

Effect of aspirin on airway inflammation and pulmonary function in patients with persistent asthma

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Background: Aspirin can cause bronchoconstriction in some asthmatic patients through increased production of proinflammatory mediators, particularly leukotrienes. However, recent *in vivo* evidence has suggested that aspirin also triggers the production of lipoxins, which act as natural antagonists of prostaglandins and leukotrienes. Aside from patients with known aspirin-sensitive asthma, physicians have avoided the use of aspirin in asthmatic patients in general because it was believed that this agent might precipitate worsening of their condition.

Objective: We sought to establish the effect of aspirin on pulmonary inflammation and function in patients with persistent asthma.

Methods: After withdrawal of their usual anti-inflammatory medication, patients with mild-to-moderate persistent asthma undertook double-blind, randomized, crossover treatment with 75 mg/d aspirin and placebo for 3 weeks each. Treatment evaluation included histamine challenge, spirometry, impulse oscillometry, total and alveolar exhaled nitric oxide measurement, and serum thromboxane B2 and 15-epilipoxin A4 levels.

Results: Fifteen patients completed the trial. Compared with placebo, there were no differences in histamine PC₂₀ values (0.17 doubling-dilution shift; 95% CI, -0.38 to 0.73; *P* = 1), exhaled nitric oxide levels (0.95-fold change; 95% CI, 0.45-2.00; *P* = 1), or any other inflammatory, spirometric, or oscillometry measurements. Aspirin led to a significant decrease in thromboxane B2 levels (17.53-fold difference; 95% CI, 5.46-56.49; *P* < .001). Baseline 15-epilipoxin A4 levels were increased at 4.88 ng/mL, and there was no increase with aspirin versus placebo (0.99-fold difference; 95% CI, 0.79-1.24; *P* = 1).

Conclusion: In this preliminary study of 15 patients, low-dose aspirin did not lead to increased 15-epilipoxin A4 synthesis or alter inflammatory markers in patients with mild-to-moderate persistent asthma. (J Allergy Clin Immunol ■■■■;■■■:■■■-■■■.)

Key words: Asthma, aspirin, inflammation, lipoxin, thromboxane, exhaled nitric oxide, histamine challenge

Abbreviation used

NSAID: Nonsteroidal anti-inflammatory drug

Aspirin is frequently used for its antipyretic, anti-inflammatory, and antithrombotic properties. Aspirin is inexpensive and has proved efficacy in cardiovascular disease, substantially reducing the primary and secondary risk of myocardial and intracranial atherosclerotic ischemic events.^{1,2} In common with other nonsteroidal anti-inflammatory drugs (NSAIDs), aspirin mediates much of its therapeutic effect through inhibition of COX-1 and COX-2, with subsequent attenuation of prostaglandin and thromboxane synthesis, both of which act as proinflammatory mediators and also as powerful stimuli to bronchoconstriction. Unlike other NSAIDs, when aspirin acetylates COX-2, a switch in enzymatic function is induced whereby prostaglandin production is halted, and in its place synthesis of lipoxin A4 and 15-epilipoxin A4 (aspirin-triggered lipoxins) is facilitated.^{3,4} These biologically active molecules belong to a family of endogenous eicosanoid cytokines termed lipoxins that are believed to act as natural “brakes” to the inflammatory cascade, antagonizing the actions of leukotrienes.^{5,6} It is speculated that the unique ability of aspirin to induce production of lipoxin analogues *in vivo* is one of the reasons why it is so clinically effective in the treatment of cardiovascular disease, which is now well recognized as having a significant inflammatory component.⁷ It has been shown that aspirin-triggered lipoxin production has a very shallow dose-response relationship with aspirin, whereby maximal production is attained with approximately 80 mg/d, which is similar to the dose found to have the greatest risk/benefit ratio for treatment of cardiovascular disease in clinical practice.⁵

There has been speculation that lipoxins might have potential as therapeutic agents in asthma after the observation that patients with severe asthma have subnormal lipoxin values in their airways compared with patients who have milder forms of the disease.⁸⁻¹⁰ Experimental data from a murine asthma model has shown that lipoxins can directly reduce airway hyperresponsiveness and also promote resolution of lung inflammation by inhibiting inflammatory cellular infiltration of the allergic airway mucosa, while simultaneously attenuating production of signaling molecules, including eotaxin, IL-5, IL-13, and the cysteinyl leukotrienes.¹¹ In both mild and severe asthma, lipoxins inhibit the release of IL-8 from peripheral mononuclear cells, further suggesting they might be important as endogenous anti-inflammatory mediators.⁶ Small trials have also shown an improvement in the airway hyperresponsiveness of asthmatic patients after inhalation of aspirin.^{12,13} Additional epidemiologic evidence from the

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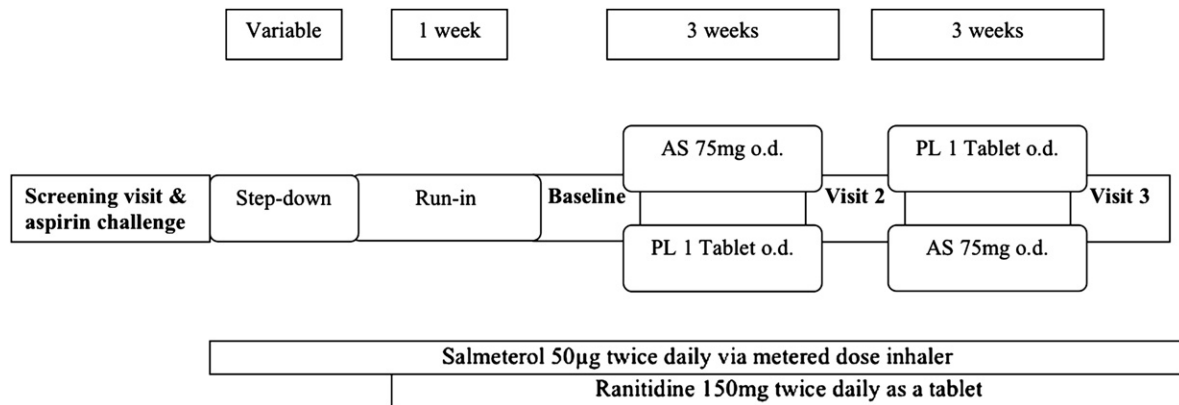


FIG 1. Study diagram. AS, Aspirin; PL, placebo to aspirin.

Physicians' Health Study has also led to speculation that aspirin can prevent asthma: in a randomized group of 22,040 participants, regular aspirin administration led to a 22% reduction in the risk of development of new-onset asthma.¹⁴

Traditionally, asthmatic patients have been advised to avoid NSAIDs, including aspirin, because of the potential for disease exacerbation secondary to COX-1 inhibition and consequent upregulation of leukotriene production. The majority of asthmatic patients are not aspirin intolerant (between 5% and 10% are intolerant), and those who are might have a genetic predisposition caused by a leukotriene receptor polymorphism.¹⁵⁻¹⁷ It remains unclear whether aspirin administration has either a deleterious or potentially therapeutic effect in asthma. We have therefore conducted a randomized controlled trial to determine the effects of therapeutic doses of aspirin on airway inflammation in asthmatic patients.

METHODS

The Tayside Committee for Medical Research Ethics gave institutional approval to the study protocol before commencement of the trial. Written informed consent was obtained from each participant before enrollment. Additional methodological information can be obtained in the [Online Repository](http://www.jacionline.org) (available at www.jacionline.org).

Participant identification and recruitment

Potential study participants were identified from a preexisting database of research volunteers with asthma. Adult (>18 years) patients with mild-to-moderate persistent asthma, an FEV₁ of greater than 60% of predicted value, and a maintenance inhaled corticosteroid dose of 1000 µg/d beclomethasone dipropionate or less were included. Any patient with reported NSAID or aspirin hypersensitivity, past or present gastrointestinal hemorrhage and ulcers, or unstable asthma requiring oral corticosteroids in the 3 months before trial commencement were excluded from participation. In addition, before entering the trial, each participant underwent a modified oral aspirin challenge to ensure they exhibited no features suggestive of aspirin intolerance (full details of the procedure are available in the [Online Repository](http://www.jacionline.org)).¹⁸

Randomized treatment

Patients had all regular maintenance asthma treatment (inhaled corticosteroids, leukotriene receptor antagonists, and theophylline) withdrawn in a stepwise fashion before discontinuing them completely for the duration of their inclusion in the trial (for more details, see the [Online Repository](http://www.jacionline.org)). Before withdrawal of preventive therapy, all participants were begun on 50 µg of salmeterol twice daily administered through a metered-dose inhaler. This process was designed to maximize any inflammatory signal that might be

present from the asthmatic airway while simultaneously palliating symptoms related to bronchoconstriction. Short-acting β_2 -receptor agonists were permitted in an unrestricted fashion throughout the trial. After successful withdrawal of anti-inflammatory therapy, all participants were started on 150 mg of ranitidine twice daily to be continued for the trial duration and then underwent a 1-week unblinded run-in period during which they received nonrandomized aspirin placebo (one tablet, once daily). The ranitidine was included to provide protection against gastrointestinal side effects (specifically gastric ulcers and gastric hemorrhage) associated with aspirin therapy. Although not a prerequisite of aspirin treatment, it was deemed ethically more acceptable to optimally minimize the risk of serious side effects in this volunteer population, in which aspirin treatment was not otherwise indicated. At the end of the run-in period, a baseline assessment was performed, at which point participants were required to have a histamine PC₂₀ value of 4 mg/mL or less to proceed to randomization; that is, patients were preselected *a priori* on the basis of active airway inflammation with documented bronchial hyperresponsiveness. Each patient then undertook double-blind, randomized treatment with 75 mg of aspirin once daily and placebo to aspirin once daily in sequential blocks of 3 weeks' duration each. A crossover design was used such that every patient received both aspirin and placebo treatment. At the end of each randomized therapy, they returned to the research department to undergo evaluation of the treatment received. The trial design is depicted schematically in [Fig 1](#).

At each scheduled visit, compliance was actively re-enforced and verified by counting the remaining tablets. The patients' supply of salmeterol and rescue medication was replenished as necessary. As a safety measure, each patient was also supplied with and shown how to use a peak flowmeter and recorded daily measurements in a diary. They were instructed to seek assistance from an investigator through a 24-hour cellular telephone number if their readings decreased to less than 60% of predicted value.

Treatment evaluation

Treatment response was assessed at baseline after the placebo run-in period and after each 3-week randomized treatment block. Salmeterol was withheld for 48 hours and salbutamol for at least 6 hours before each treatment evaluation. The following measurements were made in the order specified: impulse oscillometry,¹⁹ quantification of compartmental nitric oxide (tidal nitric oxide, bronchial flux, and alveolar nitric oxide),^{20,21} spirometry,²² histamine challenge, peripheral blood sampling to determine eosinophil count and eosinophil cationic protein serum thromboxane B2 and serum 15-epi-poxin A4 levels. Further relevant details of the techniques used to derive each of these measurements are given in the [Online Repository](http://www.jacionline.org).

Statistical analysis

SPSS version 12 (SPSS, Inc, Chicago, Ill) and Prism version 4 for Macintosh (GraphPad Software, Inc, San Diego, Calif) were used to perform the statistical analysis. A sample size of 13 patients was estimated to give 80% power to detect a 1 doubling-dilution shift in histamine PC₂₀ values (the primary outcome),

assuming a 0.8 within-patient SD in doubling-dilution shift.²³ As such, it was intended to recruit a total of 20 patients to allow for a potential 30% dropout rate. All data were assessed for normality by using distribution plots, and if appropriate, non-Gaussian data were log transformed before analysis. Treatment effect was evaluated by means of ANOVA of values at baseline and after each randomized therapy, followed by pairwise multiple range testing with the Bonferroni correction (the overall α error was set at .05) to obviate the effect of sequential comparisons.

RESULTS

A total of 32 patients were contacted and asked to participate, of whom 19 patients were enrolled and 15 completed the trial per protocol, 3 patients were withdrawn because of deterioration in their asthma control either during stepdown or during randomized intervention, and a further 1 patient was excluded from the final analysis because the histamine PC₂₀ value was greater than 4 mg/mL at baseline and therefore this patient should not have continued in accordance with the predetermined protocol. Consequentially, data from 15 patients were included in the final per-protocol analysis (Table I).

Treatment with aspirin had no effect on airway hyperresponsiveness to histamine (the primary outcome) compared with placebo (0.17 doubling-dilution shift; 95% CI, -0.38 to 0.73; $P = 1$) or baseline (0.61 doubling-dilution shift; 95% CI, -1.17 to 0.06; $P = .06$; Table II and Fig 2). Compared with placebo, exhaled nitric oxide levels were also unchanged by aspirin treatment, regardless of whether the airway was examined as a single entity (0.95-fold change in concentration of exhaled nitric oxide; 95% CI, 0.45-2.00; $P = 1$) or whether the proximal large airway (0.99-fold change in bronchial flux; 95% CI, 0.68-1.43; $P = 1$) was separated out from the distal fractional component (0.87-fold change in alveolar nitric oxide; 95% CI, 0.45-1.66; $P = 1$; Table II, Figs 3 and 4, and Fig E1 in the Online Repository at www.jacionline.org). After aspirin treatment, downstream markers of asthmatic airway inflammation were also unchanged compared with placebo, with no difference in either peripheral eosinophil count or serum eosinophilic cationic protein level (Table II). As such, localized airway-specific and systemic markers of disease activity remain unaffected, with neither an anti-inflammatory nor proinflammatory signal detected after the use of aspirin in asthmatic patients.

With regard to pulmonary function measurements, there was also no detectable effect after administration of asthma compared with placebo. All measurements of airway reactance, resistance, and compliance measured by using impulse oscillometry were unaffected by treatment, with no demonstrable improvement or decrease after aspirin administration compared with either placebo or baseline values. FEV₁, forced expiratory flow between 25% and 75% of exhalation, forced vital capacity, and peak expiratory flow variability were similarly unchanged (Table III and Fig 5). Compared with placebo, there was a marked reduction in serum thromboxane B2 levels after 3 weeks of aspirin administration (17.53-fold reduction; 95% CI, 5.46-56.49; $P < .001$). At baseline, before any randomized treatments, the geometric mean 15-epilipoxin A4 level was high at 4.88 ng/mL. However, there was no increase in 15-epilipoxin A4 levels with aspirin compared with placebo (0.99-fold change; 95% CI, 0.79-1.24; $P = 1$; Table II, Fig 6, and Figs E2 and E3 in the Online Repository at www.jacionline.org). Further *post hoc* analysis of the data failed to demonstrate any significant correlation between changes in lipoxin levels after aspirin treatment with either change in airway

TABLE I. Patient demographics

Demographics	Value
Age (y)	42.1 (3.5)
Male (female) sex	6 (9)
Dose of inhaled corticosteroid (μ g)*	400 (200-800)
FEV ₁ (% predicted)	87.5 (2.4)
FEF ₂₅₋₇₅ (% predicted)	62.8 (4.1)

FEF₂₅₋₇₅, Forced expiratory flow between 25% and 75% of exhalation.

Values are reported as means (SEMs) unless otherwise specified.

*Expressed as beclomethasone dipropionate equivalent and reported as median (interquartile range).

hyperresponsiveness (Spearman $r = 0.089$, $P = .752$) or exhaled nitric oxide levels ($r = 0.1750$, $P = .533$).

DISCUSSION

To our knowledge, this is the first prospective randomized trial designed specifically to investigate the effect of treatment with aspirin on airway inflammation in asthma. Previously published data have produced conflicting evidence as to the potential effects of aspirin in patients with asthma, although indirect evidence from epidemiologic and animal studies has suggested that this medication might be of therapeutic potential. The inflammatory process that characterizes asthma is strongly influenced by leukotriene-driven pathways. Lipoxins can act as endogenous antagonists of leukotrienes. Data from our preliminary study show that despite the theoretic potential of aspirin as an anti-inflammatory agent in patients with asthma, there were no demonstrable effects in asthmatic patients and no associated increase in lipoxin levels. The finding that low-dose aspirin does not worsen airway inflammation in asthmatic patients is perhaps not surprising, but this study is the first to provide objective evidence to support this clinical observation.

Study design

All prescribed treatment with anti-inflammatory medication was withdrawn, and then patients were preselected based on the presence of active documented airway inflammation (ie, significant hyperresponsiveness to histamine challenge) before randomization. In this way it was anticipated that any potential therapeutic signal from aspirin treatment would be maximized. Treatment effect was evaluated by using a variety of airway-specific (bronchial provocation and compartmental exhaled nitric oxide) and systemic (eosinophil count and eosinophilic cationic protein) surrogate inflammatory markers. In addition, measures of pulmonary dynamic mechanics (impulse oscillometry) and conventional spirometry were also studied. By comprehensively evaluating the inflammatory cascade, airway physiology, and geometry of asthmatic patients in whom all conventional treatment had been withdrawn, we are confident that if a therapeutic effect had been present, it would have been detected by using this study design. It is important to note that across all these measurements of disease activity, there was consistently no effect to suggest either improvement or deterioration with aspirin treatment. Because it was unclear what potential effects aspirin treatment would have and given previous safety concerns with regard to prescribing aspirin for asthmatic patients, the lack of any detectable worsening in airway inflammation is reassuring.

TABLE II. Effect of treatment on airway and peripheral markers of inflammation

	Shift from baseline with treatment					
	Placebo		Aspirin		Difference	
	Mean	95% CI (P value)	Mean	95% CI (P value)	Mean	95% CI (P value)
Exhaled nitric oxide (ppb)	1.02	0.48 to 2.15 (1)	1.07	0.51 to 2.27 (1)	0.95	0.45 to 2.00 (1)
Alveolar nitric oxide (ppb)	1.13	0.63 to 2.02 (1)	1.30	0.72 to 2.34 (1)	0.87	0.45 to 1.66 (1)
Bronchial flux (nL/s)	0.96	0.83 to 1.73 (.488)	1.21	0.84 to 1.75 (.374)	0.99	0.68 to 1.43 (1)
Histamine PC ₂₀ *	0.44	−0.11 to 0.99 (.22)	0.61	−0.06 to 1.17 (.06)	0.17	−0.38 to 0.73 (1)
Peripheral eosinophil count (cells/μL)†	53	−31 to 138 (.589)	−9	−94 to 75 (1)	63	−22 to 147 (.327)
Serum ECP (μg/L)	1.14	0.48 to 2.71 (.734)	1.04	0.44 to 2.47 (1)	1.10	0.46 to 2.63 (1)
Thromboxane-B ₂ (ng/mL)	1.38	0.43 to 4.44 (1)	0.08	0.02 to 0.25 (<.001)	17.53	5.46 to 56.49 (<.001)
15-Epilipoxin-A ₄ (ng/mL)	0.96	0.76 to 1.21 (1)	0.97	0.77 to 1.21 (1)	0.99	0.79 to 1.24 (1)

ECP, Eosinophilic cationic protein.

Data were skewed and required transformation before analysis, and as such, all differences are expressed as fold change except *histamine PC₂₀ (doubling-dilution shift) and †peripheral eosinophil count (absolute change in cell count). P values and 95% CIs are Bonferroni corrected to allow for multiple comparisons.

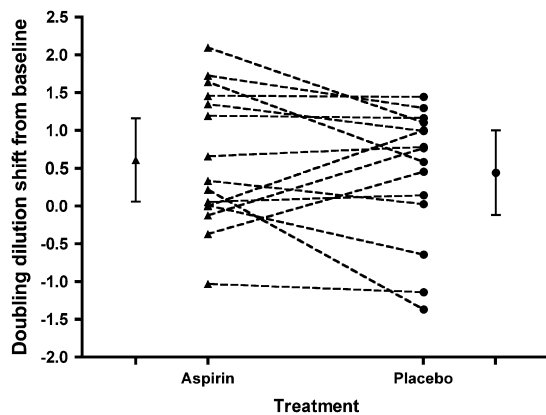


FIG 2. Effect of treatment with aspirin and placebo on histamine challenge. Doubling-dilution shift from baseline for individual patients and mean difference with 95% CI after treatment with aspirin and placebo are shown. There was no significant difference ($P = 1$) for aspirin compared with placebo.

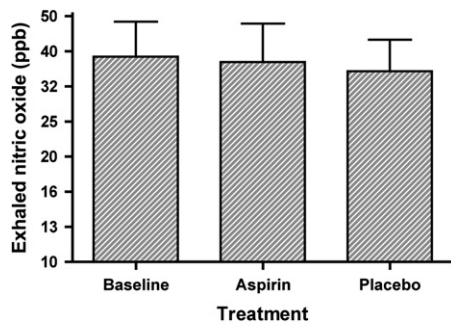


FIG 3. Effect of treatment with aspirin and placebo on tidal nitric oxide. Mean and SEM (error bars) exhaled nitric oxide levels at baseline and after treatment with aspirin and placebo on a log₁₀ scale are shown. There was no significant difference ($P = 1$) for aspirin compared with placebo.

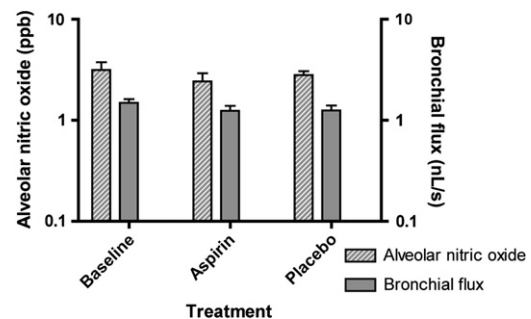


FIG 4. Effect of treatment with aspirin and placebo on alveolar nitric oxide and bronchial flux. Mean and SEM (error bars) alveolar nitric oxide (distal airway inflammation) and bronchial flux (proximal airway inflammation) at baseline and after treatment with aspirin and placebo on a log₁₀ scale are shown. There was no significant difference ($P = 1$) for aspirin compared with placebo.

on prostaglandin synthesis and lipoxin induction. In our asthmatic study population 75 mg of aspirin daily for 3 weeks led to unequivocal attenuation of thromboxane B₂ production, suggesting *in vivo* acetylation of COX did occur (ie, there was a demonstrable biologic treatment effect) but without any significant change in lipoxin synthesis. This is in contrast to previously reported data from healthy volunteers in which a similar dose of aspirin (81 mg/d) led to both a decrease in thromboxane B₂ levels and a commensurate increase in production of 15-epilipoxin A₄.⁴ The reason for the observed differing pharmacologic actions of aspirin in asthmatic patients and healthy volunteers is not clear. Vachier et al⁹ examined the relative concentrations of lipoxin in the induced sputum supernatant of asthmatic patients and found significant differences with healthy volunteers and also between asthmatic patients of differing disease severity. Patients with mild and severe asthma were shown to have the greatest concentration of lipoxin A₄ and 15-epilipoxin A₄, respectively, whereas all asthmatic patients had lipoxin levels that were greater than that of healthy volunteers. It is conceivable that lipoxin production is upregulated in asthmatic patients as an inbuilt biologic regulatory pathway in response to ongoing airway inflammation. Because such addition of aspirin to this model would have no demonstrable effect on already supranormal lipoxin levels, particularly in patients with mild disease in whom levels have been shown to

Pharmacologic actions of aspirin

Quantification of serum thromboxane B₂ and 15-epilipoxin A₄ levels was undertaken to directly evaluate the pharmacologic action of aspirin, with particular reference to any potential effects

TABLE III. Effect of treatment on spirometry and impulse oscillometry

Percent predicted	Shift from baseline with treatment				Difference	
	Placebo		Aspirin			
	Mean	95% CI (P value)	Mean	95% CI (P value)	Mean	95% CI (P value)
FEV ₁	−0.8	−8.9 to 10.5 (1)	1.3	−8.5 to 11.0 (.668)	0.5	−10.2 to 9.3 (1)
FEF ₂₅₋₇₅	2.4	−13.7 to 18.5 (.950)	3.4	−12.7 to 19.5 (.364)	−1.0	−17.7 to 15.1 (1)
FVC	−0.7	−8.4 to 7.0 (1)	−0.3	−8.0 to 7.4 (1)	−0.4	−8.1 to 7.3 (1)
PEF	3.9	−11.8 to 19.7 (1)	4.5	−11.2 to 20.2 (.681)	−0.5	−16.3 to 15.2 (1)
PEF variability*	−6.7	−12.7 to −0.6 (.048)	−6.9	−12.7 to −0.9 (.044)	0.2	−5.8 to 6.2 (1)
Total impedance	5.3	−41.0 to 51.6 (1)	5.8	−40.4 to 52.1 (1)	−0.6	−46.8 to 45.7 (1)
Total resistance	3.6	−36.4 to 43.6 (1)	5.7	−34.3 to 45.7 (1)	−2.1	−42.1 to 37.9 (1)
Central resistance	−5.3	−32.1 to 21.5 (1)	0.3	−26.5 to 27.2 (1)	−5.6	−32.4 to 21.2 (1)
Resonant frequency†	−0.6	−7.7 to 6.6 (.99)	0.9	−6.2 to 8.0 (1)	−1.5	−8.6 to 5.6 (.450)

FEF₂₅₋₇₅, Forced expiratory flow between 25% and 75% of exhalation; FVC, forced vital capacity; PEF, peak expiratory flow.

Values are expressed as percent predicted except *peak expiratory flow variability (total percentage) and †resonant frequency (Hz). P values and 95% CIs are Bonferroni corrected to allow for multiple comparisons.

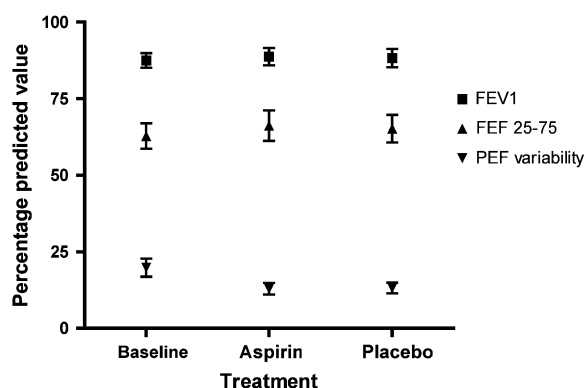


FIG 5. Effect of treatment with aspirin and placebo on spirometry and peak flow variability. Mean and SEM (error bars) FEV₁, forced expiratory flow between 25% and 75% (FEF 25-75) of exhalation (both percent predicted), and peak expiratory flow (PEF) variability (absolute percentage) at baseline and after treatment with aspirin and placebo are shown. There was no significant difference ($P = 1$) for aspirin compared with placebo.

be already maximal. This argument is supported by the observation that at baseline, before any treatment, the geometric mean 15-epilipoxin A4 value was 4.88 ng/mL (Fig 6), which is increased compared with the mean baseline level of 2.6 ng/mL previously reported in healthy volunteers by using an identical ELISA assay from the same manufacturer.⁴ There is a relative paucity of historical published data relating to serum lipoxin levels in patients with mild-to-moderate asthma. As such, it would have been desirable to include a healthy volunteer control group for this measurement to eliminate interlaboratory variation, but this issue was not anticipated at the trial conception.

Study limitations

It is possible that 3 weeks of treatment would not be sufficient time for changes in airway inflammation or pulmonary dynamics to become detectable and that a longer therapeutic trial would have been desirable. Biologic effects of aspirin treatment (attenuation of thromboxane B2) were detected after randomized treatment, and as such, we believe that if other therapeutic changes were to have occurred, they would have similarly done so in the same time period. By using a variety of techniques to assess

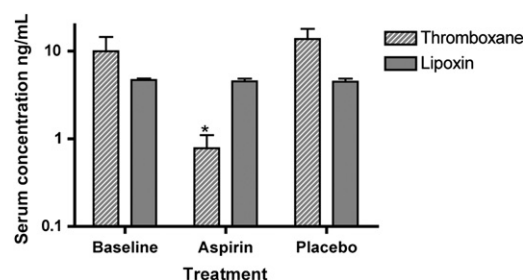


FIG 6. Effect of treatment with aspirin and placebo on thromboxane B2 and 15-epilipoxin A4. Mean and SEM (error bars) thromboxane B2 and 15-epilipoxin A4 levels at baseline and after treatment with aspirin and placebo on a log₁₀ scale are shown. Significant difference from both baseline and placebo: $P < .001$.

for treatment response and by ensuring that the inflammatory signal was maximal at the outset, we also are confident that any effect would have been detected. Although airway-specific inflammatory indices were included, it might have been desirable to measure airway rather than blood lipoxin and thromboxane levels. It is conceivable that the airway and blood compartments are biologically distinct, and as such, activity in the periphery might not accurately reflect the local inflammatory process within the bronchial mucosa. It could also be argued that a higher dose of aspirin should have been used; however, previous data have suggested that the dose-response relationship in relation to lipoxin synthesis and inhibition of prostaglandin production is precipitously steep, and as such, we believed low-dose treatment to be appropriate. Because low-dose aspirin was used in this study, it is not possible to extrapolate these results to higher doses or to the use of other NSAIDs in asthmatic patients: this is important because these medications are frequently used at higher doses in clinical practice. It should be recognized that low-dose aspirin has a biologic effect on platelet-derived lipoxin and thromboxane synthesis, but higher doses of aspirin and other NSAIDs are required to influence the production of other COX-derived mediators. It is unknown whether higher doses of aspirin can lead to clinically relevant effects through modulation of prostaglandin production or preferential leukotriene synthesis from arachidonate, which might lead to either bronchoconstriction or bronchoprotection. This study did not have a dose-ranging component and as such cannot answer this potentially important

question. Finally, asthmatic patients with more severe disease could have been included in the study, although in these patients withdrawal of conventional treatment would have been unethical and therefore made the data more difficult to interpret.

Conclusion

Despite theoretic potential, treatment with low-dose aspirin does not lead to increased 15-epilipoxin A4 synthesis and has no beneficial effects on airway inflammation or pulmonary function in patients with mild-to-moderate persistent asthma. In asthmatic patients who are not aspirin sensitive, there are also no deleterious effects on airway inflammation with aspirin treatment, and as such, clinicians can be reassured that this is safe to prescribe.

Clinical implications: Aspirin is safe to prescribe in patients who are not aspirin-sensitive.

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METHODS

Stepdown and medication withdrawal

At the beginning of the stepdown period, all participants were issued with a peak flow diary and a peak flowmeter and instructed on their use. They were requested to make daily measurements and record them in the diary. The value that corresponded to a peak expiratory flow of 60% of predicted value was determined, and participants were instructed to contact one of the investigators if their peak expiratory flow decreased to less than this point on 2 consecutive days. Regular salmeterol was begun (50 µg administered twice daily through a metered-dose inhaler), and then medication was withdrawn in a stepwise fashion, with weekly visits to the research department. Initially, all second-line asthma medication was discontinued, including antihistamines, leukotriene receptor antagonists, theophylline, and nasal steroids. After a week without these treatments, the dose of inhaled corticosteroid was sequentially halved at weekly intervals until a dose of 400 µg of beclomethasone dipropionate or equivalent was reached. At this point, inhaled steroids were discontinued completely, and the patient entered the run-in period of the trial.

Randomization and blinding

A clinical trials pharmacist independent of the study blinded medication off site (Western Infirmary, Glasgow, United Kingdom). Tablets were encapsulated in gelatin and randomized by using a computer-generated number table. Unblinding took place after the final participant had completed per protocol.

Oral aspirin challenge

An oral aspirin challenge was performed, as described by Cormican et al.^{E1} The purpose of the test was to firmly establish that none of the participants had aspirin hypersensitivity. A history of prior aspirin exposure was obtained, and only participants who had previously tolerated this medication without complication were allowed to proceed. Written consent was obtained, and baseline spirometry was then performed in accordance with joint American Thoracic Society and European Respiratory Society guidelines.^{E2} Thereafter, 30 mg of aspirin was administered and dissolved in 100 mL of water. Measurements of FEV₁ were then made every 15 minutes for the next 1½ hours, and at the same intervals, symptoms of possible hypersensitivity were documented, and external signs were assessed (rhinorrhea, urticaria, and angioedema). If there were no changes, then this procedure was repeated with 60, 100, and 300 mg of aspirin. If symptoms developed or the FEV₁ decreased by more than 15%, the test result was deemed positive. A doctor supervised the test and was on hand to administer treatment, if required.

Exhaled fractional and alveolar nitric oxide

An online chemiluminescence analyzer (Niox, Aerocrine AB, Sweden) was used to quantify exhaled nitric oxide. This was done at mouth flow rates of 50, 100, and 200 mL/s with a technically acceptable maneuver defined as one in which a sustained plateau of at least 8 seconds was observed.^{E3} The

arithmetic mean of 3 maneuvers at each flow rate was reported. Alveolar nitric oxide and bronchial flux was derived by using the 2-compartment model described by Tsoukias and George,^{E4} with the assumption that at an exhalation rate of greater than 50 mL/s, nitric oxide output becomes a linear function of flow rate.

Impulse oscillometry

Impulse oscillometry was performed by using a MasterScreen Impulse Oscillometry System (Viasys Healthcare, Leibnizstr, Germany), which is in line with current recommendations from the European Respiratory Society. Briefly, participants were asked to sit upright with their mouth on the apparatus mouthpiece. A nose clip was applied, and patients were asked to support their cheeks with the palms of their hands to avoid upper airway artifact. Once a regular tidal quiet pattern had been established, three 30-second measurements were made, with a 1-minute interval between each. Measurements were deemed acceptable if they were free from noise caused by cough, glottic closure, or leak anywhere in the system and if coherence of the signal at 20 Hz was greater than 0.9.

Eosinophilic cationic protein

Serum samples were stored at −20°C and analyzed as a single batch by using a radioimmunoassay technique to quantify eosinophilic cationic protein levels (UniCAP; Sweden Diagnostics, Milton Keynes, United Kingdom).

Thromboxane B2 and 15-epilipoxin A4

Serum samples were stored at −70°C and analyzed as a single batch by using ELISA (Neogen Europe Ltd, Ayr, United Kingdom).

RESULTS

Individual scatter plots depicting change from placebo baseline after treatment with aspirin and placebo for exhaled nitric oxide, thromboxane, and lipoxin are shown (Figs E1-E3).

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- E1. Cormican LJ, Farooque S, Altmann DR, Lee TH. Improvements in an oral aspirin challenge protocol for the diagnosis of aspirin hypersensitivity. *Clin Exp Allergy* 2005;35:717-22.
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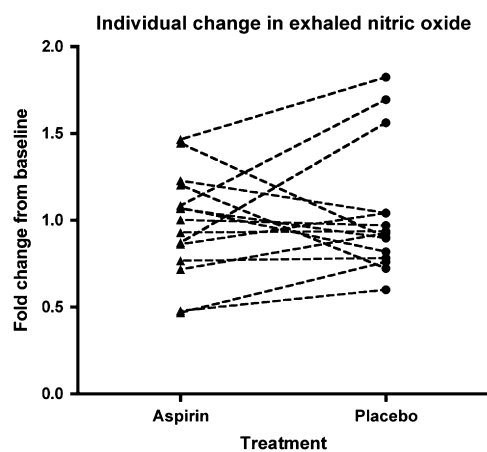


FIG E1. Individual change in exhaled nitric oxide levels.

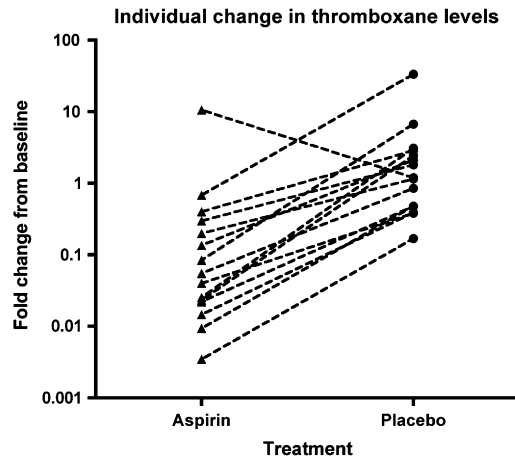


FIG E2. Individual change in thromboxane levels.

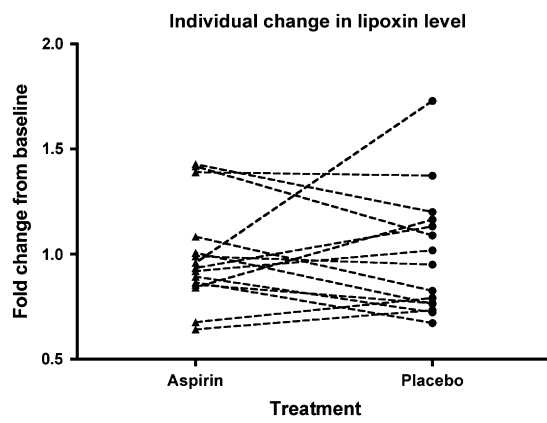


FIG E3. Individual change in lipoxin levels.