Supplementary materials

Supplementary Table of Content

Complete methods

Patients and ethics statement

Patients were sampled at the University Hospital of Lyon. Control patients were healthy volunteers for a living kidney donation, hospitalized for their pre-donation biological and clinical work-up. Hemodialysis patients were sampled within the hemodialysis unit of the Edouard Herriot hospital before the mid-week session. Inclusion criteria were age >18, undergoing hemodialysis for more than 6 months. Exclusion criteria were diabetes mellitus, ongoing inflammatory disease, liver cirrhosis, recent cardiovascular event (<3 months, cardiac myoinfarction, stroke, acute peripheral artery occlusion), uncontrolled anemia, coagulopathy and BMI higher than 35 $kg.m^{-2}$. The study was conducted in accordance with the Declaration of Helsinki and was approved by the local ethical committee (reference L16-57, Comité de Protection des Personnes Lyon Sud Est IV). A written informed consent was obtained from all subjects.

Blood collection

Blood samples were obtained by venipuncture (on EDTA coated tubes) except for dialysis blood samples that were obtained immediately before dialysis from the arterial line of the mechanical bloodstream. Blood samples were centrifuged at 3500 *x g* for 10 min to isolate plasma which were collected and stored at -80°C until use.

Animal procedures

All animal experiments were performed under the authorization no. n°69-266-0501 and according to the guidelines laid down by the French Ministry of Agriculture (n° 2013-118) and the European Union Council Directive for the protection of animals used for scientific purposes of September 22, 2010 (2010/63UE). Adult male White New Zealand rabbits (CEGAVssc, Saint Mars d'Egrenne, France) were housed in individual cages at constant ambient temperature (21-23°C) and humidity (45-50%) with a 12 h light cycle. All animals had free access to tap

water. After a 7-day period of acclimation, rabbits were randomized to either the 5/6 nephrectomy group or in the control group.

Nephrectomy was performed as described by Goitloib et al.[1] Rabbits were anaesthetized using an intramuscular injection of ketamine (50mg/kg), xylazine (5mg/kg), and acepromazine (0.5mg/kg, Centravet, Lapalisse, France). The rabbit was placed on the right lateral decubitus. The right dorsolumbar area was shaved and disinfected twice with povidone-iodine. A sterile field was applied to delimit the operative site and a sterile film was applied onto the shaved skin. Local anesthesia was performed by a subcutaneous injection of xylocaine 2% (2 ml) at the incision site. Then, a skin incision of approximately 4 cm in length was performed at 2 finger-breadths from the caudal end of the rib cage and from the lumbar muscle. The left kidney was externalized and the perirenal adipose tissue was gently dissected. The two poles of the kidney were electrocoagulated using an electric needle to produce a 2/3 reduction of the left kidney mass. The muscle and wound were sutured (Prolene 4/0, Ethicon, Somerville, NJ, USA), and the wound was painted with povidone-iodine. One week after surgery, a unilateral right nephrectomy was performed. A nylon monofilament ligature was placed on the kidney pedicle. The kidney pedicle was carefully sectioned between the kidney and the ligature. Special care was taken to avoid damage to the adrenal glands. The control animals underwent the same procedures (general anesthesia, skin, and muscle incisions) followed by a simple kidney mobilization. All rabbits were given buprenorphine (0.05mg/kg sc, 2-times a day) for 2 days to prevent post-surgical pain. Food intake and body weight were measured on a daily and twice-weekly basis, respectively.

Measurement of glomerular filtration rate

The glomerular filtration rate (GFR) was measured through the kinetics of plasma iohexol concentration decrease. Rabbits were anesthetized with an intravenous injection of 27.5 mg/kg

of sodium pentobarbital (Centravet). Then, the neck was shaved and a median incision was performed after local anesthesia with a subcutaneous infiltration of xylocaine 2% (1mL). A PE-50 catheter was introduced in the left jugular vein for the injections and PE-60 catheter was disposed in the left carotid artery for the blood sampling. A 1mL bolus of iohexol (Omnipaque 300®, GE Healthcare, Chicago, IL, USA) was performed and the timer was started after flushing the residue left in the jugular catheter with a saline solution. The syringe was weighted to the nearest milligram before and after injection to precisely calculate the amount of iohexol delivered to the rabbit. Blood was sampled at 5, 15, 30, 45, 60, 120, and 180 min on lithium-heparin coated tubes. The plasma was aliquoted after centrifugation at 5,900 *x g* for 2 min and stored at -20°C. The serum iohexol concentration was measured by HPLC as previously described[2] and GFR was calculated using a bi-compartmental model equation.

Sacrifice and necropsy

At the end of GFR measurement, rabbits were deeply anesthetized with an overdose of sodium pentobarbital (70 mg/kg iv). Blood was removed by cardiac puncture and placed in EDTA-coated tubes. After a centrifugation at 1,250 *x g* for 10 min, plasma was aliquoted and stored at -80°C. Urine were obtained with a direct bladder puncture and stored at -20°C. The kidney was dissected out, weighed, and stored in buffered formalin 10% (w/v) for histological examination.

Renal histology

Kidneys were dehydrated using ethanol, embeded in paraffin and sliced. Haematoxylin Erythrosine Saffron (HES) and Sirius Red stainings were performed.

Isolation of lipoproteins from the plasma

Lipoproteins were separated from plasma by stepwise potassium bromide (KBr) density gradient ultracentrifugation.[3] Plasma was fractionated in the Beckman ultracentrifuge with a rotor TAL 100.3 (Beckman, Brea, CA, USA). A first centrifugation at 100,000 *x g* for 3h 30min at 15°C was performed to remove the top layer corresponding to VLDL and chylomicrons. Then, the plasma density was adjusted to 1.063 $g.mL^{-1}$ with KBr (M=119.01 $g.mol^{-1}$). After a second centrifugation at 100,000 *x g* for 5h at 4°C the orange ring, corresponding to LDL, was collected. Finally, plasma density was adjusted to 1.21 $g.mL^{-1}$ with KBr and after centrifugation at 100,000 *x g* for 6 h 30 at 4°C the orange ring corresponding to HDL, was collected.

For the isolation of all the lipoproteins together, a single ultracentrifugation was performed at 100,000 *x g* for 6h 30min at 4° after an adjustment of plasma density to 1.21 $g.mL^{-1}$ with KBr.

For platelet aggregation and copper-induced HDL oxidation assay freshly isolated HDL samples were used to prevent from HDL ultrastructure modification due to freezing,40 after the ultracentrifugation, lipoproteins were extensively dialyzed against phosphate saline buffer (PBS) with 1mM EDTA for 3h, twice at room temperature and then overnight at 4°C. A last dialysis without EDTA was performed just before the platelet aggregation and copper-induced oxidation. HDL were stored at 4°C maximum 48 hours before the experiments.

Biochemistry

Serum creatinine measurement was performed using the Siemens enzymatic method (on the Dimension Vista System, Siemens Healthcare, Erlangen, Germany) traceable to National Institute of Standards and Technology Creatinine Standard Reference Materials 914 (verified with National Institute of Standards and Technology SRM 967) with calibration certified by isotope dilution mass spectrometry. Urea was measured with urease test (Vista 1500). Cholesterol and triacylglycerols levels were measured using enzymatic kits (Biomerieux, Marcy l'Etoile, France). HDL concentration was measured with an enzymatic kit (Abcam, Paris, France). Malondialdehyde (MDA) was measured by HPLC coupled to UV-visible

detection (Diode Array detector) as described by Grotto *et al*.[5]. Antioxidant activity (AOA) of the plasma was measured as described by Koracevic *et al*.[6]. Proteinuria was measured using the Bradford protein assay (BioRad, Marne-la-Coquette, France) using bovine serum albumin as a standard.

Lipoproteins assays

MDA concentration in HDL was determined by HPLC according to the method described by Therasse and Lemonnier[7]. Lipoprotein samples, mixed with thiobarbituric acid (TBA, 10 mM), acetic acid and bultyl-hydroxy-toluen (5mM) were heated for 60min at 95°C. The TBA-MDA adducts were then extracted with ethyl acetate and separated on a Nucleosil C18 column (5µm, 4.6 × 250 mm, Macherey-Nagel, Hoerdt, France) using methanol/water (20:80, v/v) as mobile phase. Detection was performed by measuring the absorbance at 532 nm.

Anti-HNE-Michael adduct (ref 393207) and anti- HHE-Michael adduct (ref NOF-N213730-EX) antibodies were purchased from Calbiochem (San Diego, CA, USA) and Cosmobio (Tokyo, Japan), respectively. The antibody used to detect HHE-adducts is reported to be highly specific for HHE Michael adducts on histidine residues and therefore enables the specific detection of HHE-histidine in protein samples[8]. Fifty micrograms of HDL were loaded directly onto a nitrocellulose membrane using the Bio-Dot apparatus (BioRad, Marne-la-Coquette, France). Following saturation with 5% bovine serum albumin (BSA), membranes were probed overnight with primary antibodies, anti-HHE-Michael adducts, or anti-HNE-Michael adducts (1:1000 dilution in a 5% BSA solution). After incubation with HRP-coupled secondary antibodies (1:3000; 5% BSA solution), membranes were processed for chemiluminescence (ECL plus, GE Healthcare, Chicago, IL, USA) and quantitated by densitometry using Image J software (NIH, Bethesda, MD, USA). The 8-isoprostane level was measured using an immunoassay (Bertin Pharma, Montigny-le-Bretonneux, France).

4-HNE was synthetized as described by Soulère *et al*.[9] and was diluted in dimethylsulfoxyde (DMSO). 4-HNE was added to HDL solutions at a final concentration of 1, 10, 50, or 100 μM. After a 16h overnight incubation at 37°C in a water bath, HDL were dialyzed three times against PBS to remove the free fraction of 4-HNE.

Platelet aggregation and activation

Blood was collected at the regional blood center from healthy volunteers who had not ingested any aspirin or any other non-steroidal anti-inflammatory drug in the previous 10 days. Platelets were prepared for the assays as described by Lagarde *et al*.[10] Platelet function test was carried out according to the Born turbidimetric method.[11] Platelet aggregation was measured in isolated platelets in a dual-channel aggregometer (Chrono-log; Coulter, Margency, France). Platelet suspensions were pre-incubated for 5 min at 37°C in the presence or absence of lipoproteins (0.025 mg of protein/mL for rabbit, 0.050 mg/mL for human) and stimulated with threshold concentrations of collagen (75 ± 9 ng/ml) while being continuously stirred at 1,000 rpm. The threshold concentration of collagen was defined as the concentration of collagen that induced a 50% increase in light transmission. The extent of platelet aggregation was expressed in terms of percentage of change in light transmission 4 min after the addition of collagen. Blocking of CD36 or SRB1 receptor was achieved by the pre-incubation with 10 μL of an anti-CD36 (Ab-CD36) or anti-SRB1 (Ab-SRB1) antibodies (dilution 1:500, Abcam, Paris, France) for 10 min at 37°C before the incubation with HDL. Blocking of SRC-kinases was achieved by the pre-incubation with Naphtyl PP1 (final concentration 1 μM, Santa Cruz Biothechnologies, Dallas, TE, USA) for 10 min at 37°C before the incubation with HDL. Aggregation values from lipoprotein assays were expressed as a percentage of the maximum aggregation induced by the collagen alone (considered as 100%).

For activation assays, platelet suspensions were pre-incubated for 15 min at 37°C in the presence or absence of lipoproteins (0.050 mg/mL for human) and stimulated with threshold concentrations of collagen (4ng/mL). Blocking of CD36 or SRB1 receptor was achieved by the pre-incubation with 10 μL of an anti-CD36 (Ab-CD36) or anti-SRB1 (Ab-SRB1) antibodies (dilution 1:500, Abcam, Paris, France) for 10 min at 37°C before the incubation with HDL. Blocking of SRC-kinases was achieved by the pre-incubation with Naphtyl PP1 (final concentration 1 μM, Santa Cruz Biothechnologies, Dallas, TE, USA) for 10 min at 37°C before the incubation with HDL. Levels of thromboxane B2 (TxB2) was measured using an immunoassay expressed as a percentage of the maximum level induced by the collagen alone (considered as 100%, Cayman Chemical, Ann Arbor, MI, USA). After pre-incubation with HDL and antibodies or pharmacological inhibitor, platelets were activated with collagen then fixed in paraformaldehyde 5% w/v and stained with anti-CD62/P-Selectin (Thermo Fisher Scientific, San Jose, CA, USA) according to the manufacturer protocol and analyzed by flow cytometry.[12] Expression of intra-platelet phosphorylated JNK was measured by Western Blot with an anti-phospho-JNK antibody (Cell signaling technologies, Danvers, MA, USA). Quantification and expression were processed as previously described.[13]

Copper-induced HDL oxidation

Aliquots of freshly dialyzed HDL (50 μg protein) were oxidized in the presence of 2.5 μM of $CuSO_4$ within the day following the last dialysis. The oxidation was monitored by measuring the formation of conjugated dienes at 234 nm[14] every 5 min for 3 h in a Kontron computer-linked spectrophotometer.

Mass spectrometry assay

List of reagents:

Sigma Aldrich (Saint Quentin Fallavier, France): Acetonitrile MS grade (ACN), Formic acid (FA), Trifluoroacetic acid (TFA), Iodoacetamide (IAA), Dithiotreitol (DTT), Sodium Deoxycholate (SDC), Ammonium Bicarbonate (AB)

MilliQ Water

Trypsin: Promega (Lyon, France)

Pierce C18 desalting spin column and Pierce quantitative fluorometric peptide assay: Thermo Scientific (San Jose, CA, USA)

In-solution digestion

30 μg of protein were diluted in 100 μL of 50 mM AB/SDC 1% buffer. They were reduced with 5 mM DTT for 1h at 57°C, and then alkylated with 10 mM IAA for 1h in the dark at room temperature and under agitation (850 rpm). Enzymatic digestion was performed with trypsin at a ratio 1/100 (enzyme/proteins) and overnight at 37°C. Endly, the samples were desalted on C18 spin column and dried on Speed-Vacuum before nanoLC-MS/MS analysis.

LC-MS/MS analysis

Prior to injection, the peptides concentrations in the samples were determined by the Pierce quantitative fluorometric peptide assay (Thermo scientific) according to the manufacturer's instructions. An aliquot containing 1μg for each sample was dried and suspended in 9 μL of FA 0.1% + 1 μL of Cytochrome C digest (2 pmol/μL) used as internal standard. 2μL of this solution was then injected. Samples were analyzed in a Label Free quantitation strategy, in triplicate using an Ultimate 3000 nano-RSLC (Thermo Scientific) coupled on line with a Q Exactive HF mass spectrometer via a nano-electrospray ionization source (Thermo Scientific, San Jose California).

Nano-LC: Samples were injected and loaded on a C18 Acclaim PepMap100 trap-column 75 μm ID x 2 cm, 3 μm, 100Å, (Thermo Scientific) for 3.0 minutes at 5 μL/min with 2% ACN,

0.05% TFA in H_2O and then separated on a C18 Acclaim Pepmap100 nano-column, 50 cm x 75 □m i.d, 2 μm, 100 Å (Thermo Scientific) with a 60 minutes linear gradient from 4% to 50% buffer B (A: 0.1% FA in H_2O, B: 100% ACN, 0.1% FA) and then from 50 to 95% of B in 2 min, hold for 10 min and returned to the initial conditions in 1 min for 14 min. The total duration was set to 90 minutes at a flow rate of 300 nL/min. The oven temperature was kept constant at 40°C.

MS: Samples were analysed with a TOP20 HCD method: MS data were acquired in a data dependent strategy selecting the fragmentation events based on the 20 most abundant precursor ions in the survey scan (300-1600 Th). The resolution of the survey scan was 60,000 at m/z 200 Th and for MS/MS scan the resolution was set to 15,000 at m/z 200 Th. The Ion Target Value for the survey scans in the Orbitrap and the MS/MS scan were set to 3E6 and 1E5 respectively and the maximum injection time was set to 60 ms for MS scan and for MS/MS scan. Parameters for acquiring HCD MS/MS spectra were as follows; collision energy = 27 and an isolation width of 2.0 m/z. The precursors with unknown charge state, charge state of 1 and 8 or greater than 8 were excluded. Peptides selected for MS/MS acquisition were then placed on an exclusion list for 20 s using the dynamic exclusion mode to limit duplicate spectra.

Data Analysis

Proteins were identified by database searching using SequestHT with Proteome Discoverer 2.2 software (Thermo Scientific) against the Swissprot Homo Sapiens database (2018-07 release, 24480 sequences). Precursor mass tolerance was set at 10 ppm and fragment mass tolerance was set at 0.02 Da, and up to 2 missed cleavages were allowed. Oxidation (M), acetylation (Protein N-terminus) and HNE (+ 156.115 Da on K or H) were set as variable modifications, and Carbamidomethylation (C) as fixed modification. Peptides and proteins were filtered with a false discovery rate (FDR) at 1% using percolator and proteins were identified with 1 unique peptide in rank 1. Protein quantitation was performed with Minora feature detector and

precursor ions quantifier node in Proteome Discoverer 2.2 software with protein quantitation based on pairwise ratios and ANOVA (individual proteins) hypothesis test.

The mass spectrometry proteomics data have been deposited to the ProteomeX change Consortium via the PRIDE partner repository with the dataset identifier PXD013301 (DOI: 10.6019/PXD013301; username: reviewer26456@ebi.ac.uk; password: oMD4gyDT).

Molecular modelling

To study the accessibility of residues, the structure of truncated human apolipoprotein A-I (pdb code 1av1) was downloaded from the PDB database. All observations were performed using PyMOL as software by representing the α-helices and histidine and lysine residues (colored in blue) in CPK to facilitate the study. The same method was achieved with the HDL model which was download from the PDB database.

Statistical analysis

Data were expressed as median and interquartile range. All analyses were performed using GraphPad Prism version 6.0 (GraphPad software, La Jolla, CA, USA). Normality was assessed using D'Agostino & Pearson test. Comparisons were performed using Mann-Whitney U-test. Differences were considered as significant at the $P<0.05$ level.

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Supplementary Table 1.

General characteristics of hemodialysis and control patients for aggregation assay

	Control	Hemodialysis (HD)	P-value
N	9	9	
General characteristics			
Age, years	41.78 ± 4.50	57.11 ± 5.60	0.049
Gender, % male	66.67 ± 16.67	55.56 ± 17.57	0.653
BMI, kg/m^2	25.42 ± 1.06	22.89 ± 1.43	0.186
Comorbidities			
HT, n	2	8	
Stroke, n	0	0	
CHD, n	0	2	
Cardiopathy, n	0	4	
PVD, n	0	1	
Therapies			
Statins, n	0	6	
PI, n	0	5	
RASi, n	1	3	
ß-blockers, n	1	5	
CCB, n	1	1	
Biological parameters			
Urea, mmol/L	6.20 ± 0.51	15.00 ± 1.62	<0.0001
Creatinine, μmol/L	78.78 ± 4.70	612.10 ± 52.95	<0.0001
mGFR, mL/min/1.73m^2	92.33 ± 4.08	-	
Total cholesterol, mg/dL	211 ± 9	153 ± 18	0.011
LDL cholesterol, mg/dL	132 ± 8	87 ± 16	0.025
HDL cholesterol, mg/dL	58 ± 2	44 ± 3	0.005
Triacylglycerols, mg/dL	105 ± 10	109 ± 9	0.749
CRP, mg/L	3.00 ± 1.13	18.83 ± 11.29	0.200

Data are expressed as mean ± SEM. BMI: body mass index; HT: hypertension; CHD: coronary heart disease; PVD: peripheral vascular disease; PI: platelet inhibitor; RASi: renin-angiotensin system inhibitor; CCB: calcium-channel blocker; eGFR: estimated glomerular filtration rate by CKD-EPI formula; mGFR: measured GFR by iohexol clearance; CRP: C-reactive protein. Creatinine: x *0.011 for mg/dl; urea:* x *2.8 for mg/dl*

Supplementary Figure 1

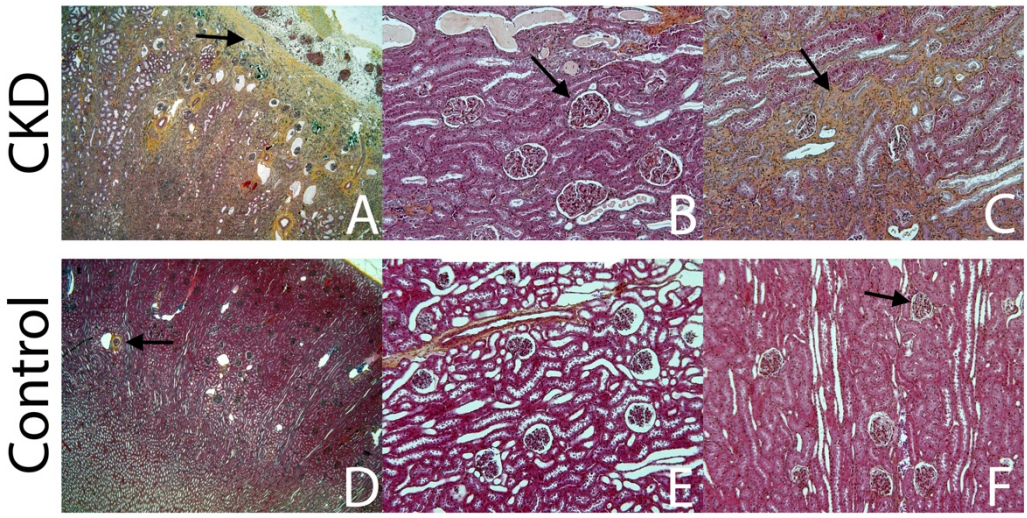
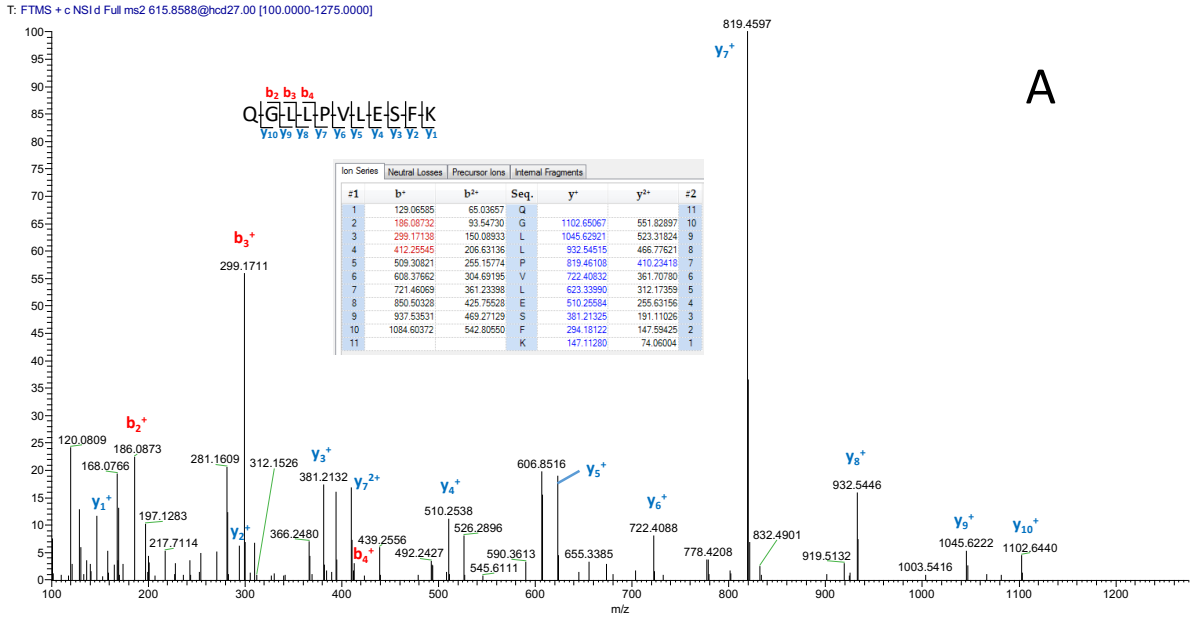


Figure 1: Histological examinations of rabbit kidneys with hematoxylin phloxine saffron (HPS) staining.
A: The electrocoagulated cortex was replaced by a fibrous layer (black arrow) and some glomeruli were surrounded by fibrosis. (x2.5)
B: Remnant glomeruli exhibiting hypertrophy (black arrow; x10). Mean glomerular radius was 23 in control group vs 29 in CKD group ($p<0.05$)
C: Peritubular fibrous involution (black arrow) (x10)
D: Note the large number of of glomeruli and the fibrosis restricted to perivascular areas (black arrow) (x2,5)
E — F: Glomeruli were smaller (black arrow) than in CKD group and fibrosis was restricted to perivascular areas (x10).

Supplementary Figure 2

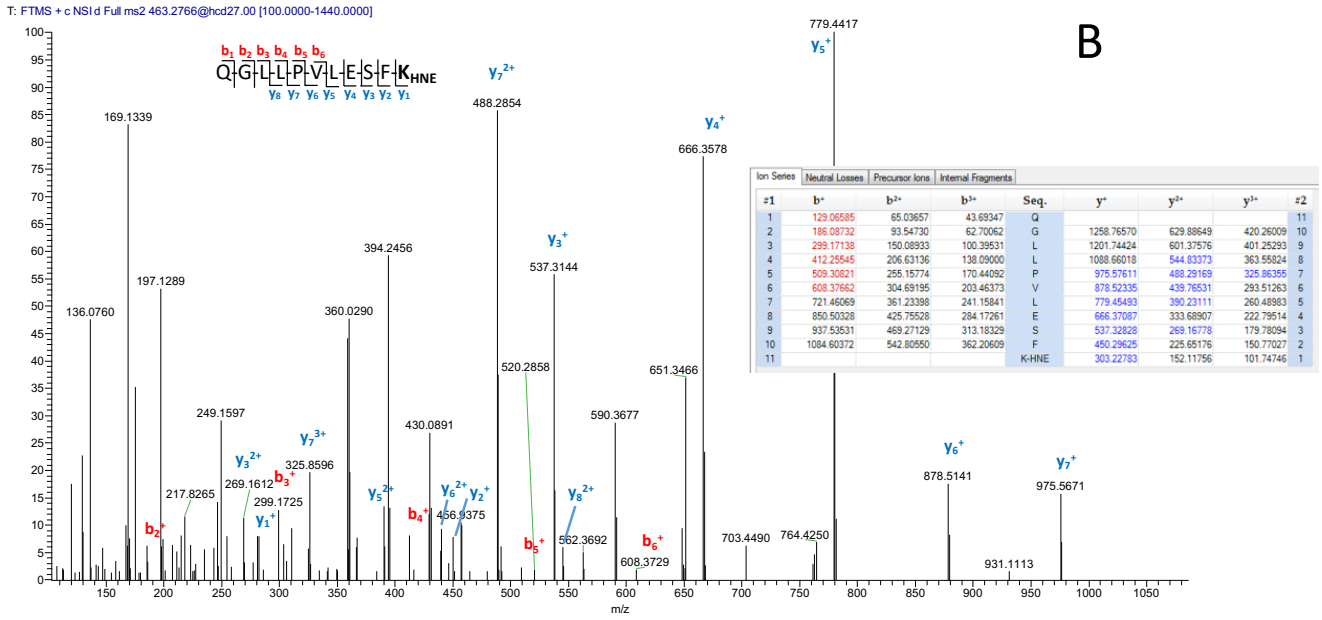
K250: A: unmodified peptide; B: modified peptide

Peptide [241-250] : MH^+ : 1230.7110 Th - **MSMS on** m/z 615.8591 (2^+ ; *+1.41 ppm*)



z1	b+	b2+	Seq.	y+	y2+	z2
1	129.06585	65.03657	Q			11
2	186.08732	93.54730	G	1102.65067	551.82897	10
3	299.17138	150.08933	L	1045.62921	523.31824	9
4	412.25545	206.63136	L	932.54515	466.77621	8
5	509.30821	255.15774	P	819.46108	410.23418	7
6	608.37662	304.69195	V	722.40832	361.70780	6
7	721.46069	361.23398	L	623.33990	312.17359	5
8	850.50328	425.75528	E	510.25584	255.63156	4
9	937.53531	469.27129	S	381.21325	191.11026	3
10	1084.60372	542.80550	F	294.18122	147.59425	2
11			K	147.11280	74.06004	1

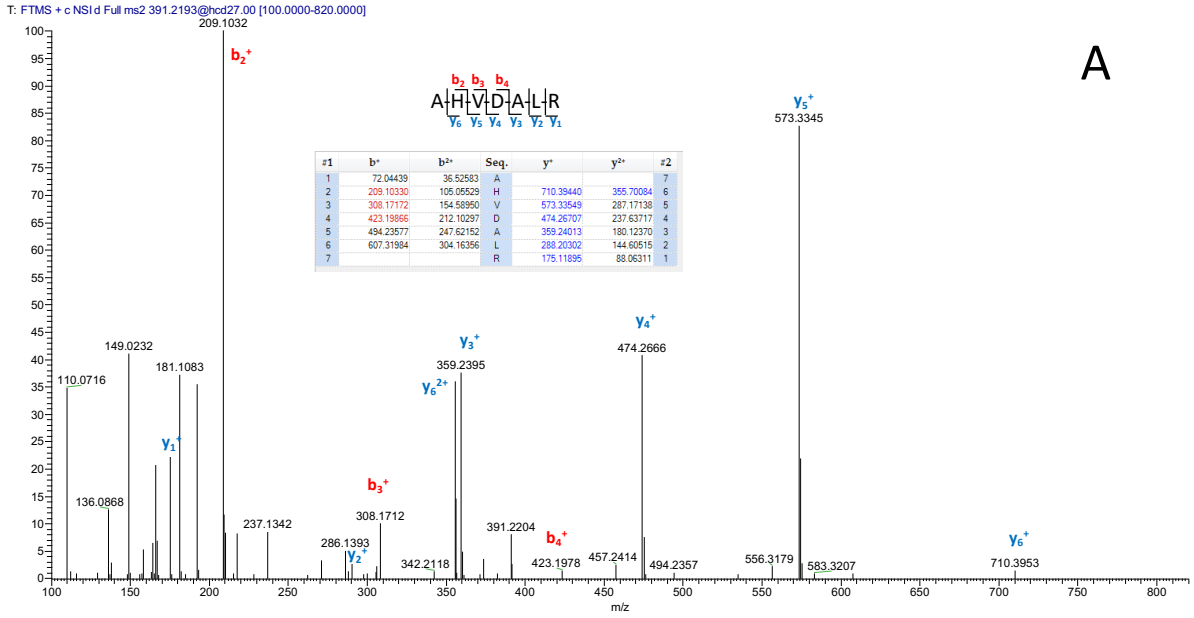
Peptide [241-250] HNE Modification : MH^+ : 1386.8163 Th - **MSMS on** m/z 462.9436 (3^+ ; *+5.79 ppm*)



z1	b+	b2+	b3+	Seq.	y+	y2+	y3+	z2
1	129.06585	65.03657	43.69347	Q				11
2	186.08732	93.54730	62.70062	G	1258.76570	629.88649	420.26009	10
3	299.17138	150.08933	100.39531	L	1201.74424	601.37576	401.25293	9
4	412.25545	206.63136	138.09000	L	1088.66018	544.83373	363.55824	8
5	509.30821	255.15774	170.44092	P	975.57611	488.29169	325.86355	7
6	608.37662	304.69195	203.46373	V	878.52335	439.76531	293.51263	6
7	721.46069	361.23398	241.15841	L	779.45493	390.23111	260.48983	5
8	850.50328	425.75528	284.17261	E	666.37087	333.68907	222.79514	4
9	937.53531	469.27129	313.18329	S	537.32828	269.16778	179.78094	3
10	1084.60372	542.80550	362.20609	F	450.29625	225.65176	150.77027	2
11				K-HNE	303.22783	152.11756	101.74746	1

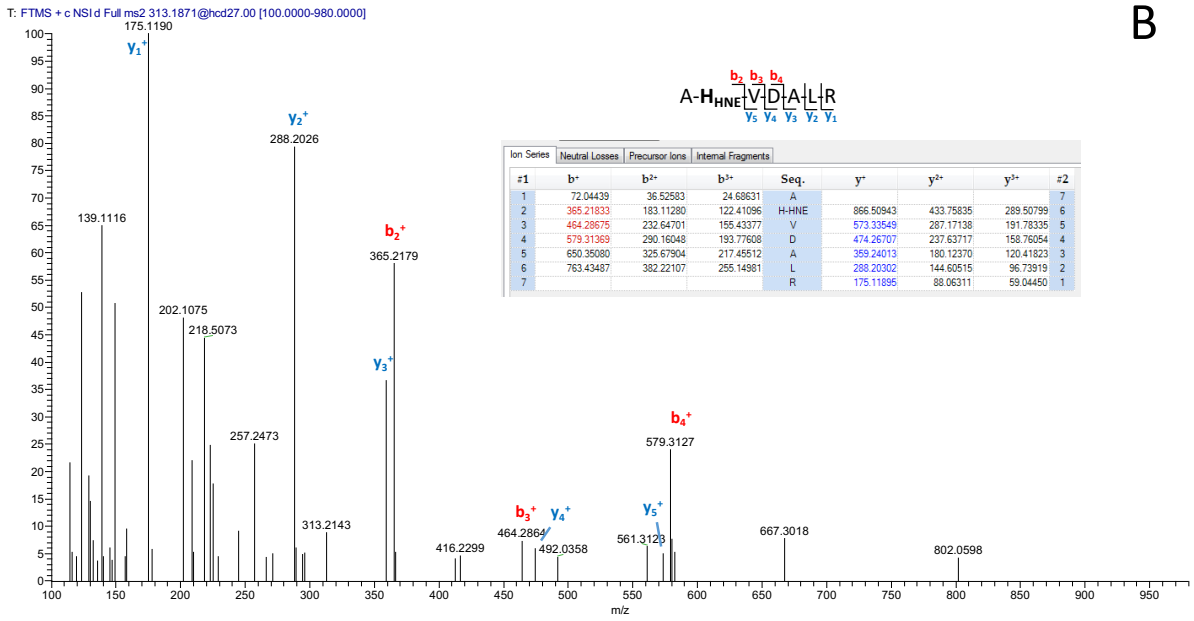
H179: A: unmodified peptide; B: modified peptide

Peptide [178-184] : MH⁺ : 781.4313 Th - **MSMS on** m/z 391.2193 (2^+ ; *-0.24 ppm*)



#1	b⁺	b²⁺	Seq.	y⁺	y²⁺	#2
1	72.04439	36.52583	A			7
2	209.10330	105.05529	H	710.39440	355.70084	6
3	308.17172	154.58950	V	573.33549	287.17138	5
4	423.19866	212.10297	D	474.26707	237.63717	4
5	494.23577	247.62152	A	359.24013	180.12370	3
6	607.31984	304.16356	L	288.20302	144.60515	2
7			R	175.11895	88.06311	1

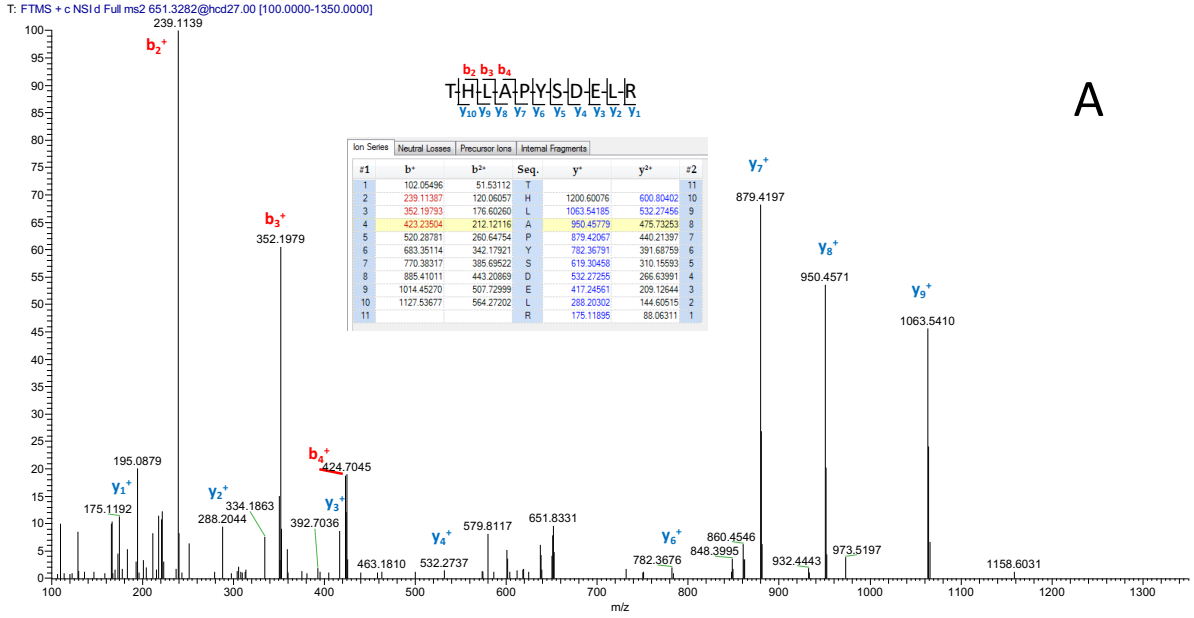
Peptide [178-184] HNE modification : MH⁺ : 937.5467 Th - **MSMS on** m/z 313.1871 (3^+ ; *+0.13 ppm*)



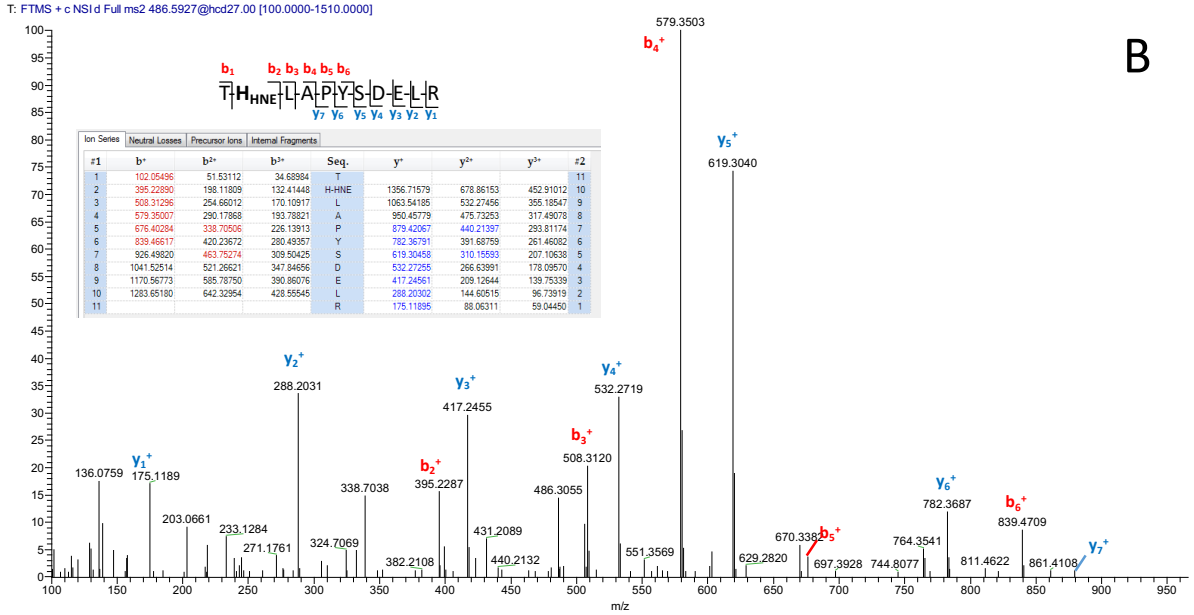
#1	b⁺	b²⁺	b³⁺	Seq.	y⁺	y²⁺	y³⁺	#2
1	72.04439	36.52583	24.68631	A				7
2	365.21833	183.11280	122.41096	H-HNE	866.50943	433.75835	289.50799	6
3	464.28675	232.64701	155.43377	V	573.33549	287.17138	191.78335	5
4	579.31369	290.16048	193.77608	D	474.26707	237.63717	158.76054	4
5	650.35080	325.67904	217.45512	A	359.24013	180.12370	120.41823	3
6	763.43487	382.22107	255.14981	L	288.20302	144.60515	96.73919	2
7				R	175.11895	88.06311	59.04450	1

H189: A: unmodified peptide; B: modified peptide

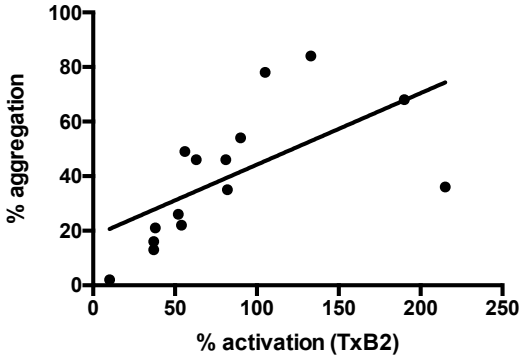
Peptide [185-195] : MH$^+$: 1301.6486 Th - **MSMS on** m/z 651.3279 (2^+ ; *+0.13 ppm*)



Peptide [185-195] HNE Modification : MH$^+$: 1457.7636 Th - **MSMS on** m/z 486.5927 (2^+ ; *+0.08 ppm*)

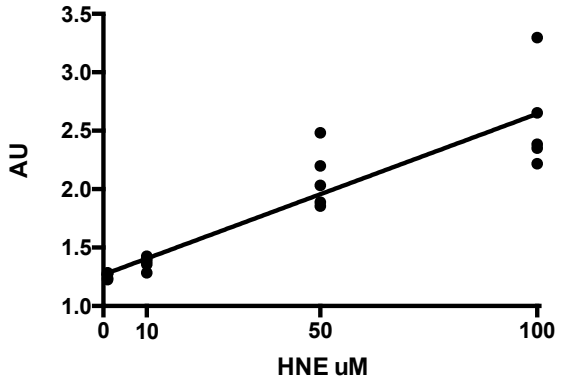


Supplementary Figure 3



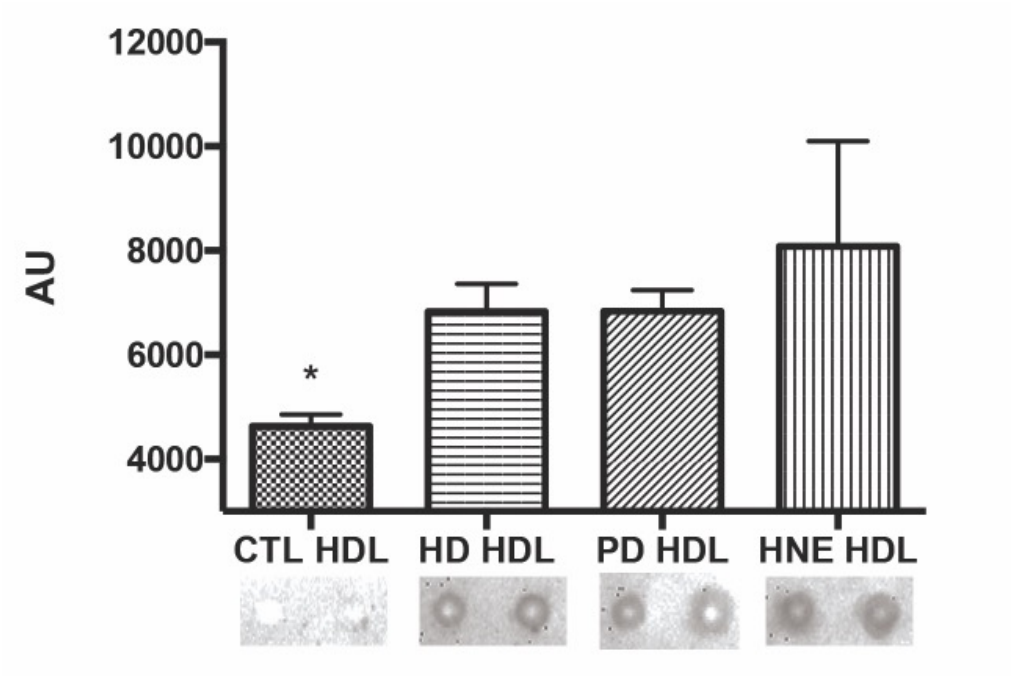
Spearman correlation of TxB2 assay and aggregation assay of 7 control patients and 8 hemodialysis patients.
r^2=0.3894, p=0.0129

Supplementary Figure 4



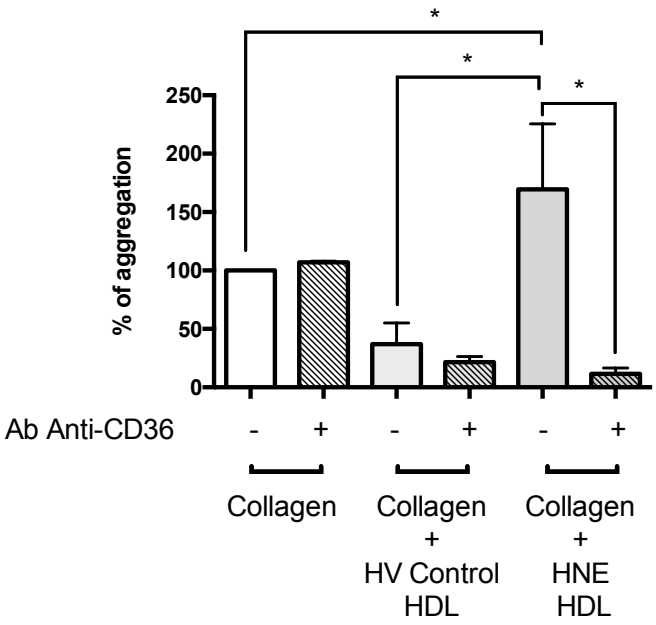
Sperman correlation of the amount of 4-HNE adducts on HDL from in vitro adduction. Significant correlation was observerd (p=0.0179)

Supplementary Figure 5



4-HNE adducts are increased in HDL from hemodialysis (HD), peritoneal dialysis (PD) patients and 4-HNE in vitro modified HDL (HNE) compared to controls (CTL). No difference were however observed in the level of adduction between HD, PD and HNE HDL. Immunoblotting (dot blot) of 4-hydroxy-nonenal (4-HNE) Michael adducts were performed as described in methods section. n=5 for each group *, $p<0.05$, Mann-Whitney U test vs HD, PD and HNE HDL

Supplementary Figure 6



Healthy volunteers (HV) HDL modified by an incubation overnight with HNE (HNE HDL) solution exhibited pro-aggregant properties compared to non-modified HDL (HV Control HDL, C). Pre-incubation with anti-CD36 antibody restored HNE HDL anti-aggregant properties like HV Control HDL (n=4 for each group). *p<0.05