Safety and Efficacy of the bi-Sulfydryl ACE-Inhibitor Zofenopril in the Management of Cardiovascular Disease

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Abstract: In the 1970s, pharmacological therapy interrupting the renin-angiotensin system was considered beneficial for patients with high-renin hypertension. Angiotensin-converting enzyme (ACE) inhibitors proved to be effective not only in patients with high renin and elevated blood pressure, but also in many hypertensive patients with normal levels of plasma renin activity. ACE inhibitors are used in a wide range of chronic illnesses such as atherosclerosis, hypertension, myocardial infarction, heart failure, diabetic complications, and stroke. To date, more than ninety controlled clinical trials evaluating the beneficial effects of 14 different ACE inhibitors were conducted. Moreover, data from experimental studies showed that ACE inhibitors can attenuate the development of atherosclerosis, oxidative stress, and vascular inflammation in a wide range of species indicating that ACE inhibition also favourably affects the vasculature. More than fifteen years ago, the bi-sulfydryl ACE-inhibitor zofenopril has shown an excellent clinical safety and efficacy in patients with hypertension and in those with myocardial infarction. More recently, this compound exhibited a potent antioxidant and antiatherosclerotic effect indicating a clinical useful vasoprotective action.

Keywords: ACE, zofenopril, atherosclerosis, acute myocardial infarction
Rationale of the use of ACE Inhibitors as Vasculoprotective Agents

Angiotensin-converting enzyme (ACE), which is responsible for conversion of angiotensin I to angiotensin II (ANG II) and degradation of bradykinin, is a component of the renin-angiotensin-aldosterone axis. ANG II is a potent vasoconstrictor and the principal active peptide of the reninangiotensin system. ANG II also regulates cellular proliferation, inflammation, oxidation-sensitive mechanisms, and endothelial function.  

The ACE inhibitor captopril has a sulfydryl group to coordinate to the active site zinc ion, enalaprilat has a carboxylate group, and fosinopril has a phosphate group; zofenopril has 2 sulfydryl groups. Enalapril, ramipril and cilazapril are ethyl-ester derivatized prodrugs that are well absorbed from the gut and, although inactive in vitro, are hydrolysed to the active diacid forms (enalaprilat, ramiprilat and cilazaprilat) in vivo by esterases in the liver, blood and other tissues. In plaques from human coronary arteries, ACE and ANG II were found to be overexpressed in unstable atherosclerotic lesions, primarily at the site of plaque rupture where macrophages and interleukin-6 colocalize. Interleukin-6 expression in VSMCs and macrophages is stimulated by ANG II. NO induces vasodilation, inhibits expression of adhesion molecules, and decreases platelet aggregation and VSMC proliferation. ANG II counteracts NO by its vasoconstricting properties and, more important, by altering NO bioactivity. Although ANG II infusion increases plasma NO, it can interfere with tissue NO bioactivity, probably via increased production of superoxide radicals (and other ROS) by VSMCs. In turn, reduced NO synthesis increases levels of superoxide and nuclear factor-kB, thereby increasing ACE expression and ANG II–receptor type 1. activation. Accordingly, chronic antagonism of nitric oxide synthase (NOS) can lead to increased ANG II-receptor type 1 gene transcription, which also suggests that endothelial dysfunction can directly increase the ANG II induced adverse vascular effects. By contrast, bradykinin stimulates NO synthesis by the endothelium. ANG II and bradykinin have opposite effects on fibrinolysis: angiotensin metabolites increase plasminogen activator inhibitor-1 activity, whereas bradykinin increases levels of tissue plasminogen activator. Because ANG II and bradykinin affect endothelial function, oxidation-sensitive mechanisms, and arterial inflammation, and because ACE activity is mainly (>90%) localized in the endothelium, using ACE inhibitors to treat atherosclerosis and its clinical sequelae is consistent with the hypothesis. 

Since then, an expanded view of RAS has gradually emerged. Local tissue RAS systems have been identified in most organs. Recently, evidence for an intracellular RAS has been reported. The new expanded view of RAS therefore covers both endocrine, paracrine and intracrine functions. Other peptides of RAS have been shown to have biological actions; angiotensin 2–8 heptapeptide (Ang III) has actions similar to those of Ang II. Further, the angiotensin 3–8 hexapeptide (Ang IV) exerts its actions via insulin-regulated amino peptidase receptors while angiotensin 1–7 (Ang 1–7) acts via mas receptors. The discovery of another ACE2 and of renin receptors has made our view of RAS unexpectedly complex. Great expectations are now generated by the introduction of renin inhibitors.

Experimental Studies in which the Sulfydryl ACE-Inhibitor Zofenopril Exerted Potent Vasoprotective Effects

Different ACE inhibitors have quite different chemical functional groups and these structural variations may account for different in vivo and in vitro effects. The ACE inhibitor captopril has a sulfydryl group to coordinate the zinc ion of the active site, enalaprilat has a carboxylate group, and zofenopril has 2 sulfydryl groups. Sulfhydryl ACE-inhibition, in particular zofenopril, stimulates the NO activity and decreases oxidative stress in human endothelial cells. Zofenopril decreases atherosclerotic development also reducing reactive oxygen species in rabbits and mice. This effect was more potent than that achieved by the sulfhydryl ACE inhibitor captopril. Consistently, zofenopril induced cardioprotective effect in perfused rat heart subjected to ischemia and reperfusion as well as attenuated hypertrophic response in rats with myocardial infarction. Moreover, the impact of zofenopril in comparison to the non-sulfhydryl ACE-inhibitor...
enalapril was studied in non non-obese diabetic mice (NOD). Insulin-dependent diabetes mellitus (IDDM) development was monitored weekly through glycosuria measurement. Zofenopril delays the onset of diabetic conditions of about 50%, and ameliorates polyuria. These data suggest that ACE-inhibitor therapy may be useful in IDDM, in particular sulfhydryl inhibition would display a better efficacy than enalapril. Finally, zofenopril, also prevents renal ischemia/reperfusion injury in rats. The interaction between antioxidant action of sulfhydryl ACE-inhibitors, nitric oxide and vascular function may explain the vascular protection afforded by these drugs.

Vasoprotective Effects in Humans

Carotid intima-media thickness (CIMT) measured by ultrasound has been shown to correlate with the presence of cardiovascular disease and is now widely accepted as a subclinical marker for atherosclerotic disease. Studies have shown that ACE-inhibitors exert anti-atherosclerotic effects, which depend to some extent on the degree of blood pressure lowering provided by these drugs. Many randomized clinical trials of antihypertensive drugs compared with placebo or no-treatment have demonstrated both a reduction of CIMT, a validated measure of subclinical atherosclerosis and predictor risk for clinical cardiovascular events, than a protection against clinical stroke events.

Specifically, some trials of patients with heart failure (HF) following acute myocardial infarction (AMI) support the benefit of ACE inhibition in this large patient population. In the SOLVD study, at 40 months, enalapril lowered significantly the risk of AMI by 23% and unstable angina by 20%, and continued to lower the rates of coronary events throughout follow-up. In the SAVE trial, AMI survivors with reduced left ventricular function were randomly assigned to receive captopril or placebo. After 42 months of followup, captopril not only reduced all-cause mortality by 19%, but also lowered significantly the risk of recurrent infarction by 25%. In the Acute Infarction Ramipril Efficacy (AIRE) and Trandolapril Cardiac Evaluation (TRACE) clinical trials, patients with HF were randomly assigned to receive an ACE inhibitor or placebo within days after AMI. The rate of recurrent infarction did not differ between groups; however, a systematic overview of these trials reported a significant reduction in recurrent infarction with ACE inhibition. Another detailed meta-analysis of trials of ACE inhibitors after AMI found that ACE inhibitor therapy lowered the risk of sudden cardiac death significantly (odds ratio = 0.80; [CI]: 0.70 to 0.92). Taken together, these clinical data can be interpreted that ACE inhibitors lower the risk of coronary events attributed to plaque instability and/or rupture.

In the Prevention of Atherosclerosis with Ramipril-2 (PART-2) study, patients with carotid atherosclerosis were assigned to receive ramipril or placebo. Throughout the 4-year follow-up, carotid artery wall thickness and plaques differed between the two groups favoring placebo. Numerous findings were reported in a trial of simvastatin and enalapril administered randomly to normocholesterolemic patients. Angiographic measures of coronary atherosclerosis did not differ betweenenalapril- and placebo-treated patients, although fewer patients receiving enalapril experienced clinical events at 4-year follow-up.

In the Study to Evaluate Carotid Ultrasound Changes in Patients Treated with Ramipril and Vitamin E (SECURE), a substudy of the Heart Outcomes Prevention Evaluation (HOPE) trial, patients at high risk of coronary events were assigned to receive ramipril, vitamin E, or both. Interestingly, at 4.5 years, ramipril reduced significantly progression of carotid intima-media thickness by 0.008 mm per year.

Zofenopril has shown clinical safety and efficacy in hypertensive patients and in those with AMI. Preliminary in vivo human evaluation showed that zofenopril accumulates in organs which express high levels of ACE, like lungs and kidneys, and in organs involved in drug metabolism such as the liver and gall bladder. A measurable concentration of zofenopril was also found in the target tissues such as the kidney and to a minor extent, the heart, where it can afford organ protection. When clinical trials conducted with zofenopril began with a vast study program called SMILE, certain questions remained unanswered, notably the long-term benefit of the ACE inhibitors beyond a short administration period. SMILE (Survival of Myocardial Infarction
Long-Term Evaluation), begun in 1995, is studying zofenopril to determine the effects of using this ACE inhibitor in the follow-up of AMI.\textsuperscript{32–34} Indeed, zofenopril is effective in reducing cardiac events after AMI and as an antihypertensive drug in the SMILE study. Zofenopril has successively demonstrated benefits on the reduction of morbidity and mortality after anterior AMI in STEMI patients, benefits that are maintained over the long term, since at 1 year, the mortality rate is significantly lower in the zofenopril group compared to the placebo group. SMILE's analyses have shown the value of zofenopril in subpopulations, particularly at-risk patients, hypertensive patients, and diabetics, whose prognosis after AMI is more severe than in patients without hypertension or diabetes. Zofenopril can be administered early, even in a more favorable situation, to AMI patients with no ST-segment elevation (NSTEMI).\textsuperscript{30,32–34} The SMILE program is continuing, notably with SMILE IV, where it is being compared to ramipril, and in the ZAAMIS trial, designed to confirm its vasoprotective effects. The results of the SMILE-ISCHEMIA study support the cardioprotective role of zofenopril when given to patients with normal left ventricular function after AMI.\textsuperscript{35} Finally, it was evaluate the clinical efficacy of the early administration of zofenopril in a group of patients with and without metabolic syndrome and anterior AMI enrolled in the Survival of Myocardial Infarction Long-Term Evaluation (SMILE) Study.\textsuperscript{36} Results demonstrated the striking benefit of early administration of zofenopril in metabolic syndrome patients with AMI.\textsuperscript{36} The antihypertensive effect of zofenopril + hydrochlorothiazide or zofenopril was similar in patients with (77%) and without metabolic syndrome. In patients with and without metabolic syndrome, however, diastolic and systolic blood pressure reductions were significantly greater with zofenopril + hydrochlorothiazide (with metabolic syndrome: 14+-8/21+/-14 mmHg; without metabolic syndrome: 15+-7/23+/-14 mmHg) than with zofenopril alone (with metabolic syndrome: 10+-9/11+/=-15; without metabolic syndrome: 12+-10/14+/=-18 mmHg).\textsuperscript{37} The safety of the two treatments was similar in patients with and without metabolic syndrome. This effect was particularly evident in patients with metabolic syndrome, in whom blood pressure control is more difficult to achieve and who are at greater risk for cardiovascular events.

Overall, in patients with mild to moderate hypertension, the efficacy and safety of zofenopril 30 mg od was compared with enalapril 20 mg od during 12 weeks of treatment.\textsuperscript{38} Both treatments significantly reduced systolic and diastolic blood pressure. However, BP reduction was significantly greater with zofenopril during the initial 4 weeks of treatment compared with enalapril.\textsuperscript{38} A similar number of patients reported adverse events in the two study groups. However, the severity of adverse events were significantly milder with zofenopril compared with enalapril. Similarly, in a multi-centre and double-blind study including 304 middle-aged to elderly patients with mild to moderate hypertension who were randomized to receive either zofenopril or atenolol for 4 weeks, BPs were substantially reduced by either treatment, but after 4 weeks, the systolic and diastolic BP reductions were significantly greater with zofenopril (P < 0.05) compared with atenolol.\textsuperscript{39} The number of subjects with adverse drug reactions possibly or probably related to the study medication was 14 (9.1%) in the zofenopril group and 30 (20.8%) in the atenolol group (P = 0.008). Moreover, zofenopril was compared also with the calcium antagonist amlodipine among 303 hypertensive patients, aged 18–75 years.\textsuperscript{40} Both drugs were well tolerated and lowered blood pressure.\textsuperscript{40} Zofenopril also induces a more rapid initial lowering of BP over the first month of therapy in comparison to the angiotensin II type 1 receptor (AT1) antagonist losartan.\textsuperscript{41} Patients aged >/= 65 years with mild to moderate essential hypertension were randomised to receive either zofenopril 30 mg or lisinopril 10 mg.\textsuperscript{42} At the end of the treatment diastolic BP was not significantly different between the two treatment groups (P = NS). Thus, in elderly hypertensive patients, treatment with zofenopril or lisinopril were effective and well tolerated.

Interestingly, we established also that zofenopril reduced potently oxidative stress in comparison to enalapril in patients with essential hypertension.\textsuperscript{43} Moreover, sulfhydryl ACE-inhibition normalizes nitrate production and potently reduces the asymmetrical dimethyl-L-Arginine (ADMA) increase
observed in hypertensive patients. Specifically, NOx levels were found to be significantly higher in zofenopril-treated compared to enalapril-treated patients. More relevantly, and consistently with preclinical studies, we also show that the carotid antiatherosclerotic effect (CIMT) of the sulfhydryl ACE inhibitor zofenopril in comparison to enalapril in hypertensive patients. The observation of higher plasma levels of ADMA in the zofenopril group might seem at odds with the reduced CIMT. In our conditions, CIMT of the right and left common carotid arteries was similar at baseline in both groups (P = NS). However, CIMT for 5 years revealed a significant reduction in the zofenopril group but not in the enalapril group (P < 0.05 vs. enalapril-treated group). The anti-atherosclerotic effect seen in our long-term study was coupled with beneficial effects on endothelial function measured by ultrasound detection of brachial artery reactivity and endothelium-dependent dilation (flow-mediated dilation, FMD). FMD was significantly increased in the zofenopril-treated group (P < 0.01).

Conclusions
We still do not know whether some parameters, such as genetic determinants, for example such of endothelial nitric oxide synthase in affording atheroprotective effect, or markers of inflammation not investigated in the HOPE study, can help to identify patients in whom ACE inhibitors would produce a more marked benefit than that found in the total trial population. Besides, improvement in endothelial function with quinapril was limited to coronary patients with the insertion allele (DI or II) of the ACE gene; and, conversely, enalapril improved endothelial function primarily in patients with the deletion allele (DI or DD genotypes). A great bulk of evidence indicates that the additional antioxidant properties of the ACE inhibitors are particularly evident in those containing a sulfhydryl group. These protective effects might include a reduction of growth factor gene expression, and reduction of polymorphonuclear cell chemoattractant release. Zofenopril which possesses 2 sulfhydryl groups is one of the most effective and vasoprotective ACE drugs.

In the absence of major contraindications (angioedema, intolerable cough or hypotension, or decline in renal function), patients with established atherosclerosis should be treated with ACE inhibitor therapy. In this regard, zofenopril showed an excellent safety and efficacy both in adult and elderly patients. Diabetic patients with an additional cardiovascular risk factor should also be on ACE inhibitor therapy.

One major problem of establishing an “antiatherosclerotic drug” is the difficulty in assessing “true” antiatherosclerotic activity over many years in human arteries. The majority of preclinical studies are based on the drug effects on atherosclerotic lesion progression. Obvious concerns related to the differences in atherosclerotic disease between humans and animal models are important to emphasize. As it is well established that atherosclerosis begins in the very early phase of human life, more questions are arising regarding the “ideal” age for starting preventive therapy for the disease. This issue is particular relevant when considering primary prevention of atherosclerotic-related diseases. If the “ideal” treatment were to begin in early life, we must consider the issue of safety. Indeed, since the follow-up of the clinical studies is usually 1–7 years, we do not know very much about the effects of long-term exposure for the majority of available drugs.

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References


