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Review

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Low-Density Lipoprotein Cholesterol Level cannot be too Low: Considerations from Clinical Trials, Human Genetics, and Biology

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LDL cholesterol is by far the best established "causal" cardiovascular risk. It is distributed normally, and the mean value ranges around 100~120 mg/dl. In terms of preventive cardiology, we now know very well that the lower the LDL cholesterol, the better. Clinical usefulness of aggressive LDL-lowering therapies using statin, ezetimibe, and proprotein convertase subtilisin-kexin type 9 (PCSK9) inhibitors have been shown in primary and in secondary prevention settings. Additionally, the idea, based on recent randomized controlled trials (RCT), that the lower LDL cholesterol the better appears to be true for LDL as low as ~ 30 mg/dl. According to those data, recent guidelines in Europe and in Japan suggest the lowering of LDL cholesterol level <70 mg/dl for high-risk patients. However, the attainment rates of such "strict" goals seem to be quite low, probably because most cardiologists still have a sense of anxiety of "low" LDL cholesterol level. But "low" indicates no more than "lower" than the "average" range, which is not always implying the optimal range. Additionally, Mendelian randomization studies focusing on individuals exhibiting "low" LDL cholesterol suggest that "normal" LDL cholesterol levels might be too much for us. Moreover, LDL cholesterol levels of other primates are substantially lower than those in humans. In this review article, based on a series of evidence from clinical trials, human genetics, and biology, we provide the idea that we need to rethink what is the optimal range of LDL cholesterol level, instead of "normal" or "average" range.

Key words: Lipoproteins, LDL, Cholesterol, Genetics, PCSK9

1. Introduction

Based on a series of evidences, cholesterol has been established as a causal factor for atherosclerosis. Firstly, cholesterol is deposited in coronary atherosclerotic plaque[1]. Secondly, cholesterol-fed animals develop atherosclerotic plaque[2]. Thirdly, epidemiological studies have shown the positive relationship between cholesterol and atherosclerotic diseases[3]. Fourthly, familial hypercholesterolemia has been shown to accompany premature coronary artery disease[4]; on the other hand, familial hypobetalipoproteinemia has been shown as having less of a prevalence of such disease[5]. Finally, lowering (LDL) cholesterol via any means has been shown to reduce atherosclerotic cardiovascular diseases (ASCVD) regardless of the patients' backgrounds[6-9]. When we think about LDL cholesterol value, the "normal" range is typically set to 70 to 139 mg/dl based on the distribution. We feel safe to see if the value is around 110 mg/dl because it is nearly the "mean" value. If the value is below 70 mg/dl, the value is reported as "abnormally low" to us even under the secondary prevention settings. This situation doesn't make sense, since there are plenty of evidences suggesting that such an "abnormally" low LDL cholesterol level is associated with better clinical outcomes, especially among the patients with ASCVD, and the current clinical guidelines are accepting this fact[10, 11]. In the current era where we can use statins, ezetimibe, and proprotein convertase subtilisin/kexin type 9 (PCSK9) inhibitors, LDL cholesterol level can be reduced far greater than in past decades, raising a critical question: How low the LDL cholesterol can be? Most of the cardiologists have a kind of hesitation to further reduce their LDL cholesterol level despite the clinical guidelines stating that

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LDL cholesterol should be lower than 70 mg/dl in a portion of high-risk patients[12]. In this review article, we would like to provide lines of evidence clearly showing that "super-aggressive" LDL cholesterol lowering is not always considered as such. Rather, we need to rethink about the optimal range of LDL cholesterol level, instead of "normal" or "average" range.

2. Considerations from Extreme Cases

It is quite easy to understand the fact that LDL cholesterol is associated with ASCVD when we have a chance to see only a single case with homozygous FH. The untreated patients with homozygous FH whose LDL cholesterol levels are quite high are exhibiting premature ASCVD without exception. Interestingly, a simple treatment; namely, LDL cholesterol lowering, regardless of strategies, has been shown to literally save their lives[13]. Additionally, we experience brothers with compound heterozygous FH where the older brother who had started treatment at the age of 23 exhibited repeated coronary events, whereas, the younger brother who had been treated since the age of 15 had been event-free for a long period, despite the similar LDL cholesterol levels based on the same mutations (NM_000527.4(LDLR):c.2054C>T (p.Pro685Leu)/NM_000527.4(LDLR):c.2431A>T (p.Lys811Ter)) **(Fig. 1)**. The phenotypic difference between them clearly indicates that earlier intervention for LDL cholesterol can be quite beneficial even for such extreme cases. Moreover, several phenocopies of this situation with extremely high LDL cholesterol, including autosomal recessive hypercholesterolemia (ARH)[14] and sitosterolemia[15] caused by different genetic mutations, exhibit similar phenotypes, including tendon/cutaneous xanthomas, and premature ASCVD, similar to those observed in homozygous FH[16]. Those cases simply indicate that LDL cholesterol is the causal factor of this situation regardless of genetic etiology. On the other hand, findings from the patients exhibiting extremely low LDL cholesterol with any genetic backgrounds could also tell us a lot about the relationship between LDL cholesterol and ASCVD. Our patient with abetalipoproteinemia (ABL) caused by microsomal triglyceride transfer protein (MTTP) mutations (LDL cholesterol=0 mg/dl) did not exhibit any coronary plaque nor aortic calcifications at the age of 51 **(Fig. 2)**, although he suffers from spinocerebellar ataxia, and retinal pigmentary degeneration due to lack of fat-soluble vitamin[17]. On the contrary, we have shown an interesting case of homozygous familial hypobetalipoproteinemia (FHBL) whose LDL cholesterol was as low as 1 mg/dl[18]. The patient did not exhibit any complications relating to fat-soluble vitamin deficiency, as described above, probably due to his preserved HDL cholesterol (HDL cholesterol ~60 mg/dl) level containing fat-soluble vitamins It would be important to see that none of the family members whose LDL cholesterol was quite low had atherosclerotic diseases. Moreover, we experience sisters working as nurse practitioners whose LDL cholesterol levels are ~40 mg/dl, caused by a loss-of function of *PCSK9* gene[19]. All of the mutation carriers exhibiting low LDL cholesterol do not have any ASCVD, or any other clinical complications, including liver dysfunction. Those individuals carrying those mutations simply showed us that very low LDL cholesterol level over a long period is not so harmful, but rather, is beneficial for their preventive cardiology. Also, it would be quite interesting to understand that novel pharmacological interventions for LDL-lowering have been developed based on the findings obtained from those extreme cases[5, 20-23] **(Table 1)**.

3. Considerations from Human Genetics

As stated above, rare genetic variations, for example, mutation(s) involving loss of function in LDL receptor gene (namely, FH), are robustly associated with elevated LDL cholesterol level and ASCVD risk. On the other hand, loss of function mutation(s) in apolipoprotein B (*APOB*) gene (namely, FHBL) are robustly associated with reduced LDL cholesterol level and ASCVD risk. The same situations are applicable to ATP-binding cassette sub-family G member 5 (*ABCG5*) (both elevated)[24], angiopoietin-like 3 (*ANGPTL3*) (both reduced)[22], and *PCSK9* (both reduced)[23]. It is interesting to note that LDL cholesterol levels are positively associated with ASCVD regardless of genes and diseases. In addition to such rare genetic variations associated with Mendelian LDL disorders, common genetic variations associated with LDL cholesterol appear to be related with ASCVD. The magnitude of the effect on ASCVD is associated with LDL cholesterol level, and also such magnitude observed in genetic studies is far greater than that observed in clinical trials, suggesting that earlier intervention on LDL cholesterol may have a greater effect for preventive ASCVD. On the other hand, genetic variants associated with HDL cholesterol were not associated with ASCVD[25], consistent with negative results of RCT targeting lower HDL cholesterol[26-28].

4. Considerations from Clinical Trials Aiming to Reduce LDL Cholesterol Aggressively

Since the establishment of clinical usefulness of statins, there are debates regarding super-aggressive

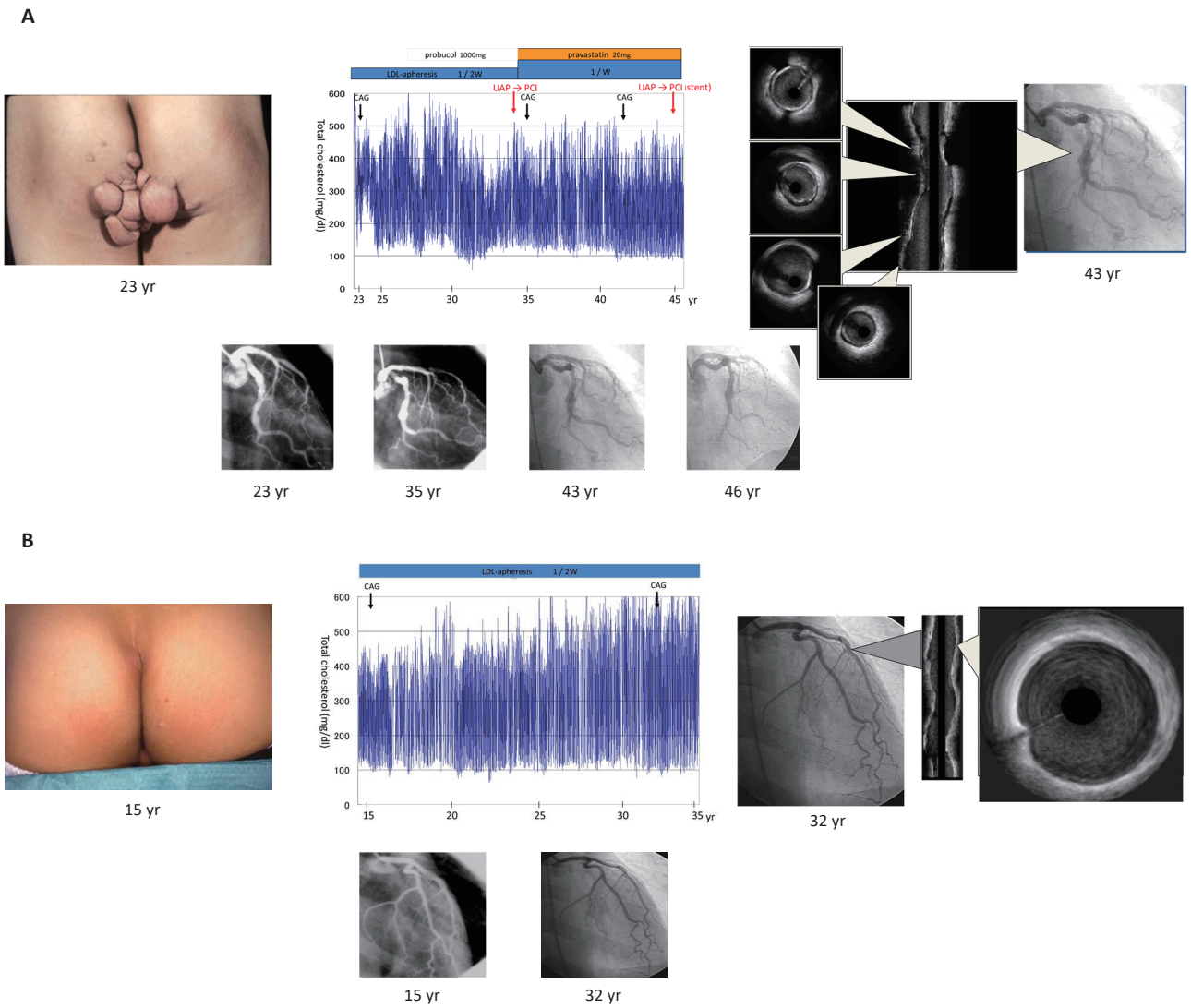

Fig. 1. Clinical course of brothers with compound heterozygous FH

A. Clinical course of older brother
Picture of buttocks with huge xanthoma is illustrated on the left. Clinical course is illustrated in the middle. Blue line indicates total cholesterol level (mg/dl). Images obtained through intravascular ultrasound are illustrated in the right.
B. Clinical course of younger brother
Picture of buttocks with small xanthoma is illustrated on the left. Clinical course is illustrated in the middle. Blue line indicates total cholesterol level (mg/dl). Images obtained through intravascular ultrasound are illustrated in the right.
UAP: unstable angina pectoris; CAG: coronary artery disease; PCI: percutaneous coronary intervention

LDL cholesterol lowering therapies, including targeting cholesterol levels much lower than 100 mg/dl, as well as the additional drugs on top of statins. Regarding the first matter, a RCT named EMPATHY study, targeting LDL cholesterol level <70 mg/dl using mainly statins among high-risk Japanese diabetic patients with primary prevention setting, revealed beneficial effect[29]. In this study, patients receiving aggressive LDL cholesterol lowering therapies (mean LDL cholesterol level was 76.5 mg/dl) exhibited significantly lower ischemic stroke events than those with standard care (mean LDL cholesterol level was 104.1 mg/dl). Moreover, high-dose statin therapy reaching to LDL cholesterol level at 76.6 mg/dl has been shown to be better than low-dose statin therapy reaching to LDL cholesterol level at 91 mg/dl among Japanese secondary prevention patients[30]. As for the second matter, recent mega RCT using ezetimibe, PCSK9 inhibitors, and a cholesteryl ester transfer protein (CETP) inhibitor on top of statins consistently revealed that additional beneficial effects could be obtained through such super-aggressive LDL choles-

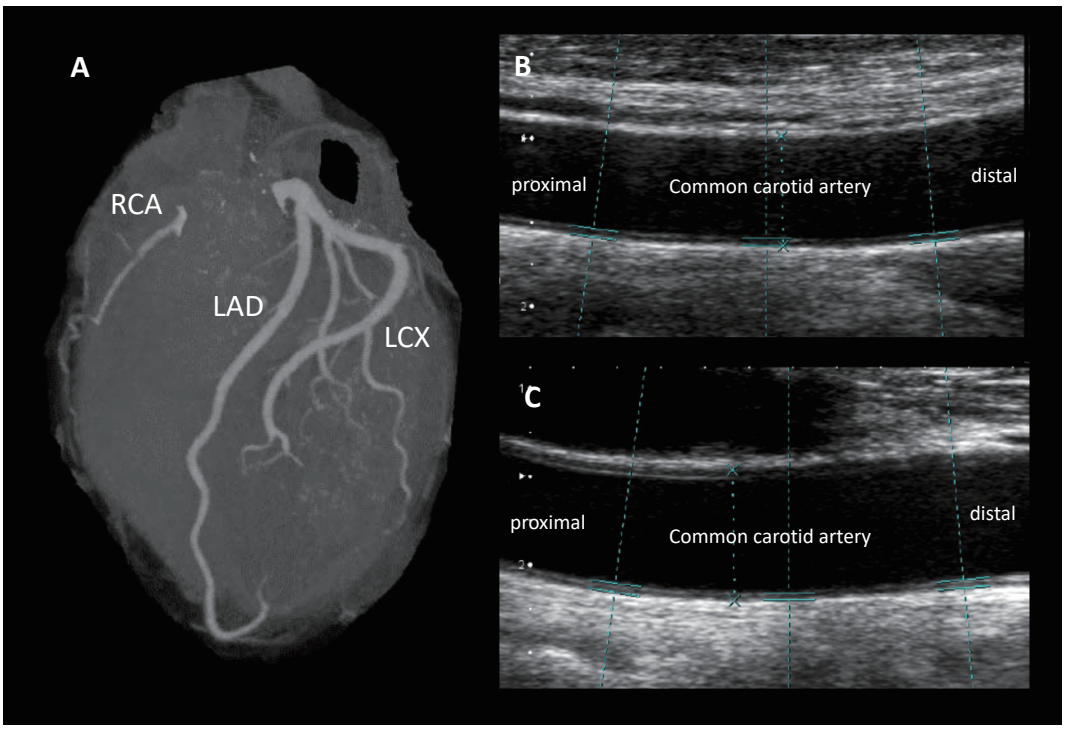

Fig. 2. Images of coronary computed tomography and carotid ultrasound in a patient with ABL

A. Coronary computed tomography obtained in a patient with ABL. There are no stenotic lesions nor any calcifications identified in coronary arteries.
B. Carotid ultrasound image obtained in a patient with ABL. There are no stenotic lesions or intima-media thickness in right common carotid artery.
C. Carotid ultrasound image obtained in a patient with ABL. There are no stenotic lesions or intima-media thickness in left common carotid artery.
ABL, abetalipoproteinemia; RCA, right coronary artery; LAD, Left anterior descending coronary artery; LCX, Left circumflex coronary artery

Table 1. Novel pharmacological interventions for LDL-lowering

Target	Deficiency or carriers of PTV	Compounds	Randomized controlled trials	Mendelian randomization
NPCL1	Heterozygous carriers (1 in 650 individuals)	Ezetimibe	IMPROVE-IT	Ref 20
PCSK9	Familial hypobetalipoproteinemia	Evolocumab Alirocumab	FOURIER ODYSSEY OUTCOMES	Ref 23
MTTP	Abetalipoproteinemia	Lomitapide	NA	NA
APOB	Familial hypobetalipoproteinemia	Mipomersen	NA	Ref 5
ANGPTL3	Familial combined hypolipoproteinemia	Evinacumab	NA	Ref 22
ACLY	NA	Bempedoic Acid	NA	Ref 21

NPC1L1, Niemann-Pick C1-Like 1; PCSK9, proprotein convertase subtilisin-kexin type 9; MTTP, microsomal triglyceride transfer protein; APOB, apolipoprotein B; ANGPTL3, Angiopoietin-like 3; Ref, reference, NA, not available; ACLY, ATP citrate lyase.

Table 2. Effects of randomized controlled trials and Mendelian randomization study in PTV on LDL-C and on ASCVD

Gene	RCT			Mendelian randomization study in PTV	
	Trial name	LDL cholesterol reduction (mg/dl)	ASCVD reduction (%)	LDL cholesterol reduction (mg/dl)	ASCVD reduction (%)
APOB	NA	NA	NA	43	72
CETP	REVEAL	26	9	12	30
NPC1L1	IMPROVE-IT	17	6	12	53
PCSK9	FOURIER/ODYSSEY	62/48	15/15	21	88

RCT, randomized controlled trial; PTV, protein truncating variant; ASCVD, atherosclerotic cardiovascular disease; APOB, apolipoprotein B; CETP, cholesteryl ester transfer protein; NPC1L1, Niemann-Pick C1-Like 1; PCSK9, proprotein convertase subtilisin-kexin type 9; NA, not available.

terol lowering therapies in proportion to the absolute degree of LDL cholesterol lowering[7-9, 31]. It is of note that ASCVD events seemed to decline with achieved LDL cholesterol, to a level of approximately 30 mg/dl in ODYSSEY OUTCOMES (using alirocumab)[32], and to a level of approximately 10 mg/dl in FOURIER (using evolocumab)[33]. Those observations collectively make us confident that the lower the LDL cholesterol, the better could be applicable, at least at the range of LDL cholesterol ~30 mg/dl in patients with ASCVD. Moreover, **Table 2** summarizing the results obtained through RCT and Mendelian randomization studies focusing on protein truncating variants (extreme situations) clearly indicates that super aggressive as well as earlier LDL-C lowering should be beneficial.

5. Lessons from Professors. Brown and Goldstein

In addition to the observations from those RCT, professors Brown and Goldstein, both of whom are Nobel laureates, suggested that the levels of cholesterol in our industrialized societies are inappropriately high[34]. This comment was derived from three different important aspects of nature: 1) a level of LDL cholesterol in serum of 25 mg/dl would be sufficient to nourish body cells with cholesterol, estimated by the experimental studies showing that LDL receptor binds LDL optimally when the lipoprotein is present at a cholesterol concentration of 2.5 mg/dl. And it has been shown that there is a 10-to-1 gradient between concentrations of LDL in plasma and interstitial fluid; 2) plasma LDL cholesterol levels of other mammals without development of atherosclerosis are generally less than 80 mg/dl; 3) LDL cholesterol level in newborn humans is approximately 30 mg/dl; 4) when humans are raised on a low fat diet, the plasma LDL cholesterol levels tend to stay in the range of 50 to 80 mg/dl.

6. Lessons from Monkeys, our Estimable Ancestors

Let me remind you that LDL cholesterol levels of monkeys, who are our estimable ancestors, have been shown to be as low as ~30 mg/dl[35]. Japanese macaque, whose life span is around 20 to 30 years, hardly exhibit ASCVD[35, 36]. Typically, wild monkeys have to survive in a natural field, requiring LDL cholesterol because of the incident of bleedings and/or infections. Accordingly, it could be skeptical that humans, especially, those living in industrialized societies, need a LDL cholesterol level as high as ~100 mg/dl. In this regard, "standard" levels are usually determined based on "average" values, not bade on "healthy" values in any biomarkers, including LDL cholesterol. Thus, it would be better to rethink "standard" levels of cholesterol.

7. Potential Concerns for Low LDL Cholesterol

In spite of a series of beneficial evidences as stated above, there are still many cardiologists who have some concerns about low LDL cholesterol, such as Alzheimer's disease, dementia, Parkinson's disease, and hemorrhagic stroke. In this regard, recent Mendelian randomization studies have suggested that low LDL cholesterol levels due to *PCSK9* and hydroxymethylglutaryl-CoA reductase (*HMGCR*) variants had no causal effect on high risk of Alzheimer's disease, vascular dementia, any dementia, or Parkinson's disease; instead, low LDL cholesterol levels may have a causal effect in reducing the risk of Alzheimer's disease[37]. On the other hand, another study showed strong positive associations of LDL cholesterol with ischemic stroke and inverse associations with hemorrhagic stroke; however, lowering LDL cholesterol appears to have net benefit for prevention of overall vascular events[38].

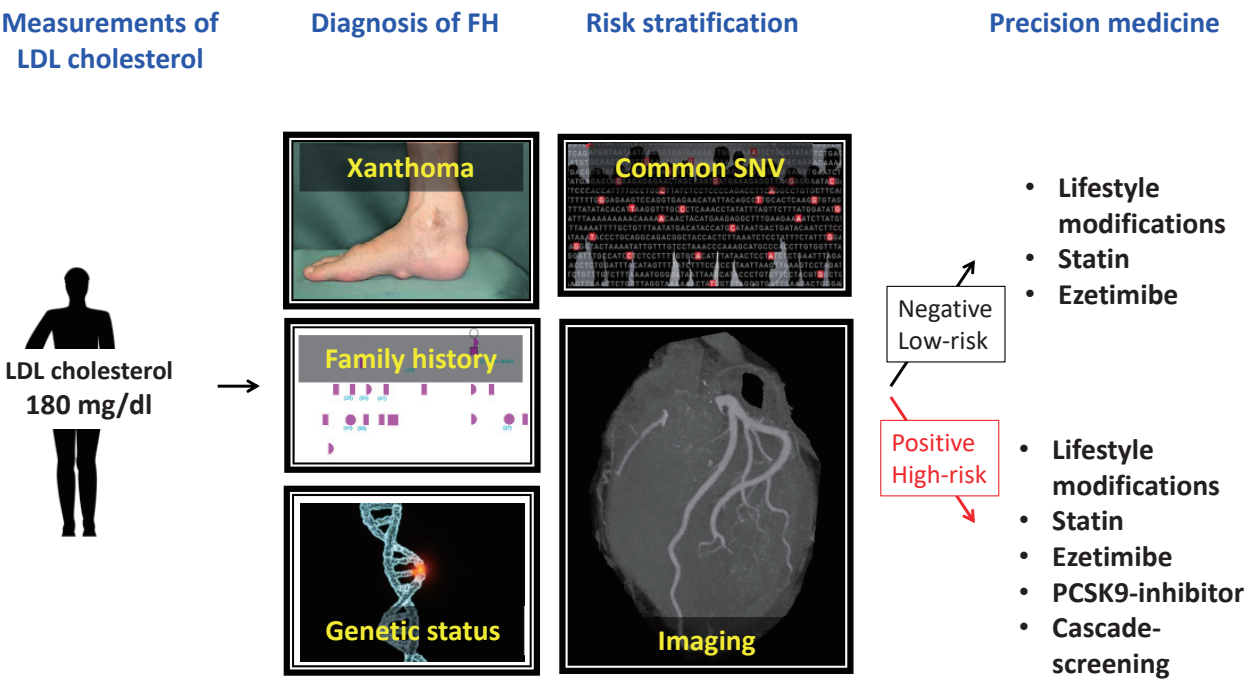

Fig. 3. Precision medicine for FH

When we encounter an individual whose LDL cholesterol level is ≥ 180 mg/dl, then we have to consider a clinical as well as genetic diagnosis of FH. Additionally, additional risk stratification can be considered based on their common genetic variations, and their imaging. According to this information, we can select the best approach for their LDL cholesterol reduction.
FH: familial hypercholesterolemia; SNV: single nucleotide variation

8. Precision Medicine of LDL Cholesterol Lowering

As stated, LDL cholesterol, as an important causal factor for ASCVD should be reduced as much as possible, especially in the secondary prevention settings. However, there is an emerging concept of precision medicine in almost all fields of medicine, including preventive cardiology. In the statin era, it has been shown that the effectiveness of this drug seems to be equal among a set of clinical subgroups, such as hypertension, diabetes, smoking, and so on[6]. On the other hand, there are a series of patients who exhibit greater responsiveness to ezetimibe, including, patients with diabetes, and those with *ABCG5* or *ABCG8* genetic mutation(s)[39-41]. Moreover, sub-analyses from recent clinical trials using PCSK9 inhibitors have suggested that there are several types of groups of patients who had greater benefit via this costly drug, including those with peripheral artery disease, elevated Lp(a) levels, or with high polygenic risk[42-45].

On the other hand, when we try to reduce LDL cholesterol among the patients with FH, we typically use the multiple LDL-lowering therapies listed above. However, there is an emerging concept of "cholesterol burden" in these particular patients. Namely, the integrated, accumulated sum of LDL cholesterol burden appears to lead them for their premature ASCVD. In other words, target LDL cholesterol level should be quite low if the patients with FH started treatment too late, whereas, that can be moderate under the situation where LDL cholesterol lowering is started early enough. To support this notion, a recent study showed that the patients with FH who had been treated moderately (LDL cholesterol level from 237 mg/dl to 160 mg/dl) since the mean age of 13 years exhibited far better prognoses compared with their age-matched relatives with FH[46]. Accordingly, "the earlier, the better" concept seems to be applicable to this extreme case, and we believe that it should also be true for non-FH hyper LDL cholesterolemia[47].

Moreover, we know that there are large variations of severity of disease (susceptibility to ASCVD) even among the patients with FH[47-51]. At least a part of it has been explained by their genetic status of FH and their physical signs of FH[52]. Another study has shown that accumulated effects of common genetic variations, in addition to rare mutation(s), which lead them to FH are contributing to their phenotypic variability[53]. Accordingly, the ideal strategy of LDL cho-

lesterol lowering should be quite individual-specific, including genetic backgrounds, and the timing of treatments **(Fig. 3)**.

9. Conclusion

In this paper, we have repeatedly emphasized that LDL cholesterol is a causal risk factor for ASCVD. Also we have learned from lines of evidence that super-aggressive LDL cholesterol lowering therapies, at least at around 30 mg/dl, are safe. We need to rethink what is the optimal range of LDL cholesterol level, instead of "normal", or "average" range, based on a series of evidences from clinical trials, human genetics, and biology.

Conflict of Interest Disclosures

None.

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