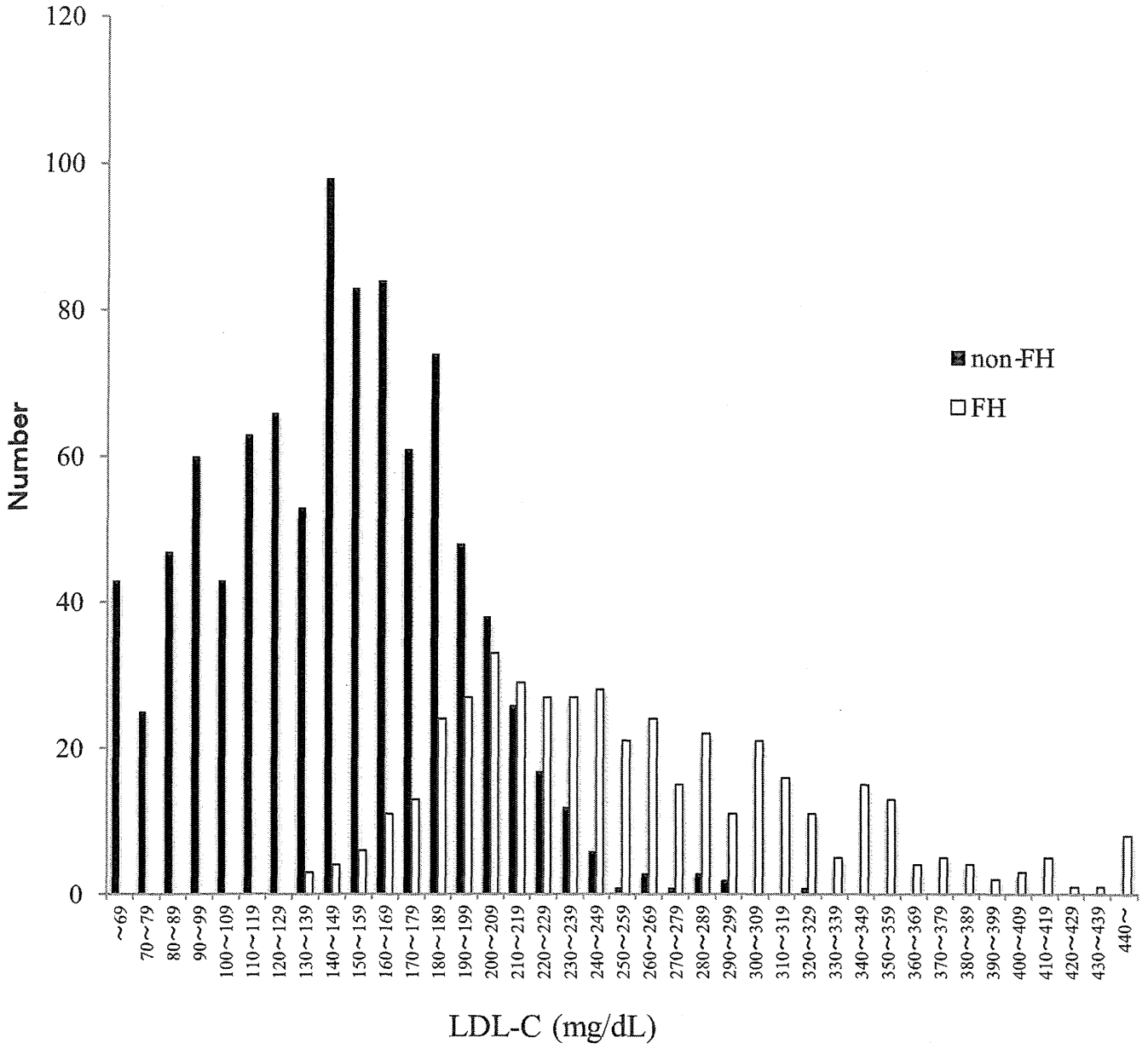

Fig. 1. Distribution of LDL-C levels before treatment in FH and non-FH patients. LDL-C levels were calculated by the Freidewald formula in patients with dyslipidemia diagnosed with FH or non-FH by specialists.

Statistical Analyses

Continuous variables are presented as the means ± SD. Categorical data are presented as numbers and percentages. Unpaired Student's *t*-test and one-way analysis of variance (ANOVA) were used to assess differences between groups in continuous variables. Differences in categorical variables were assessed by the χ^2 test.

Results

Among 1356 patients, 419 had been diagnosed with FH, while 937 with non-FH. Patient demographic data are shown in **Table 1**. FH patients were younger than non-FH patients. TC and LDL-C levels were 339 and 257 mg/dL in FH patients, respectively, and were significantly higher than in non-FH patients. The distribution of LDL-C levels in both groups is shown in **Fig. 1**. FH patients were divided into 3

Table 2. LDL-C levels in FH patients with or without genetic data

LDL-C (mg/dL)	FH (Total)	FH (Mut+)	FH (Mut−)	FH (no genetic data)	*p*-value
N	419	224	41	173	
Mean	257.4	266.2*	229.0*	252.9	
SD	67.39	69.85	60.14	63.70	
MEDIAN	244	253	216	241	0.003
IQ					
25%	205	213	189	203	
75%	300	308	244	295	

FH (Mut+): mutations in the LDL receptor or PCSK9, FH (Mut−): no mutations found, FH (no genetic data): no genetic analysis
*$p<0.005$ by Bonferroni

Table 3. Sensitivity and specificity in screening FH by physical findings and family history

	Specificity	Sensitivity
Physical findings		
ATT (+) (%)	98.6	64.1
CX (+) (%)	99.6	9.4
ATT (+) or CX (+) (%)	98.6	64.6
ATT(+) and CX(−)	99.6	11.7
Family history		
Family history of FH (+) (%)	93.6	98.2
Family history of CAD (+) (%)	96.3	28.3
Family history of FH (+) or CAD (+) (%)	91.7	98.7
Family history of FH (+) and CAD (+) (%)	98.2	27.4

ATT: Achilles tendon thickness, CX: Cutaneous nodular xanthomas
FH ($n=224$) was diagnosed by mutations in the LDL receptor and/or PCSK9. Non-FH ($n=937$) was diagnosed by specialists.

groups depending on their genetic data: FH with mutation(s) in LDL receptor or PCSK9, FH with no mutation(s) and FH with no genetic data. The mean and median of LDL-C along with SD and interquartile range of each group are shown in **Table 2**. LDL-C levels in FH with mutations were higher than those in FH without mutations.

We tried to define FH according to the screening standards as 3 major items, i.e., 1) LDL-C, 2) ATT and/or cutaneous nodular xanthomas (CX), 3) family history of FH and/or family history of premature CAD. We used LDL-C instead of total cholesterol, because LDL-C should better reflect the activity of the LDL receptor and is used for the goal of lipid management in the current Japanese guideline[8]. We incorporated "family history" as a major item because general practitioners were able to find FH by a family history of FH and/or premature CAD instead of LDL receptor activity. Sensitivity and specificity in screening FH by physical findings and family history are listed in **Table 3**. Based on these data, we decided to use 1) ATT or CX, and 2) family history of FH or CAD as 2 major items in addition to high LDL-C levels.

Next we tried to determine the cutoff levels of LDL-C. The percentage of the patients who satisfied each criterion according to LDL-C levels is listed in **Table 4**. Levels of 180 or 190 mg/dL are suggested as candidate cutoff levels. Therefore, the criteria for model 1 were set as those who satisfy 2 or more of the 3 criteria: 1) LDL-C 180 mg/dL or higher, 2) ATT (+) or CX (+), 3) Family history of FH or CAD, and for model 2, for which the cutoff point of LDL-C was changed to 190 mg/dl or higher, their sensitivity, specificity, and false positive and false negative rates were compared (**Table 5**). When we compared model 1 with model 2, higher sensitivity in model 1 than model 2 was obtained without any change in specificity, suggesting that 180 mg/dL is a better cutoff for LDL-C. The percentages were quite similar in FH with mutation (s) in LDL receptor or PCSK9, FH with no mutation (s) and FH with no genetic data. The diagnostic criteria of FH were then determined

Table 4. Percent satisfying each LDL-C level in non-FH and FH patients

	non FH	FH (All)	FH (Mut+)	FH (Mut−)	FH (No genetic data)
N	937	419	223	41	155
LDL-C ≥ 170 mg/dL (%)	30.5	94.5	96.0	85.4	94.8
LDL-C ≥ 180 mg/dL (%)	24.3	94.3	94.6	82.9	92.9
LDL-C ≥ 190 mg/dL (%)	16.6	92.1	93.7	75.6	89.7
LDL-C ≥ 200 mg/dL (%)	11.6	80.0	84.3	63.4	78.1

FH(Mut+): mutations in the LDL receptor or PCSK9, FH(−): no mutations found, FH (no genetic data): no genetic analysis

Table 5. Accuracy metrics of FH criteria using LDL-C cutoff levels of 180 or 190 mg/dL

	Sensitivity (%)	Specificity (%)	False positive (%)	False negative (%)
Model 1: Satisfying 2 or more of the following criteria: 1) LDL-C ≥ 180 mg/dL, 2) ATT(+) or CX(+), 3) Family history of FH or CAD	94.5	99.1	0.85	5.5
Model 2: Satisfying 2 or more of the following criteria: 1) LDL-C ≥ 190 mg/dL, 2) ATT(+) or CX(+), 3) Family history of FH or CAD	92.1	99.1	0.85	7.9

Table 6. Diagnostic criteria for adult (15 years or older) heterozygous FH

1	Hyper-LDL-cholesterolemia (LDL-C level before treatment: 180 mg/dL or more)
2	Tendon xanthoma (tendon xanthoma of the dorsal hands, elbows, and knees, or Achilles tendon thickening) or nodular xanthoma of the skin
3	Family history (relatives in the second degree): FH or premature CAD

-A diagnosis should be made after ruling out the possibility of secondary hyperlipidemia.
-Patients meeting 2 criteria should be regarded as having FH. Concerning those meeting 1 criterion, refer to Fig. 4. When FH is suspected, gene tests should be conducted to make a diagnosis.
-Nodular xanthoma of the skin does not include palpebral xanthoma.
-Patients with Achilles tendon thickening (9 mm or more) on radiography should be regarded as having xanthoma.
-When the LDL-C level is 250 mg/dL or more, FH should be strongly suspected.
-During drug therapy, the pretreatment lipid level should be employed as a reference value.
- CAD in males younger than 55 years old and females younger than 65 years old is defined as premature CAD.
-When a diagnosis of FH is made, the patient's family should also be investigated.
- LDL-C may be decreased after surgery, myocardial infarction, severe inflammation and so on. In these cases, LDL-C values before the diseases should be requested to give a diagnosis.
- To diagnose patients who have already been treated with statins, pretreatment levels of LDL-C should be requested; however, termination of statin treatment is not recommended to obtain pretreatment levels of LDL-C, even if the data are not available.

and are shown in **Table 6**.

Discussion

FH has the highest prevalence in genetic metabolic diseases, being heterozygous in one in 500 of the general population[1, 11]. Most young heterozygous FH patients have no symptoms other than high LDL-C levels, and those who have Achilles tendon thickness have no symptoms. The reason for undiagnosed FH patients to go to a clinic may be mainly divided into the following 4 situations: 1) a chance visit to a primary care physician due to flu or gastritis, etc., 2) recommendation of further medical examination due to high cholesterol at a health checkup, 3) transportation to the emergency room due to the development of acute coronary syndrome, 4) recommendation of medical consultation due to the presence of FH in his/

her family. The diagnostic criteria should be applied to these patients. Accordingly, conventional criteria are needed for the primary care setting.

Heterozygous FH patients show high levels of LDL-C, cutaneous and tendon xanthomas, and are complicated with myocardial infarction at young age by atherosclerosis due to intravascular exposure to high levels of LDL-C for many years. Because early diagnosis and treatment are recommended for these patients, the diagnostic criteria for FH have been reported in many countries including Japan[8, 12-17]. While some criteria give a satisfactory diagnosis of FH using specific items, others are adopting a scoring system. The Japanese criteria reported in 1988[8] were as follows. Major items included the following 3 items: (1) the patient shows the IIa or IIb phenotype at a serum cholesterol level of 260 mg/dL or above, in principle; (2) Tendinous xanthoma or xanthoma tuberosum is present; (3) Reduced or abnormal receptor activity is noted by LDL receptor analysis; however, for LDL receptor activity, even lipid specialists do not routinely measure activity. It would be even more difficult for primary care physicians to measure activity for the diagnosis of FH.

The cutoff level of serum cholesterol used in the first criterion in the criteria published in 1988 was 260 mg/dL; however, LDL-C is directly affected by dysfunction of the LDL receptor and is routinely measured in clinics by the direct method or Friedewald formula; therefore, we tried to use LDL-C as a cutoff level instead of total cholesterol. The presence of tendon and/or cutaneous nodular xanthomas was also used because of its convenience, high sensitivity and specificity. A family history of FH or premature CAD in 1st or 2nd degree relatives was proposed for the third criterion instead of measuring LDL receptor activity in the new diagnostic criteria. A family history of FH showed high sensitivity and specificity; however, primary care physicians may have difficulty obtaining this because it was not easy for them to reach a diagnosis of FH with the previous criteria. In the present study, accurate diagnosis of a family history of FH seemed to have been given because lipid specialists made the diagnosis at all the hospitals; however, the same result may not be applied to primary care physicians. Therefore, a family history of CAD, which may be easier to obtain, was added to the criteria. It should be noted that the sensitivity of a "family history of FH or CAD" was slightly higher than that of a "family history of FH". Accordingly, we chose a "family history of FH or premature CAD in 1st or 2nd degree relatives" as the third criterion.

The cutoff level of LDL-C for the diagnosis of FH should be set by its sensitivity and specificity in different cutoff points. The cutoff level of LDL-C for the diagnosis of FH was reported to be 190 mg/dL in Simon Broome[17], NICE[15] and 205 mg/dL in MEDPED[16]. In this study, 180 mg/dL was selected as the cutoff level together with the presence of xanthoma and the family history as the criteria for the diagnosis of FH because of its high sensitivity and specificity.

Reduced LDL receptor activity is direct evidence of FH and was used as one of the criteria in the previous version. Usually, LDL receptor activity is determined by the binding of fluorescent-labeled LDL to lymphocytes. The procedure of measuring LDLR activity is cumbersome and it is difficult to measure in routine clinical settings. Further, few companies can measure LDLR activity. Indeed, the specialists involved in this study measured LDLR activity only in 7 of 419 patients of FH, showing the sensitivity of the previous criteria as 60.9%. Therefore, in order to determine criteria sensitive enough to give a diagnosis of FH, the third item was changed from LDLR activity to family history.

There are some limitations in the present study. First, the patients analyzed in this study may have different characteristics from those followed by primary care physicians, because the physicians in this study are taking care of many FH patients and information about family history can be obtained more easily than by primary care physicians. Second, it is sometimes difficult for primary care physicians to take a complete family history, especially FH, and to diagnose ATT and/or the presence of CX, about which information can be missed in the primary care setting. Third, FH has been reported to have mutations in LDL receptor, PCSK9 and apolipoprotein B. Because mutations in PCSK9 may cause milder forms of FH, the sensitivity of the criteria may be reduced in these patients. Further study is required to address the applicability of the criteria for the primary care setting.

In conclusion, we have determined the cutoff of LDL-C for the diagnosis of FH by a multicenter study and proposed conventional diagnostic criteria by using high LDL-C, ATT and/or the presence of CX, and a family history of FH and/or CAD for primary care settings.

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Special Report

Guidelines for the Management of Familial Hypercholesterolemia

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Familial hypercholesterolemia (FH) is a highly prevalent autosomal dominant hereditary disease, generally characterized by three major signs, hyper-low-density-lipoprotein (LDL) cholesterolemia, tendon/skin xanthomas and premature coronary artery disease (CAD). Because the risk of CAD is very high in these patients, they should be identified at an early stage of their lives and started on intensive treatment to control LDL-cholesterol. We here introduce a new guideline for the management of FH patients in Japan intending to achieve better control to prevent CAD. Diagnostic criteria for heterozygous FH are 2 or more of 1) LDL-cholesterol ≥ 180 mg/dL, 2) tendon/skin xanthoma(s), and 3) family history of FH or premature CAD within second degree relatives, for adults; and to have both 1) LDL-cholesterol ≥ 140 mg/dL and 2) family history of FH or premature CAD within second degree relatives, for children. For the treatment of adult heterozygous FH, intensive lipid control with statins and other drugs is necessary. Other risks of CAD, such as smoking, diabetes mellitus, hypertension etc., should also be controlled strictly. Atherosclerosis in coronary, carotid, or peripheral arteries, the aorta and aortic valve should be screened periodically. FH in children, pregnant women, and women who wish to bear a child should be referred to specialists. For homozygotes and severe heterozygotes resistant to drug therapies, LDL apheresis should be performed. The treatment cost of homozygous FH is authorized to be covered under the program of Research on Measures against Intractable Diseases by the Japanese Ministry of Health, Labour, and Welfare.

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Key words; LDL cholesterol, LDL receptor, Achilles tendon thickness, Coronary artery disease, Diagnosis criteria

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Introduction

Familial hypercholesterolemia (FH) is a genetic disorder caused by mutations in the genes related to the low-density lipoprotein (LDL) receptor pathway, and is transmitted by autosomal dominant inheri-

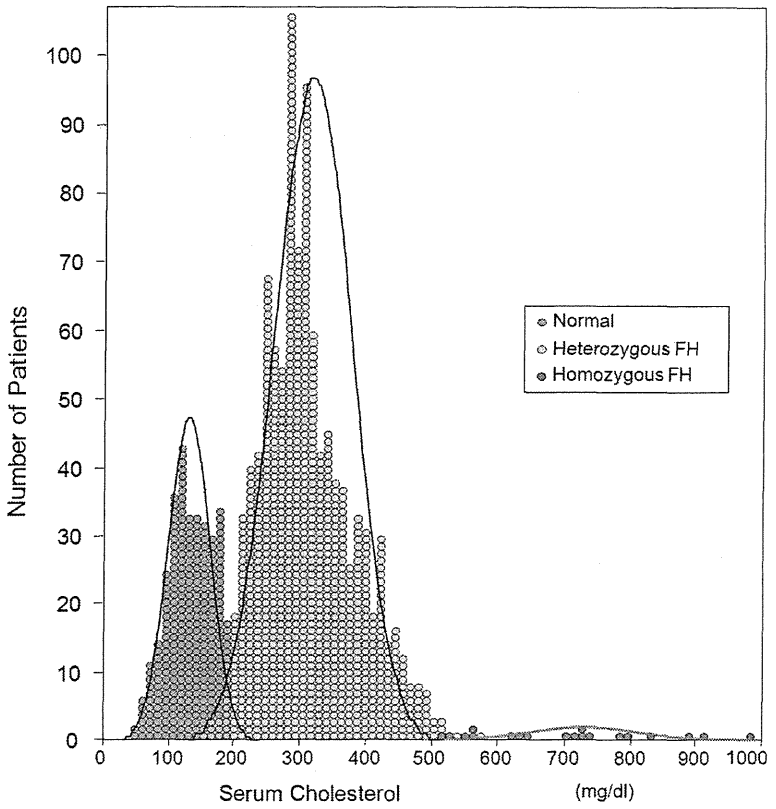

Fig. 1. Distribution of serum total cholesterol levels in normal subjects, and heterozygous and homozygous patients with familial hypercholesterolemia (modified by adding patients to Reference 85).

tance. Heterozygous patients are observed in 1 in every 500, and homozygous patients in approximately 1 in every 1 million people in Japan. The total number of FH patients has been estimated to be 250,000 or higher in Japan. Accordingly, FH is the most frequent hereditary metabolic disorder and is often encountered in daily practice. Primary manifestations of FH are hypercholesterolemia, tendon xanthoma, and premature coronary artery disease (CAD). Since atherosclerosis progresses faster and is accompanied by more severe organ disorders in FH patients than in hyperlipoproteinemic patients without a genetic background, prompt diagnosis and treatment are mandatory.

The effectiveness of reducing LDL-cholesterol (LDL-C) for primary and secondary prevention has been reported by many large-scale clinical studies using statins in high-risk groups of hypercholesterolemia and CAD. On the basis of these reports, guidelines for the prevention of atherosclerosis mainly targeting LDL-C have also been established in Japan; however, the subjects of large-scale clinical studies are not only patients with marked hypercholesterolemia, such as those with FH. In addition, as FH patients are exposed to hypercholesterolemia for a long period of time from their infancy, they exhibit more advanced atherosclerosis and more severe organ disorders than patients with typical hypercholesterolemia without a genetic background, necessitating particular caution in their diagnosis and treatment. Guidelines for the prevention of atherosclerosis classify FH patients into a high-risk group of CAD and stress the importance of early diagnosis and treatment. However, the guidelines for FH have not been sufficiently established in Japan, and no consensus has been reached; therefore, the paper proposes a guideline specific for the diagnosis and treatment of FH for the Japanese population.

Clinical Features and Gene Mutations of Familial Hypercholesterolemia

Familial hypercholesterolemia (FH) is an autosomal dominant hereditary disease characterized by three major signs: 1) hyper- LDL cholesterolemia, 2) tendon/skin xanthoma and 3) premature coronary artery disease (CAD).

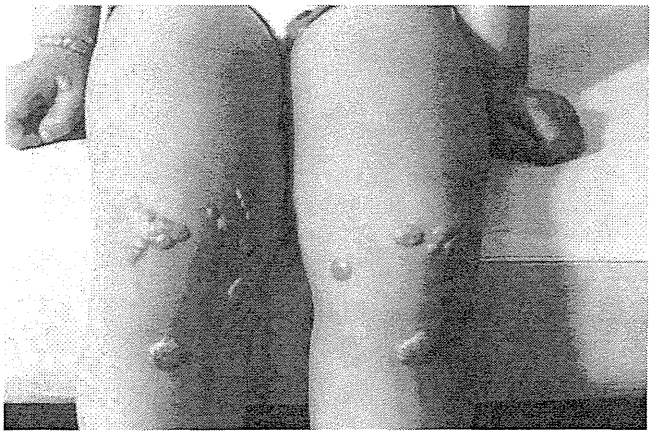

Fig. 2. Xanthoma observed in homozygous FH patients.

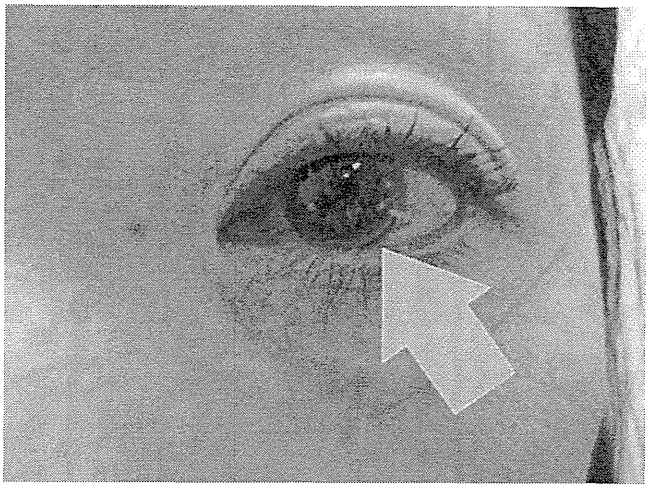

Fig. 3. Corneal arcus in FH patients.

In patients with FH, hyper-LDL cholesterolemia persists after birth, and Achilles tendon xanthoma may appear as early as in their 10s and is found in 50% of patients by the age of 30. Coronary atherosclerosis also develops at a young age, and the CAD risk is very high in FH patients. CAD events, such as myocardial infarction, which determine the prognosis of FH patients, may appear from 30 to 50 years of age in men and 50 to 70 years of age in women[1].

FH is one of the most frequently encountered hereditary diseases that may cause cardiovascular diseases (CVD) in general clinical practice. It should also be emphasized that the contribution of FH to CAD is higher in Japanese public health because of the low prevalence of CAD among the general Japanese population; however, neither diagnosis nor treatment is adequately performed for these patients. According to the literature, identification of FH may be only 20% or less in Europe, the United States and Japan[2, 3]. Health care professionals must understand that FH is a highly prevalent autosomal dominant hereditary disease and therefore surveying family members is important for the identification and treatment of patients.

Clinical Features of FH

Hyper-LDL Cholesterolemia

A patient with a mutation in one of the alleles in the gene involved in LDL metabolism, typically the LDL receptor, is termed heterozygous FH, and a patient with gene mutations in both alleles is termed homozygous FH. Serum total cholesterol (TC) in FH patients and their families shows a trimodal pattern. The mean serum TC in normal adults, heterozygous FH patients, and homozygous FH patients was 179 ± 26, 338 ± 63, and 713 ± 122 (mean ± SD) mg/dL, respectively; the mean value was 2 times higher in heterozygous FH patients and 4 times higher in homozygous FH than in normal adults[4]. There were overlaps between the normal and heterozygous FH, as well as between heterozygous and homozygous FH patients. It is sometimes difficult to differentiate these conditions based on the serum lipid level alone (**Fig. 1**). According to Bujo *et al.*, the mean LDL-C before treatment was 248 mg/dL in 641 Japanese patients with heterozygous FH (296 males and 345 females, mean age: 51 years)[5]. Although there was no gender difference in the LDL-C, triglyceride was significantly higher and HDL-cholesterol (HDL-C) was significantly lower in males than females.

Tendon/Skin Xanthoma

Physical findings such as tendon/skin xanthoma are important signs for the clinical diagnosis of FH. In homozygous FH patients, xanthoma becomes more marked than in heterozygous FH (**Fig. 2**). Xanthoma of the skin frequently develops at sites under regular mechanical stimuli, such as the extensor sides of the elbows/knees and wrist/gluteal regions. Tendon xanthoma appears as Achilles tendon thickening in many cases. It can be diagnosed based on inspection and palpation. Some FH patients may complain of inflammation-related pain of the Achilles tendon; however, FH should not be ruled out based on the absence of xanthoma. In those with a definitive diagnosis of FH by genetic analysis, xanthoma is absent in 20 to 30% of patients[5]. When xanthoma is absent, family surveys and genetic diagnosis are important. Xanthoma may be absent especially before 20 years of age, but becomes more prominent with aging. It should be noted that evaluation of xanthoma is difficult in the

Fig. 4. Cumulative number of patients with myocardial infarction by age in male and female heterozygous FH patients (patients were added to Reference 1, and the form of indication was modified).

Achilles tendon after its rupture. Palpebral xanthoma is not diagnostically valuable because it is observed in many persons without FH.

Corneal Arcus

Corneal arcus is also characteristic of FH, as shown in **Fig. 3**, and its prevalence is approximately 30% in FH. Senile arcus is observed in most people aged over 60, but the border of corneal arcus is clearer, making it distinguishable. Corneal arcus in those who are under 50 is diagnostically valuable in clinical practice.

Coronary Atherosclerosis

FH should be suspected in premature CAD patients with high LDL-C. Hyper-LDL cholesterolemia deteriorates coronary atherosclerosis, and the absence of treatment leads to cardiac death in approximately 60% of patients with heterozygous FH. Some previous studies also indicated the association of hyper-LDL cholesterolemia with cerebrovascular disorder, but the incidence is approximately 5 to 10% in heterozygous FH[4, 6]. In males, the incidence of myocardial infarction increases linearly after the age of 30. On the other hand, myocardial infarction is not very common in females under the age of 49. There is therefore a marked gender difference, as shown in **Fig. 4**.

Epidemiology

Diagnosis of FH is not definitive for most heterozygous FH; therefore, the incidence of heterozygous FH is estimated as 1/500 persons (general population) using the Hardy-Weinberg equilibrium formula, as the incidence of homozygous FH with marked clinical features is approximately 1/1,000,000[7]; however, no nationwide survey has been carried out to investigate the accurate incidence of heterozygous FH in Japanese. On the other hand, recent advances in genetic diagnostic techniques have made possible to make a definitive diagnosis in more homozygous FH patients. Mabuchi *et al.* reported that LDL-receptor mutations are more frequent and consequently the incidence of heterozygous FH was estimated to be approximately 1/200 in the Hokuriku area, based on the incidence of homozygous FH[8]. It is still unclear whether this is a region-specific phenomenon, such as seen in Quebec, Lebanon and South Africa, and there are no evident grounds to extrapolate these data to nationwide prevalence; therefore, it is not irrational to regard the nationwide FH incidence as 1/500. Nevertheless, FH is not a rare disease, and FH patients may comprise approximately 8.5% of hyper-LDL cholesterolemia patients receiving treatment[9].

Causative Genes

The diagnosis of FH is definitive when mutations of the genes involved in LDL metabolism are

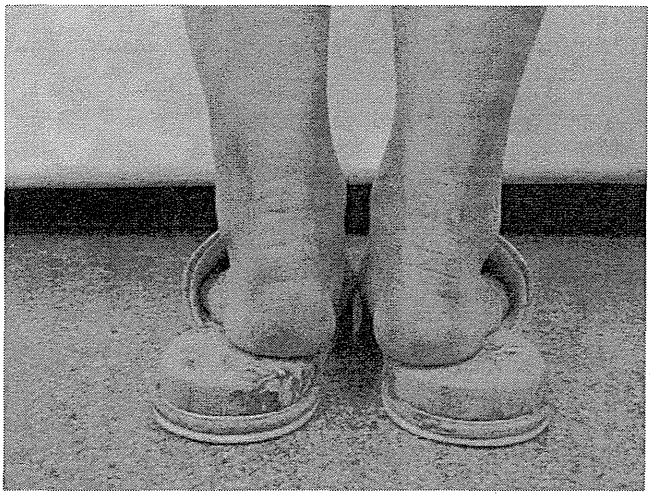

Fig. 5. Achilles tendon thickening in FH patients.

confirmed, such as LDL receptors, in addition to hyper-LDL cholesterolemia; however, institutions where genetic diagnosis is available are limited. When a genetic diagnosis of FH is made for a primary patient, the diagnosis in his/her family can be definitive.

Besides the LDL receptor, it is known that mutations in the genes for apolipoprotein B-100 (Apo B-100), and proprotein convertase subtilisin/kexin type 9 (PCSK9) cause FH[10, 11]. These molecules play an important role in LDL metabolism. The mutations of causative genes can be confirmed in 60 to 80% of clinically diagnosed heterozygous FH patients.

LDL Receptors

In the majority of FH patients, the causative mutation is in the LDL receptor gene. A number of mutation sites have been identified in this gene to cause FH; more than 1,000 sites of mutations worldwide (http://www.ucl.ac.uk/fh/) and some 100 sites in Japan[4].

Apolipoprotein B-100

Mutations in the apoB gene, a ligand of the LDL receptor, are termed familial defective apolipoprotein B-100 (FDB). The incidence of these mutations is relatively high in Caucasians in Europe and the United States but low in other races. An increase in the serum lipid level is relatively mild in comparison with LDL receptor mutations. No patient has been reported in Japan.

PCSK9

PCSK9 is a plasma protein that facilitates turnover of the LDL receptor. Gain-of-function mutation

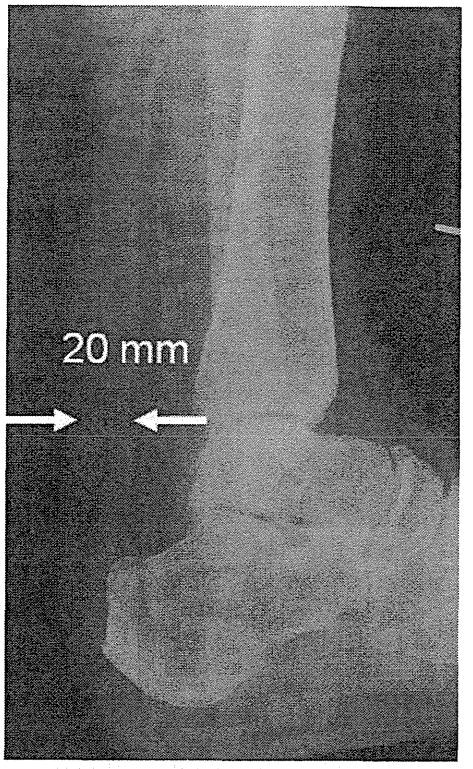

Fig. 6. Radiography of the Achilles tendon. The measurement site of the Achilles tendon is indicated by arrows.

of PCSK9 therefore decreases the number of LDL receptors, causing hyper-LDL cholesterolemia. These mutations may not always induce marked hyper-LDL cholesterolemia. In Japan, E32K mutation (slight gain-of-function mutation), which contributes to a relatively small increase in the LDL-C level, is identified in 1 to 2% of the general population and in 6% of clinically diagnosed FH patients. When E32K mutation is concomitantly present in LDL-receptor-related heterozygous FH, the clinical features may become like homozygous FH[11]; however, these patients respond to drug therapy more effectively than an FH patient homozygous for a mutation in the LDL receptor gene.

Significance of Diagnosis and Treatment

Among untreated heterozygous FH, the risk for the development of CAD is 20 times higher than in those treated. No symptom might be observed except for hyper-LDL cholesterolemia before CAD events. It is therefore important to make a definitive diagnosis at an early stage of life and to start appropriate treatment to prevent juvenile death, even when patients are still asymptomatic. FH should be positively suspected in patients with hyper-LDL cholesterolemia to make a definitive diagnosis through surveys of ten-

Table 1. Diagnostic criteria for adult (15 years or older) heterozygous FH

1. Hyper-LDL cholesterolemia (LDL-C before treatment: 180 mg/dL or more)
2. Tendon xanthoma (tendon xanthoma on the dorsal hands, elbows, and knees, or Achilles tendon thickening) or nodular xanthoma on the skin
3. Family history within the second-degree relatives FH or premature CAD

- A diagnosis should be made after ruling out the possibility of secondary hyperlipidemia.
- Patients meeting 2 items should be regarded as having FH. Concerning those meeting 1 item, refer to Fig. 4. When FH is suspected, genetic tests should be conducted to make a diagnosis.
- Nodular xanthoma on the skin does not include palpebral xanthoma.
- Patients with Achilles tendon thickening (9 mm or more) on radiography should be regarded as having xanthoma.
- When the LDL-C is 250 mg/dL or more, FH should be strongly suspected.
- During drug therapy, the pretreatment lipid level should be employed as a reference value.
- CAD in males younger than 55 years old and females younger than 65 years old is defined as premature CAD.
- When a diagnosis of FH is made, the patient's family should also be investigated.

don/skin xanthoma and of family members. When a definitive diagnosis of heterozygous FH is made, the patient's family should be examined extensively for early diagnosis/treatment.

Diagnosis of Heterozygous FH

FH is characterized by the presence of type IIa or IIb dyslipidemia, Achilles tendon thickening, skin xanthoma, and corneal arcus. Achilles tendon thickening is evaluated based on the results of inspection and palpation (**Fig. 5**); however, when diagnosis is difficult, radiography should be conducted to measure the maximum thickness of the Achilles tendon. Patients with a maximum thickness of 9 mm or more are regarded as having thickening of the tendon (**Fig. 6**). Skin/tendon xanthoma frequently develops on the extensor sides of the hands, elbows or knees. The onset of premature CAD (age at onset: less than 55 years in males, less than 65 years in females) in the family is frequent.

New Diagnostic Criteria for FH

Mabuchi *et al.* reported that the mean LDL-C levels in genetically diagnosed heterozygous FH patients and their relatives without FH were 260.8 and 114.8 mg/dL, respectively, suggesting the definition of heterozygous FH with an LDL-C level over 161 or 163 mg/dL[12].

Previous diagnostic criteria included a total cholesterol level of 260 mg/dL or more as a major symptom, but Japanese guidelines for the management of atherogenic risks use LDL-C, so that the use of LDL-C is chosen as a more appropriate parameter to diagnose and treat FH. Based on the results of previous and preliminary studies, hyper-LDL cholesterolemia, Achilles tendon thickening, skin xanthoma, and the presence of FH or premature CAD in first and/or second degree relatives were established as major symptoms, and patients with 2 or more of these symptoms were regarded as having FH. To determine the cut-off level of LDL-C for the diagnosis, we analyzed the before-treatment data obtained from 1,356 patients who had consulted the Lipid Clinics (FH patients: 419, non-FH: 937) of the National Cerebral and Cardiovascular Center, Osaka University, Kyoto University, Chiba University, Nippon Medical University, and Kanazawa University. When establishing a cut-off as 180 mg/dL or more for LDL-C, the sensitivity and specificity were 94.3 and 99.1%, respectively. When establishing the cut-off as 190 mg/dL or more, the sensitivity and specificity were 92.1 and 99.1%, respectively; therefore, 180 mg/dL was employed as a more sensitive cut-off of LDL-C (Study Group of Primary Hyperlipidemia, Research report in fiscal year 2011). In this analysis, the sensitivity and specificity of LDL-C were higher than those of total cholesterol. The new diagnostic criteria prepared in this study are shown in **Table 1**. The flow chart of diagnosis is shown in **Fig. 7**. In this analysis, non-FH patients accounted for 5% of those with an LDL-C level of 250 mg/dL or more. The subjects of this analysis were those who had been followed-up at Lipid Clinics, so that this proportion should be lower in general clinical practice; therefore, it is described in our diagnostic criteria that FH should be strongly suspected when the LDL-C level is 250 mg/dL or more. FH patients with no Achilles tendon thickening but with corneal arcus are extremely rare; therefore, corneal arcus, included in the previous diagnostic criteria, was not employed in the present diagnostic criteria. Lymphocytic LDL receptor activity assay was not employed as a standard measure because there is no data accumulation.

When serious illnesses develop concomitantly, including acute myocardial infarction, LDL-C may significantly decrease and hyper-LDL cholesterolemia

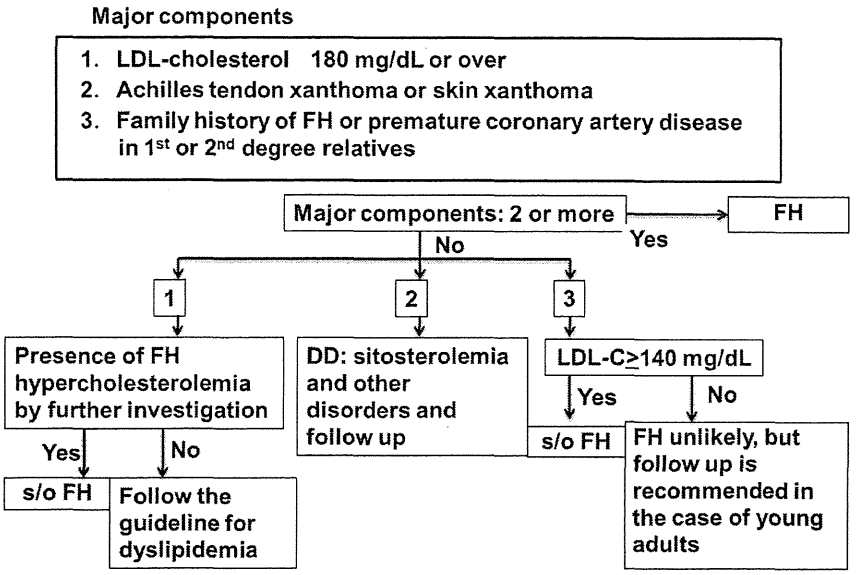

Fig. 7. Flow chart of FH diagnosis. DD: differential diagnosis

may not be apparent even in FH patients on admission. Therefore, palpation should be performed to examine the Achilles tendon and a survey of the family history must be conducted for all the patients with acute myocardial infarction.

Radiography of the Achilles Tendons

Thickening of the Achilles tendons can be measured using radiography. The angle between the lower leg bone and sole should be 90 degrees. An angle of incidence should be established involving the fibular lateral malleolus from the lateral side. The imaging distance is 120 cm. The following imaging conditions should be employed: 50 kV and 5.0 mA. Ultrasonic assessment of Achilles tendon thickening is also possible; however, it has not yet been standardized.

Differential Diagnosis

Secondary hyperlipidemia with hyper-LDL cholesterolemia (i.e. diabetes, hypothyroidism, and nephrotic syndrome), as well as familial combined hyperlipidemia (FCHL), should be differentiated from FH. FCHL can be differentiated based on the absence of tendon xanthoma, presence of small dense LDL, presence of other types of dyslipidemia (types IIa, IIb, and IV) in the family, and a less marked increase in LDL-C during childhood in comparison with FH patients. Key points to differentiate FH from FCHL are summarized in **Table 2**.

Risk Factors for Heterozygous FH and the Target Level of LDL-C in Lipid Control

The age at onset of CAD and the rate of its deterioration vary among heterozygous FH patients. Risk factors reportedly play roles in the development of CAD in Japan, as indicated by the following studies: Yagi *et al.* investigated 117 patients with heterozygous FH in the Hokuriku district and identified diabetes and hypo-HDL cholesterolemia as significant risk factors[13]. Sugisawa *et al.* reported that LDL-C 260 mg/dL or higher and/or Achilles tendon thickness 14.5 mm or thicker are useful markers for detecting patients at "very high" risk for CAD[14]. Hirobe *et al.* also indicated the involvement of hypo-HDL cholesterolemia in the onset of CAD in FH patients[15]. Yanagi *et al.* reported the involvement of diabetes and impaired glucose tolerance[16]. Nakamura *et al.* emphasized the importance of visceral fat[17]. Furthermore, the Primary Hyperlipidemia Research Group indicated that hypertriglyceridemia and hypo-HDL cholesterolemia were frequent in FH patients with CAD[18]. In The Netherlands, Rana *et al.* reported that the risk of cardiovascular events increased 1.5-fold in FH patients with metabolic syndrome, which was diagnosed using the NCEP-ATPIII criteria based on the analysis of 1,698 patients with FH[19]. Jensen *et al.* conducted a cohort study involving 2,400 patients with heterozygous FH in the Netherlands, and indicated that the risk of cardiovascular events increased 1.5-fold when lipoprotein (a) was 30 mg/dL or more[20, 21]. Holmes *et al.* performed a cohort study involving patients with heterozygous FH in Canada, and reported that the risk of

Table 2. Comparison of familial hypercholesterolemia (FH) and familial combined hyperlipidemia (FCHL)

	Heterozygous FH	FCHL
Causative gene	LDL receptors, PCSK9, Apo B-100 (single gene abnormality)	Although USF-1 and LPL have been reported as candidates, no causative gene has been identified (multi-gene abnormalities).
Frequency	1 per approximately 500 persons	1 per approximately 100 persons
Lipid profile	IIa in most patients, IIb in some patients	During the course, both patients and their families may show 3 phenotypes, IIa, IIb, and IV.
Achilles tendon thickening, skin xanthoma	Present	Absent
Juvenile corneal arcus	Present	Absent
Presence of small-dense LDL	Not frequent	Frequent
Insulin resistance	Not frequent	Frequent

PCSK9: proprotein convertase subtilisin/kexin type 9
USF-1: upstream transcription factor 1, LPL: lipoprotein lipase

cardiovascular events increased 2.5-fold in those with lipoprotein (a) of 56 mg/dL or more[22]. Recently, Nenseter *et al.* indicated that lipoprotein (a) of 35 mg/dL or more was a significant risk factor for heterozygous FH[23].

Risk factors for CAD include age, hypertension, diabetes (including impaired glucose tolerance), chronic kidney disease (CKD), family history of CAD, hypo-HDL cholesterolemia, and smoking in the guidelines for the prevention of atherosclerosis in 2012. Based on these previous studies, this guideline defines risk factors for CAD development in FH patients; an age of 30 years or older in males/45 years or older in females (or postmenopausal women), pretreatment LDL-C of 260 mg/dL or more, Achilles tendon thickening (15 mm or more), lipoprotein (a) of 50 mg/dL or more, and metabolic syndrome. Therefore, to prevent CAD in FH patients, risks other than LDL-C should also be extensively managed.

Since the CAD risk is very high in heterozygous FH, the target level of LDL-C management should be set similarly to secondary prevention; less than 100 mg/dL; however, this target may not be achievable in many cases of FH. Even when LDL-C does not reach the target level, physicians may be advised to target LDL-C reduction more than 50% of the pretreatment value. In the ASAP study, 325 patients with FH were assigned to receive high-dose atorvastatin (80 mg/day) or simvastatin (40 mg/day), and were followed-up for 2 years. LDL-C in the atorvastatin group (LDL-C: 308 to 149 mg/dL) decreased more markedly than in the simvastatin group (LDL-C: 321 to 185 mg/dL). When measuring IMT using carotid ultrasonography, intima-media thickness (IMT) increased in the latter whereas it significantly decreased in the former[24]; therefore, 50% or more decrease in LDL-C may be beneficial. As clinical studies without lipid-lowering therapy cannot be conducted on FH patients for ethical reasons, there is no evidence regarding these target levels. Even when LDL-C reaches the target goal, the absence of events may not be assured. It should be noted that the risk chart published by the Japan Atherosclerosis Society cannot be applied for risk assessment of FH patients.

The goal of the treatment in this guideline should be employed for FH patients aged 30 years or older. As a rule, treatment should be performed under specialist guidance. In particular, FH patients aged 15 to 29 years must be treated under specialist guidance. Therapeutic strategies for women who may become pregnant are described in another section.

Treatment of Heterozygous FH

As FH patients show severe hyper-LDL cholesterolemia, the direct influence of lifestyle modifications on the plasma lipoprotein profile, such as diet and exercise, and on the prognosis remains uncertain; however, lifestyle modifications may positively influence other risk management so they are recommended even when the rate of LDL-C decrease is limited to 2 or 3%. It is extremely important to instruct smokers to quit smoking.

Diet Therapy

Diet management should also be performed for

Table 3. Diagnostic criteria for heterozygous FH in children

1. Hypercholesterolemia: Pretreatment LDL-C level ≥ 140 mg/dL (When the total cholesterol level is 220 mg/dL or more, LDL-C should be measured.)
2. Family history of FH or premature CAD (second degree relative)

- As clinical symptoms such as tendon xanthoma are absent in children, FH diagnosis in their families is important.
- As there are changes in LDL-C level during the growth period, close follow-up is necessary.
- CAD in males younger than 55 years old and females younger than 65 years old is defined as premature CAD.

FH patients. There is no diet therapy regimen specific to FH, and standard diet therapy for hypercholesterolemia should be applied. Maintaining good compliance is more important for FH patients.

Exercise Therapy

Exercise should also be helpful for FH patients; however, the presence of insidious CAD should be carefully screened before starting exercise therapy, as their CAD risk is high. History taking, electrocardiography, stress electrocardiography, and echocardiography should be performed to evaluate coronary function. When the presence of CAD is suspected, exercise therapy should be conducted cautiously after appropriate treatment.

Drug Therapy

Normalization of the plasma lipid and lipoprotein profile cannot be achieved in most FH patients by lifestyle modification alone. Drug therapy is therefore required. HMG-CoA reductase inhibitors (statins) are chosen as first-line drugs. Retrospective analysis of 329 patients with heterozygous FH in Japan demonstrated that the use of statins significantly delayed the onset of CAD[25].

Statin therapy should be started with an "initial" dose, and the dose should be adjusted while observing its efficacy and side effects. The LDL-C-lowering effects of statins are potentiated in a dose-dependent manner, but the frequency and severity of side effects may also increase. It is necessary to examine the presence of myalgia by inquiry, and to evaluate liver function parameters, such as AST and ALT, as well as CK, particularly cautiously during the initial month and thereafter periodically. Rhabdomyolysis is the most serious side effect and should not be overlooked.

When patients do not respond to monotherapy with statins, combination therapy with other lipid-lowering agents may be beneficial to decrease LDL-C, such as ezetimibe, bile acid resins (cholestyramine, colestimide), probucol, fibrates, and nicotinic acid derivatives. However, it remains undermined whether these combination therapies are more effective to inhibit or delay the onset of cardiovascular events in patients with FH than monotherapy with statins. In the ENHANCE study, 720 patients with heterozygous FH were assigned to receive 80 mg/day simvastatin alone or combination therapy with 10 mg/day ezetimibe, and followed-up for 24 months. In the combination therapy group, the rate of LDL-C decrease was significantly greater (combination therapy group: 58% vs. monotherapy group: 41%, $p<0.01$). However, there was no significant difference in the carotid IMT between the two groups (combination therapy group: 0.0111 mm vs. monotherapy group: 0.0058 mm)[26]. IMT is a surrogate marker for CAD and the IMT values measured before the treatment were within the normal range in this trial, so it should be clarified in the future whether additional therapy significantly reduces thickened IMT and inhibits the onset of cardiovascular events in FH patients.

For patients with statin intolerance due to side effects such as myalgia and liver dysfunction, monotherapy or combination therapy with the above lipid-lowering agents should be performed in order to avoid or reduce the dose of statins. In Japan, the results of a retrospective study suggested that probucol delays the onset of recurrent CAD in patients with heterozygous FH[27].

To establish safe treatment for FH patients, a long-term, large-scale study is necessary. Extensive drug therapy with statins or a combination should be performed for FH patients based on carefully informed consent by the patients and/or their family after evaluating the balance between the risk of atherosclerosis and safety of the treatments. For safety evaluation of combination therapy, a study involving 248 adolescent males and females with heterozygous FH showed that there was no increase in the incidence of adverse reactions in patients receiving combination therapy with simvastatin and ezetimibe in comparison with monotherapy with simvastatin[28]; however, combination therapy with statins and fibrates is contraindicated in the presence of kidney dysfunction.

LDL Apheresis for Heterozygous FH

For heterozygous FH patients, LDL apheresis is covered by public health insurance when the total cholesterol exceeds 400 mg/dL in a steady state under diet therapy and does not decrease to 250 mg/dL or less by drug therapy in the presence of coronary lesion. It is appropriate to choose LDL apheresis for drug-resistant FH patients with severe CAD.

Screening/Follow-Up of Cardiovascular Disease (CVD)

Heterozygous FH patients develop systemic atherosclerotic disorders including CAD in the early stage of life, so early screening for these disorders is necessary. When atherosclerotic vascular disorders are found, early treatment and careful follow-up should be carried out. CAD can be fatal, so screening for CAD should be performed at 1- to 2-year intervals. For the diagnosis of CAD, history taking, electrocardiography, stress electrocardiography (master double, ergometer, and treadmill), echocardiography, and stress myocardial scintigraphy are to be performed. When CAD is suspected in these examinations, coronary multidetector-row computed tomography (MDCT) should be conducted to identify the site of coronary stenosis. When stenosis is suspected on MDCT, coronary angiography should be performed; however, FH patients frequently show calcification, which sometimes makes it difficult to make a diagnosis. Coronary angiography findings characteristic of heterozygous FH include marked stenotic lesions at the origin and dilative lesions downstream (coronary aneurysms). Coronary lesions in FH patients are often severe and multi-vessel. As systemic atherosclerosis develops in most patients with heterozygous FH, examination procedures must be performed carefully, considering complications such as thrombosis/embolism.

To evaluate carotid atherosclerosis in patients with heterozygous FH, vascular murmur hearing and carotid ultrasonography are helpful. When stenosis is suspected, MR angiography, CT angiography, or angiography should be performed. To evaluate the presence of cerebral infarction, magnetic resonance imaging (MRI) and CT should be carried out if necessary.

In some patients with heterozygous FH, peripheral arterial disease (PAD) develops concomitantly; therefore, the presence of intermittent claudication should be investigated by history taking. To evaluate arteriosclerosis of the femoral artery, the ankle-brachial blood pressure index (ABI) should be measured. In addition, when stenosis is suspected, femoral artery ultrasonography (Doppler method), CT angiography, and MR angiography should be performed.

To assess valvular disorders such as aortic valve stenosis (AS), echocardiography should be conducted. In severe cases having a decrease in the aortic valve orifice area with marked differences in the aortic valve pressure, aortic valve replacement may be performed. When selecting this procedure, the preoperative assessment of concomitant arteriosclerotic lesions, especially CAD, is necessary.

FH in Children

CAD may not be an apparent clinical manifestation in children with heterozygous FH; however, autopsy findings in the Bogalusa Heart Study[29] and Pathological Determinants of Atherosclerosis in Youth (PDAY)[30] demonstrated that atherosclerotic changes were already present in children; therefore, starting intervention for dyslipidemia in childhood is important to prevent CAD in heterozygous FH.

Diagnosis of Heterozygous FH in Children

Hyper-LDL cholesterolemia is already present at birth in heterozygous FH. In many of these patients; however, hyper-LDL cholesterolemia-related physical symptoms may not become apparent in childhood, such as Achilles tendon xanthoma and corneal arcus. Therefore, FH in children is diagnosed based on hyper-LDL cholesterolemia and the family history. To make a diagnosis of FH in children, their parents' history of FH is essential; therefore, it is important to make a definitive diagnosis of FH in the parent when either parent has hyper-LDL cholesterolemia. Diagnostic criteria for FH in children are listed in **Table 3**. As 95% of healthy children show LDL-C of 140 mg/dL or less[31], a reference value for screening was established as 140 mg/dL. Briefly, 1 per 15 to 25 children with LDL-C of 140 mg/dL or more may have FH.

FH in Screening of Children

FH can be screened during infancy[32]. Definitive diagnosis of FH should be made before the age of 10 to consider intervention, anticipating an effect of diet therapy on psychosomatic growth and the development of arteriosclerotic lesions. Ideally, FH screening should be conducted by measuring the serum lipid level once around 10 years of age in all children[33, 34]. Average Japanese children undergo a hematology and blood biochemistry check-up at least a few times before they reach 10 years old of age on various occasions, including health checks and consultations for diseases. The examination mostly includes a total cholesterol

test; therefore, the awareness of the guidelines by pediatricians should increase the chance of early identification of FH.

When a pediatric patient has LDL-C exceeding 140 mg/dL, a family member(s) diagnosed with FH, and a family history of hyper-LDL cholesterolemia or premature CAD, extensive examination must be performed to make a definitive diagnosis. The diagnostic criteria shown in **Table 3** were prepared to screen heterozygote FH children. When FH is suspected, it is necessary to refer them to a specialist.

Risk Factors for FH in Children

FH patients with risk factors for CAD have a higher risk, especially for hyper-LDL cholesterolemia and obesity, thickened IMT during childhood, coronary calcification, and vascular endothelial dysfunction during adulthood[29, 35-37]. Therefore, assessment and management of risk is important during childhood. Primary risk factors in children with heterozygous FH include a family history of CAD (within second degree relatives), obesity (degree of obesity: 20% or more), diabetes (including impaired glucose tolerance), hypertension (>125/70 mmHg), hypo-HDL cholesterolemia, and smoking. The number of these primary risk factors can be used as an index for treatment.

Diagnosis of Atherosclerotic Disease in Children

To evaluate atherosclerosis in children with heterozygous FH, non-invasive methods should be employed. IMT measurement with carotid ultrasonography is useful for evaluation of the deterioration of atherosclerosis and treatment response.

Treatment of Heterozygous FH in Children

Nutritional Guidance and Lifestyle Modification

When a diagnosis of heterozygous FH is made, the patients and parents should be advised to improve their lifestyle as early as possible. Those who smoke must stop smoking for their lifetime. It is also important to encourage their families to cease smoking. When the weight of the patients is within +20% of the standard body weight (in the absence of obesity), the dietary fat content should be less than 30% of the standard total energy intake with the saturated fatty acid 7 to 10%. Dietary cholesterol intake should be limited to 300 mg/day[38, 39]. When the body weight exceeds 20% of the standard body weight, energy intake and dietary fat/saturated fatty acid must be limited similarly to obesity treatment. Initial diet therapy should continue for 6 months to 1 year, as described in the NCEP guidelines in the United States[35]. A common standard Japanese diet is acceptable for the first step as far as it meets the requirement above, and more active dietary intervention is conducted in the second step. Lifestyle modification is useful for reducing risk factors; however, LDL-C decrease is insufficient for many patients[40-42]. The effect must be reviewed while considering potential drug therapy.

Drug Therapy

Evidence has not yet been established regarding the age at which heterozygous FH patients should start drug treatment. Nevertheless, appropriate LDL-C control should be achieved because atherosclerotic changes of coronary arteries are observed at a young age in these patients. The American Academy of Pediatrics proposed that lipid-lowering therapy should be initiated in children with LDL-C of 190 mg/dL or more, those with LDL-C of 160 mg/dL or more having a family history of premature CAD, and those with 2 or more risk factors. When the effects of lifestyle modification are not sufficient, drug therapy should be considered in boys aged 8 to 10 years or older or girls experiencing menarche[34]. In high-risk patients with tendon xanthoma/aortic valve stenosis or a family history of CAD, differentiation from homozygous FH is important and drug therapy may be initiated at a young age.

For drug therapy for children with heterozygous FH, bile acid resins have been chosen as the first-line agent as they may be considered a relatively low risk for development and growth; however, these resins do not have high potency for decreasing LDL-C, and compliance does not seem to be high. On the other hand, an increasing number of clinical studies have demonstrated the safety and efficacy of statin therapy in pediatric to adolescent patients with heterozygous FH; in children with heterozygous FH, simvastatin improved endothelial function, and 2-year pravastatin therapy reduced carotid IMT[43]. With respect to safety, several studies involving approximately 200 adolescent patients with heterozygous FH, aged 8 years or older, reported that short-term (2 years or less) therapy with lovastatin, simvastatin, pravastatin, or rosuvastatin effectively decreased LDL-C without influencing development, sexual maturation, testis volume, and blood gonadotropin and liver/muscular enzyme levels[44-47]. Based on these findings, statin therapy may be chosen when thickening of the Achilles tendon or an increase in the IMT is observed in children with FH; however, drug therapy for FH in children should be under cautious and extensive guidance by specialists.

Heterozygous FH in Women

Premenopausal Women

Although lifestyle modification is a basic strategy for female patients, it is necessary to decrease LDL-C by drug therapy[48]. The patients should consult specialists in dyslipidemia treatment after adolescence to decide the timing and types of drug therapy, considering the risks in individual patients.

There are two female-specific problems with FH drug therapy; pregnancy and oral contraceptives (OCs). During pregnancy, drug therapy other than bile acid resins therapy should be avoided due to the risk of fetal anomalies. According to the National Institute for Health and Clinical Excellence, drug therapy should be promptly discontinued when patients are identified to be pregnant. Drug administration should be discontinued for 3 months before trying to become pregnant.

OCs are administered to many patients not only for contraception, but also to relieve menstrual pain and hypermenorrhea. The influence of OCs on the risk of myocardial infarction in healthy females was investigated and showed that the increase of the odds ratios with first- and second-generation OCs was 2.21 (1.30-3.76) and 2.17 (1.76-2.69), respectively, but there was no increase with third-generation OCs, 1.27 (0.96-1.67)[49]. Another study regarding combination therapy with statins and OCs involving healthy women reported that statins exhibited lipid-lowering effects without reducing the hormonal effects of OCs[50], showing that combination therapy with OCs and statins is not always contraindicated for premenopausal women with FH. However, the scale of their study was relatively small and the confidence interval was large; therefore, when selecting combination therapy, the risks/benefits must be sufficiently explained.

Postmenopausal Women

LDL-C increases in women after menopause[51], and the increase in FH patients is greater than in healthy women. It is therefore necessary to reduce LDL-C more intensively by lifestyle modification and drug therapy with statins.

Hormonal replacement therapy (HRT) to treat climacteric disturbance also improves lipid metabolism by decreasing LDL-C and increasing HDL-C[52]. A study involving healthy postmenopausal women reported that the combination of HRT and statin therapy potently decreased LDL-C[53]. Another observational study indicated that HRT reduced the risk of myocardial infarction[54]; however, double-blind studies, the Women's Health Initiative (WHI)[55] and Heart and Estrogen/Progestin Replacement Study (HERS)[56], ruled out primary/secondary preventive effects of HRT on the risk of myocardial infarction. In fact, the results indicated that the risk increased; however, a recent observational study regarding percutaneous estrogen administration in Europe reported that the risk of myocardial infarction significantly reduced, differing from oral administration[57]. Therefore, it may be important to consider differences related to the route of administration when evaluating the risk of myocardial infarction. The influence of HRT in postmenopausal women with FH on the risk of myocardial infarction remains to be clarified.

Diagnosis of homozygous FH

Homozygous FH can be diagnosed based on clinical features: serum total cholesterol of 600 mg/dL or more, cutaneous xanthoma, premature CAD during childhood, and parents' family history of heterozygous FH. Cutaneous xanthoma during childhood is characteristic of this disease. On many occasions, patients may initially consult a dermatologist. When it is difficult to differentiate homozygous from severe heterozygous FH, it is possible to make a definitive diagnosis based on the detection of reduced LDL receptor activity in fibroblasts/lymphocytes (20% or less of the normal activity)[58] and LDL-receptor, ApoB or PCSK9 gene mutations.

A condition in which mutations of the LDL receptor or ApoB gene, or gain-of-function mutations of PCSK9 are present in both alleles is defined as homozygous FH. It is impossible to clinically differentiate homozygous FH patients with LDL-receptor mutation from those with PCSK9 mutations[59, 60]. Response to treatment may differ in the two types of homozygous FH. Since the incidences of both types are relatively high, concomitant development of LDL-receptor and PCSK9 abnormalities must be taken into account. LDL-receptor and PCSK9 mutations are present in respective alleles in these patients, so that they are not genetically homozygotes. This condition is, however, clinically undistinguishable from homozygous FH in some cases. A few other types of primary hypercholesterolemia to be differentiated exist, although cases are relatively rare. Autosomal recessive hypercholesterolemia (ARH) is one such type. Homozygous ARH is also impossible to differentiate from homozygous FH based on clinical features of the patient. LDL receptor adapter protein 1 (LDLRAP1) is a gene causing ARH[61], and heterozygous ARH is usually asymptomatic; therefore, the family history is important[62]. Sitosterolemia also causes xanthoma and

CVD in many cases, similarly to FH. It induces a marked increase in the blood vegetable sterol concentration due to ABCG5 or ABCG8 mutations[63]; however, most patients show normal LDL-C[64, 65]. For differential diagnosis, the family history and measurement of the blood levels of plant sterols (sitosterol, campesterol) are useful. Cerebrotendinous xanthomatosis (CTX) is known as an autosomal recessive disease with marked tendon xanthoma[66]. In the presence of this disease, sterol 27-hydroxylase abnormalities increase the blood cholestanol level, causing mental retardation and/or neurological symptoms as well as marked tendon xanthomatosis; however, CTX does not show hypercholesterolemia. As a disease with a similar condition, pseudohomozygous type II hypercholesterolemia is known, although its pathogenesis is genetically unclear. Patients' parents do not show marked hypercholesterolemia and favorably respond to diet therapy and bile acid resins[67, 68].

Drug Therapy for Homozygous FH

As described for patients with heterozygous FH, lifestyle modification also comprises basic treatment for patients with homozygous FH; however, the risk of CAD onset/deterioration is markedly high in these patients. Therefore, potent LDL-C-lowering therapy is required at a young age; however, most LDL-lowering agents act through increasing LDL receptor activity, such as bile acid resins and statins, so these drugs are not expected to be effective for the majority of homozygous FH where the LDL receptor is absent. Probucol exhibits limited specific LDL-C-lowering effects in patients with homozygous FH. A study reported that probucol therapy led to the reduction/disappearance of skin/Achilles tendon xanthoma[69]; therefore, LDL apheresis therapy at 1- to 2-week intervals is necessary. Drug treatment including statins generally helps to decrease the rebound rate of LDL-C after individual LDL apheresis treatment. In the statement "to decrease the risk of cardiovascular events in high-risk children" published by the American Heart Association (AHA) in 2006, it is recommended that combination therapy with high-dose statins or ezetimibe be started at a young age in addition to LDL apheresis[35].

In homozygous FH patients who wish to become pregnant, CAD/aortic valve stenosis/supravalvular stenosis screening should be conducted for the continuation of pregnancy and easy delivery. If necessary, appropriate treatment must be performed[70]. An animal experiment showed the teratogenicity of statins. As the safety of lipid-lowering agents other than bile acid resins, regarding administration to humans during pregnancy, has not been established, physicians must instruct hetero-/homozygous FH patients to discontinue therapy with lipid-lowering agents other than bile acid resins at least 3 months before trying to become pregnant and during the lactation period after delivery.

LDL Apheresis in Patients with Homozygous FH

Start of LDL Apheresis Therapy

When LDL apheresis therapy was introduced after 10 years of age, the prognosis of the patient was unfavorable[71], so treatment should be started as early as possible[72]; however, it is difficult to start this therapy until children become able to cooperate, such as resting during the treatment, usually at 4 to 6 years of age. There are some infants with coronary stenosis, complete obstruction, aortic valve stenosis, or supravalvular stenosis. During infancy, the plasma exchange method may be chosen since the extracorporeal volume of the device is small.

Effects of LDL Apheresis

Some studies have reported that LDL apheresis therapy was safe in children without influencing development/growth, and that this therapy led to the reduction/disappearance of skin xanthoma, inhibiting exacerbation of aortic valve stenosis/supravalvular stenosis, which are characteristic of homozygous FH, and coronary lesions, or resulting in improvement[73-80]. Iron-deficiency anemia is the most frequent side effect. During treatment, blood pressure may fall due to a decrease in the circulating blood volume in some patients. LDL apheresis therapy should be performed with caution, particularly in patients with aortic valve/CAD. LDL apheresis using LDL adsorption columns needs special attention as they have a negative charge, increasing the bradykinin level by activating the blood coagulation system. Therefore, combination therapy with angiotensin-converting enzyme (ACE) inhibitors may cause anaphylactic symptoms and is contraindicated.

Pregnancy/Delivery in Patients with Homozygous FH

It is important for patients with homozygous FH to plan their pregnancy. Before pregnancy, screening should be conducted using echocardiography, electrocardiography, stress electrocardiography, and carotid ultrasonography to evaluate CVD. Lipid-lowering agents other than bile acid resins must be discontinued 3 months before trying to become pregnant. In FH patients, LDL-C and triglyceride further increase during pregnancy. In particular, these two parameters increase by approximately 30 and 100%, respectively,

after week 24 of pregnancy[81]. In FH patients, the blood coagulation capacity and platelet function are enhanced during pregnancy, increasing blood viscosity[82]. In pregnant women with homozygous FH, uteroplacental blood flow is less abundant than in those with normal pregnancy. In addition, LDL apheresis therapy improves blood flow[83]. In the third pregnancy trimester, especially on delivery, high-intensity stress is added to the cardiovascular system; therefore, LDL apheresis should be performed during pregnancy. Several studies have reported that LDL apheresis therapy could be safely conducted during pregnancy, leading to delivery[72, 84, 85]. During lactation, lipid-lowering agents other than bile acid resins should also be discontinued, and periodic LDL apheresis therapy must be continued for LDL-C control.

Designation of Homozygous FH as a Specific Disease

In October 2009, homozygous FH was designated as a disease to be covered in the Specific Disease Treatment Research Business in Japan. Designation criteria include marked hypercholesterolemia, presence of skin xanthoma during childhood, and resistance to drug therapy in addition to a definitive diagnosis made on analysis of genes involved in the route of LDL metabolism or measurement of LDL receptor activity. Therefore, in addition to typical homozygous FH patients, severe hypercholesterolemic patients who resist drug therapy, even though genetic diagnosis had not led to a definitive diagnosis, are also included. These patients can receive a subsidy for the health expenditure for chronic LDL apheresis. The procedures for FH specific disease authorization are described on the homepage of the Specific Disease Treatment Research Business published by the Japan Intractable Diseases Information Center, Ministry of Health, Labour and Welfare (http://www.nanbyou.or.jp/what/nan_kenkyu_45.htm).

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