ADIS DRUG EVALUATION

Drugs 2002; 62 (1): 107-133
0012-6667/02/0001-0107/$27.50/0

Beraprost

A Review of its Pharmacology and Therapeutic Efficacy in the Treatment of Peripheral Arterial Disease and Pulmonary Arterial Hypertension

Ezequiel Balmori Melian and *Karen L. Goa*

Adis International Limited, Auckland, New Zealand.

Various sections of the manuscript reviewed by:

A. K. Andreassen, Hjertemedisinsk avdeling Rikhospitalet, Oslo, Norway; ***H. Boccalon***, Service de Medecine Vasculaire, Hopitaux de Toulouse, Toulouse, France; ***R. Ewert***, Department of Pulmonary Disease, Klinik und Poliklinik für Innere Medizin B, Greifswald, Germany; ***A. W. Gardner***, Department of Medicine, University of Maryland, Baltimore, USA; ***M. M. Hoeper***, Department of Respiratory Medicine, Hannover Medical School, Hannover, Germany; ***F. Ichida***, Toyama Medical and Pharmaceutical University, Toyama, Japan; ***K. K. Talwar***, Department of Cardiology, All India Institute of Medical Science, New Delhi, India; ***R. Verhaeghe***, Centre for Thrombosis and Vascular Research, Leuven, Belgium; ***F. Violi***, Instituto di la Clinica Medica, Universita La Sapienza Roma, Rome, Italy.

Data Selection

Sources: Medical literature published in any language since 1980 on beraprost, identified using Medline and EMBASE, supplemented by AdisBase (a proprietary database of Adis International). Additional references were identified from the reference lists of published articles. Bibliographical information, including contributory unpublished data, was also requested from the company developing the drug.

Search strategy: Medline search terms were 'beraprost' or 'ML 1129' or 'ML 129'. EMBASE search terms were 'beraprost' or 'ML 1129' or 'ML 129'. AdisBase search terms were 'beraprost' or 'ML 1129' or 'ML 129'. Searches were last updated 5 Dec 2001.

Selection: Studies in patients with peripheral arterial disease or pulmonary arterial hypertension who received beraprost. Inclusion of studies was based mainly on the methods section of the trials. When available, large, well controlled trials with appropriate statistical methodology were preferred. Relevant pharmacodynamic and pharmacokinetic data are also included.

Index terms: Peripheral arterial disease, chronic arterial occlusion, intermittent claudication, pulmonary arterial hypertension, primary pulmonary hypertension, pharmacodynamics, pharmacokinetics, therapeutic use.

Contents

Summary

Abstract

Beraprost sodium (beraprost) is a stable, orally active prostacyclin analogue with vasodilatory, antiplatelet and cytoprotective effects. Beraprost acts by binding to prostacyclin membrane receptors ultimately inhibiting the release of Ca^{2+} from intracellular storage sites. This reduction in the influx of Ca^{2+} has been postulated to cause relaxation of the smooth muscle cells and vasodilation.

Data from a large, randomised, double-blind, multicentre study indicated that beraprost was as efficacious as ticlopidine in the treatment of patients with peripheral arterial disease (Buerger's disease and arteriosclerosis obliterans). Most patients receiving beraprost exhibited reduction of ulcer size, reported improvement of granulation appearance of the tissue and showed improvement of pain at rest and sensation of cold in the extremities.

In a large pivotal clinical trial in patients with intermittent claudication, beraprost treatment was associated with statistically significant increases in pain-free and absolute walking distances compared with those in patients receiving placebo. Statistically significant differences in the incidence of critical cardiovascular events among both treatment groups were not observed but patients receiving beraprost were more likely to be satisfied with changes in their quality of life. However, while preliminary unpublished data from a large, phase III, placebo-controlled study in the US suggested a trend toward fewer critical cardiovascular events (no specific data presented), this study did not confirm the positive results from the European phase III trial and statistical significance was not achieved in the study's endpoints relating to exercise.

A series of small, noncomparative clinical trials of patients with the rare condition of pulmonary arterial hypertension (PAH) demonstrated that substantial reductions of pulmonary arterial pressure and resistance, increase of cardiac output, and increase of exercise capacity appeared to be associated with beraprost therapy; however, these data are very limited and in most instances are not fully published.

Beraprost is a well tolerated agent. Overall, the main adverse events include

headache, hot flushes, diarrhoea and nausea. However, patients with PAH showed higher incidence of adverse events than those with peripheral arterial disease.

Conclusion: Beraprost, an orally administered PGI_2 analogue, is generally well tolerated and appears to be an effective agent in the treatment of patients with Buerger's disease and arteriosclerosis obliterans. Comparative data from a large randomised trial indicated that the drug appears as effective as ticlopidine in patients with these conditions. In patients with intermittent claudication, significant benefits of beraprost compared with placebo were reported in a randomised clinical trial; however, the use of beraprost in these patients is not supported by recent preliminary unpublished data from a large, phase III, placebo-controlled study. Limited data suggest some efficacy with long-term beraprost treatment of patients with PAH, where options are few and where oral administration of the drug could be a considerable advantage over intravenous prostacyclin (PGI_2) therapy. Additional well-designed and, where possible, large trials with active comparators are necessary to define more precisely the place of beraprost in the treatment of patients with PAH, Buerger's disease and arteriosclerosis obliterans.

Pharmacodynamic Properties

Beraprost sodium (beraprost) is a stable, orally active prostacyclin (PGI_2) analogue with pharmacodynamic properties similar to those of PGI_2. The mechanisms of action of beraprost are currently under investigation and are likely to involve relaxation of vascular smooth cells, inhibition of platelet aggregation, dispersion of existing platelet aggregates, inhibition of chemotaxis and cell proliferation and cytoprotective effects.

Beraprost binds to PGI_2 membrane receptors linked to adenylate cyclase which induces the production of cyclic adenosine and guanosine monophosphates (cAMP and cGMP, respectively). It has been postulated that this effect of beraprost on cAMP and cGMP inhibits the release of Ca^{2+} from intracellular storage sites reducing the transmembrane influx of Ca^{2+}. This reduction is believed to cause relaxation of the smooth muscle cells and to induce vasodilation.

Beraprost exhibited strong antiplatelet action *in vitro* and in animal and human studies. *In vitro*, this effect was present in human, rabbit, guinea-pig, rat, dog and cat platelet rich plasma and was between 0.2 and 0.5 as potent as that of PGI_2. In animal models, beraprost improved arterial lesions induced by sodium laureate. In humans, multiple doses of beraprost appeared to inhibit platelet aggregation caused by adenosine diphosphate, collagen and epinephrine.

In animal models, beraprost significantly inhibited the formation of fibrinous thrombosis, perivascularity, fibrosis and thinning of the arterial wall and increased blood flow in electrically occluded arteries. This antithrombotic effect of beraprost was 700- and 2700-fold more potent than that of ticlopidine and cilostazol, respectively. In humans, beraprost is associated with reductions of pulmonary arterial pressure and resistance and with increases of cardiac output. Beraprost also produced vasodilation in different arterial beds in animal studies via a mechanism similar to that of PGI_2 and improved red blood cell deformability *in vitro* and *in vivo*.

In vitro, beraprost inhibited the production of the cytokines interleukin-1, interleukin-6 and tumour necrosis factor in alveolar macrophages. Moreover, beraprost inhibited in a dose-dependent manner the production of cytotoxic oxygen metabolites responsible for endothelial cell damage and reduction of endogenous PGI_2 production.

Pharmacokinetic Properties

In human studies, administration of single or multiple doses of beraprost produced peak plasma concentrations (C_{max}) of the unchanged drug within 30 to 60 minutes. C_{max} and area under the concentration curve (AUC) values ranging from 53 to 345 ng/L and 46.7 to 455.4 μg/L • h, respectively, were obtained after beraprost administration to healthy volunteers. Moreover, accumulation of beraprost and its metabolites was not observed after repeated administration. In animal studies, beraprost was primarily detected in the liver, kidney and gastrointestinal tract and to a lesser extent in the lung, blood and heart. Beraprost was excreted mainly as its main metabolite (free acid 2,3-dino-beraprost), rapidly (<72 hours) and through the faeces (73 to 82%) and urine (13 to 15%).

Therapeutic Use

Peripheral arterial disease. Results from a randomised, double-blind clinical trial of patients with peripheral arterial disease (Buerger's disease and arteriosclerosis obliterans), who had ischaemic ulcers in the extremities, indicated that beraprost (40μg three times a day, n = 84) appeared as efficacious as ticlopidine (200mg in the morning and evening and 100mg in the afternoon, n = 91). According to endpoint analyses of final global improvement and usefulness, similar effect on ulcers, rest pain and cold in the extremities was reported among patients in both treatment groups, where more than 50% of treated patients showed improvement in most parameters. Numerous, small (n = 23 to 71), noncomparative clinical trials that evaluated the improvement of objective and subjective symptoms after beraprost treatment of patients with peripheral arterial disease suggest benefit with the drug. However, these trials were not randomised, did not include comparator treatment, included relatively small numbers of patients and measured few objective parameters.

Data from a large (n = 422), randomised, double-blind, placebo-controlled, multicentre study of patients with intermittent claudication indicated that at treatment endpoint (6 months) more patients receiving beraprost showed an improvement of >50% in pain-free walking distance (91 *vs* 71, $p < 0.05$). Also, patients receiving beraprost showed a greater increase from baseline of pain-free (81.5 *vs* 52.5%, $p < 0.01$) and absolute walking distance (60.1 *vs* 35%, $p < 0.01$) although, a comparable incidence of critical cardiovascular events (4.8 *vs* 8.9%; all comparisons *vs* placebo). However, patients in the beraprost group were more likely to be satisfied with changes in their quality of life ($p < 0.05$). It is important to note, however, that recent preliminary unpublished data from a large ($n \approx 750$), phase III, placebo-controlled study do not support the use of beraprost in patients with intermittent claudication.

Pulmonary arterial hypertension. The efficacy of different dosages of beraprost has been evaluated in the treatment of patients with pulmonary arterial hypertension in small noncomparative trials (n = 11 to 12) and one retrospective comparison with historical controls. Because of the small number of patients included and limited statistical analysis reported, the data from these trials regarding the efficacy of beraprost treatment are inconclusive. However, there is some evidence that the drug decreased pulmonary arterial pressure and vascular resistance (in some cases, significantly), significantly increased cardiac output and improved New York Heart Association functional class in some patients.

Tolerability

Beraprost is generally a well tolerated agent. Combined data of 7515 patients with peripheral arterial disease indicated that adverse reactions were reported in 4.9% of patients. The main adverse events (incidence ≤1.2% in each case) were headache, facial hot flushes, hot flushes, diarrhoea and nausea. In a placebo-

controlled clinical trial of patients with intermittent claudication (n = 422), the incidence of adverse events in beraprost recipients was 16.7%, with headache (6.2%) and vasodilation (5.3%) being the most commonly reported adverse events. In this study, 8.6 and 14.5% of patients receiving beraprost and placebo, respectively, discontinued treatment.

Combined data from clinical studies in patients with pulmonary arterial hypertension receiving beraprost therapy showed a higher incidence (60% of patients) of adverse events than in patients with peripheral arterial disease, although tolerability data are only available for 40 patients. Headache (22.5%), increased lactate dehydrogenase (12.5%), increased bilirubin (10%), hot flushes, diarrhoea, nausea and increased triglycerides (all 7.5%) were the most commonly reported adverse events.

Dosage and Administration

The dosage recommendations outlined in this section focus on the use of beraprost in Japan, South Korea, The Philippines and Thailand, where beraprost has been approved for use in patients with peripheral arterial disease and pulmonary arterial hypertension (not approved for pulmonary arterial hypertension in The Philippines).

Beraprost is administered orally and should be taken after meals. In adult patients with peripheral arterial diseases beraprost 40μg three times a day is usually recommended. For adult patients with pulmonary arterial hypertension beraprost treatment starts at 60μg a day divided in three doses and can be increased gradually under careful observation up to a maximum of 180μg daily divided in three or four doses. However, in a recent 12-week clinical trial patients were administered higher beraprost dosages. In elderly patients beraprost should be prescribed with caution and it should be administered to pregnant women only if therapeutic benefits outweigh the risk of beraprost treatment. Beraprost is contraindicated in nursing women and its safety has not been defined in children.

Beraprost should be administered with care in patients receiving anticoagulant, antiplatelet or fibrinolytic agents and may also increase bleeding in menstruating patients or those with bleeding tendency. In combination with other PGI_2 preparations, beraprost may further reduce blood pressure.

1. Introduction

Beraprost sodium (hereafter referred to as beraprost) is a stable, orally active prostacyclin (PGI_2) analogue with antiplatelet, vasodilating and cytoprotective properties (sections 3.1, 3.3 and 3.4), similar to that exhibited by PGI_2 (see review by Vane and Botting[1]).

PGI_2, an endogenous prostaglandin, is the main product of arachidonic acid (AA), which is primarily produced in endothelial cells (and to lesser extent in smooth muscle cells) of all vascular tissues.[2,3] PGI_2 is the most potent endogenous inhibitor of platelet aggregation and also causes dispersion of existing platelet aggregates *in vitro* and in the systemic circulation.[4-6] PGI_2 is involved in complex interactions of the vessel wall, blood flow and platelet function and it is postulated that it antagonises the platelet aggregation and vasoconstriction caused by endogenous thromboxane A_2 (TXA_2). The mechanisms of its interactions are not fully understood but are likely to involve relaxation of vascular smooth cells, inhibition of platelet aggregation, dispersion of platelet aggregates, inhibition of chemotaxis and cell proliferation, inhibition of production and secretion of endothelin and cytoprotective effects.[1,7,8]

On the basis of the varied biological functions exhibited by PGI_2, its potential use in different therapeutic areas was postulated. Indeed, PGI_2 has

been efficacious in the treatment of patients with pulmonary arterial hypertension (PAH),[9,10] peripheral vascular disease,[11] ischaemic disease of the leg,[6] and Raynaud's phenomenon.[12] However, its short elimination half-life (approximately 3 minutes), chemical instability and intravenous route of administration hinder its therapeutic use in some indications.[13] Reconstituted solutions of PGI_2 must be stored at 2 to 8°C for no longer than 48 hours and only about 8 hours at room temperature.[14] A more stable analogue, iloprost, provided pain relief and accelerated ulcer healing in patients with stage III or IV critical leg ischaemia, but for long-term treatment its intravenous administration was impractical and could lead to clinical complications.[15,16] Moreover, an oral formulation of iloprost was not efficacious in the treatment of patients with Buerger's disease.[17]

Beraprost is an orally administered PGI_2 analogue. Under light-shielded conditions, the powder form of beraprost is stable for 42 months at room temperature and for 6 months at 50°C, and solutions of beraprost are stable for at least 10 days (pH = 4 to 10).[18] The extended half-life (section 4) and improved chemical stability of beraprost are expected to mitigate some of the problems associated with intravenous PGI_2 formulations. This review examines the role of beraprost in the treatment of patients with peripheral arterial disease and PAH.

2. Overview of Disease States

2.1 Overview of Peripheral Arterial Disease

Atherosclerosis is a multiorgan disorder characterised by localised plaque formation in selected sites of the arterial tree, particularly coronary arteries, carotid bifurcation and leg arteries, and is the most common cause of peripheral arterial diseases. Peripheral arterial disease symptoms are numerous and include intermittent claudication, pain at rest, ulceration and gangrene [as present in Buerger's disease (thromboangiitis obliterans) and arteriosclerosis obliterans].[19-22] Buerger's disease is a nonatherosclerotic inflammatory disease that most commonly affects the small- and medium-sized arteries and nerves of the extremities. The cause of this disease is unknown but it affects typically young male smokers.[23] Arteriosclerosis obliterans, on the other hand, is characterised by complete obliteration of the lumen of the artery due to proliferation of the intima of the small vessels.

Raynaud's phenomenon, a vasospastic condition, is characterised by episodic vasospasm of the finger and toes typically caused by exposure to cold.[24] However, there are differences in the definition of this condition in different countries and probably among different investigators, which might explain the high variability of the prevalence of the disease worldwide.[24,25] Also, accurate diagnosis is difficult because observation of vasospastic episodes, which rarely occur in the presence of physicians, is required. Therefore, studies that included patients with this condition are not included in this review, although, it is important to note that, so far, there is no conclusive published evidence demonstrating the efficacy of beraprost in patients with Raynaud's phenomenon.

One-third of patients with peripheral arterial disease experience the most common symptom, intermittent claudication (defined as pain in one or both legs during exercise) which worsens in 25% of the patients and causes amputation of the affected limb in 5% within 5 years.[26] Approximately 5 to 10% of patients with peripheral arterial disease exhibit critical leg ischaemia (ischaemic pain in the distal foot, ischaemic ulceration or gangrene). Although many patients do not have either intermittent claudication or leg ischaemia at rest, most patients with peripheral arterial disease have limited ability to perform daily activities.[27]

The prognosis for the limb(s) affected by intermittent claudication is therefore generally benign. Nevertheless, intermittent claudication symptoms are often a sign of systemic atherosclerosis and affected patients have a 2- to 4-fold increase in cardiovascular mortality compared with age-matched healthy individuals.[28-30] Therefore, the goals of therapy in patients with peripheral arterial disease, including intermittent claudication, are to eliminate the ischaemic symptoms, to prevent progression to vascular occlusion, to improve walking capacity and quality of life, and more importantly, to

prevent cardiovascular complications such as stroke, myocardial infarction or death.[19] For patients with more severe symptoms of the disease, the goals of therapy also include eliminating the ischaemic symptoms and preventing progression of the disease to vascular occlusion.[19,27]

Treatment of peripheral arterial diseases includes surgical (endarterectomy, bypass grafting, amputation), endovascular (transluminal angioplasty, endovascular stents, intra-arterial thrombolytic therapy) and, more commonly, nonoperative (risk factor modification, exercise, drugs) therapies. Combination of risk factor modification and exercise is to date the most effective treatment of patients with peripheral arterial diseases, although a substantial number of patients require pharmacological intervention.[21,22]

2.2 Overview of Pulmonary Arterial Hypertension (PAH)

PAH is a rare, life-threatening disease characterised by progressive pulmonary hypertension, ultimately causing right ventricular failure and death.[31-33] Clinically, PAH is defined as a mean pulmonary arterial pressure of more than 25mm Hg at rest or more than 30mm Hg during exercise.[29,33] PAH can be due to primary pulmonary hypertension (PPH, sporadic or familial) or related to collagen vascular disease, congenital systemic abnormalities to pulmonary shunts, portal hypertension, HIV infection, drugs/toxins or persistent pulmonary hypertension of the newborn, among other factors.[34]

The aetiology of PAH is unknown but genetic predisposition to the disease is likely (familial or sporadic PPH).[35,36] The locus of a gene linked to familial PPH has been identified on chromosome 2q31-32 and appears to have low penetrance since individuals carrying this gene have only a 10 to 20% likelihood of developing PPH.[34] Based on genetic studies, 94 families have been identified as carriers of mutations in the PPH gene worldwide.[34] Moreover, at least 26% of patients with sporadic PPH have these mutations.[37] The mutations are highly variable, often leading to premature termination codons (58%), indicating that additional genetic and or environmental factors may be responsible for the development of the clinical phenotype.[34,37-39]

The use of appetite suppressants (particularly for more than 3 months),[40] the use of methamphetamine and/or exposure to other stimuli (for example, solvents, cocaine, monocrotaline (MCT) extracts, rapeseed canola oil, hypoxia, oral contraceptives, HIV infection and cirrhosis) have been suggested to trigger PAH in susceptible individuals.[31,41] In Europe and the US the annual incidence of PAH has been estimated to be around one to two cases per million people per year but a necropsy study has suggested a prevalence of 1300 cases per million.[42] PAH affects mainly young populations (mean age at diagnosis of 36 years) but the disease can occur at any age.

Nonmedical treatment of PAH involves changes in lifestyle. In general, patients with PAH should be advised to avoid physical and mental stress. Female patients should avoid conception but the use of oral contraceptives is contraindicated. Since obesity also worsens the clinical course of the disease, exercise (only when possible) and a diet low in saturated fat and salt are recommended.[41]

Pharmacological treatment of patients with PAH includes calcium antagonists, vasodilators, anticoagulants (warfarin), diuretics, oxygen, nitric oxide, and PGI_2 (considered the best PAH treatment) and its analogues.[33,41,43] However, the efficacy of these agents in the treatment of patients with PAH is limited. Currently there is no cure for PPH and only 25 to 30% of patients respond to calcium antagonists, the most widely used drugs for long-term therapy. After cardiac catheterisation and acute challenge with vasodilators, patients showing a >20% reduction in pulmonary vascular resistance (PVR) and unchanged or improved cardiac output, are classified as potential responders to long-term therapy.[29,31] Heart-lung, single-lung or double-lung transplantation have been performed in patients with PPH but the 1- and 5-year survival rates are relatively poor (85 and 50%, respectively).[41] Additionally, many patients do not qualify for surgical intervention and, in those who

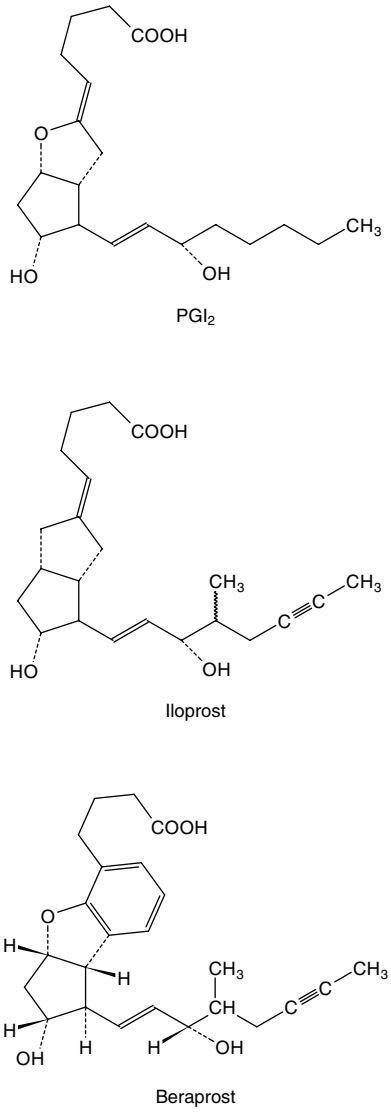

Fig. 1. Structural formulae of prostacyclin (PGI_2) and its analogues, iloprost and beraprost.

do, rapid deterioration of their condition during the waiting period associated with transplantation (several months) is not uncommon.[41]

3. Pharmacodynamic Properties

The pharmacodynamic properties of beraprost, an orally active agent with PGI_2-like structure (figure. 1), have been extensively investigated and have been the subject of several reviews.[18,44-46] These properties include antiplatelet effects,[47-53] cardiovascular effects,[54-56] antithrombotic activity,[57-59] anti-inflammatory properties,[60] effects on peripheral circulation,[57] inhibition of erythrocyte deformability[61] and cytoprotective action[62-65] (table I).

It is important to note that the activity of beraprost in different models may be the result of the combination of two or more of these effects. For example, the antiplatelet effect of beraprost may be associated with anti-inflammatory effects[56,60,63] and inhibition of neutrophil chemotaxis and superoxide generation.[62,64,65]

Beraprost binds to PGI_2 receptors on the platelet, subsequently activating adenylate cyclase and increasing cyclic adenosine and guanosine monophosphate levels (cAMP and cGMP, respectively) in the platelet membranes of humans, dogs and rats.[18,78] It has been postulated that beraprost inhibits the release of Ca^{2+} (from intracellular storage sites) and the transmembrane Ca^{2+} influx (figure 2). This results in the suppression of phospholipase A_2 activity, release of AA and eventually inhibition of TXA_2 synthesis.[18] Contrary to the antiplatelet and vasodilator activity of prostaglandin PGI_2, TXA_2 is an eicosanoid that induces platelet aggregation and vasoconstriction.

3.1 Antiplatelet Effect

3.1.1 In Vitro *Studies*

In vitro experiments showed that beraprost exhibited a strong antiplatelet effect in platelet-rich plasma of various species (human,[18,51,52] rabbit,[18,51] guinea-pig,[18,51] rat,[18,49-51] dog[18] and cat[18,51]) against different aggregation inducers [AA, thrombin, adenosine diphosphate (ADP), collagen, calcium ionophore and A23187]. In general, beraprost potency (IC_{50}, evaluated as beraprost concentration producing 50% inhibition of platelet aggregation) was between 0.1 and 0.5 times that of PGI_2 and depended, as in the case of PGI_2, on the species tested. The antiplatelet effect of beraprost was stronger in human and cat platelets. In human

Table I. Pharmacodynamic properties of beraprost

- Inhibition of platelet aggregation *in vitro*, *ex vivo* and *in vivo* in several species[18,48-53] including humans[66,67]
- Inhibition of thrombus formation in animal studies[47,57,68,69]
- Reduction of pulmonary arterial pressure and resistance, increase of cardiac output and vasodilating effects *in vitro*, *in vivo* and animal studies[18,46,54-57,59,69-75]
- Cytoprotective effect via inhibition of superoxide and cytokines generation and inhibition of chemotaxis of polymorphonuclear leucocytes *in vitro*[62-65]
- Enhancement of wound healing *in vitro* via stimulation of urokinase-type plasminogen activator in human fibroblast[76]
- Anti-inflammatory effect *in vivo* and *in vitro* via suppression of macromolecular permeability of endothelial cells[60,77]
- Inhibition of red blood cell deformability in animals caused by hypercholesterolaemia *in vitro* and *ex vivo*[61]

platelets, beraprost inhibited platelet aggregation induced by ADP and collagen (IC_{50} = 3.31 to 11.3 nmol and 9.44 to 15.98 nmol, respectively), and its potency ratio was between 0.21 and 0.53 that of PGI_2.[51] The duration of the antiplatelet effect was dose-dependent and in rabbits was 1 and >5 hours at beraprost 0.3 and 1 mg/kg, respectively.[51] Compared with no beraprost treatment, the antiplatelet effect in rat platelet-rich plasma was significant (>20% inhibition of platelet aggregate formation) up to 6 hours after beraprost ≤1 mg/kg administration.[44,51]

Additionally, beraprost was associated with marked improvement of macroscopic and histological features in the sodium laureate occlusion model in rats. Injection of sodium laureate into the artery causes injury to the endothelium followed by platelet aggregation, platelet adhesion and peripheral circulation insufficiency. Compared with placebo (water), beraprost 0.3 mg/kg (but not beraprost 1 mg/kg or ticlopidine 30 mg/kg) significantly ($p < 0.05$) improved the lesion induced by sodium laureate. It was suggested that the efficacy of beraprost in this model (similar to peripheral occlusive disease in clinical practice) was due to the antiplatelet effect of this drug in combination with suppression of leucocyte chemotaxis.[57]

3.1.2 Studies in Healthy Volunteers

A phase I study in six male volunteers receiving beraprost 25μg three times a day for 10 days showed mean reductions of platelet aggregation (from baseline) ranging from 4 to 49% (ADP-induced), 5 to 22% (collagen-induced) and 10 to 38% (epinephrine-induced) after 1, 2 or 9 days of beraprost administration. Similar reductions were also reported in six volunteers receiving 50μg three times a day.[66] Therefore, in humans, repeated administration of beraprost appears to inhibit platelet aggregation induced by ADP, collagen and epinephrine.

A similar antiplatelet effect of beraprost was also reported in a randomised, double-blind, crossover (1-week washout period after each treatment period) placebo-controlled study in healthy volunteers (n = 12), but only after 8 days of treatment.[67] Volunteers received beraprost 20, 40 or 60μg or placebo three times a day except on days 1 and 8 of each treatment period, when they received single doses of beraprost. However, the antiplatelet effect of beraprost was not observed at any of the dosages tested during the first day after beraprost administration.[67] Additionally, beraprost 20μg three times a day showed antiplatelet activity only against ADP-induced aggregation (ADP 10 μmol/ L) and all beraprost concentrations tested were not efficacious against collagen 2.5 μg/mL or AA 1 mmol/L.[67]

3.2 Antithrombotic Effects

A model of electrically induced acute thrombosis in rabbits showed the antithrombotic effect of beraprost.[57] In the control animals, the blood flow in the femoral artery started to decrease 10 minutes after electrical stimulation and after 20 minutes blood flow was obstructed almost completely indicating thrombus formation. However, beraprost 0.03 mg/kg completely prevented reduction of blood flow. Judging by the dose at which 30% reduction of blood flow occurred (0.024 mg/kg), beraprost was 700- and 2700-fold more efficacious than ticlopidine and cilostazol, respectively.[57]

Additionally, histological data indicated that, compared with no treatment, beraprost 0.3 mg/kg significantly ($p < 0.01$ for all parameters) inhibited

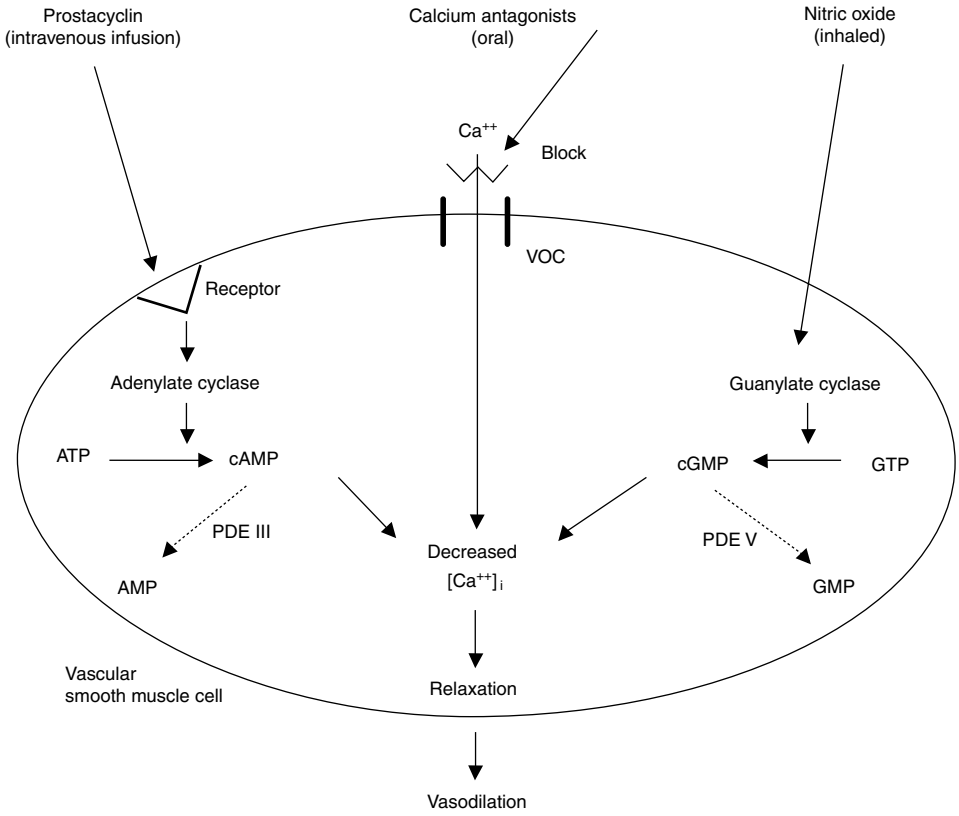

Fig. 2. Mechanisms of action of vasodilator drugs used in the treatment of patients with pulmonary arterial hypertension. Calcium antagonists inhibit the influx of Ca^{2+} into the vascular smooth muscle cell through voltage-operated calcium channels (VOC), reducing the intracellular free Ca^{2+} concentration, $[Ca^{2+}]_i$.[29] Prostacyclin (PGI_2) activates specific cell membrane receptors linked to adenylate cyclase which catalyses the formation of cGMP from GTP. cAMP and cGMP cause a reduction in $[Ca^{2+}]_i$ by a variety of mechanisms and are broken down by PDE III and PDE V, respectively. A reduction in $[Ca^{2+}]_i$ leads to relaxation of the vascular smooth muscle cell and results in vasodilation. PGI_2 and nitric oxide increase cAMP and cGMP, respectively, irrespective of the resting state of the vascular smooth muscle cell. Calcium antagonists act preferentially in cells on which there is an increase in Ca^{2+} influx and an associated elevation in vascular tone (reproduced from Wanstall and Jeffery[29]). **ATP =** adenosine triphosphate; **cAMP =** cyclic adenosine monophosphate; **cGMP =** cyclic guanosine monophosphate; **GTP =** guanosine triphosphate; **PDE =** phosphodiesterase.[29]

the formation of fibrinous thrombosis, perivascularity, and fibrosis, and thinning of the arterial wall in rats (beraprost 0.1 mg/kg, $p < 0.01$).[57] A similar antithrombotic effect of beraprost was reported in adult mongrel dogs.[68]

Beraprost 50 μg/kg/day for 60 days, but not placebo, significantly inhibited the intimal thickening (0.134 *vs* 0.205, $p < 0.01$) and luminal narrowing (0.275 ± 0.04 *vs* 0.457 ± 0.076, $p < 0.01$) of the coronary arteries caused by cardiac allograft in a cyclosporin-treated rat model.[69] The mechanism of this effect is not known but it was postulated that beraprost may protect endothelial cells, inhibit proliferation and migration of vascular smooth muscle cells or maintain vascular homeostasis through regulation of the eicosanoid cascade.[69]

3.3 Cardiovascular and Vasodilating Effects

3.3.1 Animal Studies

Intra-arterial injection of beraprost 0.1 to 3000μg in anaesthetised autoperfused dogs increased blood flow in the vertebral, coronary, mesenteric, renal, hepatic and femoral arteries.[18] The magnitude of the effect was 0.1 to 0.3 times that of PGI_2 and was greater with higher doses.[18] In vertebral arteries of adult mongrel dogs, values of ED_{40} (drug dose increasing blood flow in the artery by 40%) indicated that the vasodilating potency of beraprost (ED_{40} = 0.80μg) was inferior to that of PGI_2 (ED_{40} = 0.24μg), but superior to that of ticlopidine (ED_{40} = 600μg) and cilostazol (ED_{40} = 46μg).[79] Similar differences in ED_{40} values were obtained for beraprost, PGI_2, ticlopidine and cilostazol (0.86, 0.20, 420 and 28μg, respectively) in femoral arteries.[79] However, another study reported blood flow decreases in vertebral, carotid and femoral arteries after intravenous injection of beraprost 1 to 100 μg/kg to anaesthetised dogs.[44] Systemic blood pressure, oxygen consumption, cardiac work, cardiac output and other parameters were also decreased by beraprost but to a lesser extent than by PGI_2. No significant changes in heart rate were observed during beraprost treatment.[18]

Beraprost 10^{-9} to 10^{-6} mol/L caused vasodilation of canine isolated mesenteric, renal, coronary, femoral, basilar and middle cerebral arteries after contraction with $PGF_{2\alpha}$ or K^+.[54] This effect was greater in mesenteric and renal arteries than cerebral arteries, and was attenuated by treatment with the prostaglandin antagonist diphloretin phosphate.[54] Therefore, it appears that beraprost and PGI_2 share the same mechanism of vasodilation.[18,54]

In the rat model of tail gangrene induced by subcutaneous injection of the vasoconstrictors ergotamine and epinephrine, beraprost 0.01 to 0.3 mg/kg (but not ticlopidine 100 mg/kg or cilostazol 30 mg/kg) significantly ($p < 0.05$) inhibited the extension of gangrene in comparison with placebo treatment (water).[57] For example, among animals receiving beraprost 0.1 mg/kg (n = 19) or placebo (n = 20) the extension of gangrene was approximately 4.2 and 6.8cm, respectively ($p < 0.01$). Similar inhibition of gangrene was observed at all beraprost doses tested.[57] It was suggested that this was the result of both the vasodilating and antiplatelet effects of beraprost. In other rat models, different concentrations of beraprost treatment increased skin blood flow (3 to 10 μg/kg), skin temperature (10 to 30 μg/kg) and facilitated recovery (0.01 to 0.3 μg/kg) of skin and paw temperature after topical cooling.[44]

Moreover, the vasodilating effect of beraprost treatment may also be associated with an improvement of red blood cell (RBC) deformability.[61] Since RBCs must pass through capillaries narrower than their diameters, RBC deformability is pivotal to the rapid and homogenous perfusion of oxygen in the microcirculation. *In vitro* and *ex vivo* beraprost treatment rapidly (within 1 to 3 hours) improved the impairment of RBC deformability induced by hypercholesterolaemia. Three hours after administration of beraprost to hypercholesterolaemic rabbits, maximum pressure significantly ($p < 0.01$) decreased from 263mm Hg at baseline to 218mm Hg (beraprost 19.6 μg/kg) and 215mm Hg (beraprost 48.9 μg/kg). No effect on RBC deformability was observed in rabbits with normal levels of cholesterol.[61]

Beraprost 10 to 100 μg/kg/day decreased the degree of pulmonary hypertension in the rat MCT-induced pulmonary hypertension model.[63] The ratio of the weight of the right ventricle to that of the left ventricle plus septum (the extent of right ventricular hypertrophy) was significantly lower after beraprost 30 μg/kg/day for 2 or 4 weeks, respectively, compared with rats that did not receive beraprost (0.467 and 0.353 *vs* 0.584, $p < 0.01$). Similar reductions were reported if the start of beraprost treatment coincided with the MCT injection (as above) or if beraprost treatment was initiated 1 week or 2 weeks after MCT administration.[63,80] However, the inhibitory effect of beraprost was not observed if beraprost was started 4 weeks after MCT administration.[80]

3.3.2 Human Studies

Vasodilating effects have been observed in patients with PPH or secondary pulmonary hypertension after acute administration of beraprost.[70,71] Generally, single doses of oral beraprost (20 to 100μg) induced statistically significant reductions from baseline of pulmonary arterial pressure (ranging from –2 to –29%)[70,71] and pulmonary vascular resistance (–12 to –52%), although in some patients increases of pulmonary arterial pressure and resistance were reported.[71] Additionally, increases (11 to 32%) of cardiac output were reported in two studies after acute administration of beraprost.[70,71]

3.4 Cytoprotective Effects

The cytoprotective effect of beraprost has been demonstrated in a series of *in vitro* studies.[60,62-65,77] However, it is important to note that generally, the beraprost concentrations used were high and unlikely to be observed *in vivo*. In the MCT-induced pulmonary hypertension model in rats beraprost treatment inhibited the production of interleukin-1 (IL-1), interleukin-6 (IL-6) and tumour necrosis factor (TNF) in alveolar macrophages.[63] The inhibitory effect of beraprost on the production of these cytokines was similar to that of prostaglandin E_1.[63]

Neutrophils exposed to chemotaxins or phagocytic stimuli generate the production of superoxide that gives rise to cytotoxic oxygen metabolites. Superoxide and its metabolites cause damage to endothelial cells and decrease production of PGI_2, thereby decreasing endothelial antithrombotic function and promoting platelet aggregation.[64] In human neutrophils beraprost 1 to 100 μmol/L inhibited (by 10 to 36%) formyl-methionyl-leucyl-phenylalanine superoxide generation in a dose-dependent manner.[62] It was suggested that this inhibitory effect of beraprost was due to the suppression of Ca^{2+} influx through a cyclic AMP-dependent pathway.[62] A similar cytoprotective effect of beraprost against active oxygen metabolites or superoxide was reported earlier by Kainoh and colleagues,[64] who suggested the use of beraprost in the treatment of ischaemic diseases.

Another *in vitro* study[77] suggested that the cytoprotective effect of beraprost may also be associated with inhibition of thrombin-induced inflammatory effects causing increased permeability of vascular endothelial cells.[77] In this model, permeability was measured as fluorescein isothiocyanate conjugated (FITC) albumin transport (μg/cm^2/h) across a monolayer of cultured vascular endothelial cells treated with thrombin or thrombin plus beraprost. Beraprost (30 to 1000 nmol/L) inhibited thrombin-induced FITC-albumin transport in beraprost-treated cells compared with that of cells treated only with thrombin (12 to 19 *vs* 26.6 μg/cm^2/h, $p < 0.01$).[77] This inhibition was dose-dependent, and at the highest beraprost concentration tested, FITC-albumin transport was comparable to that observed in cells not treated with thrombin.[77]

4. Pharmacokinetic Properties

Available pharmacokinetic data are very limited, consisting of two studies in humans[18,67] and two in animals, summarised in a review.[18] Beraprost consists of two diastereoisomers, each of which is composed of racemic mixtures (D and L).[18,67] The main metabolite of beraprost in plasma, liver, kidney and lungs of rats is ^{3}H-2,3-dinor-beraprost, a free acid product of β-oxidation. Additionally, ^{3}H-15-oxo-beraprost and ^{3}H-2,3-dinor-15-oxo-beraprost (free acids) were detected.[18] It is not known whether these same metabolites are detected in humans.

4.1 Absorption and Distribution

After oral administration of a single beraprost dose (20, 40 or 60μg for 1 day) to healthy male Caucasians (n = 12), t_{max} [time to reach maximal concentration (C_{max})] values of 36 to 54 minutes were obtained for the two main D-isomers of beraprost.[67] Values of C_{max} and area under the concentration curve (AUC) increased at higher doses of beraprost and ranged from 53 to 345 ng/L and 46.7 to 455.4 μg/L • h, respectively. In general,

lower values of t_{max}, C_{max} and AUC were obtained for beraprost L-isomer. Beraprost and its metabolites did not accumulate after repeated administration (20, 40 or 60μg, three times a day for 6 days) indicating that the drug was rapidly eliminated.[67]

In healthy volunteers of Japanese descent, administration of single doses of beraprost (50, 100 and 200μg) produced C_{max} values of 0.3, 0.4 and 0.9 μg/L, respectively, at t_{max} of 30 to 60 minutes.

In rats, high levels of radioactivity were detected in the liver, kidney and gastrointestinal tract and to a lesser extent in the lung, blood and heart.[18]

4.2 Metabolism and Elimination

The elimination half-life of beraprost was approximately 60 minutes in Japanese volunteers.[18] In rats and beagle dogs, beraprost was excreted primarily as its main metabolite with virtually none excreted as the unchanged drug. Moreover, excretion of the drug was rapid (48 to 72 hours) and occurred mainly through the faeces (73 to 82%) and the urine (13 to 15%).[18]

5. Therapeutic Use

5.1 In Peripheral Arterial Disease

The efficacy of beraprost has been investigated in numerous noncomparative clinical trials of 4 to 8 weeks' duration that evaluated a variety of objective and subjective symptoms of peripheral arterial disease (section 5.1.3).[81-87]

As well, a randomised, double-blind, 6-week, clinical trial compared the efficacy of beraprost with that of ticlopidine chloride, a platelet aggregation inhibitor that has been proven efficacious in the treatment of patients with peripheral arterial diseases.[88-90] This fully published study enrolled Japanese patients with peripheral arterial disease who had ischaemic ulcers in the extremities (section 5.1.1).[91]

The largest study is a 6-month, randomised, double-blind trial conducted in France and Italy comparing the efficacy of beraprost and placebo in patients with peripheral arterial disease who all had intermittent claudication (section 5.1.2).[92]

Apart from this later study, usually clinical trials included patients with peripheral arterial disease (arteriosclerosis obliterans and Buerger's disease) who showed some ischaemic signs and symptoms, including ulceration in the extremities, pain, coldness and intermittent claudication. In general, patients were excluded from the clinical trials if they had bleeding tendency, severe hepatic or renal dysfunction, severe complicated disease, hypersensitivity to drugs or leucopenia. Additionally, pregnancy, breast feeding or potential pregnancy were also reasons for exclusion from the clinical trials.[81,82,84-87,91,93,94]

Overall, the efficacy of beraprost treatment was measured using a combination of objective [size of ulcers, nature of ulcer granulation, claudication distance, ankle-brachial index (ABI) and cyanosis] and subjective parameters (feeling of pain, coldness in the extremities and numbness). Additionally, in some studies improvement ratings on a 5-point scale were given to ulcer size (healed or markedly reduced, reduced, slightly reduced, unchanged and enlarged), appearance of granulation, subjective symptoms and patients' impressions (markedly improved, moderately improved, slightly improved, unchanged and worsened). The sum of these ratings determined the final global improvement rating and the usefulness rating of the drug treatment.

The optimum beraprost dosage has been determined to be 120 μg/day administered orally (40μg three times a day) [see section 7]. At this dosage, beraprost was found to be superior to a 20μg three times daily dosage, as assessed by the usefulness rating in a 6-week randomised dose-comparison trial in 71 evaluable patients.[94] However, there were no significant differences between the groups for any of the individual parameters tested.

5.1.1 Comparison with Ticlopidine

In a double-blind randomised study, patients (n = 84) received beraprost 120 μg/day (40μg in the morning, afternoon and evening) or ticlopidine 500 mg/day (200mg in the morning and evening

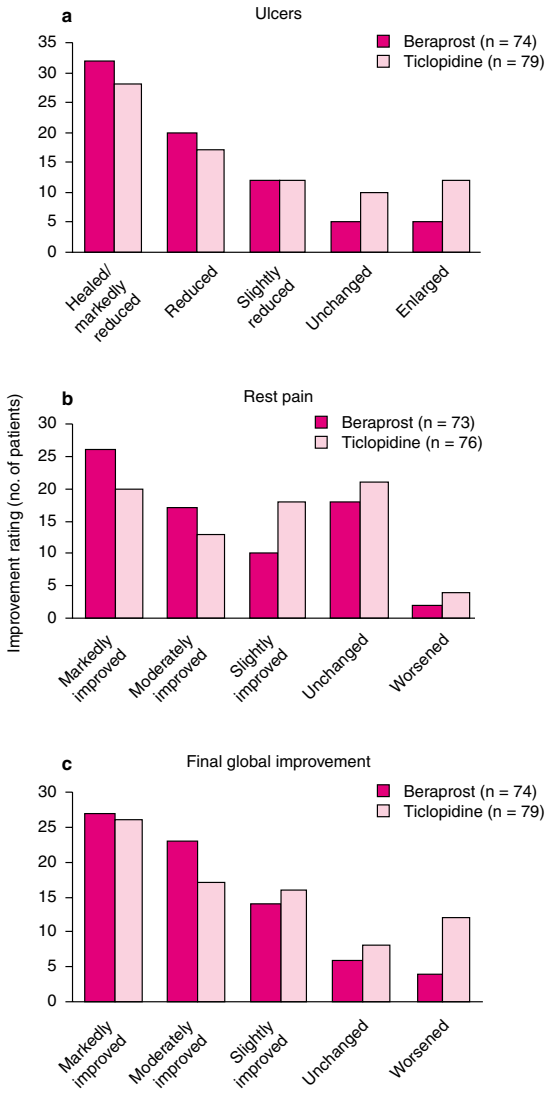

Fig. 3. Comparison of the clinical efficacy of beraprost and ticlopidine in the treatment of patients with Buerger's disease or arteriosclerosis obliterans. Patients received oral beraprost 120μg daily (40μg three times a day, n = 73) or ticlopidine 500mg (200mg in the morning and evening and 100mg in the afternoon, n = 76) for 6 weeks in a randomised, double-blind trial.[91] Efficacy is presented as **(a)** evaluation of ulcer size, **(b)** evaluation of rest pain and **(c)** final global improvement rating [the sum of scale points of objective (ulcer size and granulation appearance) and subjective (rest pain and sensation of coldness) parameters and patients' impressions].

and 100mg in the afternoon, n = 91) orally for 6 weeks.[91] Ulcer size, granulation appearance of tissue and subjective parameters (number of patients with pain at rest or cold sensation in the extremities) were recorded at baseline and treatment endpoint. In general, there were slight variations in the number of patients from both treatment groups that were included during evaluation of different parameters. The main results from this trial are shown in figure 3 and table II.

According to the improvement rating (table II), beraprost and ticlopidine had similar effects on ulcers, rest pain and sensation of cold in the extremities. For most parameters, about two-thirds of beraprost recipients and about 50 to 60% of those receiving ticlopidine showed improvement. The lowest improvement rate for both drugs was seen for 'sensation of cold extremities'.

However, significantly more beraprost than ticlopidine recipients were 'moderately or markedly improved' when appearance of granulation tissue was assessed (67.1 *vs* 50.6%, $p < 0.05$). Beraprost was also superior on the 'patient's impressions' ratings (67.6 *vs* 49.4, $p < 0.05$), but there were no significant differences between the groups in the overall endpoint analyses of final global improvement and usefulness of the drug (table II). However, it is important to note that, unlike the case of beraprost, previous double-blind trials have shown that ticlopidine is more efficacious than placebo and aspirin in the prevention of stroke, myocardial infarction or death caused by vascular events.[90]

5.1.2 Comparison with Placebo

The efficacy of beraprost in patients with intermittent claudication was studied in a pivotal, large (n = 422), randomised, double-blind, placebo-controlled, multicentre clinical trial.[92] Patients were included in the placebo run-in phase (4 weeks) if they had intermittent claudication due to noninflammatory arterial disease of the lower limbs (of >6 months' duration investigated by arteriography or Doppler scan), were stable during the previous 3 months, were not in need of invasive interventions, had a pain-free walking distance of

Table II. Improvement ratings after 6 weeks' treatment with beraprost or ticlopidine in a randomised, double-blind trial[91] of Japanese patients with peripheral arterial disease[a]

	Improvement rate (% of patients)[b]	
	beraprost[c] (n = 74)	ticlopidine[c] (n = 79)
Ulcers	70.3	57
Appearance of granulation tissue	67.1*	50.6
Subjective symptoms:		
Rest pain	58.9	43.4
Cold extremities	37.5	35.8
Patient's impressions	67.6*	49.4
Final global improvement[d]	67.6	54.4
Usefulness[e]	67.6	56.8

a Patients had Buerger's disease or arteriosclerosis obliterans.

b Patients who were 'moderately or markedly improved', except for the ulcer rating (ulcers were 'reduced' or 'healed or markedly reduced') and the 'usefulness rating' (judged according to a 100-point scale).

c Dosages were beraprost 40μg three times daily, and ticlopidine 200mg in the morning and evening and 100mg in the afternoon. Patient numbers were lower for some parameters in both treatment groups and higher for the ticlopidine group's usefulness rating (n = 81).

d Determined on a 5-point scale (markedly improved, moderately improved, slightly improved, unchanged and worsened) that included scale points of ulcer improvement, subjective symptoms and patients' impression.

e Determined on a decimal point scale from 0 ('I will never use the drug again') to 100 ('I will definitely use the drug again') and included global improvement and overall safety ratings, as judged by a physician and a subcommittee. A score ≥70 evaluated the drug treatment as 'useful'.

* $p < 0.05$ *vs* ticlopidine.

50 to 300m at the treadmill exercise test (day –28) and had no ischaemic rest pain, ulceration or gangrene. Patients were excluded from the run-in placebo phase if they were receiving antiplatelet therapy, had other diseases that affected walking, had a history of myocardial infarction or stroke in the previous 3 months, or had unstable or severe angina pectoris or type I diabetes mellitus.[92] Patients were included in the treatment phase of this pivotal trial if at the end of the placebo run-in phase (day 0) they showed a change of pain-free walking distance <25% (treadmill test) compared with results from day –28. Eligible patients were randomised to receive placebo (n = 213) or beraprost (n = 209) 40μg three times a day for 6 months (endpoint).[92]

The primary efficacy endpoint was success, defined as an improvement from baseline (day 0) of >50% in the treadmill exercise pain-free walking distance at the 6-month test and in ≥1 of the earlier tests, in the absence of critical cardiovascular events. Secondary efficacy endpoints were changes in pain-free and maximum walking distances, subjective walking distance and the incidence of critical cardiovascular events (death of cardiovascular origin, myocardial infarction, unstable angina, coronary angioplasty, coronary artery bypass graft surgery, stroke, transient ischaemic attack, critical leg ischaemia, subacute critical ischaemia, peripheral angioplasty, peripheral bypass graft surgery and amputation at any level). A tertiary efficacy endpoint was the quality of life evaluated by the use of the Quality of Life Subjective Profile questionnaire[95] that was completed at baseline (day 0) and endpoint.[92]

The main results of the trial, presented in table III and figure 4, show that beraprost was superior to placebo.

At 6 months, significantly more patients receiving beraprost were successfully treated as defined by the primary endpoint (91 *vs* 71 patients, $p = 0.036$),[92] and the percentage increases from baseline in pain-free (81.5 *vs* 52.5%, $p < 0.01$) and maximum walking distances (60.1 *vs* 35%, $p < 0.01$) were significantly higher in the beraprost group that among placebo recipients (table III). As demonstrated in figure 4, the significant difference between treatment groups in percentage increase in pain-free (figure 4a) and maximum walking distance (figure 4b) was evident at 1.5 months after starting treatment and persisted throughout treatment (except for maximum walking distance at 4.5 months, which showed no statistical difference).

At treatment endpoint, the incidence of critical cardiovascular events was 4.8 and 8.9% of patients receiving beraprost or placebo treatment, respectively. Overall, the most common critical cardiovascular event was arterial thrombosis of the leg and was reported by 8 and 14 patients in the beraprost and placebo treatment groups, respectively.

Table III. Summary of the results of a pivotal, randomised double-blind 6-month trial in patients with intermittent claudication who received oral beraprost 40μg three times daily or placebo[92]

	Beraprost (n = 209)	Placebo (n = 213)
Success rate[a] (% of patients)	43.5* [56.5][b]**	33.3 [42.2][b]
Pain-free walking distance (m)	Baseline: 130 6mo: 280 (↑81.5%)***	Baseline: 133 6mo: 245 (↑52.5%)
Maximum walking distance (m)	Baseline: 275 6mo: 467 (↑60.1%)**	Baseline: 271 6mo: 378 (↑35%)
Incidence of critical cardiovascular events (% of patients)	4.8	8.9

a Defined as an improvement from baseline of >50% in the treadmill exercise pain-free walking distance at the 6-month test and in ≥1 earlier test, in the absence of critical cardiovascular events.

b Improvement from baseline of >50% in the treadmill exercise pain-free walking distance at the 6-month test.

* $p < 0.05$, ** $p < 0.005$, *** $p \leq 0.001$ *vs* placebo.

In general, patients in the beraprost group were more likely to be satisfied with global changes in their quality of life than patients in the placebo group ($p < 0.05$). The difference was also statistically significant ($p < 0.05$) in favour of patients receiving beraprost when analysing individual items such as 'going out', 'general condition', 'relationships with people' and 'concerns about health'.[92] However, it is important to note that at baseline, there were statistically significant differences between the beraprost and placebo treatment groups regarding lifestyle modification prescriptions (57.9 *vs* 47.4%, $p = 0.032$) and level of physical activity (higher among patients receiving beraprost, $p = 0.030$). These two factors may have influenced the final results of this study.[92] In this context, it is important to note, that although recent preliminary unpublished data from a large ($n \approx 750$), phase III, placebo-controlled study conducted in the US suggested a trend toward fewer cardiovascular events (no specific data presented), this study did not confirm the positive results of the European phase III trial and statistical significance was not achieved in the study's endpoints relating to exercise.[96]

5.1.3 Noncomparative Trials

Overall, the data from noncomparative trials seem to indicate that beraprost treatment may be of benefit in patients with peripheral arterial diseases represented in most studies by patients with Buerger's disease and arteriosclerosis obliterans. However, these trials were not randomised, did not include an active comparator or placebo treatment, included relatively small numbers of patients (23 to 70) and few objective parameters.

Reductions from baseline of 37 to 53% in ischaemic ulcer size were reported in several studies.[81,82,87] Additionally, most patients (>60%) receiving beraprost treatment showed at least 'mild improvement' in the size of the ulcers.[81,82] The largest study (n = 70) reported reduction in ulcer size from 7.5 at baseline to 3.5mm after at least 6 weeks of treatment (mean values, $p < 0.05$). Improvement in the appearance of ulcer granulation was also reported in a high proportion of patients receiving beraprost (61 to 82%).[81,82] For example, the largest study reported that at baseline the appearance of granulation was 'slightly bad' (66.7%) or 'bad' (33.3%) in all patients with ischaemic ulcers (n = 16). However, at treatment endpoint (week 6) the appearance of granulation in a high proportion of patients was 'cured' (37.5%), 'good' (12.5%) or 'slightly good' (18.8%).[82] Similar improvement rates have been reported in other clinical trials.[81,87] Also, two studies reported increases (222 and 95%) in claudication distance from 163 and 468m (at baseline) to 526 and 914m (treatment endpoint), respectively, after 6 and 8 weeks of beraprost treatment.[82,85]

Evaluation of subjective symptoms also appears to show a positive effect of beraprost treatment in patients with peripheral arterial disease. Compared with baseline, the proportion of patients reporting pain at rest, coldness in the extremities, numbness and cyanosis decreased after several weeks of beraprost treatment.[81,82,84-87] For example, at baseline 57.9, 36, 39.4 and 39.1% of the patients in the

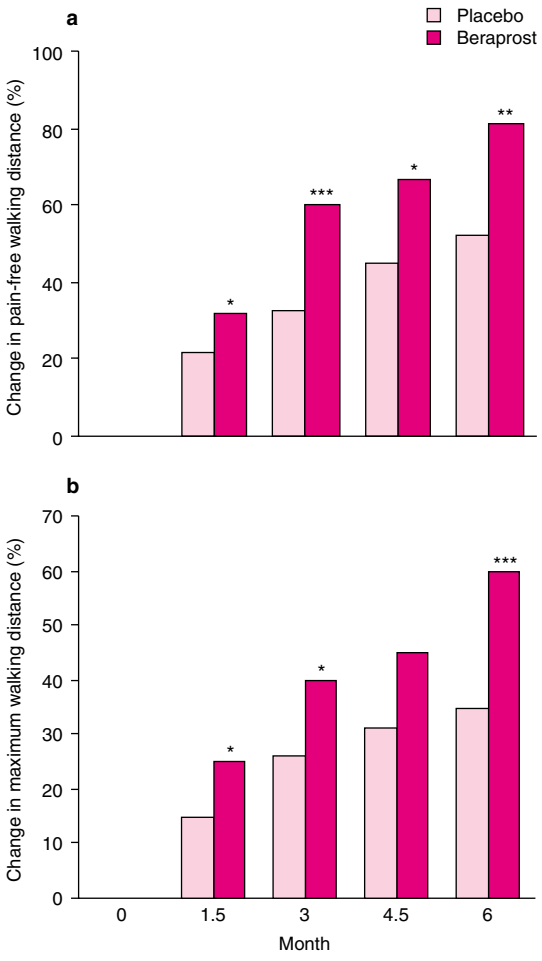

Fig. 4. Effects of beraprost on walking distance. Main results from a large (n = 422) double-blind, randomised, placebo-controlled clinical trial of patients with intermittent claudication who received beraprost 40μg three times a day or placebo for 6 months.[92] Efficacy was evaluated at 0, 1.5, 3, 4.5 and 6 (treatment endpoint) months as percentage change from baseline of **(a)** pain-free walking distance (claudication distance) and **(b)** maximum walking distance. * $p < 0.05$, ** $p < 0.01$, *** $p \leq 0.001$ *vs* placebo.

largest study exhibited 'no symptom' or 'mild symptom' regarding pain at rest (n = 38), coldness (n = 50), numbness (n = 32) and cyanosis (n = 23), respectively.[82] After 6 weeks of beraprost 40μg three times a day the proportion of patients reporting 'no symptom' or 'mild symptom' increased to 68.4, 70, 87.5 and 73.9%, respectively ($p < 0.001$ for all symptoms).[82] Similar improvement was reported in other clinical trials.[81,84-87]

Final global improvement ratings (markedly improved, moderately improved, slightly improved, unchanged and worsened), which included evaluation of objective and subjective symptoms, also suggested that patients may benefit from beraprost treatment.[82,84-87] The response to beraprost treatment of at least 27% (27 to 70%) of patients was evaluated as 'moderately improved' or better. Response of 'slightly improved' or better was recorded in at least 68.6 to 90.9% of the patients.

5.2 In PAH

The efficacy of various dosages of beraprost in the treatment of patients with moderate to severe PPH (New York Heart Association (NYHA) functional class III to IV) has been evaluated in several small noncomparative trials (n = 11 to 12)[70,72,97,98] and one retrospective comparison with historical controls (n = 58)[99] [table IV]. Several trials also included patients with other types of PAH.[70,97,98]

As all trials were reported as abstracts or short communications, study and patient details were few and statistics were not always provided. The studies evaluated cardiovascular and exercise parameters, and effect on NYHA functional class after 1 to 12 months' treatment. The retrospective study followed patients treated with beraprost for a mean of 30 months and calculated survival rates using Kaplan-Meir curves for both the beraprost group and the historical controls who were given usual therapy (calcium antagonists, nitrates, digitalis and diuretics).[99]

Available data from these small trials are insufficient to draw any firm conclusions regarding the efficacy of beraprost in PAH. However, there is some evidence that the drug may be beneficial in this condition. Pulmonary arterial pressure and pulmonary vascular resistance decreased during beraprost treatment; in some, but not all, instances these decreases were significant (table IV). Car-

Table IV. Efficacy of beraprost in patients with moderate to severe pulmonary arterial hypertension (NYHA functional class III to IV)

Reference	No. of evaluable patients	Dosage regimen (duration)	Effects on cardiovascular/exercise parameters (change from baseline)				NYHA class
			PAP (mm Hg)	PVR (μ)	CO (L/min)	exercise	
Noncomparative prospective studies							
Nakayama et al.[70]a	11[b]	20-40μg tid (1-2mo)	–22.5 (32%)[c]	–5.8 (29%)[c]	+0.5 (17%)[c]		
Okano et al.[72]a	12[d] (severe)	80-180 μg/day (average 2mo)	–8 (12%)	–5 (26%)			Improved in 8/12 pts (67%)
Shimizu et al.[97]a	14[e] (severe)	Average 96 μg/day (3mo)				Maximal load ↑12watts (14%)** peak VO_2 ↑115 ml/min (12%)*	
Vizza et al.[98]a	12[f]	100-360 μg/kg in 3-4 doses/day + usual therapy (12 mo)	–7 (8%) NS			6MWT: ↑102m (48%)***	↓ from 3.5 to 2.6***
Retrospective comparison with historical controls							
Nagaya et al.[99]	24[g]	60-180 μg/day + usual therapy (mean follow-up 30mo)	–13%*** (at 53d)	–25%*** (at 53d)	+17%** (at 53d)		Improved in 16/24 pts (67%)

a Abstract[70,97,98] or short communication.[72]

b Included patients with moderate to severe PPH or inoperable thromboembolic pulmonary hypertension.

c Difference from baseline stated to be significant but no p-values given.

d Included patients with PPH unresponsive to calcium antagonists and inhaled nitric oxide.

e Included patients with PPH (6) and thromboembolic pulmonary hypertension (8).

f Included patients with PPH (7), chronic pulmonary thromboembolism (4) and Eisenmenger's syndrome. These patients received beraprost in adjunction with oral anticoagulants, digoxin, furosemide, enalapril and/or nifedipine.

g 34 historical controls received usual therapy only (calcium antagonists, nitrates, digitalis and diuretics); no data were provided for this group.

6MWT = 6-minute walk test; **CO =** cardiac output; **NS =** not significant; **NYHA =** New York Heart Association; **PAP =** pulmonary arterial pressure; **PPH =** primary pulmonary hypertension; **PVR =** pulmonary vascular resistance; **tid =** three times a day; ↑ = increased; ↓ = decreased; * $p < 0.05$, ** $p < 0.01$, *** $p < 0.001$ *vs* baseline.

diac output increased significantly in both studies measuring this parameter. NYHA functional class also improved in 67% of patients in one study[72] and significantly decreased (improved) in another.[98]

Beraprost therapy [1 to 3 μg/kg daily or highest dose tolerated (96μg daily)] may also be associated with improvement in the exercise capacity of patients with PAH. Significant increases from baseline values of total distance in a 6-minute walk test, maximal load and peak VO_2 were reported in patients receiving beraprost treatment for 3[97] or 12[98] months (table IV).

Beraprost treatment appeared to be associated with greater 1-, 2- and 3-year survival rates (96, 86 and 76%, respectively) than conventional therapy (77, 47 and 44%) in the retrospective study. During a mean follow-up period of 30 ± 20 months, 4 of 24 patients from the beraprost treatment group died of cardiopulmonary causes compared with 27 of 34 in the conventional therapy treatment group (44 ± 45 months follow-up period).[99] It is important to note, however, that the time point for the enrolment and the follow-up period differed between the two treatment groups, which might bias the results of this study,[99] and the trial was not designed as a mortality study.

6. Tolerability

Tolerability data from clinical trials and unpublished studies indicate that in general beraprost is a well tolerated agent.[92,93,100,101] Data in this section are derived from a combined analysis of patients with peripheral arterial disease (n = 7515) or PAH (n = 40) presented in the manufacturer's prescribing information[100] and from individual clinical trials.

6.1 Peripheral Arterial Disease

Adverse events were reported in 370 of the 7515 patients (4.9%) in the combined analysis.[100] The most commonly reported adverse events were vasodilatory (headache, facial hot flushes, hot flushes) or gastrointestinal (diarrhoea and nausea), and each occurred in ≤1.2% of patients (figure. 5a). Drug related rash was noted rarely.[91] Laboratory abnormalities were also reported in some studies.[81,82,91,94] No significant differences were observed in the overall incidence of adverse events in patients from the beraprost (15.6%) and ticlopidine (24.4%) treatment groups. A comparison of beraprost (n = 77) and ticlopidine (n = 86) treatment reported abnormal hepatic function (one patient), abnormal lipid levels (three patients) and decreased erythrocyte levels (one patient) among patients receiving beraprost.[91] However, as in other studies,[81,82,94] these symptoms were mild, disappeared during or after completion of beraprost therapy, did not require treatment and did not cause treatment withdrawals.[91]

The main tolerability data in patients with intermittent claudication receiving beraprost treatment are from the randomised, double-blind, placebo-controlled clinical trial described in section 5.1.2 (figure. 5b).[92] Adverse events (including critical cardiovascular events) considered to be drug-related were recorded in 16.7 and 7% of patients from the beraprost (n = 209) and placebo (n = 213) treatment groups, respectively. Additionally, 18 (8.6%, beraprost) and 31 patients (14.5%, placebo) discontinued treatment because of adverse events. The most common adverse events in the beraprost group were headache (6.2%) and vasodilation (5.3%).[92]

6.2 PAH

According to the limited data available, the incidence of adverse events in patients with PAH receiving beraprost appears considerably higher than in patients with peripheral arterial disease. Combined data from clinical studies in patients with PAH receiving beraprost treatment indicated occurrence of adverse events (including laboratory abnormalities) in 24 of 40 patients (60%, total of 65 events).[100] The most frequent adverse events were headache (22.5%), increased lactate dehydrogenase (12.5%), increased bilirubin levels (10%), hot flushes (7.5%), diarrhoea (7.5%), nausea (7.5%) and increased triglycerides levels (7.5%) [figure. 5c]. Combined data are not available on the severity of these symptoms and their effect on compliance with beraprost treatment. However, in 12 patients receiving beraprost and adjunctive standard therapy for up to 12 months no major beraprost-related adverse events were reported.[98]

7. Dosage and Administration

The efficacy of beraprost has been evaluated for the treatment of numerous indications related to peripheral vascular disorders (see section 5). Beraprost has been approved for use in patients with PAH and peripheral arterial disease in Japan (1992), South Korea (1997) The Philippines (2001, only peripheral arterial disease) and Thailand (2001). Currently, beraprost is being considered for the treatment of patients with intermittent claudication and PAH in the US and Europe.

Beraprost is administered orally and should be taken after meals. In adult patients with peripheral arterial disease the recommended dosage of beraprost is 120μg daily divided in three doses. For adults with PAH beraprost treatment should be started at 60μg divided in three doses. For these patients beraprost dosage can be increased gradually under careful monitoring up to a maximum dosage of 180μg daily divided in three to four doses.[100] However, in patients with PAH included in the recent 12-week ALPHABET trial, the dosage was increased from 20μg four times a day up

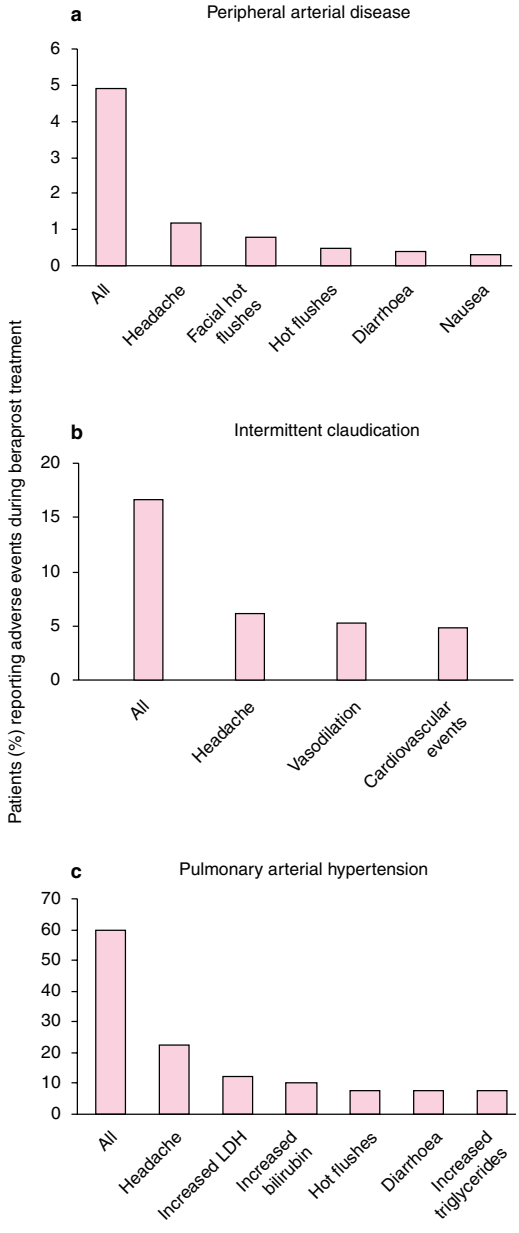

Fig. 5. Combined data of the most frequently reported adverse events (including laboratory abnormalities) during beraprost therapy among patients with **(a)** peripheral arterial disease (n = 7515),[100] including **(b)** intermittent claudication[92] (n = 422) or **(c)** pulmonary arterial hypertension (n = 40).[100] Data from the large pivotal, randomised, double-blind study[92] (section 5.1.2) in patients with intermittent claudication presented in figure 5b may also be included in the data presented in figure 5a. **LDH =** lactate dehydrogenase.

to a mean and maximum dosage of 80 and 120μg four times a day, respectively.[102]

The manufacturer recommends exercising caution when prescribing beraprost in the elderly. The safety profile of beraprost in pregnant women has not been established so it should be administered only when therapeutic benefits outweigh the risk of beraprost treatment. Beraprost is contraindicated in nursing women (beraprost is secreted in milk of rats) and its safety has not been established in children.[100]

Beraprost should be administered with care in patients receiving anticoagulant (e.g. warfarin), antiplatelet (e.g. aspirin, ticlopidine) or fibrinolytic (e.g. urokinase) agents, since it may intensify the effect of these drugs. As beraprost may increase bleeding tendency, caution is recommended in menstruating patients or those with a bleeding tendency.[100] Also, beraprost in combination with other PGI_2 preparations may further reduce blood pressure.

8. Place of Beraprost in the Management of Peripheral Arterial Diseases and PAH

8.1 Peripheral Arterial Disease

Primary prevention of peripheral arterial disease, estimated to have an age-adjusted prevalence of approximately 12%,[20] consists of modifying lifestyle factors (i.e smoking) and managing comorbid conditions such as diabetes mellitus and hypertension[19,20,27] to reduce the risk of cardiovascular morbidity (see section 2).[27]

Pharmacological agents for the treatment of patients with peripheral arterial diseases are numerous and their use has been recently reviewed.[27,103] These include antiplatelet, vasodilator and anticoagulant agents. However, the efficacy of many of these agents is questionable and current data do not support their widespread use.[27,103] PGI_2 and iloprost have shown efficacy when given intravenously but this route of administration can be problematic and costly (section 2). Although the American College of Chest Physicians recommends aspirin 81 to 325 mg daily for the treatment of pa-

tients with peripheral arterial disease, a US Food and Drug Administration (FDA) panel of experts did not find sufficient evidence to approve its use for this indication.[27] Ticlopidine and clopidogrel may reduce the severity of claudication and the need for vascular surgery, but ticlopidine and rarely clopidogrel also are associated with risk of thrombocytopenia, neutropenia and thrombotic thrombocytopenic purpura. Pentoxifylline use in patients with intermittent claudication (approved by the FDA) has also been questioned, since several studies have not found significant differences between pentoxifylline treatment and placebo.[27,103] Moreover, a recent evaluation of drugs approved in Belgium for the treatment of patients with intermittent claudication concluded that there was scarce evidence for the use of cinnarizine, cyclandelate, isoxsuprine, naftidrofuryl, pentoxifylline, xanthinol nicotinate and buflomedil.[103] In patients with intermittent claudication, the type III phosphodiesterase inhibitor cilostazol in dosages of 100mg twice a day was significantly superior to placebo and pentoxifylline treatment, although this drug is not recommended in patients with heart failure.[27,104-106]

Beraprost, a stable oral analogue of PGI_2, appears to be efficacious in the treatment of patients with Buerger's disease and arteriosclerosis obliterans. Among these patients, beraprost was as efficacious as ticlopidine.[91] In general, the mechanism of action of beraprost in these indications is not fully understood and is probably the result of the simultaneous action of different pharmacodynamic characteristics of the drug (section 3).

Data from the large 6-month pivotal trial discussed in section 5.1.2 appeared to indicate that beraprost treatment (40μg three times a day for 6 months) improved the condition of patients with intermittent claudication. Compared with patients receiving placebo, patients in the beraprost treatment group exhibited significantly greater improvement in pain-free and absolute walking distance. Additionally, patients receiving beraprost were significantly more likely than placebo recipients to report satisfaction with changes in their quality of life (see section 5.1.2).[92] However, while preliminary unpublished data from a large ($n \approx 750$), placebo-controlled, multicentre study, with similar entry criteria and study endpoints suggested a trend toward fewer critical cardiovascular events (no specific data presented), this study did not confirm the positive results of the European phase III trial and statistical significance was not achieved in the study's endpoints relating to exercise.[96]

Evaluation of ulcers among patients with Buerger's disease or arteriosclerosis obliterans receiving beraprost treatment (40μg three times a day) in the 6-week comparison with ticlopidine indicated that 70.3% of patients showed 'reduced' or better response to treatment. Compared with those receiving ticlopidine treatment, a greater proportion of patients receiving beraprost exhibited 'moderately improved' or better granulation appearance of the tissue (67.1 *vs* 50.6%, $p < 0.05$). Similar improvement rates were reported in several noncomparative studies of 4 to 8 weeks' duration discussed in section 5.1.3. Additionally, adverse events due to beraprost treatment among patients with peripheral arterial disease were uncommon, and all laboratory abnormalities observed were mild and disappeared during or after termination of beraprost treatment.

There are, however, several shortcomings concerning the study of patients with peripheral arterial diseases. The diagnostic criteria of some peripheral arterial diseases vary in different parts of the world, which hinders the correct evaluation of their prevalence worldwide. For example, Buerger's disease appears to be more prevalent in the Middle East and Far East than in North America and western Europe, but these differences are likely to be influenced by differences in diagnostic criteria.[23] Also, clinical efficacy is often evaluated using a wide range of subjective parameters (section 5), which makes comparison across different clinical trials inaccurate. Moreover, in patients with intermittent claudication, changes in exercise performance are not always evaluated using the graded treadmill test (constant speed of 3.2 km/h with increases of grade of 2 to 3% every 2 or 3 minutes). Unlike the constant-load treadmill test

(traditional test, 2.4 to 3.2 km/h at a defined grade of 0 to 12%), results using the graded treadmill test correlate with improved patient ambulatory function, and small percentage improvements are more likely to be clinically relevant.[107]

At present, however, there are few well designed clinical trials in the literature that included patients with peripheral arterial diseases who received beraprost therapy, and most studies were of short duration. Also, direct comparison of beraprost treatment with patients undergoing supervised exercise therapy and lifestyle changes or with patients receiving cilostazol have not been reported.

8.2 PAH

PAH is a progressive disease that until recently was considered universally fatal, with transplantation being the only option for patients with this condition.[29,43] Currently, the treatment of PAH is complex, controversial and potentially dangerous, but recent advances and refinements in the use of pharmacological agents (vasodilators in conjunction with anticoagulant agents; see section 2.2) and PGI_2 in particular, have markedly improved the quality of life and extended survival of patients with PAH.[14,29,43,102,108] For example, compared with baseline values, long-term (>12 months) aerosolised iloprost treatment of patients with PAH increased the distance in the 6-minute walk test and cardiac output, and reduced mean pulmonary arterial pressure and resistance.[109] Improvement of these parameters were also observed after administration of subcutaneous treprostinil (a PGI_2 analogue) for 12 weeks[110] and oral bosentan (endothelin-1 antagonist) for 8 weeks.[102,111]

Continuous intravenous delivery of PGI_2 is currently considered the most efficacious treatment of patients with severe forms of PAH (NYHA class III or IV) who do not respond to conventional treatment (calcium antagonists).[112-116] Acute challenge with PGI_2 induces a dose-related decrease in pulmonary artery pressure and an increase of cardiac output and systemic oxygen delivery in a substantial proportion of patients with PAH (≈50%). Additionally, patients who do not show these haemodynamic improvements after acute administration can respond to long-term intravenous administration of PGI_2.[41,116-118] Long-term therapy with PGI_2 has shown sustained clinical benefits and improved long-term survival in patients with PAH.[112-115,119]

There are, however, limitations to PGI_2 therapy. Because of its chemical instability (half-life of 3 to 5 minutes), PGI_2 must be administered by continuous intravenous infusion. Infection, catheter thrombosis and pump failure are associated with this method of delivery. Particularly, interruption of the infusion or underdosing of PGI_2 may induce acute return of the symptoms that can be life-threatening.[33,120] Additionally, the high cost of long-term PGI_2 therapy (cost of the drug, the ambulatory pump and intravenous lines and supplies) must be considered.[120,121] Because of the severity and rarity of PAH, large, randomised, double-blind and placebo-controlled trials evaluating the long-term use of intravenous prostacyclin have not been conducted.[43,120]

Beraprost is a more stable PGI_2 analogue with a half-life in healthy adults of 0.9 to 1.1 hours.[66] This added stability is of critical importance because allows for oral administration of the drug. It has been suggested that intermittent doses of beraprost do not provide consistent levels of the drug in the bloodstream needed to treat the most critically ill patients with PAH.[122] Therefore, development of a slow-release formulation of beraprost may improve its efficacy.

The strongest evidence of the efficacy of beraprost in the treatment of patients with PAH comes from a long-term noncomparative study of 12 consecutive patients with severe PPH (see section 5.2). After 2 months of beraprost treatment pulmonary arterial pressure and resistance decreased from baseline values (12 and 26%, respectively). Improvement in NYHA functional class was also recorded in most patients (9 of 12) and after 12 to 18 months receiving the same dose of beraprost 8 patients were still alive.[72] These results are promising, since the patients included in this study previously did not respond to calcium antagonists or

inhaled nitric oxide. Other studies appeared to indicate that beraprost may have beneficial effect on the survival[99] (60 to 180 μg/day) and exercise capacity[97,98] (96 μg/day) of patients with PAH.

Based on this evidence it seems that beraprost treatment may be of use for patients with PAH before starting intravenous PGI_2 therapy or patients with less severe forms of the disease (not NYHA functional class IV), who may respond to long-term beraprost treatment. Indeed, a recent randomised, double-blind, placebo-controlled multicentre study, the ALPHABET trial, included patients (n = 130) with PAH of NYHA class II and III who received beraprost for 12 weeks. This is the largest study of patients with PAH, who received beraprost doses much higher than those recommended by the manufactures, but its results have not been published yet (see section 7).

However, PAH clinical trials included small number of patients, lacked patient details and often did not provide relevant statistical data (see section 5.2). Also, studies directly comparing the efficacy of oral beraprost with that of intravenous PGI_2 treatment are necessary to define the place of beraprost in the treatment of patients with PAH.

8.3 Conclusions

There are two factors that confound the precise definition of the place of beraprost in clinical settings. Firstly, the mode of action of beraprost (and generally PGI_2 and its analogues) in peripheral arterial diseases and PAH is not fully understood; hence establishment of the correct dosage has been difficult, particularly in patients with PAH. Secondly, there is a limited amount of large, well designed studies in the literature evaluating the clinical efficacy of beraprost in patients with these conditions.

Beraprost, an orally administered PGI_2 analogue, is generally well tolerated and appears to be an effective agent in the treatment of patients with Buerger's disease and arteriosclerosis obliterans. Comparative data from a large randomised trial indicated that the drug appears as effective as ticlopidine in patients with these conditions. In patients with intermittent claudication, significant benefits of beraprost compared with placebo were reported in a randomised clinical trial; however, the use of beraprost in these patients is not supported by recent preliminary unpublished data from a large, phase III, placebo-controlled study. Limited data suggest some efficacy with long-term beraprost treatment of patients with PAH, where options are few and where oral administration of the drug could be a considerable advantage over intravenous PGI_2 therapy. Additional well designed and, where possible, large trials with active comparators are necessary to define more precisely the place of beraprost in the treatment of patients with PAH, Buerger's disease and arteriosclerosis obliterans.

References

1. Vane JR, Botting RM. Pharmacodynamic profile of prostacyclin. Am J Cardiol 1995; 75 (3): 3A-10A
2. MacIntyre DE, Pearson JD, Gordon JL. Localisation and stimulation of prostacyclin production in vascular cells. Nature 1978 Feb 9; 271: 549-51
3. Weksler BB, Marcus AJ, Jaffe EA. Synthesis of prostaglandin I2 (prostacyclin) by cultured human and bovine endothelial cells. Proc Natl Acad Sci U S A 1977 Sep; 74 (9): 3922-6
4. Moncada S, Gryglewski R, Bunting S, et al. An enzyme isolated from arteries transforms prostaglandin endoperoxides to an unstable substance that inhibits platelet aggregation. Nature 1976 Oct 21; 263: 663-5
5. Ubatuba FB, Moncada S, Vane JR. The effect of prostacyclin (PGI_2) on platelet behaviour. Thrombus formation *in vivo* and bleeding time. Thromb Haemost 1979; 41: 425-35
6. Szczeklik A, Nizankowski R, Skawinski S, et al. Successful therapy of advanced arteriosclerosis obliterans with prostacyclin. Lancet 1979 May 26: 1111-4
7. Graf H. Endothelial control of cell migration and proliferation. Eur Heart J 1993; 14 Suppl. 1: 183-6
8. Prins BA, Hu R-M, Nazario B, et al. Prostaglandin E_2 and prostacyclin inhibit the production and secretion of endothelin from cultured endothelial cells. J Biol Chem 1994 Apr 22; 269: 11938-44
9. Higenbottam T. The place of prostacyclin in the clinical management of primary pulmonary hypertension. Am Rev Respir Dis 1987 Sep; 136 (3): 782-5
10. Long WA, Rubin LJ. Prostacyclin and PGE_1 treatment of pulmonary hypertension. Am Rev Respir Dis 1987 Sep; 136 (3): 773-6
11. Virgolini I, Fitscha P, Linet OI, et al. A double-blind placebo controlled trial of intravenous prostacyclin (PGI2) in 108 patients with ischaemic peripheral vascular disease. Prostaglandins 1990; 39 (6): 657-65

12. Dowd PM, Martin MFR, Cooke ED, et al. Treatment of Raynaud's phenomenon by intravenous infusion of prostacyclin (PGI_2). Br J Dermatol 1982; 106: 81-9
13. GlaxoWellcome. Epoprostenol prescribing information [online]. Available from URL: http://www.flolan-center.com/pages/flolan [Accessed 2001 Mar]
14. Herner SJ, Mauro LS. Epoprostenol in primary pulmonary hypertension. Ann Pharmacother 1999 Mar; 33: 340-7
15. Fiessinger JN, Schafer M. Trial of iloprost versus aspirin treatment for critical limb ischaemia of thromboangiitis obliterans. TAO Study. Lancet 1990 Mar 10; 335: 555-7
16. Grant SM, Goa KL. Iloprost: a review of its pharmacodynamic and pharmacokinetic properties, and therapeutic potential in peripheral vascular disease, myocardial ischaemia and extracorporeal circulation procedures. Drugs 1992; 43 (6): 889-924
17. European TAO Study Group. Oral iloprost in the treatment of thromboangiitis obliterans (Buerger's disease): a double-blind, randomised, placebo-controlled trial. Eur J Vasc Endovasc Surg 1998 Apr; 15: 300-7
18. Toda N. Beraprost sodium. Cardiovasc Drug Rev 1988; 6 (3): 222-38
19. Weitz JI, Byrne J, Clagett P, et al. Diagnosis and treatment of chronic arterial insufficiency of the lower extremities: a critical review. Circulation 1996 Dec 1; 94: 3026-49
20. Caspary L. Epidemiology of vascular disease. Dis Manage Health Outcomes 1997; 2 Suppl. 1: 9-17
21. Haustein KO. State of the art – treatment of peripheral occlusive arterial disease (POAD) with drugs vs. vascular reconstruction or amputation. Int J Clin Pharmacol Ther 1997; 35 (7): 266-74
22. McNamara DB, Champion HC, Kadowitz PJ. Pharmacologic management of peripheral vascular disease. Surg Clin North Am 1998; 78 (3): 447-64
23. Olin JW. Thromboangiitis obliterans (Buerger's disease). N Engl J Med 2000 Sep 21; 343 (12): 864-9
24. Block JA, Sequeira W. Raynaud's phenomenom. Lancet 2001 Jun 23; 357 (9273): 2042-8
25. Belch JJF. Prostaglandins in Raynaud's phenomenon. In: Vane JR, O'Grady J, editors. Therapeutic applications of prostaglandins. London: Edward Arnold, 1993: 219-39
26. Imparato AM, Kim G-E, Davidson T, et al. Intermittent claudication: its natural course. Surgery 1975; 78: 795-9
27. Hiatt WR. Medical treatment of peripheral arterial disease and claudication. N Engl J Med 2001 May 24; 344 (21): 1608-21
28. Reunanen A, Takkunen H, Aromaa A, et al. Prevalence of intermittent claudication and its effect on mortality. Acta Med Scand 1982; 211: 249-56
29. Wanstall JC, Jeffery TK. Recognition and management of pulmonary hypertension. Drugs 1998 Dec; 56: 989-1007
30. Jelnes R, Gaardsting O, Hougaard JK, et al. Fate in intermittent claudication: outcome and risk factors. BMJ 1986; 293 (Clin Res Ed): 1137-40
31. Gaine SP, Rubin LJ. Primary pulmonary hypertension. Lancet 1998 Aug 29; 352: 719-25
32. Blythe D, van HPV, Power BM. Pulmonary hypertension and selective pulmonary vasodilators in acute lung injury. Anaesth Intensive Care 1998 Feb; 26: 26-39
33. Rubin LJ. Primary pulmonary hypertension. N Engl J Med 1997 Jan 9; 336: 111-7
34. WHO. World Symposium on Primary Pulmonary Hypertension 1998 [online]. Available from URL: http://www.who.int/ncd/cvd/pph.html [Accessed 2001 Nov]
35. Edwards BS, Weir EK, Edwards WD, et al. Coexistent pulmonary and portal hypertension: morphologic and clinical features. J Am Coll Cardiol 1987 Dec; 10 (6): 1233-8
36. Loyd JE, Primm RK, Newman JH. Familial primary pulmonary hypertension: clinical patterns. Am Rev Respir Dis 1984; 129: 194-7
37. Thomson JR, Machado RD, Pauciulo MW, et al. Sporadic primary pulmonary hypertension is associated with germline mutations of the gene encoding BMPR-II, a receptor member of the TGF-B family. J Med Genet 2000; 37: 741-5
38. Machado RD, Pauciulo MW, Thomson JR, et al. BMPR2 haploinsufficiency as the inherited molecular mechanism for primary pulmonary hypertension. Am J Hum Genet 2001; 68: 92-102
39. Deng Z, Morse JH, Slager SL, et al. Familial primary pulmonary hypertension (gene PPH1) is caused by mutations in the bone morphogenetic protein receptor-II gene. Am J Hum Genet 2000; 67: 737-44
40. Abenhaim L, Moride Y, Brenot F, et al. Appetite-suppressant drugs and the risk of primary pulmonary hypertension. N Engl J Med 1996 Aug 29; 335 (9): 609-16
41. Ricciardi MJ, Rubenfire M. How to manage primary pulmonary hypertension; giving hope to patients with a life-threatening illness. Postgrad Med 1999 Mar; 105 (3): 45-56
42. McDonnell PJ, Toye PA, Hutchins GM. Primary pulmonary hypertension and cirrhosis: are they related? Am Rev Respir Dis 1983; 127: 437-41
43. Brij S, Peacock AJ. Pulmonary hypertension: its assessment and treatment. Thorax 1999; 54 Suppl. 2: S28-32
44. Takeo S. Pharmacodynamics and clinical studies with beraprost sodium, a drug for peripheral vascular disease. Cardiovasc Drug Rev 1993; 10 (4): 392-403
45. Murata T, Murai T, Kanai T, et al. General pharmacology of beraprost sodium. 2nd communication: effect on the autonomic, cardiovascular and gastrointestinal systems, and other effects. Arzneimittelforschung 1989 Aug; 39: 867-76
46. Murata T, Sakaya S, Hoshino T, et al. General pharmacology of beraprost sodium. 1st communication: effect on the central nervous system. Arzneimittelforschung 1989 Aug; 39: 860-6
47. Ueno Y, Kawashima A, Koike H, et al. Effect of beraprost sodium, a stable prostacyclin analogue, on pulmonary thromboembolism in mice. Thromb Res 1995 Jan 15; 77: 193-8
48. Okada Y, Marchevsky AM, Kass RM, et al. A stable prostacyclin analog, beraprost sodium, attenuates platelet accumulation and preservation-reperfusion injury of isografts in a rat model of lung transplantation. Transplantation 1998 Nov 15; 66: 1132-6
49. Umetsu T, Murata T, Nishio S. Studies on the antiplatelet effect of the stable epoprostenol analogue beraprost sodium and its mechanism of action in rats. Arzneimittelforschung 1989 Jan; 39: 68-73
50. Hirasawa Y, Nishio M, Maeda K, et al. Comparison of antiplatelet effects of FK409, a spontaneous nitric oxide releaser, with those of TRK-100, a prostacyclin analogue. Eur J Pharmacol 1995 Jan 5; 272: 39-43

51. Nishio S, Matsuura H, Kanai N, et al. The *in vitro* and *ex vivo* antiplatelet effect of TRK-100, a stable prostacyclin analog, in several species. Jpn J Pharmacol 1988 May; 47: 1-10
52. Yang L, Yatomi Y, Satoh K, et al. Inhibitory effects of beraprost on platelet aggregation: comparative study utilizing two methods of aggregometry. Thromb Res 1999 Apr 1; 94: 25-32
53. Kim D-I, Kambayashi J, Shibuya T, et al. Suppression of pseudointimal hyperplasia by a novel prostacyclin analogue: beraprost. Surg Today 1995; 25: 722-8
54. Akiba T, Miyazaki M, Toda N. Vasodilator actions of TRK-100, a new prostaglandin I2 analogue. Br J Pharmacol 1986 Dec; 89: 703-11
55. Matsumoto T, Wada A, Tsutamoto T, et al. Vasorelaxing effects of atrial and brain natriuretic peptides on coronary circulation in heart failure. Am J Physiol 1999 Jun; 276 (6 Pt 2): H1935-42
56. Alexandrou K, Hata Y, Matsuka K, et al. The effect of washout perfusion with beraprost sodium on the no-reflow phenomenon in the rat island flap. Eur J Plast Surg 1996; 19 (2): 73-6
57. Murai T, Muraoka K, Saga K, et al. Effect of beraprost sodium on peripheral circulation insufficiency in rats and rabbits. Arzneimittelforschung 1989 Aug; 39: 856-9
58. Yamada N, Isogaya M, Ueno Y, et al. Synergic effect of beraprost sodium, a PGI2 analogue, and aspirin on canine carotid artery thrombosis model induced by electrical stimulation [abstract]. Thromb Haemost 1993 Jun 30; 69: 591
59. Hirano T, Yamori Y, Kanai N, et al. The effects of beraprost Na, a stable prostacyclin analog, on animal models of stroke. Mol Chem Neuropathol 1992 Aug; 17: 91-102
60. Ueno Y, Koike H, Annoh S, et al. Anti-inflammatory effects of beraprost sodium, a stable analogue of PGI2, and its mechanisms. Prostaglandins 1997 Apr; 53: 279-89
61. Hayashi J, Ishida N, Sato H, et al. Effect of beraprost, a stable prostacyclin analogue, on red blood cell deformability impairment in the presence of hypercholesterolemia in rabbits. J Cardiovasc Pharmacol 1996 Apr; 27: 527-31
62. Okuyama M, Kambayashi J, Sakon M, et al. PGI2 analogue, sodium beraprost, suppresses superoxide generation in human neutrophils by inhibiting p47phox phosphorylation. Life Sci 1995; 57: 1051-9
63. Miyata M, Ueno Y, Sekine H, et al. Protective effect of beraprost sodium, a stable prostacyclin analogue, in development of monocrotaline-induced pulmonary hypertension. J Cardiovasc Pharmacol 1996 Jan; 27: 20-6
64. Kainoh M, Nishio S, Nakadate T. Cytoprotective action of beraprost sodium against peroxide-induced damage in vascular endothelial cells. Pharmacology 1992; 45: 61-70
65. Kainoh M, Imai R, Umetsu T, et al. Prostacyclin and beraprost sodium as suppressors of activated rat polymorphonuclear leukocytes. Biochem Pharmacol 1990 Feb 1; 39: 477-84
66. Kato R, Uji Y, Matsumoto K. Phase I study of beraprost sodium (TRK-100), a prostacyclin derivative – repeated oral administration study for 10 days [in Japanese]. Jpn J Clin Pharmacol Ther 1989; 20 (3): 529-39
67. Nony P, Ffrench P, Girard P, et al. Platelet-aggregation inhibition and hemodynamic effects of beraprost sodium, a new oral prostacyclin derivative: a study in healthy male subjects. Can J Physiol Pharmacol 1996 Aug; 74: 887-93
68. Cunningham MR, Yamada N, Kumagai H, et al. Prevention of experimental coronary artery thrombosis – effects of beraprost [abstract]. Circulation 1991 Oct; 84 Suppl. 2: II-599
69. Kurisu Y, Orihashi K, Sueda T, et al. Protective effect of beraprost sodium, a stable prostacyclin analogue, on cardiac allograft vasculopathy in rats. Hiroshima J Med Sci 1997 Mar; 46: 11-9
70. Nakayama Y, Okano Y, Satoh T, et al. Beneficial effects of beraprost sodium in patients with pulmonary hypertension [abstract]. J Am Coll Cardiol 1995 Feb Spec Issue: 221A
71. Saji T, Ozawa Y, Ishikita T, et al. Short-term hemodynamic effect of a new oral PGI2 analogue, beraprost, in primary and secondary pulmonary hypertension. Am J Cardiol 1996 Jul 15; 78: 244-7
72. Okano Y, Yoshioka T, Shimouchi A, et al. Orally active prostacyclin analogue in primary pulmonary hypertension [letter] [published erratum appears in Lancet 1997 Nov 8; 350 (9088):1406]. Lancet 1997 May 10; 349: 1365
73. Alexandrou K, Hata Y, Matsuka K, et al. Effect of beraprost sodium (Procylin), a stable prostaglandin I2 analogue, on a dorsal skin flap model in rats. Scand J Plast Reconstr Hand Surg 1996 Mar; 30: 17-22
74. Tanaka H, Matsui S, Shigenobu K. Cardioprotective effects of beraprost sodium against experimental ischemia and reperfusion as compared with propranolol and diltiazem. Res Commun Mol Pathol Pharmacol 1998 Mar; 99: 321-7
75. Ishikawa M, Namiki A. The contractile mechanism of beraprost sodium, a stable prostacyclin analog, in the isolated canine femoral vein. Heart Vessels 1994; 9: 14-8
76. Hatane T, Yoshida E, Kawano J, et al. Prostaglandin I2 analog enhances the expression of urokinase-type plasminogen activator and wound healing in cultured human fibroblast. Biochim Biophys Acta 1998 Jun 22; 1403: 189-98
77. Imai-Sasaki R, Kainoh M, Ogawa Y, et al. Inhibition by beraprost sodium of thrombin-induced increase in endothelial macromolecular permeability. Prostaglandins Leukot Essent Fatty Acids 1995 Aug; 53: 103-8
78. Kajikawa N, Nogimori K, Murata T, et al. Specific binding of the new stable epoprostenol analogue beraprost sodium to prostacyclin receptors on human and rat platelets. Arzneimittelforschung 1989 Apr; 39: 495-9
79. Ishikawa J, Asano M, Ida H. Vasodilating action of beraprost sodium in anesthetized dogs. Gendai Iryo 1992; 24 (Spec.): 153-8
80. Terao G. Inhibitory effects of beraprost sodium on the progression of pulmonary hypertension induced by monocrotaline in rats: a comparison with aspirin, nifedipine, and enalapril. J Juzen Med Soc 1997; 106 (3): 311-8
81. Suh BY, Kwun KB, Kwun TW, et al. Open-label, uncontrolled, 6-week clinical trial of beraprost in patients with chronic occlusive arterial disease. Curr Ther Res Clin Exp 1998 Sep; 59: 645-53
82. Kondo J, Imada T, Adachi R, et al. Utility of beraprost sodium, an oral PGI2 analogue preparation, in patients with chronic arterial occlusive diseases [in Japanese]. J New Remedies Clinics 1996; 45 (9): 1822-32
83. Kondo H, Imada T, Adachi R, et al. Efficacy and safety of an oral PGI2 analogue, beraprost sodium, in patients with chronic arterial obliterans. J New Remedies Clinics 1996; 45 (9): 194-204

84. Katsumura T, Masaki H, Komoto Y, et al. Clinical evaluation of the oral form of PGI2 (beraprost sodium) in a multicentre trial for chronic arterial occlusive diseases. Shinyaku to Rinsho 1995; 44 (12): 2057-65
85. Sakuma M, Yasuda K, Matsui Y, et al. Clinical evaluation of beraprost sodium on chronic arterial occlusive disease [in Japanese]. Geriat Med 1995; 33 (3): 357-63
86. Ohshiro T, Okuda H, Tanaka T, et al. Clinical study of long-term therapy of PGI2 analogue (Procylin) in patients with chronic arterial obliterans [in Japanese]. Igaku Yakugaku 1995; 34 (6): 1012-26
87. Kojima K. Therapeutic efficacy of beraprost sodium in patients with chronic arterial occlusion [in Japanese]. Igaku Yakugaku 1993; 30 (1): 205-23
88. Katsumura T, Mishima Y, Kamiya K, et al. Therapeutic effect of ticlopidine, a new inhibitor of platelet aggregation, on chronic arterial occlusive disease, a double-blind study versus placebo. Angiology 1982 Jun; 33 (6): 357-67
89. Andreozzi GM, Signorelli SS, Cacciaguerra G, et al. Three-month therapy with calcium-heparin in comparison with ticlopidine in patients with peripheral arterial occlusive disease at Leriche-Fontaine IIb class. Angiology 1993 Apr; 44 (4): 307-13
90. Ito MK, Smith AR, Lee ML. Ticlopidine: a new platelet aggregation inhibitor. Clin Pharm 1992 Jul; 11: 603-17
91. Sakaguchi S, Tanade T, Mishima Y, et al. An evaluation of the clinical efficacy of beraprost sodium (a PGI2 analogue) for chronic arterial occlusion [in Japanese]. Rinsho to Kenkyu 1990; 67 (2): 575-84
92. Lievre M, Morand S, Besse B, et al. Oral Beraprost sodium, a prostaglandin I(2) analogue, for intermittent claudication: a double-blind, randomized, multicenter controlled trial. Beraprost et Claudication Intermittente (BERCI) Research Group. Circulation 2000 Jul 25; 102: 426-31
93. Sakaguchi S. Clinical studies of beraprost sodium, a newly developed oral prostacyclin (PGI2) derivative [in Japanese]. Gendai-Iryo 1992; 24: 119
94. Sakaguchi S, Tanade T, Mishima Y, et al. Clinical evaluation of the prostacyclin derivative beraprost sodium in chronic arterial occlusive disease. Shinryo to Shinyaku 1989; 26 (9): 1498-515
95. Gerin P, Dazord A, Boissel JP, et al. Quality of life assessment in therapeutic trials: rationale and presentation of a more appropriate instrument. Fundam Clin Pharmacol 1992; 6: 263-76
96. United Therapeutics. United Therapeutics announces phase III beraprost clinical trial results [online]. Available from URL: www.unither.com/pressReleases [Accessed 2001 Oct 15]
97. Shimizu Y, Nagaya N, Satoh T, et al. Orally active prostacyclin analogue improves exercise capacity in patients with primary or thromboembolic pulmonary hypertension. J Am Coll Cardiol 2000 Feb; 35 (2) Suppl. A: 286A
98. Vizza CD, Sciomer S, Morelli S, et al. Beraprost sodium in patients with severe pulmonary hypertension: long-term results [abstract]. Eur Respir J 2000: 16: 395s
99. Nagaya N, Uematsu M, Okano Y, et al. Effect of orally active prostacyclin analogue on survival of outpatients with primary pulmonary hypertension. J Am Coll Cardiol 1999 Oct; 34: 1188-92
100. Toray Industries I. Oral preparation of prostacyclin (PGI2) derivative – Dorner tablets 20mg; beraprost sodium prescribing information. Tokyo, Japan, 1999
101. Lievre M, Azoulay L, Lion L, et al. A dose-effect study of beraprost sodium in intermittent claudication. J Cardiovasc Pharmacol 1996; 27 (6): 788-93
102. News from the ESC XXIII Congress [online]. Pulmonary arterial hypertension: new hopes for a severe condition [online]. Available from URL: http://www.escardio.org/newscasts/stock2001/galie2.htm [Accessed 2001 Nov]
103. De Backer TL, Stichele RHV, Warie HH, et al. Oral vasoactive medication in intermittent claudication: utile or futile? Eur J Clin Pharmacol 2000; 56 (3): 199-206
104. Money SR, Herd JA, Isaacsohn JL, et al. Effect of cilostazol on walking distances in patients with intermittent claudication caused by peripheral vascular disease. J Vasc Surg 1998 Feb; 27 (2): 267-75
105. Dawson DL, Cutler BS, Meissner MH, et al. Cilostazol has beneficial effects in the treatment of intermittent claudication; results from a multicenter, randomized, prospective, double-blind trial. Circulation 1998 Aug 18; 98: 678-86
106. Beebe HG, Dawson DL, Cutler BS, et al. A new pharmacological treatment for intermittent claudication: results of a randomized, multicenter trial. Arch Intern Med 1999 Sep 27; 159: 2041-50
107. Hiatt WR, Hirsch AT, Regensteiner JG, et al. Clinical trials for claudication. Circulation 1995; 92 (3): 614-21
108. Nauser TD, Stites SW. Diagnosis and treatment of pulmonary hypertension. Am Fam Physician 2001 May 1; 63 (9): 1789-98
109. Hoeper MM, Schwarze M, Ehlerding S, et al. Long-term treatment of primary pulmonary hypertension with aerosolized iloprost, a prostacyclin analogue. N Engl J Med 2000 Jun 22; 342 (25): 1866-70
110. Barst RJ, Simonneau G, Rich S, et al. Efficacy and safety of chronic subcutaneous infusion UT-15 in pulmonary arterial hypertension (PAH). Circulation 2000 Oct 31; 102 (18) Suppl. 2: II-100
111. Channick R, Badesch DB, Tapson VF, et al. Effects of the dual endothelin receptor antagonist bosentan in patients with pulmonary hypertension: a placebo-controlled study. J Heart Lung Transplant 2001 Feb; 20 (2): 262-3
112. Rubin LJ, Mendoza J, Hood M, et al. Treatment of primary pulmonary hypertension with continuous intravenous prostacyclin (epoprostenol). Ann Intern Med 1990 Apr 1; 112 (7): 485-91
113. Higenbottam TW, Spiegelhalter D, Scott JP, et al. Prostacyclin (epoprostenol) and heart-lung transplantation as treatments for severe pulmonary hypertension. Br Heart J 1993; 70: 366-70
114. Long W, Rubin L, Barst R, et al. Randomized trial of conventional therapy alone (CT) vs. conventional therapy + continuous infusion of prostacyclin (CT + PGI2) in primary pulmonary hypertension (PPH): a 12 week study [abstract]. Am Rev Respir Dis 1993; 147: A538
115. Barst RJ, Rubin LJ, McGoon MD, et al. Survival in primary pulmonary hypertension with long-term continuous intravenous prostacyclin. Ann Intern Med 1994 Sep 15; 121 (6): 409-15
116. Cremona G, Higenbottam T. Role of prostacyclin in the treatment of primary pulmonary hypertension. Am J Cardiol 1995 Jan 19; 75: 67A-71A

117. Higenbottam T, Wheeldon D, Wells F, et al. Long-term treatment of primary pulmonary hypertension with continuous intravenous epoprostenol (prostacyclin). Lancet 1984 May 12: 1046-7
118. McLaughlin VV, Genthner DE, Panella MM, et al. Reduction in pulmonary vascular resistance with long-term epoprostenol (prostacyclin) therapy in primary pulmonary hypertension. N Engl J Med 1998 Jan 29; 338 (5): 273-7
119. Barst RJ, Rubin LJ, Long WA, et al. A comparison of continuous intravenous epoprostenol (prostacyclin) with conventional therapy for primary pulmonary hypertension. N Engl J Med 1996 Feb 1; 334 (5): 296-301
120. Magnani B, Galie N. Prostacyclin in primary pulmonary hypertension. Eur Heart J 1996 Jan; 17: 18-24
121. Trent Institute for Health Services Research. Prostacyclin in the treatment of primary pulmonary hypertension (March 1997) [online]. Available from URL: http://www.shef.ac.uk/uni/academic/r-z/tiwgap/summs/GN9702.htm [Accessed 2001 Aug]
122. GlaxoSmithKline. Overview of Flolan treatment [online]. Available from URL: http://www. flolan-center.com/pages/flolan [Accessed 2001 Dec]

Correspondence: *Ezequiel Balmori Melian,* Adis International Limited, 41 Centorian Drive, Private Bag 65901, Mairangi Bay, Auckland 10, New Zealand.
E-mail: demail@adis.co.nz