

## **Chapter 13. Anti-atherosclerotic Drugs**

**Jean-Claude Tardif, M.D.** <sup>1</sup>

**Therese M. Heinonen, D.V.M.** <sup>2</sup>

**Marie-Claude Guertin, Ph.D.** <sup>2</sup>

<sup>1</sup> Research Center  
Montreal Heart Institute  
5000 Belanger Street  
Montreal, Quebec H1T 1C8  
CANADA  
Tel 514-376-3330 x 3612  
Fax: 514-593-2500  
Email [jean-claude.tardif@icm-mhi.org](mailto:jean-claude.tardif@icm-mhi.org)

<sup>2</sup> Research Center  
Montreal Heart Institute  
5000 Belanger Street  
Montreal, Quebec H1T 1C8  
CANADA

## **I. INTRODUCTORY REMARKS**

At this moment, atherosclerotic disease places a great burden on society. Not only is atherosclerosis currently one of the leading causes of death around the world, but it also has a major impact on an individual's quality of life as a result of chronic pain, activity restriction, unemployment and disability. In 2002 in the United States alone, it was estimated that over 62 million individuals had one or more type of cardiovascular disease and that the direct and indirect cost of atherosclerotic coronary artery disease (CAD) was over 110 billion dollars.

A variety of risk factors for the development of atherosclerosis have been identified. These risk factors include smoking, high blood pressure, high cholesterol, diabetes, obesity, male gender, genetic predisposition, and physical inactivity. Lifestyle modifications and pharmacological therapy, including cholesterol-lowering medication, have become the standard of care provided to patients with coronary artery disease. While these interventions have been effective, they have failed to keep pace with the expanding "at risk" population. In particular, the increased proportion of elderly individuals and the rise in prevalence of risk factors such as metabolic syndrome, diabetes, and obesity are cause for concern. It is important to recognize that cardiovascular disease not only affects the elderly but is also a leading cause of premature death in individuals under the age of 75. Furthermore, hospitalization rates by age groups indicate that acute myocardial infarction (MI) and ischemic heart disease become important diseases by the fourth and fifth decades of life. As a result of this trend, there has been an increase in the number of deaths due to cardiovascular disease worldwide with the expectation that this trend will continue for the next fifteen years.

A complex series of events leads to the formation of atherosclerotic plaques in human arteries. Atherosclerosis is now understood to be a systemic chronic inflammatory disease characterized by the excess accumulation of lipid-laden macrophages (foam cells) within the arterial wall. The steps involved in the development of atherosclerotic lesions include endothelial expression of adhesion molecules, release of cytokines and chemokines, involvement of reactive oxygen species, macrophage accumulation in the arterial wall, and incorporation of oxidized LDL. The resulting atheroma is comprised of cholesterol, inflammatory cells, and matrix. Atherosclerotic lesions are heterogeneous and the clinical manifestation of atherosclerosis is dependent on the lesion composition and affected vascular bed, as well as other factors. When the atherosclerotic plaque ruptures the clinical presentations include MI, unstable angina, stroke, transient ischemic attack (TIA), and cardiac death.

The complex pathophysiology of atherosclerotic disease highlights the fact that many processes contribute to lesion development, and suggests that cholesterol modification is not the only mechanism by which the condition may be positively affected. It is well accepted that high serum levels of cholesterol, and particularly low-density lipoprotein cholesterol (LDL-C), increase the risk for CAD. The use of HMG-CoA reductase inhibitors (statins) became widespread as data from numerous clinical trials supported their safe and efficacious use in a broad population base. With a reduction in combined cardiovascular morbidity and mortality of 30 – 35% resulting from their use, statin drugs have had a substantial positive benefit on health care. However, the majority of clinical events are not prevented by statins and the persistently high level of cardiovascular disease is far from satisfying.

Attention has therefore been focused on non-lipid risk factors of CAD including some of the key systems involved in the formation and progression of atherosclerotic plaque. As a result of the ubiquitous and chronic nature of atherosclerosis, as well as the persistently high incidence of cardiovascular morbidity and mortality, research efforts in this area remain vital. In recent years these efforts have changed our understanding of atherosclerosis significantly. A process which was once thought of as a static accumulation of fat in the arteries is now realized to be a very dynamic process in which numerous environmental, genetic, and individual factors contribute. Research which at one time focused almost exclusively on cholesterol metabolism has now branched out to provide further insight into the role of

other processes such as inflammatory contributors. A number of chemokines and cytokines involved in the inflammatory process have now been identified, and potential markers of disease progression and regression are under evaluation. Additionally, our ability to understand the process of atherosclerosis and the causative factors has gained sophistication with the experimental and clinical employment of various high-technology tools. These tools include the use of genomics, proteomics, the identification of soluble plasma markers, as well as imaging technologies such as quantitative coronary angiography, intravascular coronary and peripheral ultrasound, B-mode carotid ultrasound, positron emission tomography, computed tomography, and magnetic resonance imaging.

The development of atherosclerotic plaque changes both vascular structure and function. The endothelial lining of the blood vessels is important in the regulation of vascular tone and the accommodation toward greater blood supply in circumstances of greater demand. Modification of endothelial function is one of the earliest clinically demonstrated changes within the vasculature during the course of atherosclerotic progression. Subsequent lesion progression with the accumulation of cholesterol and matrix leads to clinically detectable changes in vascular anatomy and further functional changes. The vascular wall becomes thickened with accumulating debris, and the lumen eventually narrows despite remodelling changes to accommodate expanding plaque volume. The convergence of these factors, along with the inflammatory components, results in supply/demand mismatch as well as plaque instability and rupture – all accumulating in clinical disease.

While progress has been made in the field of atherosclerosis, there remain many challenges in developing an agent which will work by a non-lipid anti-atherosclerotic mechanism. There are currently no regulatory guidelines for the development of such drugs and no therapies approved for the treatment of atherosclerotic coronary disease that work by non-lipid mechanisms. Additionally, despite the recent generation of data identifying potential biomarkers such as C-reactive protein, clear and independent links between such markers and coronary artery disease still need to be strengthened in order to maximize their utility in drug development programs. Other developmental hurdles include the chronic nature of CAD, trial duration, ethical considerations, concomitant medications, and plaque heterogeneity. Since atherosclerotic plaques develop slowly over decades, in most instances, clinically meaningful changes using even the most sensitive methods have required relatively long-term therapy of 12-18 months. Due to the effectiveness of current therapy, including statins, at decreasing cardiovascular risk in a wide patient population, true placebo-controlled trials in patients with cardiovascular risk have become unethical. Also due to the demonstrated effectiveness of current therapy, any potential new therapy is expected to be evaluated as an adjunct to standard care. Standard care may involve several medications for lipid-lowering, as well as hypertension and glucose control. Care must therefore be given to potential interactions between new agents and these concomitant medications. While the development of new anti-atherosclerotic agents that work by non-lipid mechanisms will continue to present a developmental challenge, the new tools available, and the diligence of those undertaking this challenge is expected to make significant inroads in our ability to successfully identify the next wave of effective therapy.

## **II. PHASE II STUDIES FOR REGISTRATION OF NEW ANTI-ATHEROSCLEROTIC DRUGS**

### **II.1. Outline of a typical development plan**

Although atherosclerosis is a systemic condition, which leads to stroke and other serious peripheral circulatory disorders, the development of anti-atherosclerotic agents typically focuses on coronary disease indications. Standard exploratory studies conducted during this stage of development include those designed to assess the maximally tolerated dosages over a short to moderate treatment durations, pharmacokinetics and drug interactions. The length of treatment should be sufficient to obtain the safety and efficacy information needed to determine the feasibility of the long-term evaluations needed in Phase III. Additionally, since these agents work by a non-lipid mechanism, it is important for the early clinical

trials to explore the utility of other potential biomarkers. Two types of biomarkers may be included in these trials. The first type of biomarker is one which will provide pharmacodynamic information on the biochemical system targeted. For example, an agent which activates enzyme X should be evaluated at this stage for the degree of activation conferred across an acceptable dose range, if possible. The second type of biomarker is one which will provide a reasonable indication of clinical efficacy (ie. vascular improvement). The selection of potential biomarkers for evaluation should be based on a good understanding of the agent's mechanism of action, as well as the pathophysiology of atherosclerotic disease. The appropriate set of biomarkers for each evaluated agent may be quite different depending on the mechanism and anticipated response. Therefore the development plan for each agent may also be unique. Additionally, since lipid-lowering therapy has clearly demonstrated effectiveness and safety, it is important to assess at an early stage the influence of the study drug on current therapy through the use of well designed drug interaction studies. The results of these carefully designed early studies are critical for appropriate design of pivotal studies, which may be significant in terms of size, duration, and resource needs.

There are several different approaches employed in Phase II to obtain clinical efficacy. One approach is to evaluate a population with a clinical indication for cardiac catheterization in order to enable the use of sensitive but invasive imaging tools such as intravascular ultrasound (IVUS) to detect drug-induced changes. Such an approach has the advantages of directly assessing the coronary circulation, minimizing sample size and detecting small but potentially important changes in plaque burden. However, the obvious disadvantages are the invasiveness of the assessment and the limitations regarding patient selection. Another approach is to conduct initial evaluations in individuals with stable moderate atherosclerotic disease using non-invasive tools such as B-mode ultrasound in peripheral vessels (carotid and brachial). Such an approach has the advantage of non-invasive assessments in a widely available patient population, but relies on correlations between peripheral changes and coronary disease. Additionally, this approach may require a larger sample size to detect drug effect changes. Often, both of these approaches are included in the Phase II development plan.

The typical pivotal trial will utilize a double-blind, multi-center, placebo-controlled, randomized, adjunctive therapy, parallel group design. At least 2 or 3 dose levels should be explored, preferably within the same trial. Early proof-of-concept monotherapy studies or single-center studies may be included as part of the phase II development program. Patients included in short-term studies should be allowed to enter long-term open-label follow-up for collection of additional safety information.

## **II.2. Short-term studies**

### **II.2.i. Adjunctive therapy trial in patients with clinical indication for cardiac catheterization - IVUS**

#### **II.2.i.A. Objectives**

To evaluate short-term efficacy and tolerability during adjunctive therapy use

#### **II.2.i.B. Primary endpoints**

- a. The percent change in plaque volume (follow-up – baseline)/baseline x 100) measured by 3D IVUS.

#### **II.2.i.C. Secondary endpoints**

- a. Absolute change in plaque volume on three dimensional (3-D) IVUS
- b. Change in percent plaque volume on 3-D IVUS
- c. Changes in plaque volume in anatomically comparable 5-mm segments centered on the sites with lowest and highest plaque burden at baseline by 3-D IVUS
- d. Changes in plaque characterization indices assessed by IVUS

- e. Coronary score assessed by quantitative coronary angiography (QCA) defined as the per-patient mean of the minimal lumen diameter for all lesions measured.
- f. Incidence of adverse events

#### **II.2.i.D. Exploratory endpoints**

Changes from baseline in markers of biochemical target activation and markers of atherosclerosis pathophysiology (ie. inflammatory markers).

#### **II.2.i.E. Study design**

A multicenter, randomized, placebo-controlled, parallel-group design is typically used. Inclusion criteria usually require patients to be at least 18 years of age and with a need for diagnostic coronary angiography for clinical indication (with or without percutaneous coronary intervention). Patients are usually required to have stable background treatment following standard care practice. Patients are screened and the initial IVUS examination is performed in a target coronary artery which must not have undergone previous percutaneous coronary intervention (PCI) nor be a candidate for intervention at the time of the baseline catheterization. The baseline phase should be of sufficient duration to allow for scheduling of catheterization procedures within an acceptable time and stabilization of concomitant medication (4 weeks). Patients are then randomized to treatment groups including placebo and typically 2-3 doses of active study medication. A treatment phase of variable length (1-12 months) is initiated based on the characteristics of the drug and the mechanism of action. During the active treatment period, background medication must remain as stable as possible. The follow-up IVUS procedures are performed from 6 weeks to 12 months (see below for longer term IVUS studies) following the baseline IVUS.

#### **II.2.i.F. Planned sample**

Assuming that the standard deviation of the % change in plaque volume is around 6%, a sample size of approximately 65 patients per treatment group will provide 80% power to detect a 3% difference in plaque volume between the placebo group and one of the active treatment groups using a two-sided significance level of 0.05. Approximately 50 patients per treatment group are needed to detect a difference between the placebo group and two combined active groups in a 3-arm trial given the same assumptions. The final sample size must also account for loss-to-follow up of approximately 20 – 25%, given the cardiac catheterization procedures required.

#### **II.2.i.G. Study population**

Adults who require diagnostic coronary angiography for clinical indication, and stabilized to other background medications according to standard of care practices.

#### **II.2.i.H. Specific inclusion criteria**

- a. Male or female aged  $\geq 18$  years
- b. Female patients who are not of childbearing potential (at least two years postmenopausal, surgically sterile, or practicing adequate contraception)
- c. Scheduled for clinically indicated coronary angiography (with or without PCI)
- d. Presence of at least one luminal diameter stenosis of 20% or more in one coronary artery by visual (angiographic) estimation
- e. Presence of a non-PCI target coronary artery in which IVUS examination can be performed (target vessel)
- f. The target vessel must not have undergone previous PCI nor be a candidate for intervention at the time of the baseline catheterization

#### **II.2.i.I. Specific exclusion criteria**

- a. Previous or planned coronary artery bypass surgery

- b. Any medical or psychiatric condition which in the opinion of the investigator makes the patient an unsuitable candidate for the study
- c. History of alcohol or drug abuse within the past year

#### **II.2.i.J. Tools for assessing primary endpoints**

Assessment of the main outcome measures of atherosclerosis regression is performed by 3-D reconstruction of IVUS images. All the IVUS images should be interpreted by experienced technicians supervised by a cardiologist blinded to treatment assignment. The baseline and end-of-treatment studies should be viewed together. The use of reproducible IVUS landmarks (i.e. aorto-ostial junction, branches) and a known pullback speed (0.5 mm/sec) facilitate comparison of the same 30-mm segment on both studies and permit volumetric (3-D) analysis. Frame-by-frame review of the images is also systematically used to confirm matching of segments. The images are digitized and quantitative analysis performed. The lumen and external elastic membrane (EEM) borders can be traced manually or using an edge detection algorithm if all tracings are visually verified.

Plaque, lumen and total vessel volumes are computed by multiplying the corresponding areas of each of the cross-section by the distance between neighbouring slices and by then adding all the products. Cross-sections are analyzed in the 30-mm segment of interest at both baseline and follow-up. Plaque, lumen and total vessel volumes are first computed for the entire length (30 mm) of the analyzed segment. Volumes are also calculated on 5-mm segments centered on the sites with a) smallest plaque burden at baseline and b) largest plaque burden at baseline. In addition to the absolute plaque volume, percent plaque volume is also calculated as plaque volume divided by total vessel volume times 100. Detailed analysis of plaque composition is then also performed with IVUS at baseline and follow-up using plaque characterization indexes.

For the quantitative analysis of coronary angiograms (QCA), all angiograms from a given patient should be viewed together and analyzed by experienced technicians supervised by a cardiovascular radiologist blinded to the patients' treatment assignments.

#### **II.2.i.K. Specific criteria for early withdrawal and discontinuation**

Standard criteria for early withdrawal include withdrawal of consent, or adverse events that render the continued treatment of patient(s) medically unacceptable.

#### **II.2.i.L. Data analysis method**

The data analysis methods used may be dependent on the objective of the trial. One method is to base the analysis of efficacy variables on the intention-to-treat (ITT) population (including all randomized patients who have follow-up data). However, for exploratory purposes, the per-protocol population (patients with no major protocol violations) may also be investigated. Statistical tests are generally two-sided and p values  $\leq 0.05$  are often considered statistically significant. Percent change is analyzed using analyses of variance (ANOVA) models. Analyses of covariance (ANCOVA) models adjusting for baseline values are used for absolute change. Coefficients of correlation are computed to assess the relationship between changes in inflammatory markers and primary and secondary endpoints. The safety analysis includes adverse events presented by treatment group using descriptive statistics (frequencies and counts).

### **II.2.ii. Adjunctive therapy trial in patients with stable atherosclerosis – brachial artery reactivity**

#### **II.2.ii.A. Objectives**

To evaluate short-term efficacy and tolerability during adjunctive therapy use

#### **II.2.ii.B. Primary endpoints**

- a. Change in flow mediated dilation (FMD) defined as the percent change from brachial artery diameter (mm) at rest to maximal brachial artery diameter during reactive hyperemia.

### **II.2.ii.C. Secondary endpoints**

Secondary endpoints are designed to distinguish between vasodilatory changes resulting from changes in endothelial function as opposed to smooth muscle function.

- a. Change in nitroglycerin-mediated dilation (percent change from brachial artery diameter before administration to maximal brachial artery diameter after administration of sublingual nitroglycerin, an endothelium-independent vasodilator)
- b. Change from baseline in normalized brachial artery diameter
- c. Change from baseline in blood pressure, heart rate and velocity versus time integral

### **II.2.ii.D. Exploratory endpoints**

- a. Changes from baseline in markers of biochemical target activation and markers of atherosclerosis pathophysiology (i.e. inflammatory markers).

### **II.2.ii.E. Study design**

A randomized, placebo-controlled, parallel-group design is generally used. Single-center or small multi-center studies are often preferable due to inter-operator variability. There are typically three phases, a phase for stabilization of concomitant medication and diet, a single-blind placebo-baseline phase, and a double-blind treatment phase. Inclusion criteria usually require patients to be at least 18 years of age with objective evidence of coronary artery disease, peripheral artery disease or carotid artery disease. At screening, patients are asked to refrain from significant changes in dietary, smoking, and exercise habits for the duration of the trial as these may affect their endothelial function. Following the baseline period (typically 4 weeks), patients have vascular reactivity assessed through ultrasound procedures performed on the brachial artery. Patients are then randomized to treatment groups including placebo and typically 2-3 doses of active study medication. A treatment phase of 8-12 weeks is initiated based on the characteristics of the drug and the mechanism of action. During the active treatment period, background medication must remain as stable as possible. The follow-up brachial assessments are performed at the end of the treatment period.

### **II.2.ii.F. Planned sample**

A sample size of approximately 45 patients per treatment group will provide 80% power to detect a 2.5% difference in change in FMD between the placebo group and one of the active treatment groups assuming that the standard deviation of the change is 4% and using a two-sided significance level of 0.05.

### **II.2.ii.G. Study population**

Adults with objective evidence of coronary artery disease, peripheral artery disease or carotid artery disease and stabilized to other background medications according to standard of care practices.

### **II.2.ii.H. Specific inclusion criteria**

- a. Males or females aged  $\geq 18$  years
- b. Female patients who are not of childbearing potential (at least two years postmenopausal)
- c. Other specific inclusion criteria are based on defining objective evidence of atherosclerotic disease as follows:
  - Stable angina pectoris for which frequency, severity, duration, time of appearance, and precipitating events have not changed for 60 days prior to screening;
  - Myocardial ischemia as evidenced by any of the following
    - ✓ Stress ECG showing ischemic ST-segment response
    - ✓ Stress echocardiography showing myocardial wall motion abnormality, or
    - ✓ Myocardial perfusion scan showing a myocardial perfusion defect
  - At least 50% occlusion of the lumen of one or more coronary arteries, as evidenced by coronary angiography, IVUS, or other methods
  - ECG evidence of Q-wave myocardial infarction

- Iliac, femoral or carotid artery atherosclerosis as evidenced by angiography, ultrasound duplex scan, IVUS or other methods
- Previous carotid endarterectomy, peripheral bypass surgery, or abdominal aneurysm.

#### **II.2.ii.I. Specific exclusion criteria**

- a. History of myocardial infarction or unstable angina within 4 weeks prior to screening
- b. History of PCI, coronary artery bypass, cerebrovascular accident, or diagnosis of heart failure within 3 months prior to screening
- c. Uncontrolled hypertension
- d. Uncontrolled diabetes
- e. Concomitant administration of supplemental antioxidants, L-arginine supplements, dipyridamole, pentoxifylline, angiotensin-converting enzyme (ACE) inhibitors, or ACE receptor inhibitors.
- f. Any medical or psychiatric condition which in the opinion of the investigator makes the patient an unsuitable candidate for the study
- g. History of alcohol or drug abuse within the past year

#### **II.2.ii.J. Tools for assessing primary endpoints**

High resolution B-mode ultrasound imaging is used to determine the diameter of the right brachial artery before and immediately after reactive dilation induced by ischemia which is produced by inflating a blood pressure cuff to 200 mmHg or to 50 mmHg greater than the systolic blood pressure (whichever is higher) for 5 minutes. Arterial diameter at baseline and at 60 seconds after cuff deflation is recorded. The relative difference is the measure of reactivity. Image analysis should be performed by a central laboratory with experienced technicians blinded to study treatment.

#### **II.2.ii.K. Specific criteria for early withdrawal and discontinuation**

The investigator will discontinue a patient before completion of the study if consent is withdrawn or if it is medically unacceptable to continue treatment due to adverse events.

#### **II.2.ii.L. Data analysis method**

The analysis of efficacy variables may be based on the intention-to-treat (ITT) population (including all randomized patients who have follow-up data). All statistical tests are two-sided and p values  $\leq 0.05$  are considered statistically significant. Analysis of variance or analysis of covariance adjusting for the baseline values could be used to compare the primary and secondary endpoints between groups. In any case, data analysis methods should be planned a priori and chosen to answer the specific objectives of the trial.

### **II.3. Long-term studies**

Since the accumulation of lipid rich plaque material develops slowly over several decades, it has generally been accepted that chronic therapy would be required in order to detect the effects of these agents on measures of plaque burden or subsequent risk of cardiovascular events. For example, while some recent IVUS trials have demonstrated efficacy of exploratory agents when administered over a period of weeks, approved lipid-lowering agents with a demonstrated ability to decrease cardiovascular morbidity and mortality have generally required treatment durations of 18 months or more in order to demonstrate plaque volume changes detectable with IVUS or carotid ultrasound. Therefore, trials may need to be designed to evaluate long-term administration in order to detect significant treatment effects. The primary objective of these long-term studies is to provide an opportunity for the drug effect to be realized and to obtain data on tolerability and safety during long-term use. The studies described above as short-term studies may also be considered long-term studies depending on the study drug mechanism of action and the anticipated time required for treatment effect. In addition to the use of QCA and IVUS in Phase II long-term studies to detect anti-atherosclerotic effects, carotid imaging is also an effective imaging tool.

### **II.3.i. Adjunctive therapy trial in patients with clinical indication for cardiac catheterization - IVUS**

#### **II.3.i.A. Objective**

To evaluate long-term efficacy and tolerability during adjunctive therapy use

Study description is similar to short-term study description, except that long-term studies using IVUS generally provide for follow-up evaluations after 12-24 months.

### **II.3.ii. Adjunctive therapy trial in patients with stable atherosclerosis – carotid intima-media thickness (IMT) evaluations**

#### **II.3.ii.A. Objective**

To evaluate long-term efficacy and tolerability during adjunctive therapy use

#### **II.3.ii.B. Primary endpoints**

- a. The change over time in mean maximum IMT across 12 pre-selected carotid arterial segments.

#### **II.3.ii.C. Secondary endpoints**

- a. The difference in the slope of left and right common carotid artery far wall mean IMT progression (up to 200 far-wall measurements over a distance of approximately 2 cm beginning 1 cm below the bifurcation and using a standard angle of imaging)
- b. The change in mean far wall IMT
- c. The change in maximum far wall IMT
- d. The rate of carotid artery progression measured as linear slope over annual ultrasound examinations
- e. The average of the maximum carotid IMT of the far wall of up to 4 arterial segments (right and left distal common and right and left carotid bulb)
- f. The rate of progression in the far wall of the left and right carotid bulb IMT
- g. The rate of progression of the mean of maximum IMTs of the left and right common carotid artery
- h. Change from baseline in mean IMT of the left and right common carotid arteries
- i. Change from baseline in the maximum IMT of the left and right common carotid arteries
- j. Incidence of adverse events.

#### **II.3.ii.D. Exploratory endpoints**

Changes from baseline in markers of biochemical target activation and markers of atherosclerosis pathophysiology (i.e. inflammatory markers)

#### **II.3.ii.E. Study design**

A multicenter, randomized, placebo-controlled, parallel-group design is generally used. Inclusion criteria usually require patients to be at least 18 years of age with evidence of carotid and coronary atherosclerosis. Patients are usually required to have stable background treatment following standard care recommendations. Patients are screened and the initial carotid ultrasound examination is performed bilaterally on the common and internal arteries. Patients are then randomized to treatment groups including placebo and typically 2-3 doses of active study medication. It is generally thought that treatment effects on the common carotid artery mean IMT could be seen after one year of treatment, but an 18-24 month treatment period would enhance the likelihood of this. Due to the non-invasive nature of carotid IMT assessment, serial examinations of the carotid wall are often undertaken.

#### **II.3.ii.F. Planned sample**

The sample size calculation is based on an average expected effect of 0.02 mm/yr change from baseline in carotid IMT measurements in at least one treatment group (for a total change of 0.04 mm in a 24-month

study). No change is expected in the placebo group. According to the literature, 0.20 mm would be a reasonable estimate of the standard deviation of the change in IMT from baseline to 24 months. Under these assumptions, a sample size of approximately 400 patients per treatment group would provide 80% power to detect a difference of 0.04 mm in change in IMT between the placebo group and one of the active treatment groups using a two-sided significance level of 0.05.

#### **II.3.ii.G. Study population**

Patients with evidence of atherosclerotic disease

#### **II.3.ii.H. Specific inclusion criteria**

- a. Patients may be selected based on a pre-specified minimal baseline IMT measurement (> 0.8 mm),
- b. Patients may be selected based on other evidence of atherosclerotic cardiovascular disease
- c. Patients may be selected based on both of these criteria.

#### **II.3.ii.I. Specific exclusion criteria**

- a. History of carotid revascularization
- b. Patients in whom a screening IMT is suboptimal
- c. Any medical or psychiatric condition which in the opinion of the investigator makes the patient an unsuitable candidate for the study
- d. History of alcohol or drug abuse within the past year

#### **II.3.ii.J. Tools for assessing primary endpoints**

Images of the right and left common carotid and internal carotid arteries are captured, including images of the near and far wall, using high-resolution B-mode ultrasound. Ultrasound methodology should be specifically designed to include procedures to quality control the critical components of measurement variation including instrumentation, and ultrasound operations. Standardization of ultrasound machines at all sites is optimal, but not necessary. Image analysis is performed centrally at a center with experienced technicians.

#### **II.3.ii.K. Data analysis method**

The analysis of efficacy variables may be based on the intention-to-treat (ITT) population (including all randomized patients who have follow-up data). The continuous efficacy endpoints are analyzed using an analysis of covariance (ANCOVA) model with treatment and center as effect and the parameter's baseline as covariate. If serial examinations of the carotid wall are undertaken, repeated measures ANOVA could also be conducted to study the way IMT changes over time. Chi-square tests or Cochran-Mantel-Haenszel tests using center as a stratification factor are used for categorical endpoints. Other data analysis methods more suitable for exploratory evaluations may be employed, depending on the objectives of the trial.

### **III. PHASE III STUDIES FOR REGISTRATION OF NEW ANTI-ATHEROSCLEROTIC DRUGS**

#### **III.1. Outline of a typical development plan**

Phase III studies are conducted to establish the overall risk-benefit relationship of the drug and to provide adequate information for drug labeling. Therefore, for an atherosclerosis indication, the endpoints, patient population, and patient numbers should be consistent with these goals. It is important to recognize that no anti-atherosclerotic drugs working by a non-lipid mechanism have yet been approved. Additionally, there are no official guidelines written to direct the development of such agents. Three concerns have previously been identified for drugs working through novel, non-LDL cholesterol mechanisms. The first concern is that use of true placebo-controlled trials is not ethical in high-risk patients. Secondly, the administration of background "usual care" consisting of statins and other lipid-lowering therapy may add to the complexity of trial design and interpretation. Finally, with the need to demonstrate the effectiveness

of adjunctive therapy, sample size may become prohibitively large. The use of the imaging technologies outlined in Phase II trial descriptions may some day provide sufficient data to support the use of these endpoints as a surrogate for the reduction of cardiovascular risk. However, to date, the ability to rely solely on imaging endpoints in Phase III is yet theoretical. Therefore the development plan outlined in this document consists of a multicenter (usually multi-national), double-blind, randomized, placebo-controlled trial of two parallel groups designed to assess the combined incidence of cardiovascular morbidity and mortality.

## **III.2. Long-term adjunct therapy trial**

### **III.2.i. Adjunctive therapy trial in patients at high risk for a major cardiovascular event**

#### **III.2.i.A. Objectives**

To assess the effect of the investigational drug versus placebo on the combined incidence of cardiovascular morbidity and mortality

The definition of cardiovascular morbidity may include only “hard” endpoints such as non-fatal myocardial infarction, and stroke, or be expanded to include other “soft” endpoints, such as the need for coronary revascularization procedures, worsening angina requiring hospitalization, objective evidence of ischemia, and incidence of peripheral arterial disease.

#### **III.2.i.B. Primary endpoint**

- a. Combined incidence of cardiovascular morbidity and mortality

#### **III.2.i.C. Secondary endpoints**

- a. Incidence of all cause mortality
- b. Incidence of cardiovascular mortality
- c. Incidence of cardiovascular morbidity
- d. Combined incidence of a subset of the events within the cardiovascular morbidity and mortality definitions.
- e. Incidence of adverse events.

#### **III.2.i.D. Exploratory endpoints**

Changes from baseline in markers of biochemical target activation and markers of atherosclerosis pathophysiology (ie. inflammatory markers).

#### **III.2.i.E. Study design**

A multicenter, randomized, adjunctive therapy, double-blind, placebo-controlled parallel-group design could be utilized. Patients should receive standard care, including treatment of underlying diabetes, hypercholesterolemia and hypertension, prior to entry into this trial. There are several reasonable approaches to overall study design. One approach is to design the trial so that it will be complete with a fixed number of patients completing a minimal, fixed treatment period. Another approach is to design the trial so that it will complete when a predetermined number of patients have experienced a primary event based on expected control group rates and anticipated treatment effects.

#### **III.2.i.F. Planned sample**

The sample size will be based on the expected event rate in the selected patient population as well as the anticipated magnitude of treatment effect as demonstrated in Phase II trials. Other factors to consider in determining sample size include standard of care in country/region where trial is being conducted, planned trial duration, and time to effect based on an individual agent’s mechanism of action. A sample size of 2,000

– 6,000 moderate to high-risk patients per treatment group with minimally 18-month follow up would be a reasonable expectation.

### **III.2.i.G. Study population**

Patients at moderate to high risk of cardiovascular morbidity and mortality based on the presence of one or more risk factors. Patients should receive treatment for modifiable risk factors prior to entry into trial.

### **III.2.i.H. Specific inclusion criteria**

- a. Specific inclusion criteria are selected based on identification of patients with moderate to high risk of cardiovascular morbidity and/or mortality.
- b. Inclusion criteria may include one or more of the following major risk factors:
  - Cigarette smoking
  - Hypertension (BP > or = 140/90 mm Hg)
  - Low HDL cholesterol (< 40 mg/dL)
  - Family history of premature CHD (CHD in male first degree relative < 55 years; CHD in female first degree relative < 65 years)
  - Age ( $\geq$  55 years)
  - Diabetes
- c. Inclusion criteria may include one or more of the following life-habit risk factors
  - Obesity
  - Physical inactivity
  - Atherogenic diet
- d. Inclusion criteria may include one or more of the following emerging risk factors
  - C reactive protein
  - Lipoprotein (a)
  - Homocysteine
  - Prothrombotic and proinflammatory factors
  - Impaired fasting glucose
  - Evidence of subclinical atherosclerotic disease

### **III.2.i.I. Specific exclusion criteria**

- a. Any medical or psychiatric condition which in the opinion of the investigator makes the patient an unsuitable candidate for the study
- b. History of alcohol or drug abuse within the past year

### **III.2.i.J. Tools for assessing primary endpoints**

An independent Clinical Endpoint Committee (CEC) is established to review and classify all suspected major cardiovascular events according to pre-defined guidelines and definitions. The CEC remains blinded to treatment group assignment for the duration of the trial. The CEC is typically composed of 5 cardiologists experienced in patient care. Reviewers are assigned at random to review each case and classify the event. For the classification to be considered final, consensus between two primary reviewers is required. If consensus between two primary reviewers is not reached, the case is typically decided by simple majority of the entire committee.

### **III.2.i.K. Specific criteria for early withdrawal and discontinuation**

The investigator will discontinue a patient before completion of the study if consent is withdrawn or if it is medically unacceptable to continue treatment due to adverse events. An independent Data Safety Monitoring Board (DSMB) is typically established and responsible for assessing patient safety during the course of the trial. A trial may be discontinued if such a recommendation is made by the DSMB based on a periodic review of the generated data.

### **III.2.i.L. Data analysis method**

The analysis of efficacy variables is based on the intention-to-treat (ITT) population. Statistical tests are 2-sided at the 0.05 level of significance. Survival analysis including Kaplan-Meier curves and log-rank tests to compare survival curves across groups are used. Multivariate analysis using Cox proportional hazards models may also be performed.

## **IV. OTHER STUDIES (SPECIAL INDICATIONS)**

Other studies may be considered for inclusion in the development plan for anti-atherosclerotic agents that work by non-lipid mechanisms. Patient populations that may warrant focused evaluation in separate clinical trials include patients with traditional or emerging risk factors, or genetically defined conditions which confer high risk of CAD. Separate trials in patients with diabetes, metabolic syndrome, genetic predisposition to CAD, peripheral arterial disease, etc. may provide focused and useful information for further development. There may be also other imaging technologies that may have an important role in drug development aside from those mentioned previously. Magnetic resonance imaging, computed tomography, positron emission tomography and other technologies may all have an important role in determining the effectiveness of potential anti-atherosclerotic agents. These additional studies may have utility in early stage development to help establish proof of concept and better define the mechanism of action. Alternately further evaluations of specific populations and technologies may have greatest utility in Phase IV trials to further define established efficacy.

## **V. EXAMPLES OF LANDMARK WELL DESIGNED TRIALS**

### **Intravascular Ultrasound**

1. Tardif JC, Gregoire J, Schwartz L, Title L, Laramée L, Reeves F, Lesperance J, Bourassa MG, L'Allier PL, Glass M, Lambert J, Guertin MC; Canadian Antioxidant Restenosis Trial (CART-1) Investigators. Effects of AGI-1067 and probucol after percutaneous coronary interventions. *Circulation* 2003;107:552-558.
2. Nissen SE, Tsunoda T, Tuzcu EM, Schoenhagen P, Cooper CJ, Yasin M, Eaton GM, Lauer MA, Sheldon WS, Grines CL, Halpern S, Crowe T, Blankenship JC, Kerensky R. Effect of recombinant ApoA-I Milano on coronary atherosclerosis in patients with acute coronary syndromes: a randomized controlled trial. *JAMA* 2003;290:2292-2300.
3. Cote G, Tardif JC, Lesperance J, Lambert J, Bourassa M, Bonan R, Gosselin G, Joyal M, Tanguay JF, Nattel S, Gallo R, Crepeau J. Effects of probucol on vascular remodeling after coronary angioplasty. Multivitamins and Protocol Study Group. *Circulation*. 1999;99:30-35.
4. Tardif JC, Gregoire J, Lesperance J, Lambert J, L'Allier PL, Rodes J, Anderson T, Blue JW, Imus J, Heinonen T. Design features of the Avasimibe and Progression of coronary Lesions assessed by intravascular UltraSound (A-PLUS) clinical trial. *Am Heart J* 2002;144:589-596.

### **Brachial artery reactivity**

1. Cohen JD, Drury JH, Ostdiek J, Finn J, Babu BR, Flaker G, Belew K, Donohue T, Labovitz A. Benefits of lipid lowering on vascular reactivity in patients with coronary artery disease and average cholesterol levels: a mechanism for reducing clinical events? *Am Heart J* 2000;139:734-738.
2. Dupuis J, Tardif JC, Cernacek P, Theroux P. Cholesterol reduction rapidly improves endothelial function after acute coronary syndromes. The RECIFE (reduction of cholesterol in ischemia and function of the endothelium) trial. *Circulation* 1999;99:3227-3233.

### **Carotid intima-media thickness (IMT)**

1. De Groot, E, Jukema W, Montauban AD, et al. B-mode ultrasound assessment of pravastatin treatment effect on carotid and femoral artery walls and its correlations with coronary arteriographic

findings: a report of the regression growth evaluation statin study (REGRESS). *J Am Coll Cardiol* 1998;31:1561-1567.

2. Smilde TJ, van Wissen S, Wollersheim H, Trip MD, Kastelein JJ, Stalenhoef AF. Effect of aggressive versus conventional lipid lowering on atherosclerosis progression in familial hypercholesterolaemia (ASAP): a prospective, randomised, double-blind trial. *Lancet* 2001;357:577-581.

#### **Major cardiovascular morbidity and mortality studies**

1. Randomised trial of cholesterol lowering in 4444 patients with coronary heart disease: the Scandinavian Simvastatin Survival Study (4S) *Lancet* 1994;344:1383-1389.
2. Shepherd J, Cobbe SM, Ford I, Isles CG, Lorimer AR, MacFarlane PW, McKillop JH, Packard CJ. Prevention of coronary heart disease with pravastatin in men with hypercholesterolemia. West of Scotland Coronary Prevention Study Group. *N Engl J Med* 1995;333:1301-1307.

#### **VI. SUGGESTED READINGS**

1. Orloff DG. Use of surrogate endpoints: a practical necessity in lipid-altering and antiatherosclerosis drug development. *Am J Cardiol* 2001;87(4A):35A-41A.
2. Aurecchia S, Orloff D, Sobel S. Regulatory concerns at various phases of drug development. *Am J Cardiol* 1998;81(8A):2F-4F.
3. Black DM. Documenting regression of atherosclerosis: practical approaches in drug development. *Am J Cardiol* 2002;89(4A):1B-3B.
4. Feinstein SB, Voci P, Pizzuto F. Noninvasive surrogate markers of atherosclerosis. *Am J Cardiol* 2002;89(5A):31C-43C.
5. Naghavi M, Libby P, Falk E, et al. From vulnerable plaque to vulnerable patient: a call for new definitions and risk assessment strategies: Part I. *Circulation* 2003;108:1664-1672.
6. Naghavi M, Libby P, Falk E, et al. From vulnerable plaque to vulnerable patient: a call for new definitions and risk assessment strategies: Part II. *Circulation*. 2003;108:1772-1778.
7. Heinonen TM. Acyl coenzyme A: cholesterol acyltransferase inhibition: potential atherosclerosis therapy or springboard for other discoveries. *Expert Opin Investig Drugs* 2002;11:1519-1527.