SERVIZIO SANITARIO REGIONALE
EMILIA-ROMAGNA
Azienda Unità Sanitaria Locale di Reggio Emilia

Arcispedale S. Maria Nuova
Istituto in tecnologie avanzate e modelli assistenziali in oncologia
Istituto di ricovero e cura a carattere scientifico

RUOLO DEL LABORATORIO NELLA DIAGNOSI DELLE PIÙ FREQUENTI DISLIPIDEMIE

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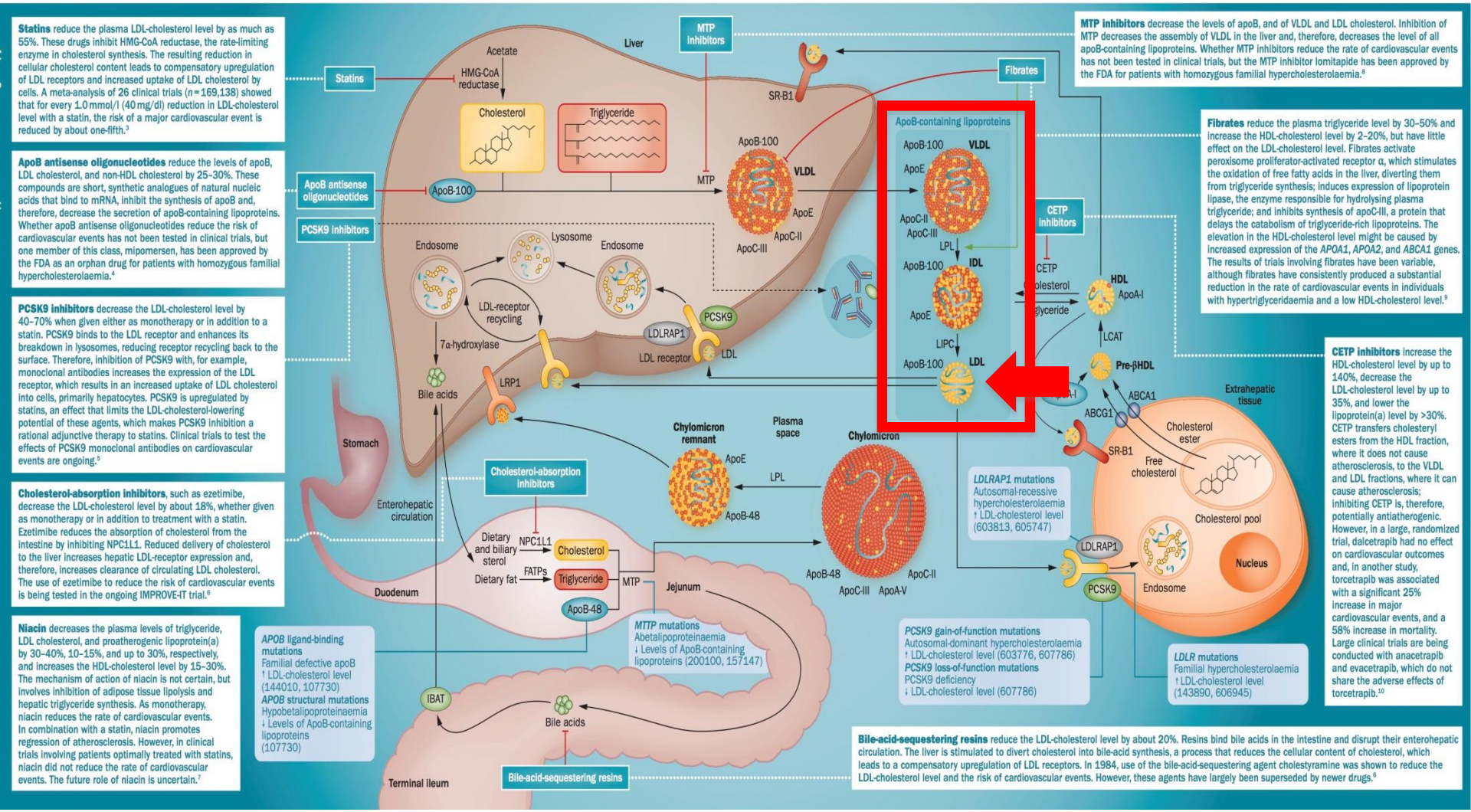
Presente e futuro della Medicina di Laboratorio

Oratorio San Filippo Neri, Bologna - 6 Dicembre 2019

Main lipid-metabolism pathways in the body



RUOLO DEL LABORATORIO NELLA DIAGNOSI DELLE PIÙ FREQUENTI DISLIPIDEMIE

Topics:

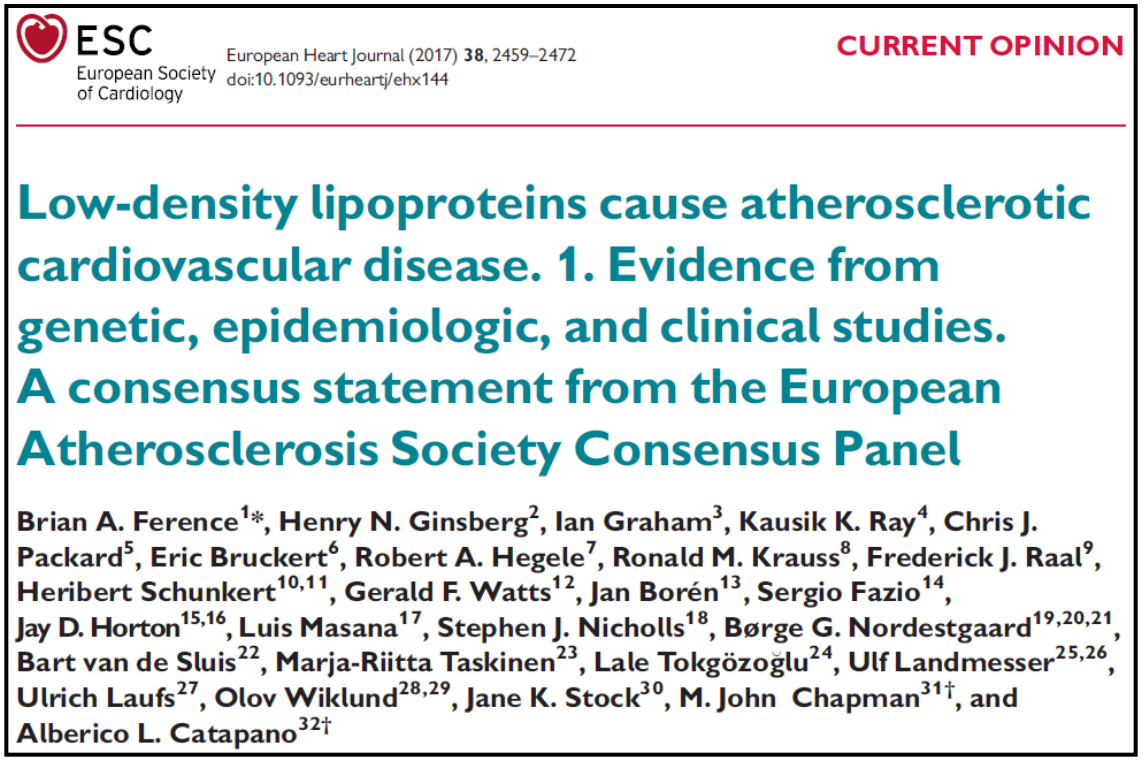
1- Why do we measure Low-Density Lipoprotein (LDL) cholesterol?

2- Which atherogenic lipoproteins should be measured?

3- Role of Clinical Chemistry Laboratory in the diagnosis of patients with Familial Hypercholesterolemia (FH):

- opportunistic screening for FH using laboratory database

1- Why do we measure Low-Density Lipoprotein (LDL) cholesterol?



ESC European Society of Cardiology

European Heart Journal (2017) **38**, 2459–2472
doi:10.1093/eurheartj/ehx144

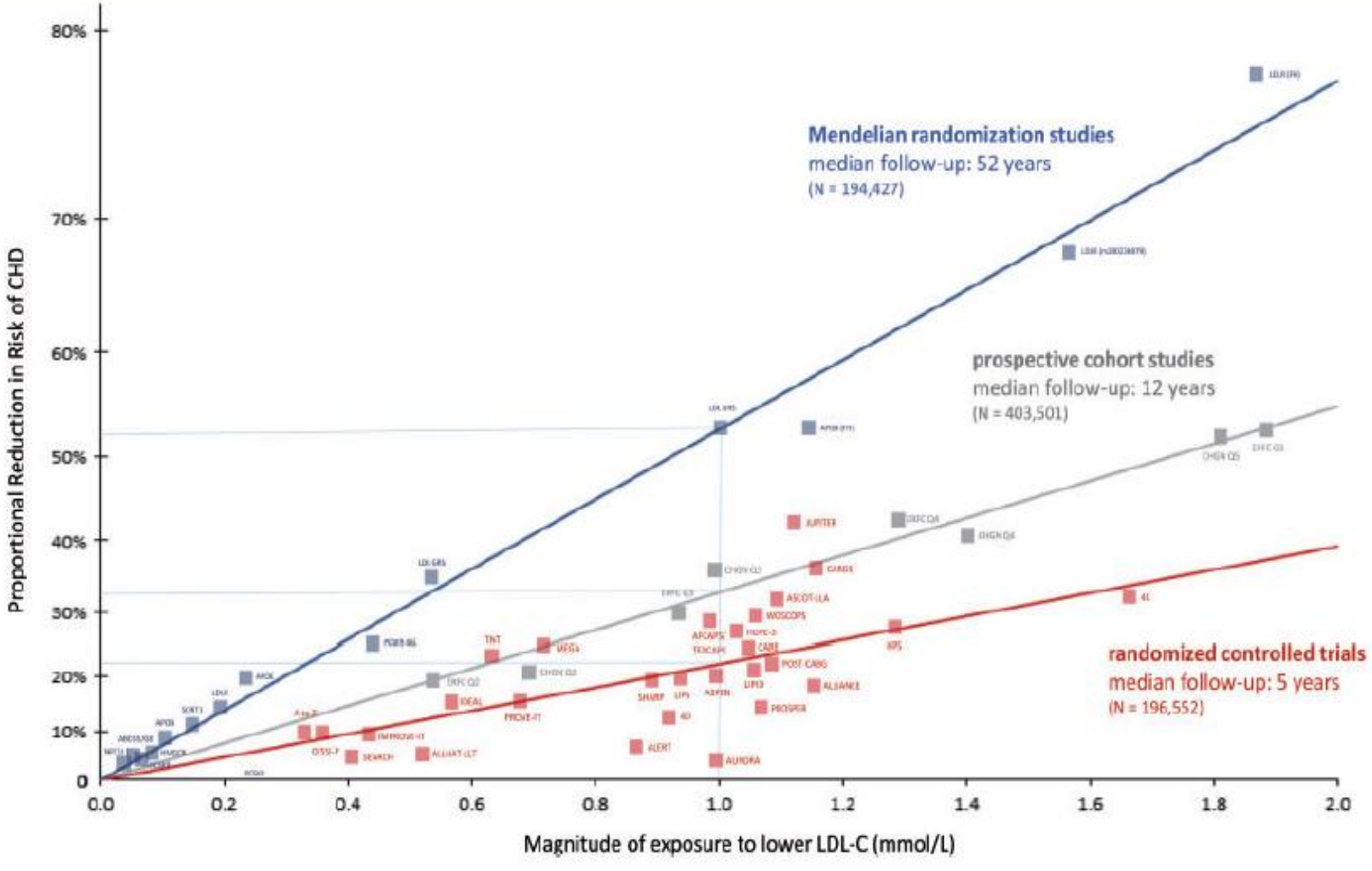
CURRENT OPINION

Low-density lipoproteins cause atherosclerotic cardiovascular disease. 1. Evidence from genetic, epidemiologic, and clinical studies. A consensus statement from the European Atherosclerosis Society Consensus Panel

Brian A. Ference[1]*, Henry N. Ginsberg[2], Ian Graham[3], Kausik K. Ray[4], Chris J. Packard[5], Eric Bruckert[6], Robert A. Hegele[7], Ronald M. Krauss[8], Frederick J. Raal[9], Heribert Schunkert[10,11], Gerald F. Watts[12], Jan Borén[13], Sergio Fazio[14], Jay D. Horton[15,16], Luis Masana[17], Stephen J. Nicholls[18], Børge G. Nordestgaard[19,20,21], Bart van de Sluis[22], Marja-Riitta Taskinen[23], Lale Tokgözoğlu[24], Ulf Landmesser[25,26], Ulrich Laufs[27], Olov Wiklund[28,29], Jane K. Stock[30], M. John Chapman[31]†, and Alberico L. Catapano[32]†

- Evidence from inherited disorders of lipid metabolism
- Evidence from prospective epidemiologic studies
- Evidence from Mendelian randomization studies
- Evidence from randomized controlled trials

Continuous, dose-dependent, and log-linear causal association between magnitude of the absolute change in LDL-C level and lifetime risk of CHD.



2- Which atherogenic lipoproteins should be measured?

Clinical Chemistry 64:7
1006–1033 (2018)

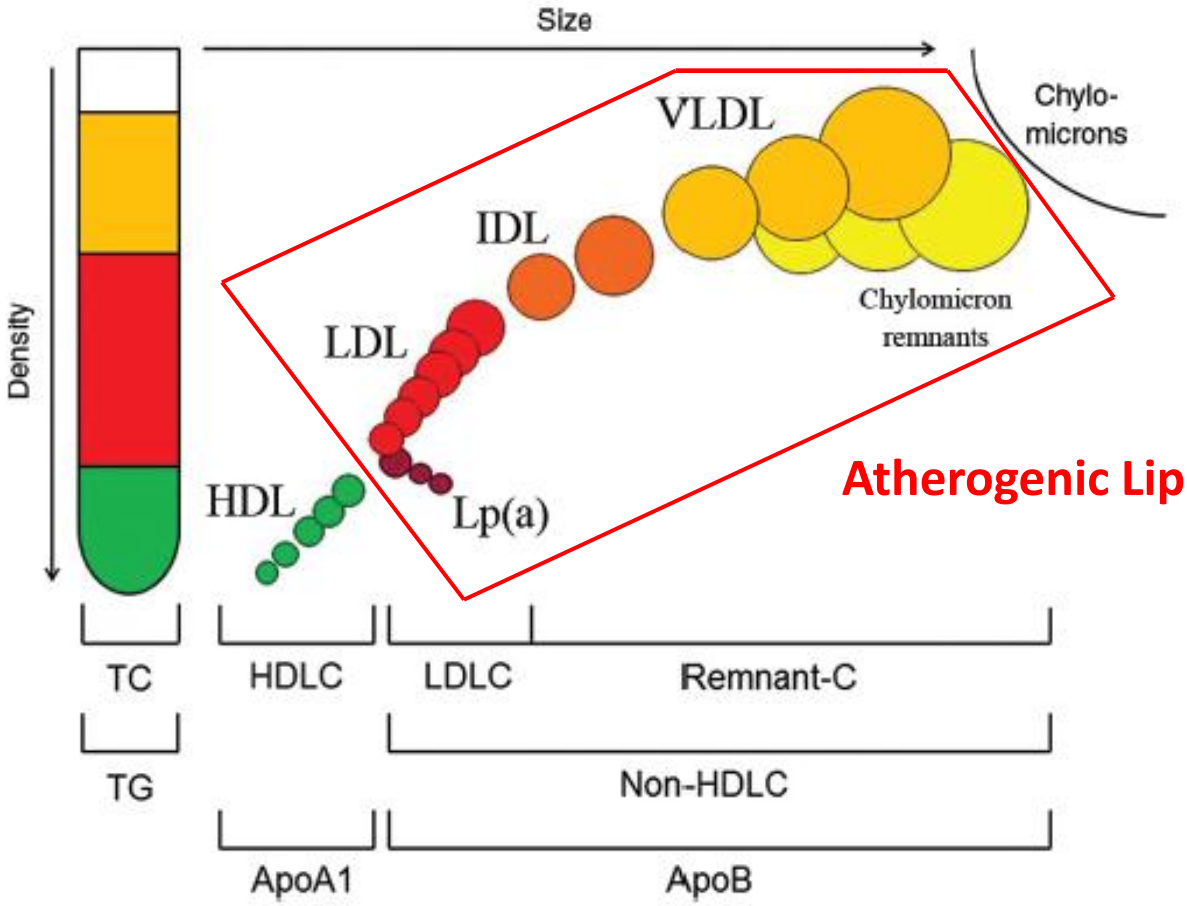
Special Report

Quantifying Atherogenic Lipoproteins: Current and Future Challenges in the Era of Personalized Medicine and Very Low Concentrations of LDL Cholesterol. A Consensus Statement from EAS and EFLM

Michel R. Langlois,[1*] M. John Chapman,[2] Christa Cobbaert,[3] Samia Mora,[4] Alan T. Remaley,[5] Emilio Ros,[6] Gerald F. Watts,[7] Jan Borén,[8] Hannsjörg Baum,[9] Eric Bruckert,[10] Alberico Catapano,[11] Olivier S. Descamps,[12] Arnold von Eckardstein,[13] Pia R. Kamstrup,[14] Genovefa Kolovou,[15] Florian Kronenberg,[16] Anne Langsted,[14] Kari Pulkki,[17] Nader Rifai,[18] Grazyna Sypniewska,[19] Olov Wiklund,[8] and Børge G. Nordestgaard,[14] for the European Atherosclerosis Society (EAS) and the European Federation of Clinical Chemistry and Laboratory Medicine (EFLM) Joint Consensus Initiative

European Atherosclerosis Society and
European Federation of Clinical Chemistry and Laboratory Medicine
provide recommendations to optimize atherogenic lipoprotein quantification for cardiovascular risk management.

2- Which atherogenic lipoproteins should be measured?



Lipoproteins separated according to density and size and their representative laboratory markers measured in a blood sample.

2- Which atherogenic lipoproteins should be measured?

Table 1. Current challenges for LDLC quantification.

Cause	Problem	Recommendation[a]
Analytical		
Novel therapies: very low LDLC concentrations	Magnification of measurement and calculation errors (e.g., Friedewald)	CBR2, CBR3, CBR4
Nonfasting lipid testing	Postprandial variation of TG in LDLC calculation	CBR4, CBR5
Increasing prevalence of obesity, diabetes, and moderate or major increases in TG	Nonspecificity bias in hypertriglyceridemic (>175 mg/dL; >2 mmol/L) and dyslipidemic samples	CBR2, CBR3, CBR4, CBR9, FR1, FR2
High Lp(a)	Overestimation of LDLC	CBR10
Clinical		
Increasing prevalence of obesity and diabetes	LDLC is a less predictive marker	CBR1, CBR5, CBR6, CBR7, FR3
Residual (on-treatment) CVD risk	Residual risk unexplained by LDLC	CBR8, FR3, FR4
Personalized medicine	LDLC has low or no diagnostic and predictive performance in certain patients	CBR1, CBR8, FR4, FR5

[a] CBR and future research recommendation (FR) to address the problem, listed in Table 2 (CBR) and Table 8 (FR).

Accumulating evidence suggests that a focus solely on the assessment and management of LDL-C is not an optimal strategy for all patients. Emerging evidence has established that VLDL, their remnants and Lp(a) likewise are causally related to CVD.

2- Which atherogenic lipoproteins should be measured?

Table 3. CBRs for the clinical indication for atherogenic lipid and lipoprotein quantification.

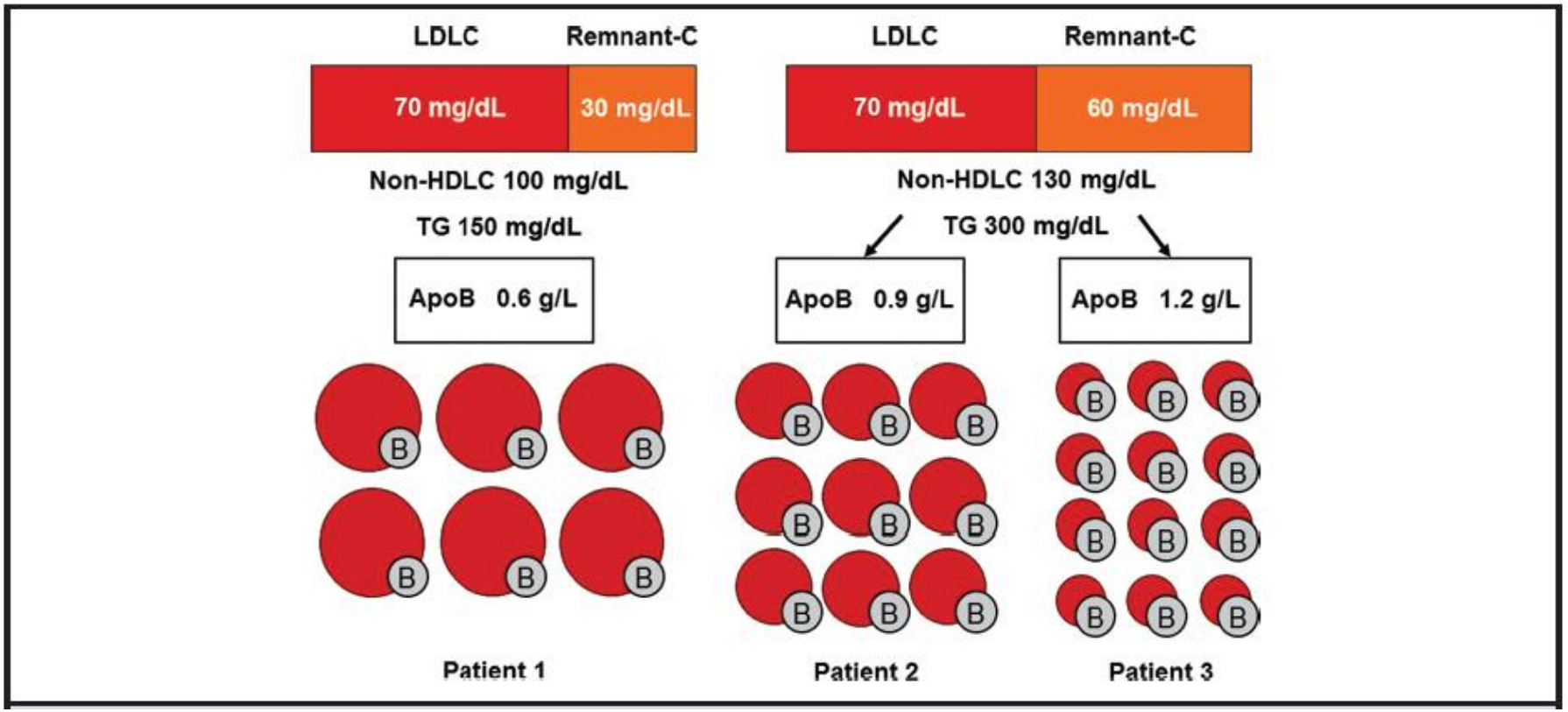
	CVD risk estimation	Dyslipidemia characterization	Treatment choice	Treatment target	Desirable value
TC	Yes[a]	Optional[b]	Optional[b]	Optional[b]	<190 mg/dL (5.0 mmol/L)
LDLC	Yes	Yes	Yes	Yes	Low to moderate risk <115 mg/dL (3.0 mmol/L)
					High risk <100 mg/dL (2.5 mmol/L)
					Very high risk <70 mg/dL (1.8 mmol/L)
TG	Yes	Yes	Yes	No	Fasting <150 mg/dL (1.7 mmol/L)
					Nonfasting <175 mg/dL (2.0 mmol/L)
Non-HDLC	Yes	No	No	Yes[c]	Moderate risk <145 mg/dL (3.8 mmol/L)
					High risk <130 mg/dL (3.3 mmol/L)
					Very high risk <100 mg/dL (2.5 mmol/L)
ApoB[d]	Optional[c]	Yes[c]	No	Optional[c]	High risk <100 mg/dL (1.0 g/L)
					Very high risk <80 mg/dL (0.8 g/L)

[a] In combination with HDLC.
[b] To be considered when LDLC is not available.
[c] In patients with mild to moderate hypertriglyceridemia (2–10 mmol/L; 175–880 mg/dL).
[d] Or LDLP if available.
To convert mmol/L to mg/dL, multiply by 38.6 for cholesterol and 88.5 for TG.

Comprehensive testing of atherogenic lipoproteins should use a biomarker, or a panel of multiple markers, that can be measured in either the fasting or nonfasting state and assesses the risk associated not only with LDL particles but also with remnant particles and Lp(a).

2- Which atherogenic lipoproteins should be measured?

Three patients with identical LDLC but with discordant non-HDLC and ApoB.



Patient 1 has all 3 targets at goal and normal numbers of LDL particles (70 mg/dL).
Patient 2 with moderate hypertriglyceridemia and discordant non-HDLC above target (100 mg/dL).
Patient 3 with moderate hypertriglyceridemia and increased non-HDLC but higher ApoB concentration than patient 2 (small dense LDL particles).

2- Which atherogenic lipoproteins should be measured?

Table 5. Examples of interlaboratory uncertainty when plasma lipid parameters are determined by different methods.

Assay	Assumed total error	Defined concentration in model patient	Range of uncertainty
TC	9%[a]	200 mg/dL (5.2 mmol/L)	182–218 mg/dL (4.7–5.7 mmol/L)
TG	15%[a]	250 mg/dL (2.8 mmol/L)	212–288 mg/dL (2.4–3.3 mmol/L)
HDLC	−20% to +36%[b]	40 mg/dL (1.0 mmol/L)	32–54 mg/dL (0.8–1.4 mmol/L)
Non-HDLC	Derived from TC and dHDLC	160 mg/dL (4.1 mmol/L)	128–186 mg/dL (3.3–4.8 mmol/L)
Measured LDLC	−26% to +32%[b]	110 mg/dL (2.8 mmol/L)	82–145 mg/dL (2.1–3.8 mmol/L)
Estimated LDLC (Friedewald)	Derived from TC, TG, and dHDLC	110 mg/dL (2.8 mmol/L)	70–144 mg/dL (1.8–3.7 mmol/L)
ApoB	12%[c]	110 mg/dL (1.1 g/L)	97–123 mg/dL (0.9–1.2 g/L)

[a] Based on NCEP analytical performance criteria *(41)*.

[b] Total error ranges observed by Miller et al. *(44)* across different dLDLC and dHDLC methods in dyslipidemic samples. The total error combines systematic bias and random imprecision. The tables are not relevant for the monitoring of a patient by the same laboratory/method over time. In this situation, the bias remains constant and only the (inevitable) imprecision is relevant. It will be considerably lower than the total error, at least for dHDLC and dLDLC (<10%), but not for TC, TG, and the derived measures cLDLC or non-HDLC.

[c] Based on AACC Lipoprotein and Vascular Diseases Division–Working Group on Best Practices assessment *(38)*.

There are various dLDLC and dHDLC assays available based on different principles from different manufacturers to selectively isolate and measure cholesterol in these lipoproteins.

- Substantial nonselectivity errors have been reported for many of the direct assays.
  - According to National Cholesterol Education Program (NCEP) criteria, the total error of LDLC measurements should be within 12% of the true value.

2- Which atherogenic lipoproteins should be measured?

Table 2. Key CBR to improve the clinical use of atherogenic lipoprotein assays.

CBR1	Comprehensive assay(s) of atherogenic lipoproteins should assess the risk conferred by LDL particles, remnant particles, and Lp(a).
CBR2	Laboratories and clinical trial centers should report lipid profiles with declaration of the assay method/manufacturer used.
CBR3	Follow-up of lipid profiles of a patient, from baseline at diagnosis to on-treatment measurements, should be ideally performed with the same assay method (and preferably the same laboratory and instrument).
CBR4	Values near the treatment decision cutpoints should be confirmed by ≥2 repeated measurements by the same method and then averaged.
CBR5	Laboratories should automatically calculate and report non-HDLC on all lipid profiles.
CBR6	Non-HDLC adds Remnant-C to LDLC and can be calculated in the fasting and nonfasting state, independent of TG variability.
CBR7	ApoB assay can estimate LDLP (~95% of apoB) plus Remnant-P and Lp(a) particle numbers in the fasting and nonfasting state.
CBR8	LDLC is the primary target of lipid-lowering therapy. When LDLC goal is achieved, then non-HDLC or apoB should be preferred as secondary treatment targets in patients with TG >175 mg/dL (>2 mmol/L), obesity, metabolic syndrome, or type 2 diabetes.
CBR9	When LDLC is unavailable because of an invalid Friedewald equation (TG >400 mg/dL; 4.5 mmol/L), follow-up calculation of non-HDLC should be used at higher TG concentrations rather than additional direct LDLC measurement.
CBR10	Lp(a)-corrected LDLC should be assessed at least once in patients with suspected or known high Lp(a), or if the patient shows a poor response to LDL-lowering therapy.

Familial Hypercholesterolemia

Estimated prevalence in population: 1:250

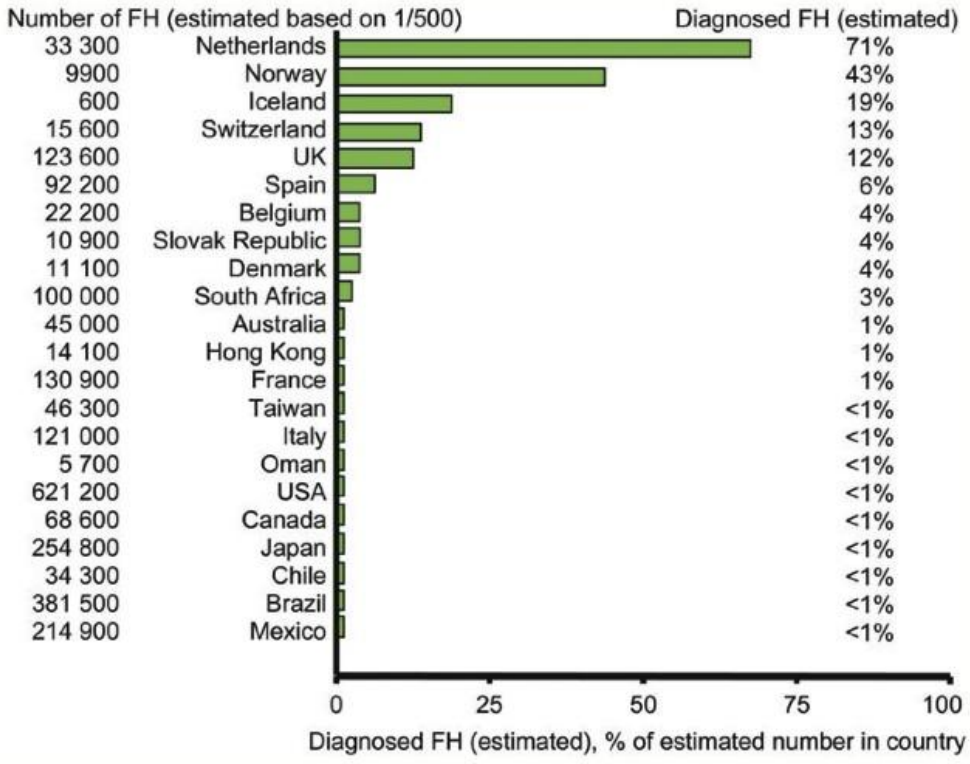
Emilia-Romagna: 18000 FH individuals

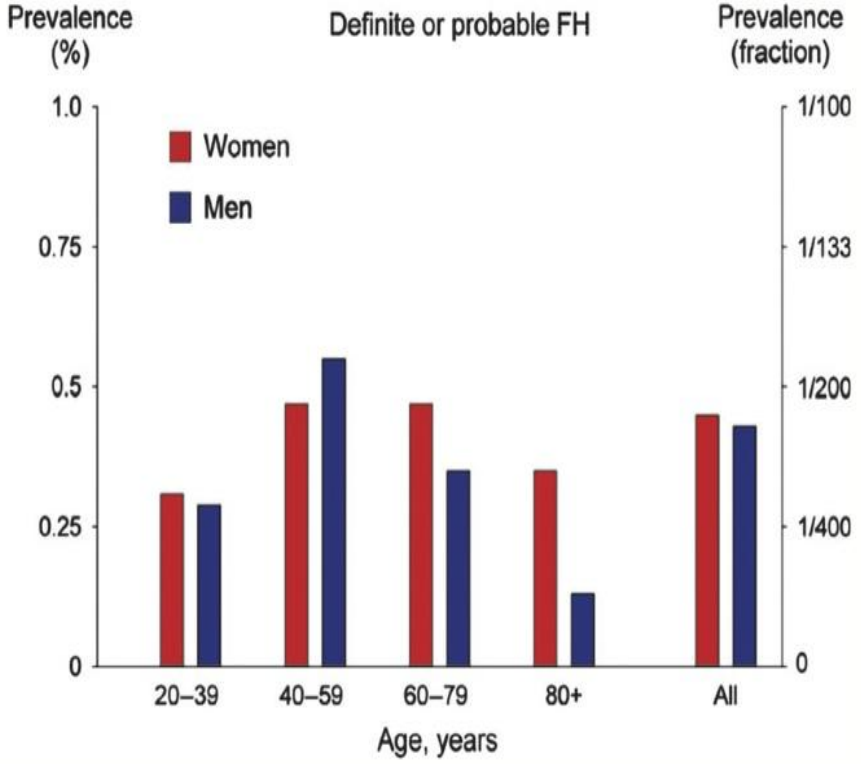
European Heart Journal (2013) **34**, 3478–3490
doi:10.1093/eurheartj/eht273

CURRENT OPINION

Familial hypercholesterolaemia is underdiagnosed and undertreated in the general population: guidance for clinicians to prevent coronary heart disease

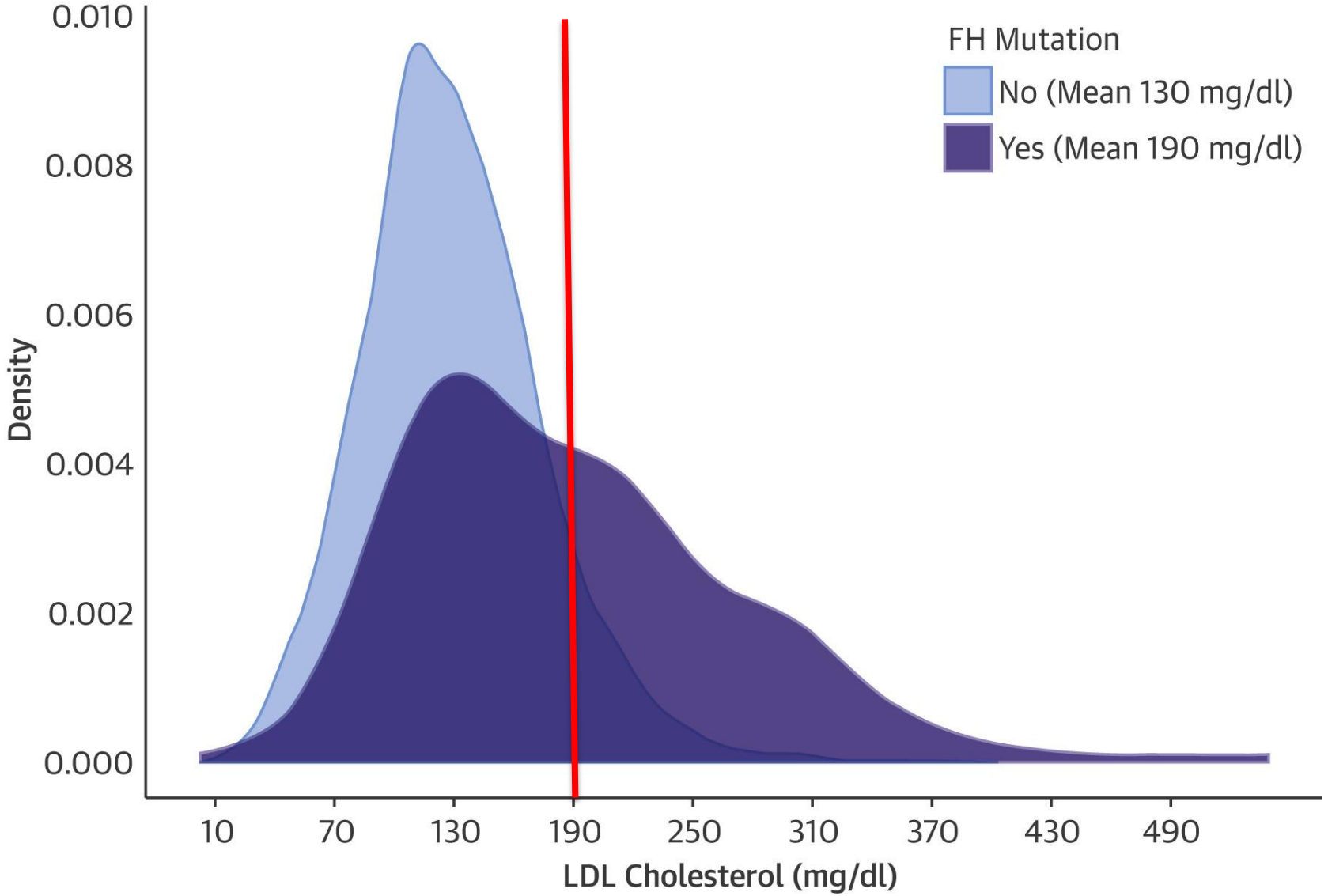
Consensus Statement of the European Atherosclerosis Society





3- Role of Clinical Chemistry Laboratory in the diagnosis of FH patients

FH is mainly characterized by a biochemical phenotype: ↑**TC and** ↑↑**LDL**



Clinical Scoring Systems for Familial Hypercholesterolemia

Criteria	Simon Broome Register[18]	Dutch Lipid Clinic Network[19]		MED-PED[20a]	AHA[21]	Canadian Criteria[22,144]
Lipids						
Total cholesterol (mmol/l)	• >7.5 (adult) [a] • >6.7 (child) [a]	NA		NA	NA	NA
LDL cholesterol (mmol/l)	• >4.9 (adult) [a] • >4.0 (child) [a]	• >8.5 [8] • 6.5–8.4 [5] • 5.0–6.4 [3] • 4.0–4.9 [1]	> 330 mg/dl 250 - 330 mg/dl 190 - 250 mg/dl 155 - 190 mg/dl	>5.7–9.3[b]	• >5.0 (adult) [a] • >4.0 (child) [a]	• >4.0 (child) [a] • >4.5 (18–39 years) [a] • >5.0 (>40 years) [a] • >8.5 [b]
Physical stigmata						
Personal	Tendon xanthoma [b]	• Tendon xanthoma [6] • Arcus cornealis[c] [4]		NA	NA	Tendon xanthoma [c]
Family	Tendon xanthoma in one relative [b]	Tendon xanthoma or arcus cornealis [2]		NA	NA	NA
Family history						
CAD	MI aged <50 years in two relatives or aged <60 years in one relative [d]	• Premature CAD[d] [2] • Premature CVD or PVD[d] [1]		NA	Premature CAD in one relative [b]	Premature CAD in one relative[d] [d]
LDL cholesterol (mmol/l)	>7.5 in one or two relatives [e]	Child with LDL cholesterol >95th percentile [2]		NA	One affected relative [c]	One relative with high LDL-cholesterol level [d]
Genetics	NA	NA		Known FH in a relative	NA	FH mutation in one relative [c]
Genetics						
Genetic mutations	*APOB*, *LDLR*, or *PCSK9* mutation [c]	*APOB*, *LDLR*, or *PCSK9* mutation [8]		NA	*APOB*, *LDLR*, or *PCSK9* mutation [d]	*APOB*, *LDLR*, or *PCSK9* mutation [c]
Diagnosis						
Diagnosis of FH	• Definite: a + (b or c) • Probable: (a + d) or (a + e)	• Definite: >8 • Probable: 6–8 • Possible: 3–5		Meets adjusted LDL-cholesterol cut-off point	a + (b or c) or d	• Definite: (a + c) or b • Probable: a + d

Role of Clinical Biochemistry Laboratories

- **Clinical Biochemistry Laboratories** are ideally placed to augment the **opportunistic detection of FH**.
- Primary care physicians request the majority of lipid profiles in clinical chemistry laboratories.
- Important **role of interpretative comments** with specific recommendations to improve the **detection of FH patients**.
- **Expert computer systems** may further optimise detection of FH by incorporating information on clinical and familial hystory and previous laboratory results.
- Possibility to increase FH identification for inpatients through **interaction with electronic health records (EHR)**.

Interpretative comments on lipid profile to highlight risk of FH

European Heart Journal
doi:10.1093/eurheartj/ehw152

CURRENT OPINION

Fasting is not routinely required for determination of a lipid profile: clinical and laboratory implications including flagging at desirable concentration cut-points—a joint consensus statement from the European Atherosclerosis Society and European Federation of Clinical Chemistry and Laboratory Medicine

Table 8 Life-threatening and extremely abnormal concentrations with separate reporting and consequent direct referral to a lipid clinic or to a physician with special interest in lipids

	Life-threatening concentrations	Refer patient to a lipid clinic or to a physician with special interest in lipids for further assessment of the following conditions
LDL cholesterol	>13 mmol/L >500 mg/dL[a]	Homozygous familial hypercholesterolaemia with extremely high cardiovascular risk[44]
LDL cholesterol	>5 mmol/L >190 mg/dL[a]	Heterozygous familial hypercholesterolaemia with high cardiovascular risk[43]
LDL cholesterol in children	>4 mmol/L >155 mg/dL[a]	Heterozygous familial hypercholesterolaemia with high cardiovascular risk[45]

Commento Interpretativo sul Profilo Lipidico:

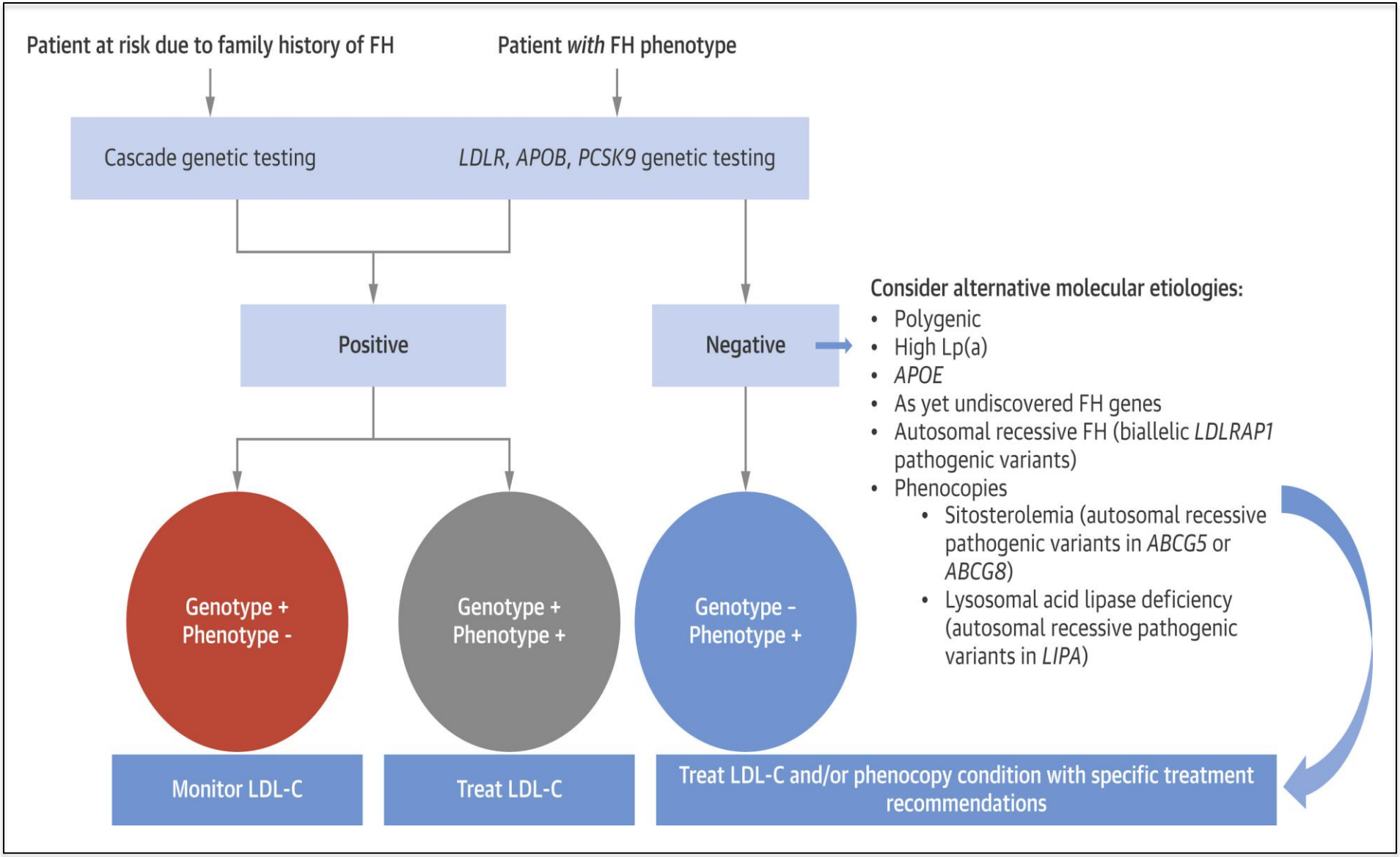
- **LDL-C > 190 mg/dl in adulti > 18 anni:** Valore di LDL significativamente elevato.

  Si consiglia accurata valutazione degli eventuali fattori di rischio cardiovascolare associati.

- **LDL-C > 250 mg/dl in adulti o LDL > 190 mg/dl in soggetti < 18 anni:** I valori rilevati possono essere indicativi di una forma di Ipercolesterolemia Familiare (FH) ad elevato rischio cardiovascolare. Si consiglia una valutazione specialistica lipidologica.

334 Referti → 68% Richiedenti Esterni → 287 Pazienti

Patients with FH phenotype identified through DLCN score may undergo genetic testing




GRAZIE PER L’ATTENZIONE!