Nova Biomedical

STATINS

Indications and Uses, Safety and Modes of Action

Gunnar N. Holmqvist
Editor

NOVA

STATINS: INDICATIONS AND USES, SAFETY AND MODES OF ACTION

GUNNAR N. HOLMQVIST
EDITOR

Nova Science Publishers, Inc.
New York

LIBRARY OF CONGRESS CATALOGING-IN-PUBLICATION DATA

Available upon request

ISBN: 978-1-60692-103-6

Published by Nova Science Publishers, Inc. † *New York*

STATINS: INDICATIONS AND USES, SAFETY AND MODES OF ACTION

PREFACE

This new book focuses on statins which are a relatively new group of drugs used to lower blood cholesterol levels. A high cholesterol level increases a person's risk of having a heart attack or stroke. The long-term use of statins reduces the risk of such an event and can increase the life expectancy of people with a history of heart disease. The statins work by blocking an enzyme in the body that is involved in the production of LDL cholesterol, especially in the liver. This enzyme is known as HMG coenzyme A reductase. The statins are the most effective group of drugs for lowering the levels of LDL cholesterol in the body. Potential side-effects include muscle cramps and gastrointestinal upsets. These are usually resolved on temporarily lowering the dose. Liver enzyme derangements may occur, which generally return to normal after briefly discontinuing the drug. Some report headaches. Other side-effects occur rarely.

Chapter 1 - The authors propose a new concept, 'Vascular Failure,' to detect early stage atherosclerosis, which is characterized as integration of endothelial dysfunction, smooth muscle cell dysfunction and metabolic abnormality of the vessel wall. Also, 'Vascular Failure' occurs not only in atherosclerosis, but also in vasculitis as well as systemic inflammatory disorders, which is of great interest. Statins targeting 'Vascular Failure' should be appled in the earlier stage, even when anatomical vascular abnormalities are not present.

Chapter 2 - Statins have been shown to be remarkably potent in reducing cardiovascular events and improving patient's survival. Overall, the statins show a rather good safety profile. Apart from reports of rare life threatening side effects, mainly from rhabdomyolysis (particularly in combination treatment with fibrates), knowledge and understanding on a great variety of different side effects occurring quite frequently is limited until now. This review on side effects in statin monotherapy attempts to analyse whether there are predisposing factors or specific properties of one or the other of the compounds concerning the development of side effects such as hepatic and renal function, erectile dysfunction, muscular problems and others. Types, prevalence and association with concurrent diseases are described. Underlying biochemical aspects, such as oxidation injury and eventual therapeutic interventions, are discussed. It seems that the rare severe statin side effects can completely be avoided by adhering to 5 simple rules, while the mild side effects probably are much more prevalent than considered so far.

Chapter 3 - The statins are widely used for the prevention of cardiovascular diseases thanks to their lipid lowering effects. Recent reports suggest that statins have antihypertensive and antiinflammatory properties besides the lipid lowering one. Moreover, in animal models of hypertension and kidney disease, the statins induced both the eNOS up-regulation and the iNOS, LOX-1 and NFkB down-regulations in the renal endothelium promoting the increase of the perfusion lowering the sclerosis as well. All these effects of statins might be helpful in reducing the incidence of cardiovascular diseases in human hypertensives whose clinical picture is often characterized by risk factor such as dyslipidemia, elevated serum C-reactive protein levels and nephrosclerosis with reduced renal vascular reserve and microalbuminuria.

Aim of this prospective, self-controlled, interventional study was to assess the trends of the renal hemodynamics, the blood pressure, the lipid panel and the inflammation during a short course of atorvastatin therapy, in essential hypertensives (EH).

After a 5-days run-in period, the patients began atorvastatin therapy (10 mg q.d.). At T_0 (before the first tablet of atorvastatin) and T_{30} (after 30 days of therapy) the patients underwent the following assessments: systolic, diastolic and mean blood pressure (SBP, DBP and MAP respectively), glomerular filtration rate (GFR) and effective renal plasma flow (ERPF) (inulin and p-aminohippurate clearance respectively), filtration fraction (FF) (GFR/ERPF) and total renal vascular resistances (TRVR) (using ERPF, hematocrit and MBP), microalbuminuria and serum levels of lipids and C-reactive protein (hs-CRP). The patients followed a standard diet (35 Kcal/kg bw/day, 1 g of protein/kg bw/day and 6 g of sodium/day). Never treated patients with I stage EH were included. Patients with heart, kidney or liver diseases were excluded as well as patients taking any other medication.

This report provides preliminary data about 6 EH (3M/ 3F), 45.5 ± 14.8 (mean ± SD) years old so far enrolled. The patients have been compliant with the atorvastatin therapy as showed by the reduction of serum lipids (mg/dL) (total cholesterol: T_0: 226.8 ± 33.7 vs T_{30}: 164.5 ± 46.5, p=0.01; triglycerides: T_0: 157.8 ± 50.2 vs T_{30}: 89.8 ± 16.5, p=0.05; apo-B: T_0: 110.3 ± 24.2 vs T_{30}: 75.5 ± 28.8, p=0.003). Mean daily protein and sodium intake as well as serum hs-CRP levels, microalbuminuria and renal hemodynamics were unchanged during the study instead of blood pressure which was significantly reduced (mmHg) (SBP: T_0 : 150.3 ± 10.6 vs T_{30}: 139.1 ± 6.9, p=0.01; DBP: T_0 : 97.4 ± 11.5 vs T_{30}: 88.6 ± 9.4, p=0.04; MAP: T_0 : 115.0 ± 11.0 vs T_{30}: 105.5 ± 7.9, p=0.02).

This preliminary experience on EH suggest that in EH a short course of atorvastatin therapy is associated with the statistically significant reduction of both the serum lipids levels and the blood pressure and the restoration of the glomerular autoregulation. The absence of changes of the serum hs-CRP levels and microalbuminuria during atorvastatin therapy might mean that the pleiotropic effects of statins demonstrated in animal models might be subdivided in short-term and long-term effects, depending on the type, the dose and the timing of therapy.

Chapter 4 - *Objective*: The objective of the present investigation was to find out whether the addition of fenofibrate to statin monotherapy produced any synergistic or additive beneficial effects in reducing risk factors, especially plasma fibrinogen, in patients of Acute Coronary Syndromes (ACS) requiring Percutaneous Coronary Interventions (PCI).

Methods: This was a randomized, non-blind, prospective study with parallel group design, conducted in 102 patients who had angiographically documented Coronary Artery Disease (CAD). All had undergone angioplasty. The patients were randomized to atorvastatin (20mg/day, n=25), simvastatin (40mg/day, n=27), atorvastatin(10mg/day)-fenofibrate (200mg/day) combination (n=25) and simvastatin (20mg/day)-fenofibrate(200mg/day) combination (n=25). The serum lipid profile and plasma fibrinogen were recorded before initiation of therapy and after 3 months of the respective treatments.

Results: All the patients already had desirable lipid levels as per the NCEP ATP III guidelines. The addition of fenofibrate to statin monotherapy produced additional benefits on reduction in triglyceride (TG) and very low density lipoprotein (VLDL) levels, and an increase in high density lipoprotein (HDL) levels. All the treatment groups showed a significant decrease in the plasma fibrinogen levels. This did not correlate with any of the study parameters like age, body weight, hemodynamic characteristics and lipoprotein levels. Statin monotherapy produced a significant decrease in the fibrinogen levels and the addition of fenofibrate further enhanced the reduction.

Conclusions: Addition of fenofibrate to statins seems to be beneficial in patients with ACS. Statins, contrary to various reports, were found to decrease plasma fibrinogen significantly.Furhter, in combination with fenofibrate there was enhanced reduction of the novel risk factor, fibrinogen.

Chapter 5 – The beneficial effect of statins on the reduction of cardiovascular events can be only partly attributed to their cholesterol lowering effect. The antiproliferative, anti-inflammatory, and immunomodulatory properties of statins appear to be largely unrelated to lipid-lowering but may be explained by affecting post-translational modification or isoprenylation essential for membrane localization and biologic activity of several proteins including adapter proteins and enzymes involved in signal transduction pathways. The present data reinforce the hypothesis that statins may represent innovative pharmacological tools not only for the prevention of cardiovascular related disease in normolipidemic patients but also for diseases where a reduced isoprenylation of regulatory proteins reveals benefit effects.

Chapter 6 - It has been repeatedly shown that statins (3-hydroxy-3-methylglutaryl coenzyme A reductase inhibitors) are very effective in primary and secondary prevention of ischemic heart disease. In the settings of acute coronary syndrome (ACS), different pathological pathways are triggered that are known to be inhibited by statins, including endothelial dysfunction and activation of inflammation and coagulation; the idea to use these drugs also under conditions of ACS seems to be, therefore, fully justified. Recently, several prospective controlled clinical trials have been presented, showing safety and in some points also efficacy of statins, when administered early after ACS. An increasing number of publications demonstrates, however, that statins may express a positive effect not only in the early secondary prevention but also directly in the therapy of ACS, i.e. when statin treatment is started as a first-line care in clinically unstable patients. This therapeutic option is supported by (i) experimental studies, showing a protective effect of statins under the condition of acute ischemia, (ii) analysis of different registers and trials, demonstrating better prognosis of statin-treated patients, and (iii) small clinical trials, describing a lower peri-procedural infarction rate during coronary intervention or lower level of C-reactive protein

and other inflammatory markers as a result of statin therapy. Nevertheless, confirmation of this hypothesis by large prospective controlled clinical trials will be necessary before introduction of statins as the first line therapy in unstable patients with ACS, even without knowledge of the blood cholesterol level.

Chapter 7 - Lowering high levels of low-density lipoprotein cholesterol (LDL-C) is the primary aim in the prevention of cardiac events. However, low levels of high-density lipoprotein cholesterol (HDL-C) are also associated with an increased risk of ischemic heart disease.

Lipid-lowering drugs are known to decrease LDL-C and to increase HDL-C slightly. However, not all patients benefit from this effect. Some patients have lower HDL-C during statin treatment than before the treatment.

These patients were first described in a case report in 2002 as 'bad HDL-C responders to statins'. In the case of one man, HDL-C and the ratios of total cholesterol (TC) to HDL-C and LDL-C to HDL-C worsened dramatically during pravastatin treatment. After 3 years, pravastatin was replaced by fenofibrate. The result was spectacular. The HDL-C increased to at least twice the level obtained during pravastatin.

Bad HDL-C responders are characterized by HDL-C levels which decrease below 40 mg/dl during the treatment, despite higher HDL-C levels before the treatment.

The existence of bad HDL-C responders to statins was confirmed by a prospective survey of 2,259 patients treated with a statin or a fibrate for hyperlipidaemia. The proportion of bad HDL-C responders is higher for statins (6%) than for fibrates (4%).

In a review of the guidelines, almost all selected guidelines consider low HDL-C as a marker of increased risk for coronary heart disease. However, only few guidelines use the level of HDL-C as a threshold or target level for the treatment of dyslipidemia. The guidelines provide only little information on the management of patients with treatment-induced low HDL-C. Instead of using TC or LDL-C we consider the use of the ratios of TC to HDL-C or LDL-C to HDL-C as a threshold as well as a target for treatment.

Treatment with fibrates was studied in 14 bad HDL-C responders to statins. Far better levels for HDL-C, TC to HDL-C and LDL-C to HDL-C were obtained with fibrates compared to statins. For bad HDL-C responders to statins with low or normal LDL-C, treatment with fibrates instead of statins should be considered. For those with high LDL-C, fibrates should be added to statins.

Treatment for bad HDL-C responders should be studied in randomized controlled trials. Such a trial with simvastatin and fenofibrate has been initiated to corroborate the findings.

Chapter 8 - The 3-hydroxy-3-methylglutaryl-coenzyme-A (HMG-CoA)-reductase inhibitors (statins) are the most commonly prescribed agents for the treatment of hypercholesterolemia, due to their efficacy in lowering LDL-cholesterol and ability to reduce clinical outcome in both primary and secondary prevention of coronary artery disease. In addition to their serum lipid-lowering action, statins display non lipid-lowering pharmacological activities known as pleotropic actions. The pleiotropic effects include significant anti-inflammatory and immunomodulatory actions and many essential cellular functions including cell proliferation, differentiation, and survival and participate in the regulation of cell shape and motility.

This chapter is about the intracellular pathways involved in the pathogenesis of systemic lupus erithematosus (SLE) and the possible effects of the statins in those pathways which could be modulating their therapeutic effects, including their beneficial effects on primary and secondary prevention of cardiovascular diseases, anti-inflammation, and immunomululation.

CONTENTS

In: Statins: Indications and Uses, Safety and Modes of Action ISBN 978-1-60692-103-6
Editor: Gunnar N. Holmquist

Chapter 1

STATIN THERAPY FOR CORONARY ARTERY DISEASE BEYOND LIPID LOWERING EFFECT

Teruo Inoue and Koichi Node
Department of Cardiovascular and Renal Medicine, Saga
University Faculty of Medicine, Saga, Japan

ABSTRACT

We propose a new concept, 'Vascular Failure,' to detect early stage atherosclerosis, which is characterized as integration of endothelial dysfunction, smooth muscle cell dysfunction and metabolic abnormality of the vessel wall. Also, 'Vascular Failure' occurs not only in atherosclerosis, but also in vasculitis as well as systemic inflammatory disorders, which is of great interest. Statins targeting 'Vascular Failure' should be appled in the earlier stage, even when anatomical vascular abnormalities are not present.

INTRODUCTION

3-hydroxy-3-methylglutaryl coenzyme A (HMG-CoA) reductase inhibitors, or statins, are es lished as the principal and the effective class of drug to reduce serum cholesterol levels. Although several different statins are available for treatment of hypercholesterolemia and their pharmacokinetic profiles are different, all statins have one characteristic in common. Statins inhibits the conversion of HMG-CoA to mevalonic acid with consecutive attenuation of the biosynthesis of cholesterol, reducing cellular cholesterol content in hepatocytes. Hepatocytes respond to sterol depletion by activating nuclear sterol regulatory element-binding protein-2, which upregulates the transcription of key genes implicated in cholesterol metabolism including HMG-CoA reductase and the low density lipoprotein (LDL) receptor. Thus, the cholesterol-lowering effect of statins is principally mediated by the up-regulation of

LDL-receptor activity, which leads to enhanced hepatic uptake of atherogenic apo B-containing lipoproteins, such as very low density lipoprotein (VLDL), VLDL remnant, intermediate density lipoprotein (IDL), and LDL.

There are increasing numbers of evidences that statins reduce cardiovascular events such as coronary artery disease in hypercholesterolemic patients in both primary and secondary prevention. The striking benefit achieved with statin treatments in patients with a wide range of cholesterol levels cannot be merely attributed to their cholesterol lowering effect. Recent substantial data has accumulated showing that statins exert various effects on multiple targets, namely pleiotropic effects, especially targeting blood vessels, which are considered to be derived from suppression of the small GTP-binding protein Rho and Rho kinase signaling, independently of cholesterol lowering properties. These effects include the improvement of vascular endothelial function, inhibiting vascular smooth muscle cell proliferation and migration, anti-inflammatory actions, anti-oxidative effects or stabilizing vulnerable plaques.

Atheroscrelosis is a progressive disease characterized as a response of the vessel wall to chronic, multifactorial injury and leads to the formation of atheromatous or fibrous plaques. These plaques are regions of thickened intima and are composed of various mixtures of fibrous tissues, cells, and lipid. Vessel wall injury promote endothelial dysfunction and increased adhesion of leukocytes and platelets to endothelium, leading to release of various inflammatory mediators that potentiate vascular smooth muscle proliferation, to accumulation of peroxidized lipid and thereby to plaque formation. In these processes, endothelial dysfunction is thought to be an initial stage of atherosclerosis. In addition to endothelial dysfunction, smooth muscle cell dysfunction, metabolic abnormality of the vessel wall including inflammation, oxidative stress, breakdown of neurohormonal balance occur in early stage of atherosclerosis process. Patients who have only risk factors as dyslipidemia as well as hypertension, diabetes mellitus, smoking or others are known to have endothelial dyfunction, smooth muscle dyfunction and abnormalities of the vessel wall metabolism. In light of early intervention in atherogenic risk factors to prevent disease progression, however, we believe these vascular functions should be integrated. Now, we propose a new concept 'Vascular Failure' to detect early stage atherosclerosis, which is characterized as integration of endothelial dysfunction, smooth muscle cell dysfunction and metabolic abnormality of the vessel wall. Also, 'Vascular Failure' not only in atherosclerosis but also in vasculitis or systemic inflammatory disorders is of great interest. We should try to apply the statins targeting 'Vascular Failure' in earlier stage even when anatomical vascular abnormalities are not present.

PREVENTION OF CORONARY EVENTS BY STATINS IN CLINICAL TRIALS

European WOSCOPS (West of Scotland Coronary Prevention Study) [1] and US AFCAPS/TexCAPS (Air Force/Texas Coronary Atherosclerosis Prevention Study) [2] are representative large clinical trials for the primary prevention of coronary events by statins. WOSCOPS is a randomized placebo-controlled trial in a double blind fashion that evaluated the effects of 40 mg/day of pravastatin on preventing the onset of coronary events in 6595

hyperlipidemic male subjects aged 45 to 65 years by the 4.9 year's observation. Although there were no significant differences in non-cardiac death, the onset of non-fatal myocardial infarction or death resulted from coronary artery disease was seen in 5.9 % in the pravastatin group that was significantly lower than 7.9 % in the placebo group, and the results showed 31 % (95 % CI; 17-45 %) of the relative risk reduction by pravastatin. In AFCAPS/TexCAPS [2], 20-40 mg/day of lovastatin or placebo was prescribed in a double blind fashion in total of 6605 subjects (5608 male aged 45-73 years and 997 menopaused females aged 55 to 73 years) including normolipidemic subjects. The results of 5.2 year's observation showed that the major coronary events appeared in 6.8 person/1000 person every year in lovastatin group and 10.9 person/1000 person every year in placebo group, indicating reduction of the coronary events by lovasatin with a 0.63 (95 % CI; 0.50-0.79) of the relative risk.

The clinical trials for secondary prevention of coronary artery disease includes 4S (Scandinavian Simvastatin Survival Study) [3], LIPID (Long-term Intervention with Pravastatin in Ischemic Disease Study) [4] and CARE (Cholesterol and Recurrent Events) [5]. The 4S evaluated the effects of simvastatin administration for 5.4 years in 4444 hypercholesterolemic subjects. As a result, in the simvastatin group, 34 %, 42 % and 39 % reductions of recurrent coronary artery disease, coronary death and total death, respectively, were evident in association with lipid lowering effects, compared with the control group. In the LIPID trial that evaluated the effects of pravastatin in 9014 subjects including nomolipidemic and mild to moderate hyperlipidemic subjects, recurrent coronary artery disease, coronary death and total death were also reduced. The CARE observed the effects of pravastatin in 4159 post myocardial infarction subjects with normal total cholesterol level and with normal to mild elevation of LDL cholesterol level. The 5 years' observation showed that pravastatin could reduce occurrence of non-fatal myocardial infarction or fatal coronary artery disease by 24 % and 27 %, respectively.

The AVERT (Atorvastatin Versus Revascularization Treatment) [6] is a moderate scale but very unique trial, in which aggressive lipid lowering therapy using 80 mg/day of atorvastatin was compared with percutaneous coronary intervention (PCI) with conventional medical treatments in 341 stable coronary artery disease subjects. The results of 18 months follow-up showed 36 % reduction of coronary events in atorvastatin group, compared with PCI group. In the MIRACL (Myocardial Ischemia Reduction with Aggressive Cholesterol Lowering) trial [7], in 3086 acute coronary syndrome (ACS) patients the initiation of aggressive lipid lowering by 80 mg/day of atorvastatin within 24-96 hrs from the onset reduced the ischemic events requiring readmission within 16 weeks from the onset. The LIPS (Lescol Intervention Prevention Study) [8] is a trial that compared the early initiation of 80 mg/day of fluvastatin with that of placebo in 1677 patients getting initial success of PCI. The results of 3 years' observation showed that 22 % reduction of the relative risk for major cardiac events in the fluvastatin group, compared with placebo group.

STATINS' LIPID-INDEPENDENT EFFECTS FOR VASCULAR FAILURE

Statins' Actions for Vascular Endothelial Dysfunction

Endothelium is a flat monolayer of cells that cover vascular lumina throughout the body. Endothelial cells not merely are constitutes of the vessel wall, but play various biological roles, such as maintaining vascular tone and structure, regulating intravascular hemostasis and permeability, protecting from oxidative stress, and inhibiting cell adhesion and migration, i.e., anti-inflammatory properties [9]. Recent progress in vascular biology has revealed that the endothelium release a large number of vasactive substances. These substances are divided into two classes: endothelium-derived relaxing factors (EDRFs), and endothelium-derived contracting factors (EDCFs). It has been shown that EDRFs such as nitric oxide (NO), endothelium-derived hyperpolarizing factors (EDHF), or prostacyclin (PGI2) protect vasculature from atherogenic insult, whereas EDCFs such as endothelin-1 (ET-1) or thromboxane A2 (TXA2) have opposite effects and participate in the progression of cardiovascular diseases. Endothelial dysfunction is characterized by a reduction of the bioavailability of EDRFs, in particular, NO, whereas EDCFs increase [10]. This imbalance leads to an impairment of endothelium-dependent vasodilation, which represents the functional characteristics of endothelial dysfunction. On the other hand, endothelial dysfunction, aside from denoting impaired endothelium-dependent vasodilation, also comprises a specific state of "endothelial activation", which is characterized by a proinflammatory, proliferative, and procoagulatory milieu that favors all stages of atherogenesis [11]. Given this relationship between endothelial dysfunction and atherosclerosis, it is likely that the status of endothelial function may reflect the propensity of an individual to develop atherosclerotic disease, and thereby, the presence of endothelial dysfunction may serve as an indicator to detect the initial step of 'Vascular Failure'.

One of the best documented effects of statins for coronary vascular failure is improvement in parameters associated with endothelial dysfunction. Hypercholesterolemia reduces NO production and enhances its degradation in vascular endothelial cells. Statins' lipid-lowering effects improve these endothelial dysfunctions, increasing endothelial nitric oxide synthase (eNOS) gene expression via the reduction of degradation of mRNA and decreasing ET-1 production [12]. However, lipid-independent statins' effects on endothelial function may be more important, and such effects have been experimentally evident in the coronary artery. It has been recently demonstrated that simvastatin preserves coronary endothelial function in experimental porcine hypercholesterolemia in the absence of any lipid lowering effects [13], which translates into preservation of myocardial perfusion response and coronary microvascular integrity during episodes of increased cardiac demand [14]. In accordance with these results, pravastatin was shown to improve coronary endothelial function in cynomolgus monkeys, which were pretreated with an atherogenic diet for 2 years, independent of serum lipoprotein concentrations [15]. Since endothelial dysfunction is characterized by an imbalance between vasodilating and vasoconstricting substances, with an impairment of EDRFs and a predominance of EDCFs, statin-induced improvement in

endothelial function is likely achieved by both enhancement of vasodilator and attenuation of vasoconstrictor activity in the vascular wall.

Table 1. Clinical Assessment of Vascular Endothelial Function

Endothelium-dependent vasodilatory response-
Drug response: acetylcholine (ACh)
Shear stress: reactive hyperemia
1 Coronary artery: coronary angiography
Conduit vessel: change in vessel diameter after ACh provocation assessed by quantitative coronary angiography
Resistance vessel: change in coronary flow assessed by Doppler coronary flow velocimetry
2 Peripheral artery: coronary angiography
Conduit vessel: change in vessel diameter of brachial artery after reactive hyperemia assessed by high resolution ultrasonography
-------flow-mediated vasodilation (FMD)
Resistance vessel: change in blood flow of brachial artery after ACh or reactive hyperemia assessed by strain gouge plethysmography

Clinical Assessment of Vascular Endothelial Function and Statins' Effects

Vascular endothelial function can be clinically assessed using physiological methods in both coronary as well as peripheral arteries (Table 1). Since the first description of endothelial dysfunction in atherosclerotic epicardial coronary arteries in 1986 by Ludmer and colleagues [16], invasive assessment of coronary endothelial function by quantitative coronary angiography along with graded intracoronary infusions of endothelium-dependent vasodilator such as acetylcholine (ACh), may be considered the "golden standard" for endothelial function testing. ACh has both actions of vasodilatation by promoting endothelial NO release and vasoconstriction by direct action to vascular smooth muscles. ACh dilates normal blood vessels that have intact endothelium, but paradoxically constricts the vessels if their endothelium is damaged [16-20]. Thus, the observation of ACh-induced vasomotion is very sound methods for evaluating vascular endothelial function. Coronary artery vasospasm is considered to be an ultimate feature of endothelial dysfunction [21] as well as to be caused by hyperconstriction of vascular smooth muscle [22]. Nowadays, intracoronary injection of ACh is also widely applied to diagnose vasospastic angina. On the other hand, coronary Doppler flow measurements with intracoronary injection of ACh provides us an information regarding endothelial function of coronary resistance vessel level [23,24].

During the last decade, less-invasive or non-invasive techniques such as the forearm blood flow measurement (FBF) by strain gauge plethysmography using the venous occlusion technique or flow-mediated vasodilation (FMD) by high resolution ultrasonography has developed to assess endothelium-dependent vascular function of the forearm arteries, namely peripheral vascular endothelial function. The former can detect mainly endothelial function of resistance vessel levels, while the latter mainly detects that of conduit artery levels [25]. Using strain gauge plethysmography, endothelial function can be evaluated by an ACh-induced blood flow increase or post-ischemic reactive hyperemia [26]. In this case, however, ACh must be administrated by direct intra-arterial infusion, which is somewhat invasive. On the other hand, post-ischemic reactive hyperemia is also mediated by endothelial NO [27]. A 5 minutes occlusion of flow to the upper extremity produced by inflation of a blood pressure cuff, followed by release of the occlusion, results in an immediate 5-10-fold increase in blood flow, namely reactive hyperemia. However, the technique using venous occlusion plethysmography with reactive hyperemia is a little complex. In contract, since reactive hyperemia increases the vessel wall shear stress in the proximal artery with subsequent blood flow-dependent vasodilation, FMD of the brachial artery is nowadays the most frequently used as a non-invasive surrogate of endothelial function. The non-invasive nature of this technique allows repeated measurements over time to study the effectiveness of various interventions that may affect vascular health, although there are several limitations such as reproductibility or confounding factors in this method. Uehara et al. [28] observed brachial artery FMD by non-invasive assessment was compared with coronary endothelial function testing by assessing the conduit vessel vasomotor response to ACh as a part of coronary angiography in the same patients and demonstrated that both were correlated. This result suggests that non-invasively measured peripheral arterial endothelial function can be a simple surrogate for coronary endothelial function.

Using above-mentioned techniques, statins' effects for vascular endothelial function of the coronary as well as peripheral arteries were extensively assessed in various clinical setting. We have investigated endothelial function of the brachial artery in patients with hypercholesterolemia by the assessment of reactive hyperemic blood flow increase using the strain gauge plethysmography and evaluated the effects of statin treatments on the endothelial function [29]. As a result, administration of 20 mg/day fluvastatin but not 10 mg/day pravastatin for 16 weeks improved the endothelial function (Figure 1). Jarvisalo et al. [30] observed the statin's effects on peripheral endothelial function by a study comparing FMD in a group of 23 men with coronary artery disease without lipid-lowering medication with that in 22 age- and blood pressure-matched coronary artery disease patients with similar lipid level but ongoing statin therapy. In this study, FMD of the brachial artery was significantly higher in patients receiving statins than in those without any treatment. Moreover, multivariate analysis revealed the statin use as the only significant predictor of the FMD. The statins' effects was also evaluated on the endothelial function of coronary microvasculatures. Egashira et al. [31] demonstrated using coronary Doppler flow measurements with intracoronary ACh injection an evidence that exercise-induced myocardial ischemia even in patients without hemodynamically relevant coronary artery disease is associated with impaired endothelium-dependent vasodilation of coronary resistance vessels, namely endothelial dysfunction of coronary microvessels and that pravastatin administration