Clinical Pharmacology of Platelet, Monocyte, and Vascular Cyclooxygenase Inhibition by Naproxen and Low-Dose Aspirin in Healthy Subjects

Marta L. Capone, PharmD; Stefania Tacconelli, PharmD; Maria G. Sciulli, PhD; Marilena Grana, MD; Emanuela Ricciotti, PharmD; Pietro Minuz, MD; Patrizia Di Gregorio, MD; Gabriele Merciriao; Carlo Patrono, MD; Paola Patrignani, PhD

Background—The current controversy on the potential cardioprotective effect of naproxen prompted us to evaluate the extent and duration of platelet, monocyte, and vascular cyclooxygenase (COX) inhibition by naproxen compared with low-dose aspirin.

Methods and Results—We performed a crossover, open-label study of low-dose aspirin (100 mg/d) or naproxen (500 mg BID) administered to 9 healthy subjects for 6 days. The effects on thromboxane (TX) and prostacyclin biosynthesis were assessed up to 24 hours after oral dosing. Serum TXB₂, plasma prostaglandin (PG) E₂, and urinary 11-dehydro-TXB₂ and 2,3-dinor-6-keto-PGF₁α were measured by previously validated radioimmunoassays. The administration of naproxen or aspirin caused a similar suppression of whole-blood TXB₂ production, an index of platelet COX-1 activity ex vivo, by 94±3% and 99±0.3% (mean±SD), respectively, and of the urinary excretion of 11-dehydro-TXB₂, an index of systemic biosynthesis of TXA₂ in vivo, by 85±8% and 78±7%, respectively, that persisted throughout the dosing interval. Naproxen, in contrast to aspirin, significantly reduced systemic prostacyclin biosynthesis by 77±19%, consistent with differential inhibition of monocyte COX-2 activity measured ex vivo.

Conclusions—The regular administration of naproxen 500 mg BID can mimic the antiplatelet COX-1 effect of low-dose aspirin. Naproxen, unlike aspirin, decreased prostacyclin biosynthesis in vivo. (Circulation. 2004;109:1468-1471.)

Key Words: aspirin ■ naproxen ■ thromboxanes ■ epoprostenol ■ platelets

Aspirin is the only nonsteroidal antiinflammatory drug (NSAID) known to react covalently with the cyclooxygenase (COX) channel of prostaglandin (PG) G/H synthase-1 and -2 (also referred to as COX-1 and COX-2) through a selective acetylation of a single serine residue (Ser⁵²⁹ in human COX-1 and Ser⁵¹⁶ in human COX-2) that results in the permanent loss of the COX activity of the enzyme.¹ ² The consistency in dose requirement and saturability of the effects of aspirin in acetylcating platelet COX-1, inhibiting thromboxane (TX) A₂ formation, and preventing atherothrombotic complications constitutes the best evidence that the antithrombotic effect of aspirin is largely caused by the suppression of platelet TXA₂ production.³ ⁴ However, it is uncertain whether other NSAIDs that act as competitive, reversible inhibitors of both COX-1 and COX-2 share an aspirin-like cardioprotective effect. This question has received considerable attention after publication of the Vioxx Gastrointestinal Outcome Research (VIGOR) trial,³ a study of approximately 8000 patients with rheumatoid arthritis randomized to receive rofecoxib 50 mg/d or naproxen 500 mg BID with a mean duration of follow-up of 9 months. The rates of myocardial infarction were 0.5% and 0.1% in the rofecoxib- and naproxen-treated groups, respectively, raising the possibility of a thrombogenic effect of rofecoxib, a cardioprotective effect of naproxen, and/or the play of chance.⁵ Six of 8 recent observational studies and a meta-analysis of these studies suggest that regular use of naproxen might be associated with a somewhat reduced risk of a first myocardial infarction versus nonuse (L.A. García Rodríguez, Centro Espanol de Investigacion Farmacoepidemiologica, Madrid, Spain, personal communication, October 2003). Because of the paucity of data on the clinical pharmacology of platelet and vascular prostanooid inhibition by naproxen,⁷ ⁸ we performed a crossover, open-label study of low-dose aspirin (100 mg/d) or naproxen (500 mg BID) administered to healthy subjects for 6 days. The primary aim of the study was to compare the extent and duration of steady-state inhibition of platelet COX-1 activity ex vivo by low-dose aspirin and...
naproxen. The secondary aim was to evaluate the effects of these drugs on systemic biosynthesis of TXA₂ and prostacyclin in vivo and on monocyte COX-2 activity ex vivo.

Methods

Study Subjects

The study protocol was approved by the Ethics Committee of the G. d’Annunzio University of Chieti. Informed consent was obtained from each subject. The volunteers were 9 healthy subjects (8 men, 1 woman, 23 to 58 years old) with a negative medical history and physical examination and with routine hematological and biochemical parameters within the normal range. Smokers and subjects with a bleeding disorder, allergy to aspirin or any other NSAID, or a history of any gastrointestinal disorder were excluded. Subjects abstained from the use of aspirin and other NSAIDs for at least 2 weeks before enrollment.

Design of the Study

This was a crossover, open-label study of low-dose aspirin (100 mg/d in an enteric-coated formulation, Bayer SpA) or naproxen (500 mg BID, Recordati SpA) for 6 consecutive days, with a washout period of at least 14 days. The inhibition of platelet COX-1 was assessed by measurements of whole-blood TXB₂ production. Monocyte COX-2 activity was assessed through the measurement of lipopolysaccharide (LPS)-induced PGF₂ₑ production in whole blood. Measurements were performed before and at 1, 12, and 24 hours after the last dose of aspirin and at 3, 12, and 24 hours after the last dose of naproxen. Urinary samples were collected for 12 hours before dosing and in 3 postdosing aliquots: 0 to 6, 6 to 12, and 12 to 24 hours to evaluate the excretion of 11-dehydro-TXB₂ and 2,3-dinor-6-keto-PGF₁α, major enzymatic metabolites of TXA₂ and PGI₁, respectively, that are indexes of their systemic biosynthesis in vivo.

Biochemical Analyses

Immunoreactive TXB₂, PGF₂ₑ, 11-dehydro-TXB₂, and 2,3-dinor-6-keto-PGF₁α were measured by previously validated radioimmunoassay techniques.

Statistical Analysis

The data are expressed as mean±SD. Statistical comparisons were made by ANOVA followed by Student-Newman-Keuls test. A probability value of P<0.05 was considered to be statistically significant. Assuming an intersubject coefficient of variation (CV) of 25% for serum TXB₂ (primary end point) in healthy subjects, 8 subjects would allow detection of a difference of 40% in its postdosing concentrations versus baseline with a power of 90%, on the basis of 2-tailed tests, with probability values less than the type I error rate of 0.05. Thus, 9 healthy volunteers were enrolled, but 1 male subject refused to take aspirin after completing treatment with naproxen.

Results

We compared the time course of recovery from steady-state inhibition of platelet COX-1 activity by low-dose aspirin (100 mg/d) and naproxen (500 mg BID) administered for 6 days to 9 healthy subjects. As shown in Figure 1A, at 1, 12, and 24 hours after the last dose of aspirin, platelet COX-1 activity was suppressed by 99±0.3%, 99±0.3%, and 99±1% (mean±SD, n=8, P<0.01 versus predrug values reported in the Table). The persistent suppression of platelet TXB₂ production by low-dose aspirin up to 24 hours reflects the irreversible inactivation of platelet COX-1 activity. At 3 and 12 hours after the last dose, naproxen caused comparably profound suppression of platelet COX-1 activity (94±8% and 94±3%, respectively; mean±SD, n=9) (Figure 1A). Therefore, a slow recovery of platelet COX-1 activity was detectable (at 24 hours after dosing, serum TXB₂ was reduced by 80±9%) (Figure 1A) that is consistent with the reversible interaction of naproxen with COX-1.

As shown in Figure 1B, both aspirin and naproxen caused a comparable and persistent inhibition of the biosynthesis of TXA₂ in vivo, as reflected by the urinary excretion of 11-dehydro-TXB₂. In 3 consecutive urine collections performed after the last dose of aspirin or naproxen (ie, 0 to 6, 6 to 12, and 12 to 24 hours after dosing), urinary 11-dehydro-TXB₂ was reduced by 76±9%, 79±7%, and 74±16%, and 85±5%, 85±8%, and 79±11%, respectively.

As shown in Figure 2A, the administration of low-dose aspirin did not affect LPS-induced PGF₂ₑ production in whole blood, an index of monocyte COX-2 activity, to any statistically significant extent. In contrast, naproxen significantly reduced COX-2 activity at 3, 12, and 24 hours after the last dose; COX-2 activity was reduced by 68±18%, 62±9%, and 56±21%, respectively (P<0.01 versus predrug values). As shown in Figure 2B, aspirin and naproxen had markedly different effects on systemic prostacyclin biosynthesis, as reflected by urinary 2,3-dinor-6-keto-PGF₁α excretion. In the 3 consecutive urine collections, 2,3-dinor-6-keto-PGF₁α levels were not significantly affected by aspirin, whereas naproxen reduced 2,3-dinor-6-keto-PGF₁α by 78±9%
(P<0.001), 77±19% (P<0.001), and 66±17% (P<0.001), respectively (Figure 2B).

Discussion

The aim of the present study was to explore the pharmacodynamic plausibility of an aspirin-like cardioprotective effect of naproxen, a nonselective NSAID with a long half-life. We found that the chronic administration of a therapeutic antiinflammatory dose of naproxen (500 mg BID) to healthy subjects caused persistent and almost complete suppression of platelet TXB₂ production throughout the 12-hour dosing interval that was indistinguishable from that of low-dose aspirin (100 mg/d). However, whereas aspirin pharmacokinetics is dissociated from pharmacodynamics and 100 mg represents approximately a 3-fold excess versus the lowest effective dose to saturate platelet COX-1 activity,³ naproxen pharmacodynamics is strictly related to its systemic bioavailability, as shown by the significant recovery of platelet COX-1 activity at 24 hours after dosing. Moreover, 500 mg BID is probably close to but not quite at the top of the dose-response curve for COX-1 inhibition. This suggests that compliance and daily dose are likely to represent the main determinants of the clinical efficacy of naproxen for cardiovascular protection. Thus, the apparently conflicting results of a randomized clinical trial, like VIGOR,⁵ and observational studies¹⁴–¹⁸ may reflect markedly different rates of compliance and regular use of a high dose of the drug in the 2 settings.

The results of 2,3-dinor-6-keto-PGF₁α measurements confirm earlier studies¹⁹,²⁰ suggesting that an important component of the basal rate of PGI₂ biosynthesis is COX-2–dependent, as reflected in the present study by the consistent differential effects of naproxen versus low-dose aspirin on monocyte COX-2 activity and urinary 2,3-dinor-6-keto-PGF₁α excretion.

The inhibitory effects of naproxen on PGI₂ biosynthesis are unlikely to counteract its potential cardioprotective effect, in light of the demonstrated efficacy of aspirin at high doses having a similar effect on PGI₂.³,⁴ However, the clinical relevance of simultaneous suppression of TXA₂ and PGI₂ by naproxen in patients with cardiovascular disease remains to be determined. Moreover, the impact of naproxen on COX-2–dependent sources of thromboxane biosynthesis might contribute to its clinical effects in preventing myocardial infarction.

In conclusion, the present study demonstrates the pharmacodynamic plausibility of a COX-1–dependent cardioprotective effect of naproxen and contributes to the interpretation of the VIGOR cardiovascular findings. Although our results are mechanistically informative of what may happen under the best-case scenario of a randomized clinical trial of regular, prolonged use of a high-dose reversible COX inhibitor, practicing physicians should not assume that the same holds true in the less-than-ideal circumstances of real-life use of these drugs, which is neither regular nor continuous nor necessarily at high doses.

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References


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