LIPOPEROXIDATION IN CHRONIC PANCREATITIS - preliminary data

M. Vecka[1,2], A. Žák[1,2], L. Vávrová[1,2], J. Kodydková[1,2], J. Macášek[1], M. Urbánek[1], T. Krechler[1], B. Staňková[1,2]

[1/]Charles University in Prague, 1st Faculty of Medicine, 4th Department of Internal Medicine, and [2/] Institute of Clinical Biochemistry and Laboratory Diagnostics, General Teaching Hospital in Prague, the Czech Republic

Background

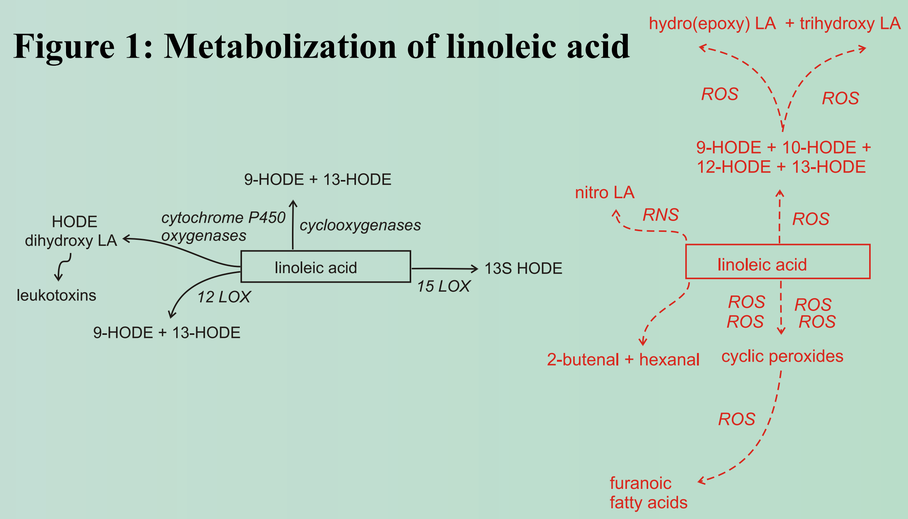
Oxidative stress is connected with pathogenesis of many diseases of gastrointestinal tract, including chronic pancreatitis (ChP). In the idiopathic form of ChP, nutritional effects could also play a role. It is known that increased levels of various classes of fatty acids exhibit lipotoxic effects on pancreas. The initial stages of ChP include damage of pancreatic acinar cells and activation of stellar cells, which is caused by fat (VLDL). The role of oxidative stress (OS) in the development of ChP was confirmed by the findings of activated antioxidant system indices in patients with ChP , though the indirect markers of OS are not usually elevated.

Fatty acids, the important part of molecules in lipid classes in VLDL particles, are metabolized into many derivatives (Figure 1), some of them being useful indices of OS. Recent data indicate that metabolites of linoleic acid (18:2n-6), HODEs, are superior to analogues from arachidonic acid (20:4n-6), HETEs, in predicting the OS damage in ChP.

Aim of the Study

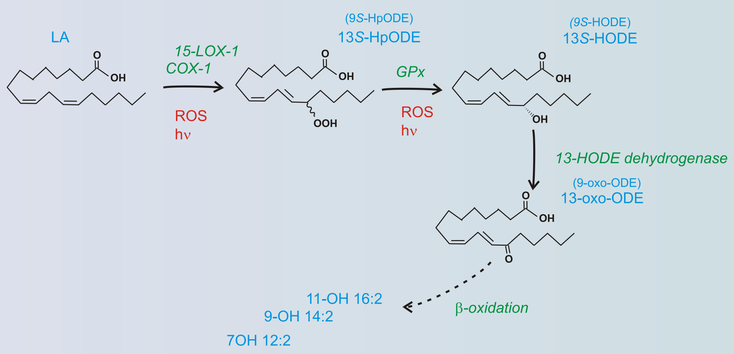
We analysed selected parameters of lipoperoxidation in patients with ChP, including lipoperoxidation products of linoleic acid, hydroxyoctadecadienoic acids (HODEs).

Figure 1: Metabolization of linoleic acid

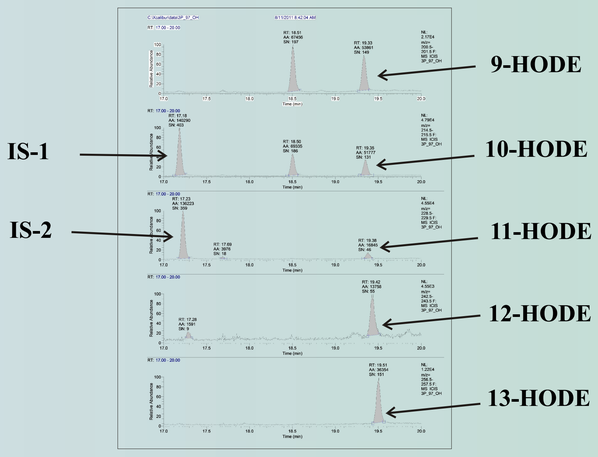


In the Figure, the metabolic pathways for linoleic acid are summarized. Linoleic acid can be modified either by enzymatical systems (left, black part), or nonenzymatically (right, red part). Abbreviations: HODE - hydroxyoctadecadienoic acids, LA - linoleic acid, LOX - lipoxygenase, ROS - reactive oxygen species, RNS - reactive nitrogen species.

Figure 2: Metabolization of LA into HODE



Scheme 1: HODE analysis by GC-MS



Participants

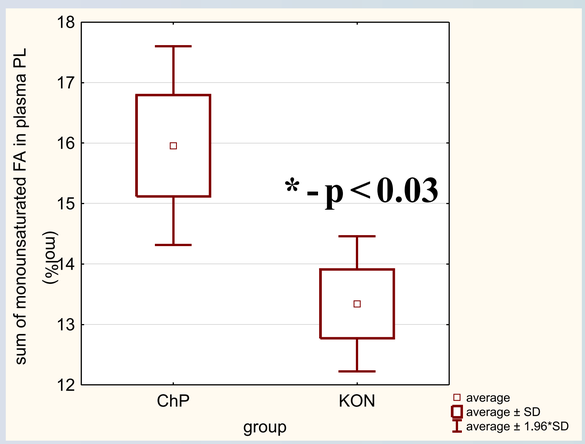
The study involved 12 control (CON) subjects (10M/2F) and 20 patients (19M/1F) with morphologically confirmed ChP (ultrasonography, ERCP) and determination of exocrine capacity (FELA test). Of these patients, 18 were smokers and 8 had type II DM.

Methods

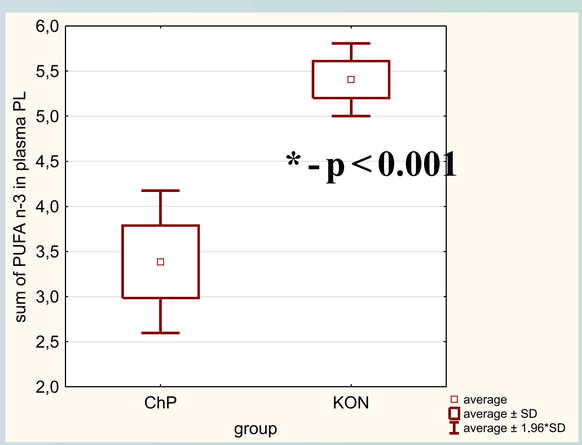
Common biochemical parameters were analyzed by routine methods (enzymatic-colorimetric, EIA, RIA), the level of conjugated dienes in LDL particles was assayed spectrophotometrically at 234 nm, the activities of antioxidant enzymes with colorimetric methods.

The fatty acid profile of plasma phospholipids was assessed by gas chromatography, the concentrations of HODE were assayed with GC-MS method. Statistical analysis was performed with STATISTICA® for Windows (StatSoft, Inc., Tulsa, OK, U.S.A., 1999).

Figure 3: Fatty acid content in plasma lipid classes



monounsaturated FA



PUFA n-3

Results

The CON group did not differ from ChP in age, BMI, nor in basic biochemical parameters except for higher plasma triacylglycerols ($p<0.05$), and glucose ($p<0.01$) (Tables 1 and 2). The patients with ChP had higher plasma concetrations of total HODEs ($p<0.01$), mainly due to the elevated concentrations of 9-HODE ($p<0.001$), see Table 3. In the profiles of plasma lipid classes, we observed lower content of n-3 polyunsaturated fatty acids in phospholipids ($p<0.001$) and higher content of monounsaturated FA ($p < 0.05$)(Figure 3) . The concentrations of HODEs correlated negatively with the activities of antioxidant system enzyme - paraoxonase, and concentrations of reduced glutathione in ChP group (Table 4).

Table 1: Basic characteristics of studied groups I

parameter	CON - controls (n = 12)	ChP - chronic pancreatitis (n = 20)
age (years)	61 ± 9	55 ± 8[§]
BMI (kg.m^{-2})	26.4 ± 4.9	24.3 ± 5.5
TC (mmol/L)	4.77 ± 0.72	5.00 ± 0.96
HDL-C (mmol/L)	1.46 ± 0.26	1.57 ± 0.51
LDL-C (mmol/L)	2.88 ± 0.68	2.78 ± 0.68
TAG (mmol/L)	0.95 ± 0.46	1.46 ± 0.83*
glucose (mmol/L)	4.95 ± 0.74	7.36 ± 2.90**
NEFA (mmol/L)	0.55 ± 0.21	0.72 ± 0.56
bilirubin (umol/L)	12.8 ± 5.8	12.3 ± 5.8

data are in average ± SD format;[§] - Welch test; *p < 0.05, **p < 0.01

Table 2: Basic characteristics of studied groups II

parameter	CON - controls (n = 12)	ChP - chronic pancreatitis (n = 20)
LDL-CD (umol/L)	47.5 ± 11.9	50.5 ± 16.2[§]
oxLDL (U/L)	93 ± 27	90 ± 28
CRP (mg/L)	4.53 ± 6.80	7.70 ± 6.70[x]
procalcitonin (ug/L)	3.2 ± 1.1	6.1 ± 2.7***
vitamin A (mg/L)	0.53 ± 0.33	0.90 ± 0.48
vitamin E (mg/L)	12.4 ± 9.4	12.6 ± 6.0
homocysteine (umol/L)	16.5 ± 4.0	16.9 ± 6.6
vitamin B_{12} (nmol/L)	433 ± 225	400 ± 143

data are in average ± SD format;[§] - Welch text; [x] - Wilcoxon unpaired test; *p < 0.05, **p < 0.01, ***p < 0.001; LDL-CD - conjugated dienes in LDL, CRP - C reactive protein; oxLDL - oxidatively modified LDL

Table 3: Concentrations of HODEs

parameter	CON - controls (n = 12)	ChP - chronic pancreatitis (n = 20)
9-HODE (nmol/L)	229 ± 43	530 ± 286***[§]
10-HODE (nmol/L)	143 ± 35	353 ± 272**
11-HODE (nmol/L)	52 ± 19	213± 208*
12-HODE (nmol/L)	124 ± 70	148 ± 83
13-HODE (nmol/L)	121 ± 55	189 ± 174
9+13-HODE (nmol/L)	349 ± 88	717 ± 454*
sum of HODE (nmol/L)	668 ± 140	1270 ± 1056

data are in average ± SD format;[§] - Welch test: *p < 0.05, **p < 0.01, ***p < 0.001

Table 4: HODEs and selected parameters of oxidative stress

parameter	CRP	CD-LDL	oxLDL	GPx	PON	GSH
9-HODE (nmol/L)	0.54*	0.02	-0.09	-0.33	-0.41*	-0.49*
10-HODE (nmol/L)	0.60*	-0.03	0.17	-0.16	-0.52*	-0.53*
11-HODE (nmol/L)	0.43*	0.07	0.26	-0.05	-0.23	-0.43*
12-HODE (nmol/L)	0.36	0.16	0.06	-0.02	-0.50*	-0.33
13-HODE (nmol/L)	0.49*	-0.13	0.04	-0.18	-0.26	-0.47*

Spearmann coefficient of correlation: *p < 0.05, n = 20 (group ChP)

Conclusions

Although in our studied ChP group, there are not elevated commonly analyzed markers of oxidative stress, these patients exhibit raised concentrations of HODEs, which are probably connected with enzymatic antioxidant system.

Acknowledgement

The work was supported by the project RVO–VFN64165/2013 and Research project of Charles University in Prague, 1st Faculty of Medicine – PRVOUK-P25/LF1/2

Marek Vecka, Ph.D.; email: marvec@volny.cz