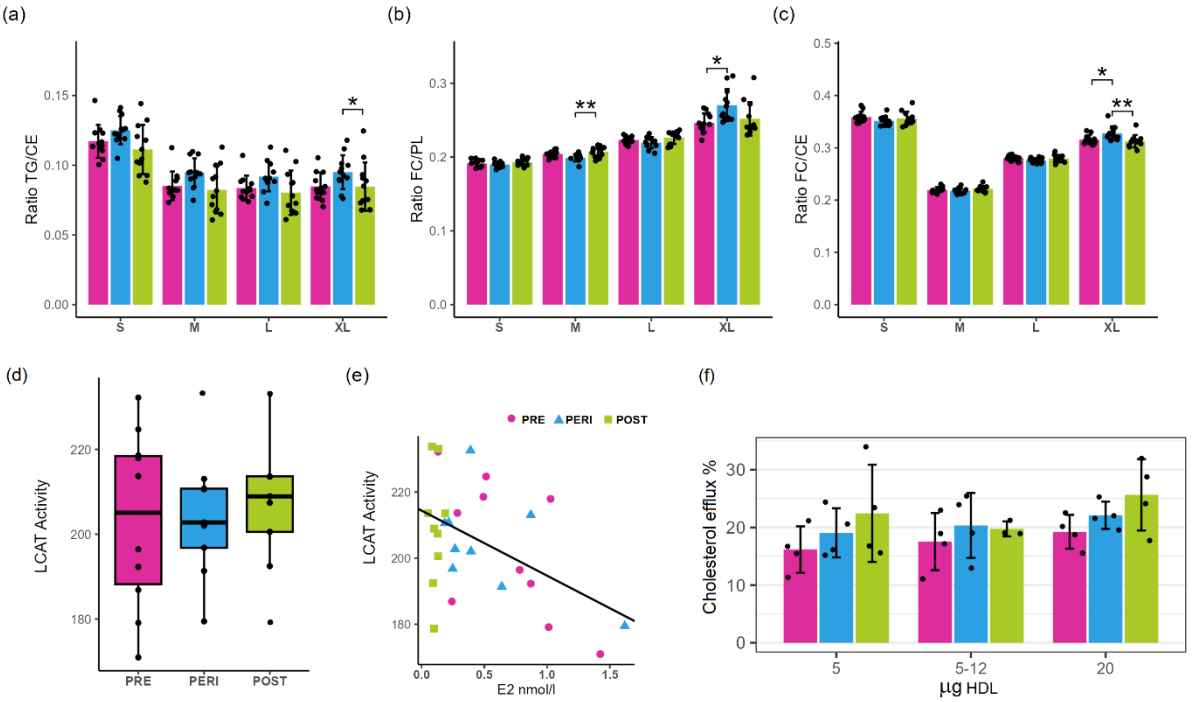
Supplemental Figure S4. Violin plots of unnormalized HDL protein abundances (raw data). All protein abundances were measured with LC-MS method from the isolated HDL particles. Statistical significance * < 0.05, **<0.01. Abbreviations: PRE premenopause, PERI perimenopause, POST postmenopause, HDL high-density lipoprotein, and LC-MS liquid chromatography-mass spectrometry.

Supplemental Figure S5



Supplemental Figure S5. Violin plots of HDL protein abundances normalized to apoA-I. All protein abundances were measured with LC-MS method from the isolated HDL particles. Statistical significance * < 0.05, **<0.01. Abbreviations: PRE premenopause, PERI perimenopause, POST postmenopause, HDL high-density lipoprotein, and LC-MS liquid chromatography-mass spectrometry.

Supplemental Figure S6



Supplemental Figure S6. Functional assessments of the HDL particles across menopausal stages. (a). The ratio of lipid classes TG/CE in HDL particle size categories. (b). The ratio of lipid classes FC/PL in HDL particle size categories. (c) The ratio of lipid classes FC/CE in HDL particle size categories. All lipid class ratios were calculated from lipid classes measured with NMR. (d) LCAT activity assay with serum samples representing PRE (n = 10), PERI (n = 9) and POST (n = 9) women. (e) Correlation plot of the measured LCAT activity and estradiol concentration. (f) Cholesterol efflux assay with 5 µg, 12.5 µg and 20 µg of purified HDL originating from the pooled serum samples of PRE, PERI and POST women. Statistical significance * < 0.05, **<0.01. Abbreviations: HDL high-density lipoprotein, PRE premenopause, PERI perimenopause, POST postmenopause, TG triglycerides, CE cholesteryl ester, FC unesterified cholesterol, PL phospholipid, NMR nuclear magnetic resonance, LCAT lecithin:cholesterol acyltransferase, and E2 17β-estradiol.