Analysis of HTG gene score in patients with familial dysbetalipoproteinemia

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Introduction – definition

- **FD**, **hyperlipoproteinemia III**
- **AR** (10% AD), **polymorphisms *APOE***
- **genotype** - ***apoE2/E2*** (90%)
- **phenotype** - mixed DLP in context of another trigger factors (genetics?, T2D, NS, ...) – 5-15%

Koopal CH at al. DOI: 10.1097/MED.0000000000000316
Mahley RW et al. Annu Rev Genomics Hum Genet. 2000;1:507–537

Importance of FD (ASCVD and GIT)

- **prevalence:** 0.12 – 0.4%, up to 25% diagnosed
- in the **CR up to 10 000** patients with FD
- **10x higher risk** of premature manifestation of **ASCVD** – **CAD, PAD** (CAD – 3 %, 29 %, 47 %, PAD – 20 %)
- relationship between ***APOE* genotype, LDL-C and risk of ASCVD**: ***apoE2/E2*** > *apoE2/E3* > *apoE2/E4* > *apoE3/E3* > *apoE3/E4* > ***apoE4/E4***
- HTG causes up to **4% of all acute pancreatitis**

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Ramasamy I. DOI: 10.1016/j.cca.2015.10.033

Pitfalls of diagnostics

- gold standard – **ultracentrifugation of lipoproteins** (only 11 patients), **PGGE**

- **mixed DLP - dg. criteria**

Red flags - FD awareness

- **TC/TG**................1(2):1 (TC > 5mmol/l, TG > 3mmol)
- **nonHDL/apoB**....> 5
- **apoB/TC**............< 0.15
- **apoB algorithm** (apoB < 1.2g/l, TG > 2.3mmol/l, TG/apoB < 10, TC/apoB > 6.2)
- **clinical findings** – PAD, palmar xanthomas

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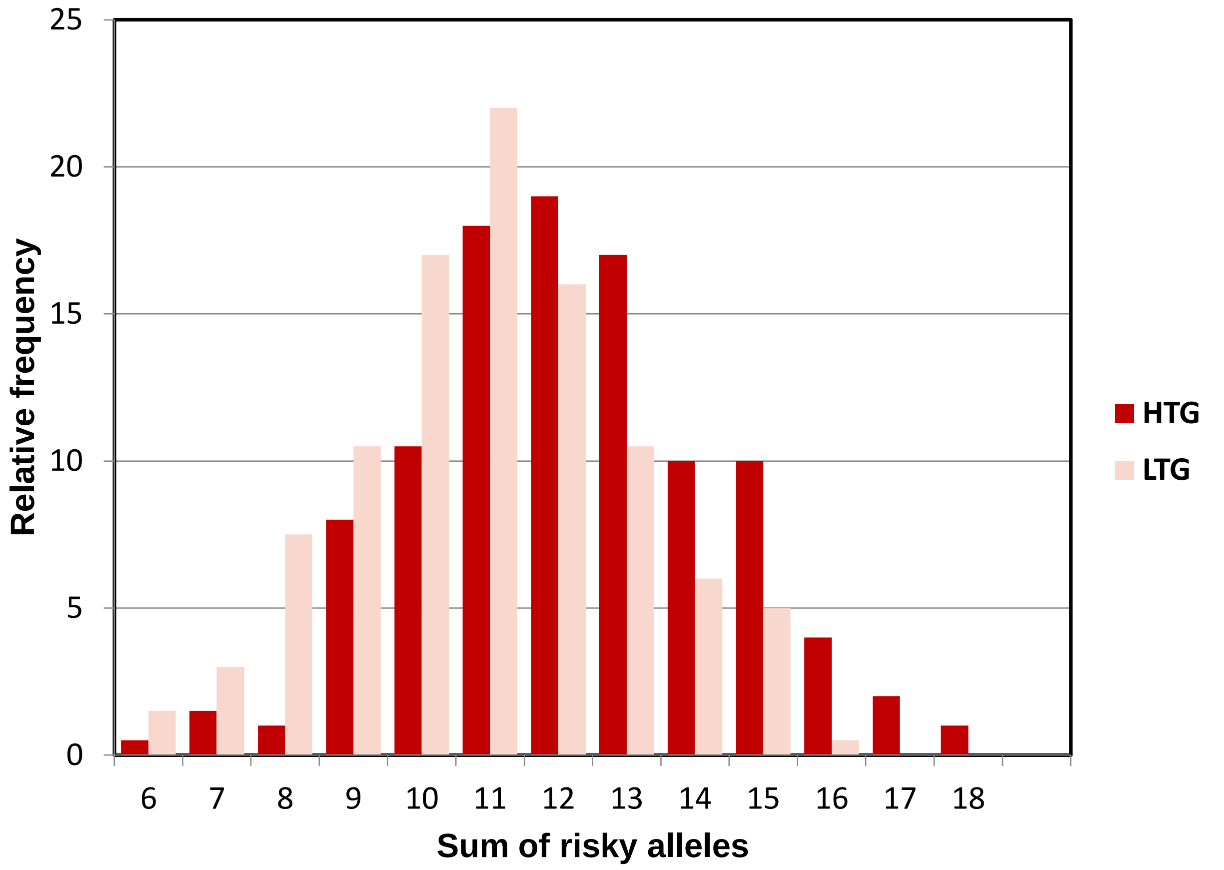
Study goal

- evaluation of the dg. criteria
- analysis of HTG gene score in patients with *apoE2/E2* as a possible determinant of FD development

Background

Gen	**SNP**	**P**	**OR (95%CI)**
APOA5	rs964184	.000001	14.9 (8.9-25.2)
FRMD5	rs2929282	.03	2.0 (1.2-3.5)
GCKR	rs1260326	.00001	2.7 (1.7-3.7)
CAPN3	rs2412710	.000005	2.8 (1.2-4.4)
TRIB1	rs2954029	.00001	1.7 (1.2-2.2)
MLXIPL	rs7811265	.05	1.4 (1.0-2.0)
LPL	rs12678919	.005	2.3 (1.3-4.4)
LIPC	rs261342	.00001	2.1 (1.5-2.6)
GALNT2	rs1321257	n.s.	-
TYW1B	rs13238203	.01	3.9 (1.2-12.8)
ANGPLT3	rs2131925	.03	1.5 (1.1-2.3)
COBLL1	rs10195252	.05	1.2 (1.0-1.6)
CILP2	rs10401969	.05	1.5 (1.0-2.3)
NAT2	rs1495743	.01	1.5 (1.1-2.1)
PLPT	rs4810479	.05	1.4 (1.0-2.6)
FADS1/2/3	rs174546	n.s.	-
TIMD4	rs1553318	.05	1.3 (1.0-2.1)
CETP	rs7205804	.01	1.7 (1.1-2.6)



Hubacek JA, Satny M, Vrablik M et al. The Gene Score for Predicting Hypertriglyceridemia: New Insights from a Czech Case–Control Study. Molecular Diagnosis & Therapy [online]. 2019, 23(4), 555-562. DOI: 10.1007/s40291-019-00412-2.

Methods

- **71** patients with **apoE2/E2 + DLP** x **90** controls **apoE2/E2 without DLP** (biobank post-MONICA a HAPIEE studies)
- analyzed polymorphisms: **CYP26A1, LRP1, MAP3K1, APOE, CTF1, GCKR, APOA5, LIPC, LPL, TRIB1, NAT2, CILP2, GALNT2, CETP, HLA, TYW1B, FRMD5, CAPN3**
- genetic analysis of SNPs - **PCR-RFLP** or **realtime-PCR**
- data processed by descriptive statistics methods

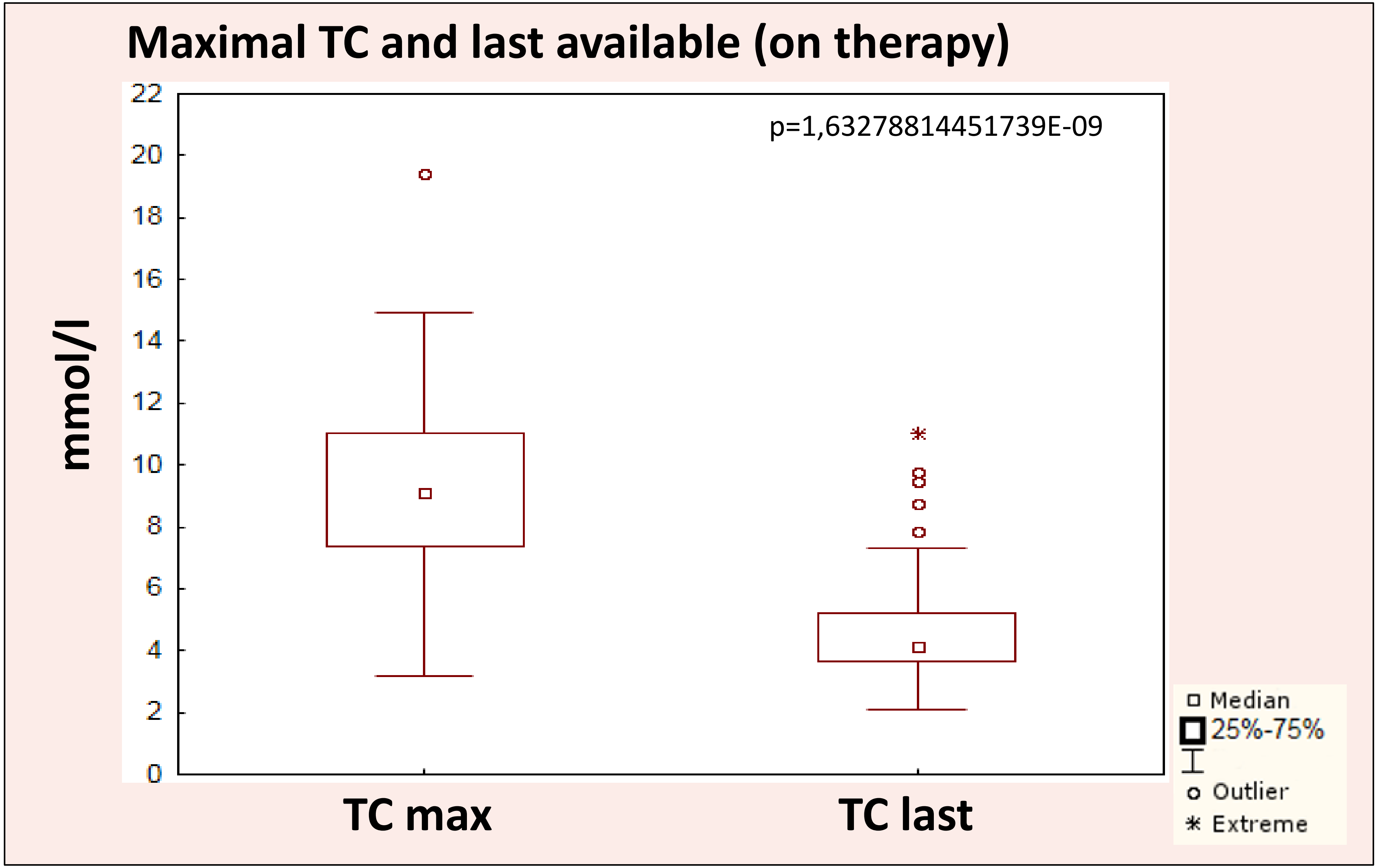
Characteristics of study population - FD

- **71** patients - **apoE2/E2 with DLP**

	Median (± SD)
Age (years)	62 ± 16,5
Weight (kg)	84,6 ± 16,0
WC (cm)	97,0 ± 10,8
BMI (kg/m^2)	30,6 ± 4,5

	No. (%)
Alcohol abuse	16 (22.8 %)
Smoking	25 (35.7 %)
Family history ASCVD	23 (32.9 %)
T2D	19 (27.2 %)
Art. hypertension	32 (47.2 %)
CAD	14 (20 %)
PAD	9 (12.9 %)
Stroke	4 (5.7 %)

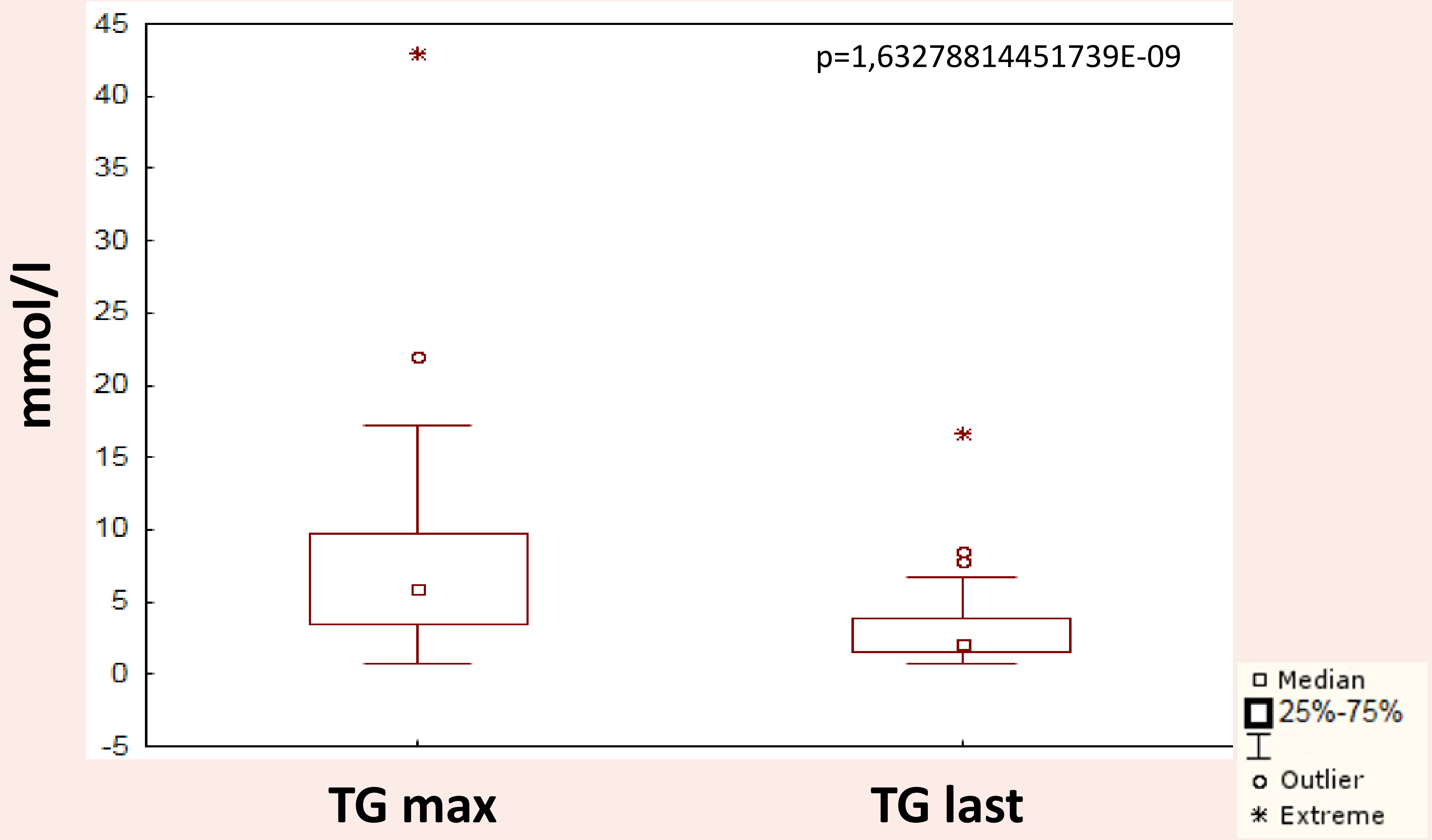
Laboratory findings - FD



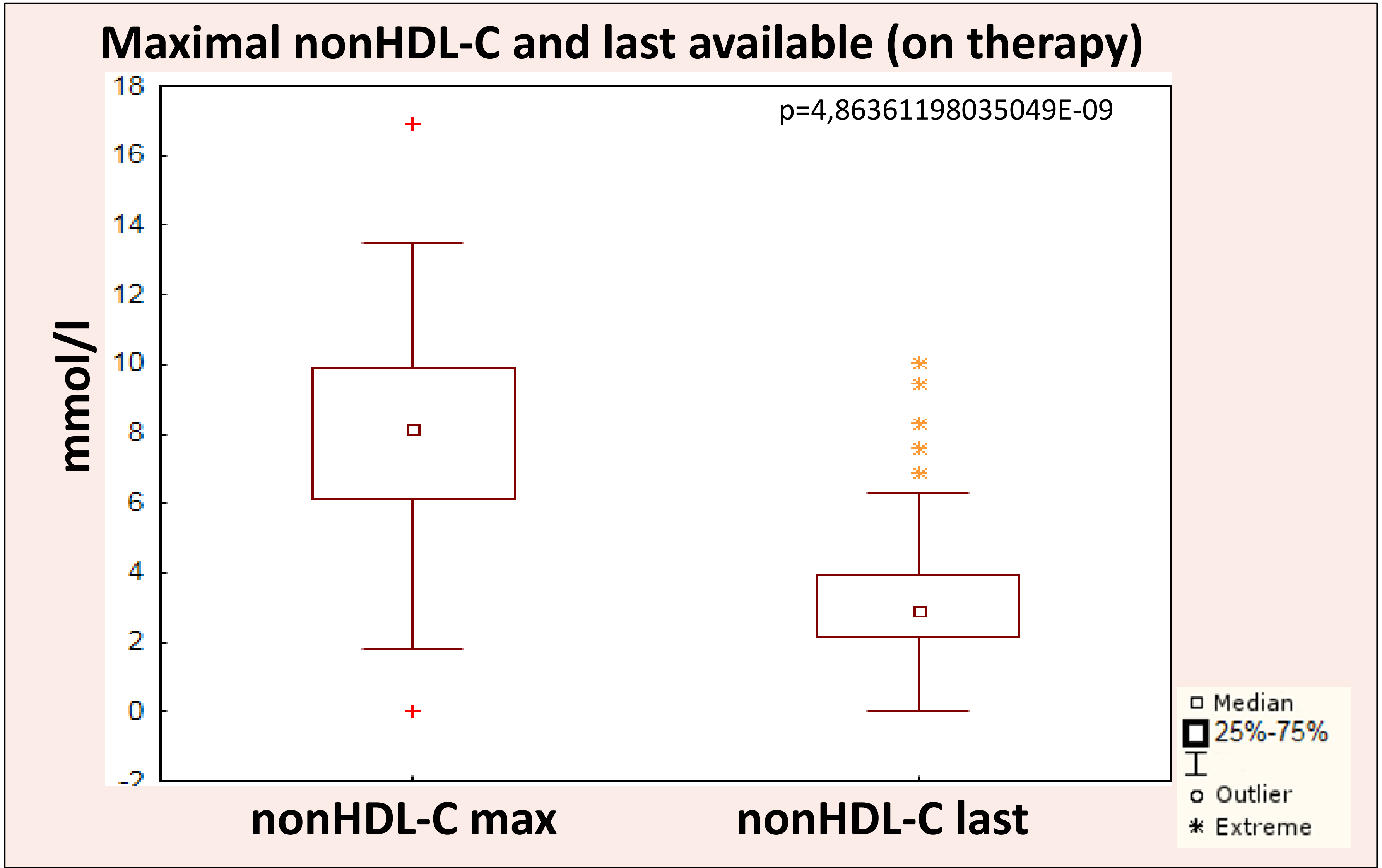
Maximal TC and last available (on therapy)

Laboratory findings - FD

Maximal TG and last available (on therapy)



Laboratory findings - FD



Maximal nonHDL-C and last available (on therapy)

p=4,86361198035049E-09

Characteristics of population – controls

- **90** controls **apoE2/E2 without DLP**

	Controls
Number (% males)	90 (50)
Age (years)	52.6 ± 7.4
Alcohol abstinence	- -
Active smoking	32%
Arterial hypertension	- -
Type 2 diabetes	6%
CAD in anamnesis	0%
PAD	0%
Ischemic stroke	0%
BMI (kg/m^2)	28.1 ± 4.6
Total cholesterol (mmol/l)	4.9 ± 1.2
Triglycerides (mmol/l)	2.0 ± 1.0

Evaluation of dg. criteria

- patients with definitive FD (ultracentrifugation, *apoE* - 11) and non-definite FD (*apoE*, clinical dg. - 60)

	non-definite FD	**definite FD**	**significance**
TC/TG	84%	100%	p > 0,05
nonHDL/apoB	96%	100%	p > 0,05
apoB/TC	85%	90%	p > 0,05
apoB algorithm	43%	60%	p > 0,05

Analysis of HTG gene score

Analysis of HTG gene score

- analysed **18 SNPs,** used **5 SNPs**

- for score calculation:
  - **risky homozygote** = 2
  - **heterozygote** = 1
  - **non-risky homozygote** = 0

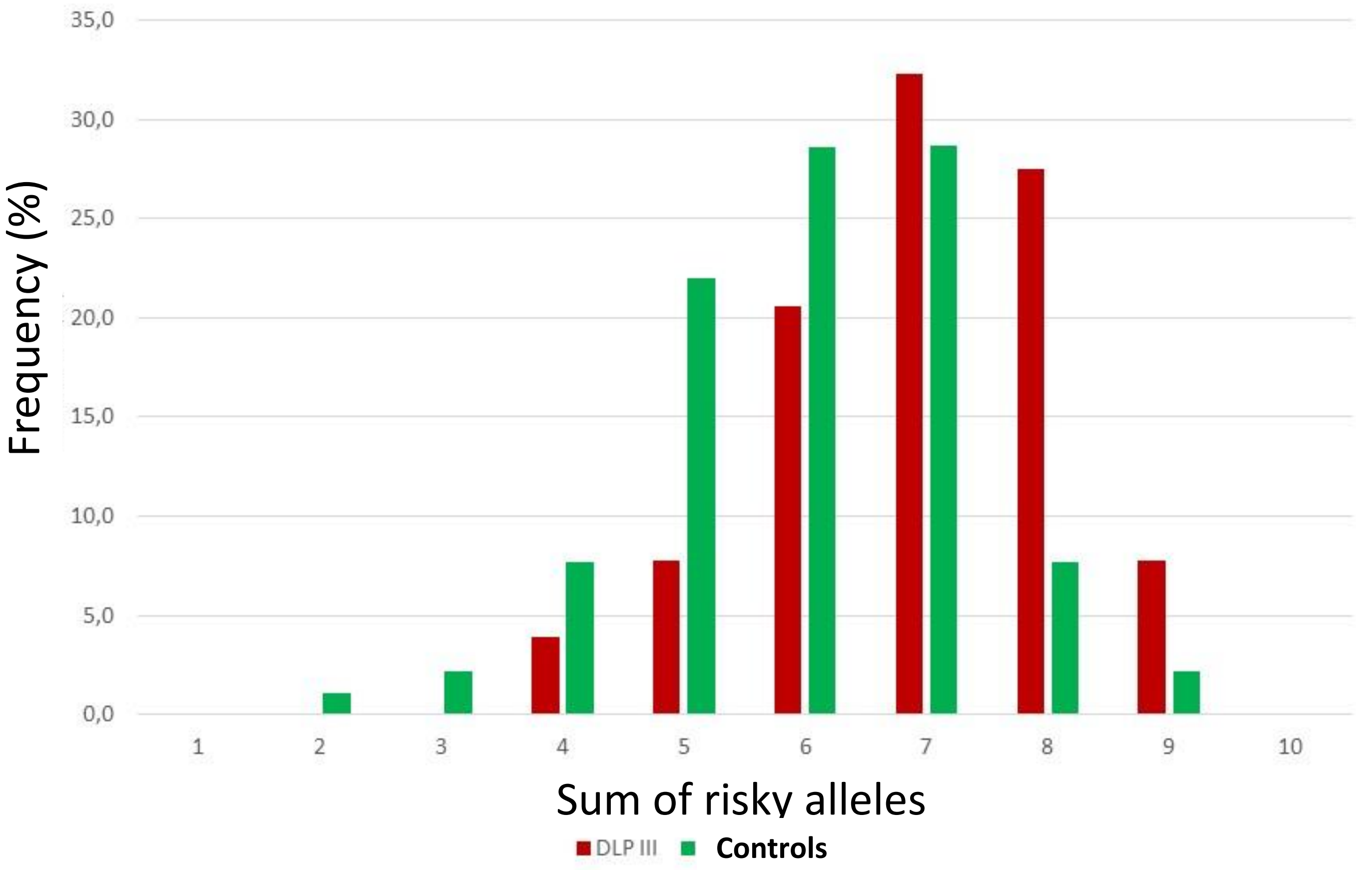
- only **variants with OR > 1.75** were used for score analysis

- created as **unweighted score**

Analysis of HTG gene score

Gen	SNP/Enzym	Varianta	OR	95% CI
APOE	rs439401	CC vs TC	9.09	2.01-41.20
APOA5	Bsp143I	CC vs GG	7.13	1.52-33.52
CTF1	rs11649653	GG vs CC	2.32	0.91-5.91
LPL	rs12678919	AA vs G+	1.96	0.84-4.58
MAP3K1	rs9686661	CC vs TT	1.76	0.35-7.54
LRP1	rs11613352	TT vs CC	1.49	0.34-6.51
HLA	rs2247056	TT vs CC	1.48	0.50-4.35
CETP	rs7205804	GG vs AA	1.46	0.69-3.09
LIPC	TaiI	GG vs CC	1.35	0.58-3.15
NAT2	rs1495743	GG vs CC	1.35	0.40-4.52
CYP26A1	rs2068888	GG vs AA	1.29	0.56-2.95
GCKR	MspI	CC vs TT	1.25	0.56-2.81
GALNT2	rs1321257	GG vs AA	1.16	0.48-2.82
TRIB1	rs2954029	AA vs TT	1.01	0.47-2.21
CILP2	TaaI	TT vs +C	0.59	0.25-1.41

HTG gene score (5 SNPs)



P < 0,00003

OR 95CI

4.97 (2.24 – 11.056)

Summary

- Diagnosed up to 25%, significantly higher risk of ASCVD and AP
- Selection of patients suitable to futher testing – **nonHDL/apoB ratio**
- **HTG gene score (5 SNPs involved in TG metabolism) is another significant predictor of FD development in patients with *apoE2/E2***
- HTG gene score in **all patients with *apoE2/E2* – patients at FD risk**

Thank you for your attention!