

A review of picotamide in the reduction of cardiovascular events in diabetic patients

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Abstract: Picotamide is an antiplatelet drug with a dual inhibitory action. Thus, picotamide inhibits both thromboxane A₂ (TxA₂) receptors and TxA₂ synthase and, at variance with aspirin, does not interfere with endothelial prostacyclin (PGI₂) production. Two large randomized trials have been performed to assess the clinical efficacy of picotamide in patients at risk of atherothrombosis. The ADEP study compared peripheral artery disease (PAD) patients randomized to picotamide or placebo. This study did not show a significant reduction of cardiovascular events by picotamide but a subgroup analysis showed its potential usefulness in patients with diabetes. To investigate this issue further, the DAVID study recently enrolled diabetic patients with PAD randomized to picotamide versus aspirin; the results showed a significant reduction of overall mortality in the picotamide group. Moreover long-term picotamide treatment in diabetes promotes the reduction of microalbuminuria and the inhibition of growth of carotid plaques. These data suggest that picotamide may represent an interesting drug to be further investigated in future trials in the atherothrombotic setting.

Keywords: picotamide, aspirin, diabetes, cardiovascular events, peripheral artery disease

Introduction

Picotamide, a derivative of methoxy-isophthalic acid, is an antiplatelet drug that inhibits both thromboxane A₂ (TxA₂) receptors and TxA₂ synthase. As concentrations of the molecule needed to inhibit both pathways are almost equivalent (Modesti et al 1994), picotamide may exert a dual pharmacological action in vivo and be potentially useful in various clinical settings characterized by atherosclerotic disease. Picotamide has been investigated in two large clinical trials in patients suffering from cardiovascular events, ie, in patients with peripheral artery disease (PAD) (Balsano and Violi 1993) and in patients with diabetes (Neri Serneri et al 2004). This review will focus on the results of these two trials and on the future perspective of picotamide in the setting of cardiovascular disease.

Pharmacology

Upon platelet activation, arachidonic acid is released from platelet membrane by phospholipase A₂ (PLA₂)-mediated degradation of membrane phospholipids and is converted to prostaglandin endoperoxides, as prostaglandin G₂ (PGG₂) and prostaglandin H₂ (PGH₂), via cyclooxygenase-1 (COX-1) activation. PGH₂ is then converted by TxA₂ synthase to TxA₂. Furthermore, PGH₂ participates in cycles of amplification signals for platelet activation by recruitment of other platelets through interaction with the same receptors of TxA₂ (Parise et al 1984; FitzGerald 1991).

TxA₂ has detrimental effects on the arterial tone by virtue of its vasoconstrictor property, an effect that is counteracted by endothelial molecules, such as nitric oxide and prostacyclin (PGI₂), a cyclo-oxygenase-derived substance.

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Aspirin is the most widely used antiplatelet drug to prevent cardiovascular events in patients with acute or chronic cardiovascular diseases (Antithrombotic Trialists' Collaboration 2002). Aspirin irreversibly acetylates COX-1, thereby preventing TxA₂ in platelets and PGI₂ in the endothelium. Although endothelial production of PGI₂ is inhibited only in part by aspirin (Caughey et al 2001), since the 1980s a number of drugs has been developed to interfere selectively with TxA₂ activity, without inhibiting endothelial PGI₂ production (Landolfi et al 1988). Among these drugs picotamide attracted the attention of researchers for its ability to inhibit both TxA₂ platelet synthase and platelet TxA₂ receptors and, in turn, platelet aggregation. In particular, Violi et al (1988) showed that picotamide at micromolar concentrations inhibited platelet aggregation induced by various agonists, such as ADP, arachidonic acid, and collagen. Furthermore the reduction of platelet aggregation was directly related to the inhibition of TxA₂ production. Unlike aspirin, picotamide was not able to reduce the release of PGI₂ by endothelium. Taken together these results demonstrated that picotamide reduced platelet function by inhibiting TxA₂ production without affecting cyclo-oxygenase activity. These data were confirmed by the *in vivo* study, which showed a consistent reduction of platelet aggregation and TxA₂ production in platelets from eight healthy volunteers taking picotamide 1200 mg/die (Violi et al 1988). Moreover Gresele et al (1989) showed that picotamide, apart from reducing the synthesis of TxA₂, enhanced the formation of PGE₂ in platelets and favored the formation of PGI₂ by aspirinated endothelial cells.

The evidence that picotamide almost completely inhibits platelet aggregation induced by the TxA₂ analogous U46619 suggested that this drug also has an inhibitory effect on TxA₂ receptors (Berrettini et al 1990). In this regard, an *in vitro* study showed that picotamide displaced a selective radioligand of TxA₂ receptor from platelets, so indicating that it directly inhibits this receptor (Modesti et al 1989). Other pharmacological studies demonstrated that picotamide binding to thromboxane A₂ receptors is initially reversible and becomes progressively non-displaceable (Modesti et al 1991, 1994).

The reduction of circulating platelet-derived TxA₂ levels induced by picotamide not only inhibits platelet aggregation, but also has important consequences for the inflammatory processes of atherosclerotic plaque. TxA₂ has an important role in regulating vascular tone, as it induced endothelin-1 (ET-1) in both endothelium and smooth muscle cells (Chua

et al 1996). As a consequence, the reduction of TxA₂ platelet formation induced by picotamide administration decreases vessel tone by reducing the circulating levels of endothelin-1 (Saitta et al 1998). Moreover picotamide reduces smooth muscle cell proliferation as TxA₂ is a mythogenic stimulus: *in vitro* studies with picotamide demonstrated that it inhibits DNA synthesis of smooth muscle cells incubated with U46619 (a TxA₂ analog) alone and also with other mythogenic molecules such as EGF or PDGF (Ratti et al 1998). Finally, it should be mentioned that *in vitro* as well as *in vivo* studies documented an antiplatelet and antivasoconstrictor property of picotamide that is unrelated to inhibition of TxA₂ formation, such as inhibition of serotonin-induced platelet aggregation (Veza et al 1997).

Platelet activation and aspirin in diabetes

Diabetes is a clinical setting characterized by enhanced platelet activation that depends in part on increased platelet production of thromboxane A₂. In fact, Davi et al (1990) showed that urinary thromboxane B₂ (Tx_B2), a stable metabolite of TxA₂, was significantly higher in patients with type 2 diabetes than in controls. Moreover, tight diabetic control with insulin therapy reduced urinary 11-dehydrothromboxane B₂ by almost 50%, while aspirin therapy reduced this urinary metabolite by 80%. Recovery of impaired excretion of such molecules occurred after an aspirin wash-out of 10 days, suggesting the platelet origin of thromboxane A₂.

Aspirin has been used in patients with diabetes to prevent cardiovascular complications, but the results are disappointing because only a slightly reduction in clinical outcomes has been observed compared with patients affected by other cardiovascular risk factors (Antithrombotic Trialists' Collaboration 2002). The reason why diabetic patients are less sensitive to aspirin than non-diabetic ones is unclear. An enhanced turnover of platelets has been observed in diabetic patients, a phenomenon that could limit the antiplatelet effect of aspirin. However, more data are necessary to support such a hypothesis (Csiszar et al 2002; Di Minno and Violi 2004; Gresele and Migliacci 2004).

A chronic inflammatory state is often associated with diabetes and may be responsible for platelet activation via oxidative stress-mediated formation of isoprostanes (Patrono and FitzGerald 1997). Thus, these molecules are products of non-cyclooxygenase oxidative modifications of arachidonic acid due to oxidative stress-mediated modification of membrane phospholipids or circulating LDLs, and are characterized by potent vasoconstrictor and pro-aggregatory

effects. Their effects in activating platelets are mediated by receptors closely related to TxA₂ receptors and may be potentially counteracted by TxA₂ receptor antagonists such as picotamide. So far, however, no data are available on the effect of picotamide on isoprostanes-induced platelet activation.

Platelet activation and aspirin in peripheral arterial disease

Platelet activation as assessed by urinary excretion of TxB₂ has been studied in patients with PAD and matched controls (Davì et al 1997). This study demonstrated enhanced values of urinary TxB₂ in PAD patients compared with control. Such differences, however, seemed to be attributable essentially to the coexistence of risk factors for atherosclerotic disease such as hypercholesterolemia, diabetes, or smoking habit. Thus, in PAD patients without such risk factors, urinary excretion of TxB₂ was similar to that in controls, suggesting that the risk factors rather than atherosclerotic disease per se are responsible for enhanced production of TxB₂.

Despite these data suggesting a role for COX-1 in the pathophysiology of PAD, the clinical efficacy of aspirin in this clinical setting is uncertain. Prospective studies with adequate sample size have never been performed. Data on the effects of aspirin in preventing cardiovascular disease stem essentially from meta-analysis, which have not provided definite findings on its potential efficacy (WAVE Investigators 2006). Therefore further studies with adequate sample size should be performed to investigate the clinical efficacy of aspirin in preventing cardiovascular disease in this clinical setting.

Clinical studies

Since the 1970s, prospective studies have shown a strong association between cardiovascular events and type 1 or type 2 diabetes mellitus (Garcia et al 1974; Panzram 1987). Moreover in recent decades the beneficial effects of aspirin and other antiplatelet drugs against myocardial infarction, stroke, and other vascular events have been well documented (Antithrombotic Trialist's Collaboration 2002). However, aspirin treatment seems to be less useful in diabetic patients compared with patients with other risk factors in primary or secondary prevention of cardiovascular complications. In this regard a recent meta-analysis analysed 4961 subjects among 9 trials and showed only a 7% odds reduction of vascular complications in diabetic patients treated with aspirin versus placebo. Although this reduction hardly reached statistical

significance, it is consistently less when compared with the proportional reduction over 195 studies analyzed by the meta-analysis (22% versus 7%) (Antithrombotic Trialist's Collaboration 2002).

As reported above, evidence for any benefits of aspirin treatment in patients with PAD is insufficient (WAVE Investigators 2006). Thus, in contrast to other authorities (Clagett et al 2004), the Food and Drug Administration expert panel did not provide any indication of aspirin treatment for patients with PAD (Food and Drug Administration 1998). This strongly suggests the need for developing new antiplatelet drugs potentially useful in diabetes and PAD.

The first large randomized trial (ADEP trial) on picotamide investigated its clinical usefulness in patients with PAD (Balsano and Violi 1993). For this study 2304 patients were consecutively enrolled, allocated to either placebo or picotamide (300 mg bid), and followed for 18 months. Endpoints of the study were major events (ie, cardiovascular death, myocardial infarction, stroke, or amputation) and minor events (unstable angina, transient ischemic attack, hypertension, renal failure, deterioration of PAD). The "intention to treat analysis" showed a risk reduction (18.9%) in the combined endpoints, major plus minor events, in the picotamide group compared with the controls, which, however, did not reach statistical significance; conversely, "on treatment analysis" showed a higher and statistically significant reduction (22.8%) in the same endpoints. Side-effects such as bleeding were almost identical in the two groups. As the authors suggested, the lack of any beneficial effects of picotamide against major events could have been related to the low occurrence of these events during the follow-up; this phenomenon may be related to a bias in patient selection, which excluded high-risk patients.

The capacity of picotamide to prevent vascular complications was, however, magnified when claudicant patients affected by diabetes were taken into account. Thus, a sub-study of the ADEP trial retrospectively analyzed 438 diabetic patients and observed a risk reduction of 45.2% of combined major and minor events in those treated with picotamide compared with those treated with placebo (Milani et al 1996). On the basis of this post-hoc analysis a new randomized trial (the DAVID trial) was specifically designed for diabetic patients with PAD (Neri Serneri et al 2004). Thus, 1209 patients were enrolled and randomly assigned to picotamide (600 mg bid) or aspirin (320 mg/day) and followed for 2 years. The primary endpoint was the overall mortality and the secondary one was the combined incidence

of death and major cardiovascular events. Mortality was significantly lower in picotamide-treated patients than in those treated with aspirin, showing a relative risk of reduction of 45%; furthermore the incidence of gastrointestinal bleeding was much lower in the picotamide group than in the aspirin group. The secondary endpoint did not show any significant difference between two populations, showing only a non-significant trend in favor of patients taking picotamide (Tables 1 and 2). As pointed out by the authors, a possible bias relative to the high proportion of patients (about 20% in each group) who discontinued the trial because of a non-fatal events may have underestimated the real incidence of the secondary endpoints; moreover it is possible that the sample size of the study was insufficient to detect any difference in these end-points between the two groups (Tables 1 and 2). Comparison of the results achieved by the ADEP and DAVID trials also raises the question as to whether the differences seen are dependent on the fact that TxA₂ production is more relevant for atherosclerotic progression in PAD patients with diabetes compared with PAD without diabetes, or whether the different dosage of picotamide (600 mg vs 1200 mg in the ADEP and DAVID respectively) has a different impact on clinical outcome. Further studies are therefore necessary to explore the relationship between picotamide dosage and TxA₂ inhibition *in vivo*.

In conclusion, despite the interesting findings relative to the reduction of overall mortality and the better safety

of picotamide compared with aspirin, other clinical trials are necessary in order to clarify the real clinical benefit of picotamide for ischemic non-fatal events in PAD patients with diabetes.

The beneficial effects of long-term picotamide in diabetic patients are also corroborated by its capacity in reducing microalbuminuria and in inhibiting growth of carotid plaques.

Thirty type 2 diabetic, normotensive patients, all characterized by microalbuminuria at rest, were randomized to picotamide 300 mg/day or placebo for 1 year. Results showed a reduction of microalbuminuria at rest and after exercise without significant changes in metabolic control in the picotamide-treated patients compared with control subjects. Moreover, a linear correlation was found between urinary thromboxane excretion and microalbuminuria after stress-test (Giustina et al 1998). These findings suggest an interesting relationship between thromboxane formation and vascular complication of type 2 diabetes. Thus, the fact that picotamide is more effective in reducing microalbuminuria induced by stress-test than at rest suggests that the vasoconstrictor properties of TxA₂ may be responsible for its physiopathological action on glomerular activity.

The effects on plaque progression are also supported by a 2-year prospective study that demonstrated a reduction of carotid plaque evolution in diabetic patients, affected by non-stenotic and asymptomatic carotid atherosclerosis. In

Table 1 Baseline characteristics and side-effects of both ADEP and DAVID trials

	ADEP study		DAVID study	
	Picotamide 300 mg bid	Placebo	Picotamide 600 mg bid	Aspirin 320 mg od
Clinical characteristics				
Dosage				
n = 1150	n = 1154	n = 603	n = 606	
Age	63.4 + 7.3	62.9 + 7.4	63.8 + 7.2	64.6 + 7.3
Diabetes	20%	18%	100%	100%
PAD	100%	100%	100%	100%
Smoke	39.6%	37.1%	30.5%	28.4%
Hypertension	34.5%	37.5%	58.2%	55.6%
Dyslipidemia	36.5%	35.8%	38%	38.4%
Previous stroke	1.4%	1.7%	10.4%	10.2%
Coronary heart disease	15%	12.7%	19.4%	19%
Side-effects				
Gastro-intestinal	11.1%	12%	10.9%	18.3%
Total	14.3%	13.5%	25.6%	33.9%

Abbreviations: bid, twice a day; od, once a day; PAD, peripheral artery disease.

Table 2 Duration of follow-up, endpoints and statistically relevant results of both ADEP and DAVID trials

	ADEP study		DAVID study	
Follow-up	18 months		24 months	
Primary endpoints	Major events: death, MI, stroke, amputation		Overall mortality	
Secondary endpoints	Minor events: UA, TIA, hypertension, renal failure, deterioration of PAD		Stroke, MI, amputation, other death	
Statistical significance	Major plus minor events ("on treatment analysis")		Overall mortality	
Percentage of event in statistically relevant endpoints	picotamide	placebo	picotamide	aspirin
	10.1%	13%	3%	5.5%

Abbreviations: MI, myocardial infarction; PAD, peripheral artery disease; TIA, transient ischemic attack; UA, unstable angina.

extension, 50 type II normotensive diabetic patients with asymptomatic mild or moderate non-stenotic (<50%) carotid atherosclerotic plaque were randomly given picotamide 300 mg/day or placebo; after 2 years, lesion numbers and percentage stenosis in the picotamide group were significantly lower than in the placebo group (Cocozza et al 1995).

Conclusion

The data so far reported on the effect of picotamide in patients at risk of atherothrombosis are of particular interest overall because the clinical efficacy of picotamide has emerged in patients with diabetes, who seem to be less sensitive to the effect of other antiplatelet drugs such as aspirin. Although the number of patients included in trials with picotamide was not limited, trial results are still insufficient to provide definite conclusions on its clinical efficacy. Therefore the results of the above-reported trials should represent a useful background to further test the hypothesis that an antiplatelet drug with a dual mechanism of action such as picotamide may be beneficial in preventing the cardiovascular events in patients with atherothrombosis. In this context it is interesting to mention a recent experimental study in which a combination of drugs inhibiting COX-1 and TxA₂ receptor antagonist has been investigated in an animal model of atherosclerosis (Cyrus et al 2006). The study showed that such a combination possesses a greater anti-atherosclerotic property than the single drug, so providing further support for the potentially clinical usefulness of drugs with dual COX-1 and TxA₂ receptor inhibition. The DAVID study suggests that in patients with PAD and diabetes, antiplatelet

drugs with similar pharmacological characteristics should be investigated. The study should be prospective and include more than 1500 patients (the sample size primarily considered by the DAVID committee as number of patients necessary to detect a significant difference between picotamide and aspirin), and should obviously compare picotamide versus aspirin in a follow-up of 2 years. Such a study could provide more definite data on the clinical efficacy of picotamide in this clinical setting and potentially open new avenues in an atherosclerotic sub-setting where the clinical efficacy of aspirin is still debated.

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